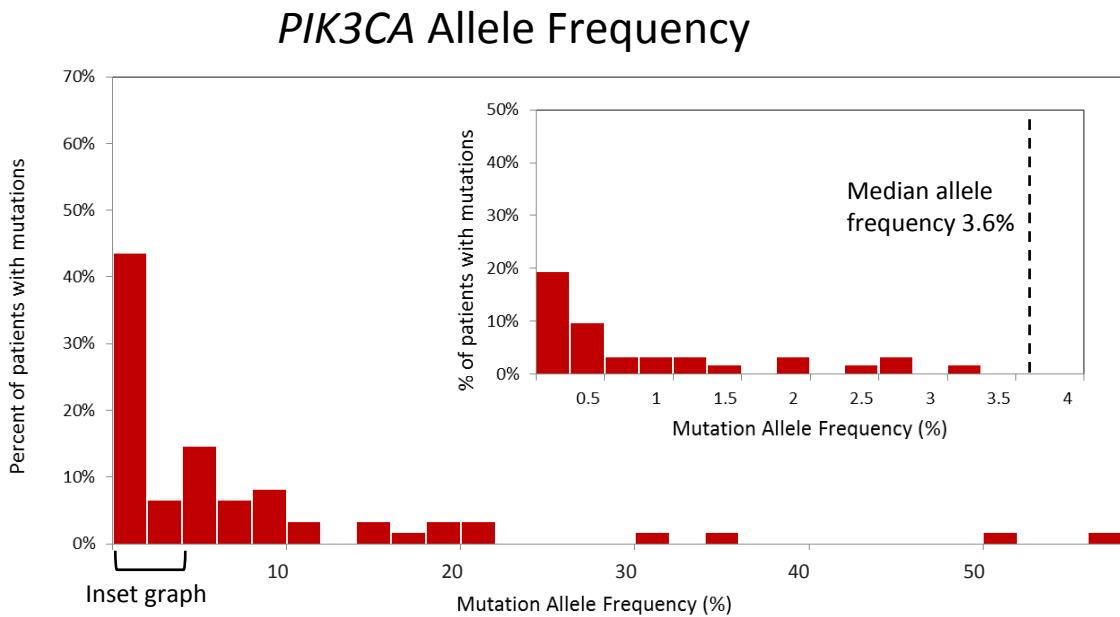
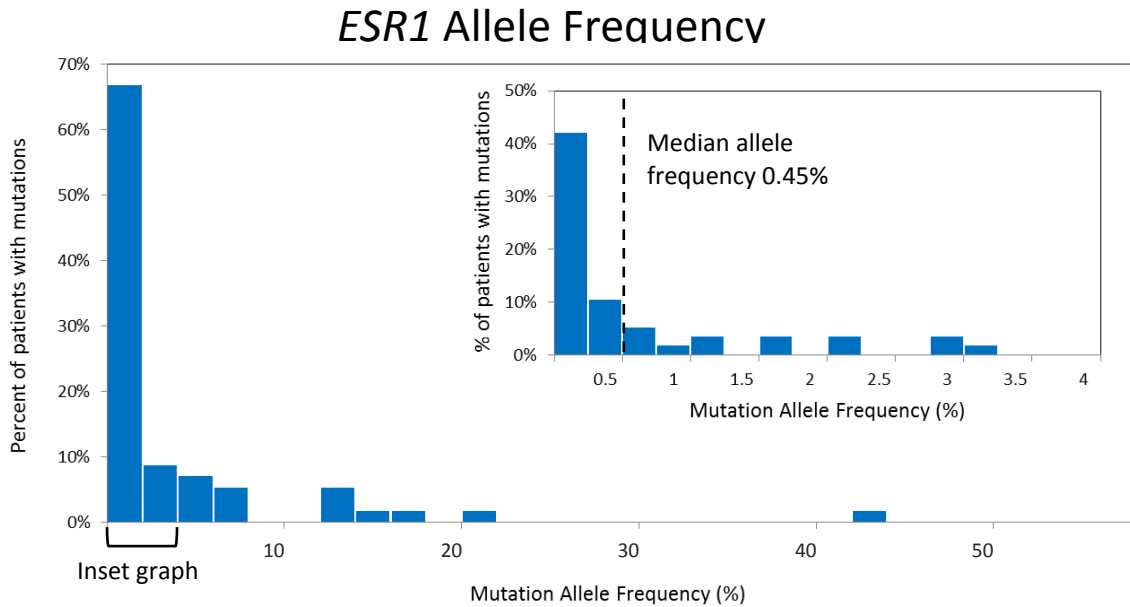


Supplementary Figure 1

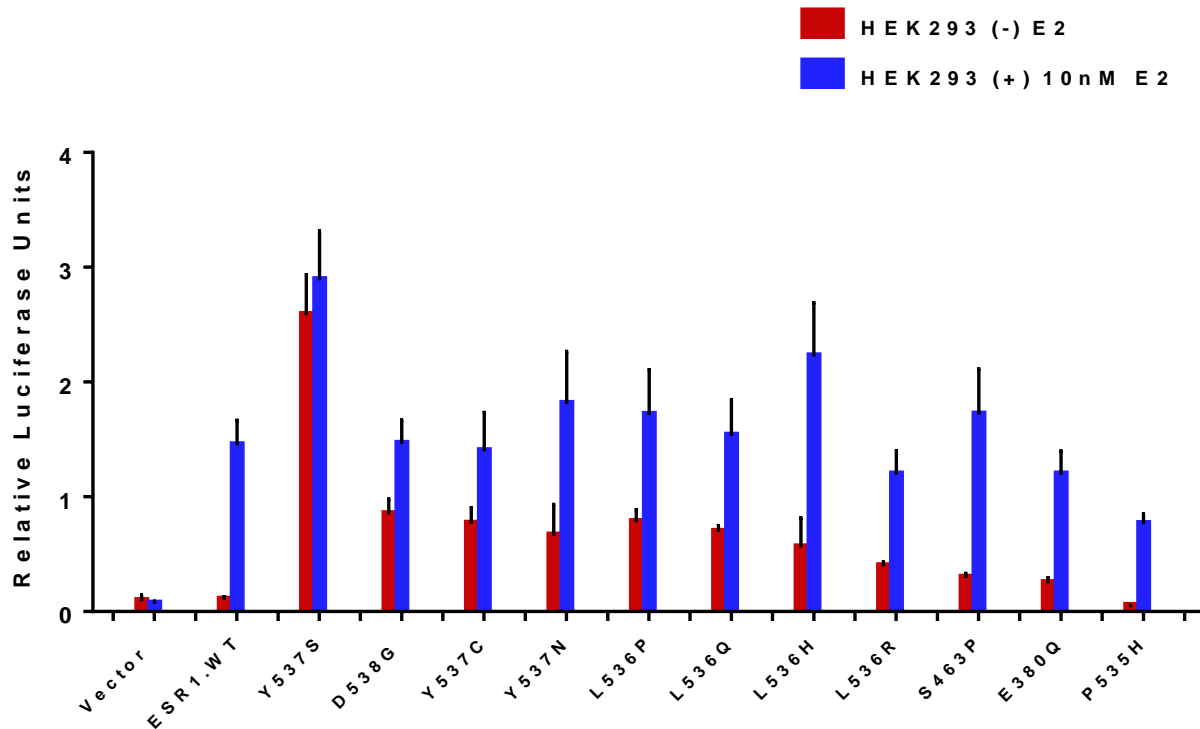
Distribution of mutant allele frequencies



Supplementary Figure 1 Histogram of *ESR1* (blue) and *PIK3CA* (red) ctDNA mutant allele frequencies for part 1 patients (*ESR1* n=57, *PK3CA* n=62). Each bar represents the percent of patients with a mutation allele frequency within the specified bin, for example 0.02% to 2% for the first bin, >2% to 4% for the second bin, etc. The inset graphs are histograms for patients with mutant allele frequencies less than 4%, which represents half or more of the patients for both *ESR1* and *PIK3CA*.

Supplementary Figure 2

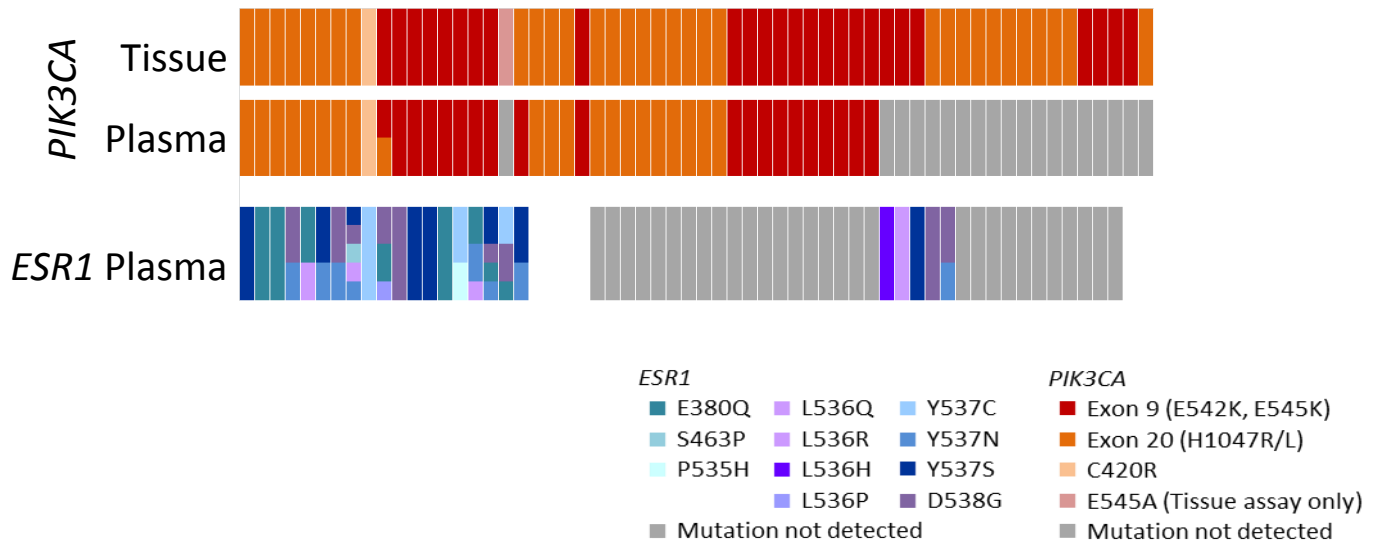
Estrogen Response Element (ERE) reporter assay indicates relative levels of ligand-independent transcriptional activity of ER α mutants



Supplementary Figure 2

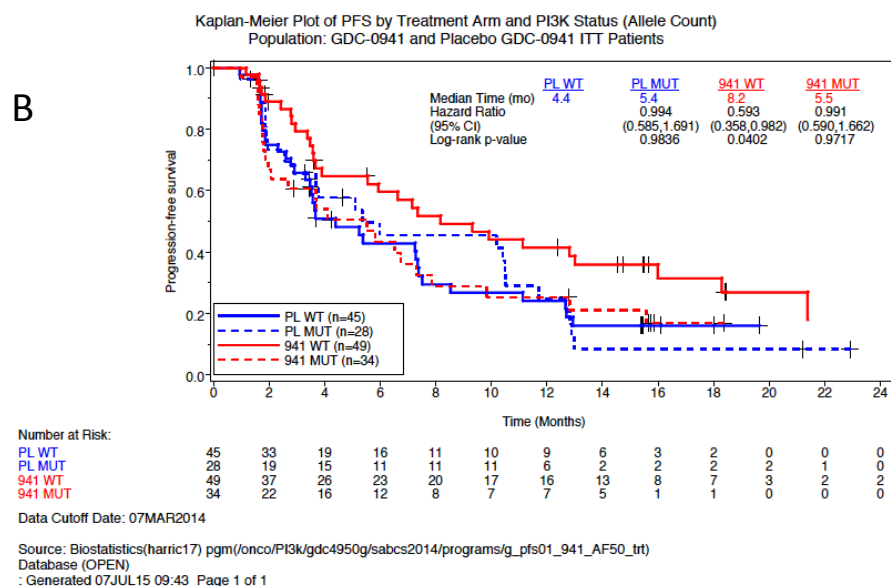
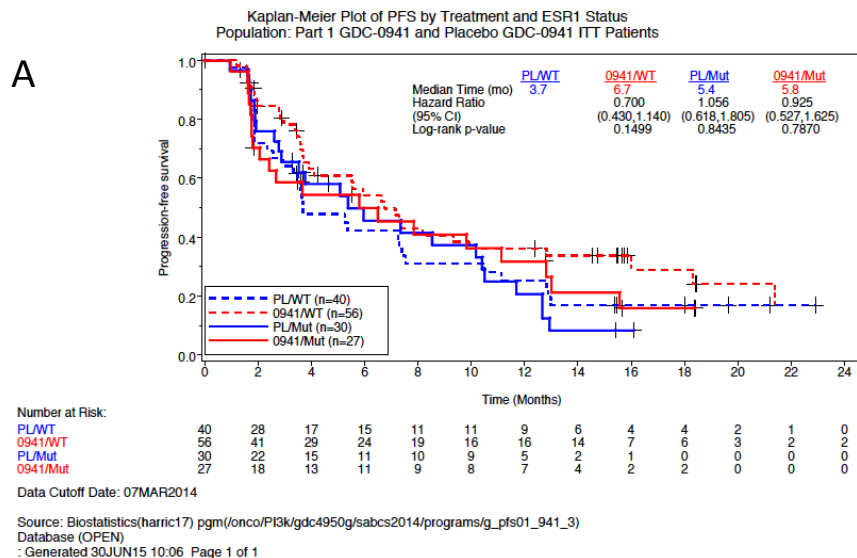
Relative ERE transcriptional activity levels of ER α proteins. HEK293 cells obtained from ATCC were cotransfected with an ERE–firefly luciferase reporter plasmid, a plasmid constitutively expressing Renilla luciferase as an internal control, and pcDNA3.1+ constructs containing WT ESR1 cDNA sequence or various *ESR1* mutations. Cells were grown in steroid hormone-deprived conditions and treated with DMSO or 10 nM β -estradiol (E2) for 24 h. Relative luciferase units were measured using a Dual-Glo[®] Luciferase Assay (Promega) and Firefly luciferase activity was normalized to Renilla luciferase activity. Data are representative of three independent experiments; error bars indicate the standard deviation of three replicates.

Supplementary Figure 3 – Part 2 *PIK3CA* tissue vs plasma status;
ESR1 plasma status



Supplementary Figure 3 *PIK3CA* and *ESR1* mutation analysis of matched tumor tissue and plasma samples from n=60 patients from part 2 of the clinical study. Mutations are colored according to exon (*PIK3CA*) or amino acid (*ESR1*). Grey indicates mutations assessed were negative, and blank indicates data not available.

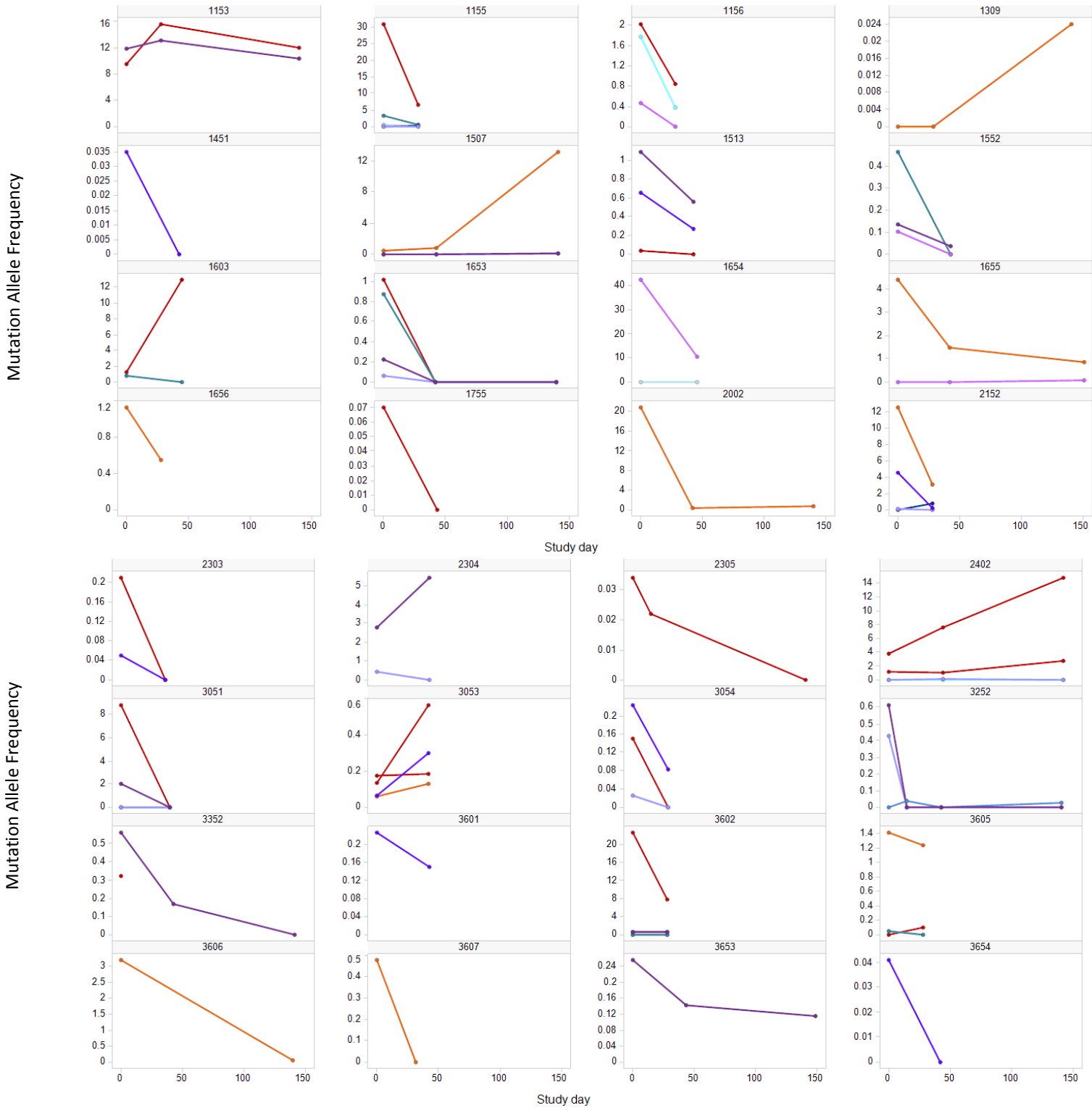
Supplementary Figure 4 Kaplan-Meier plots of progression free survival by treatment



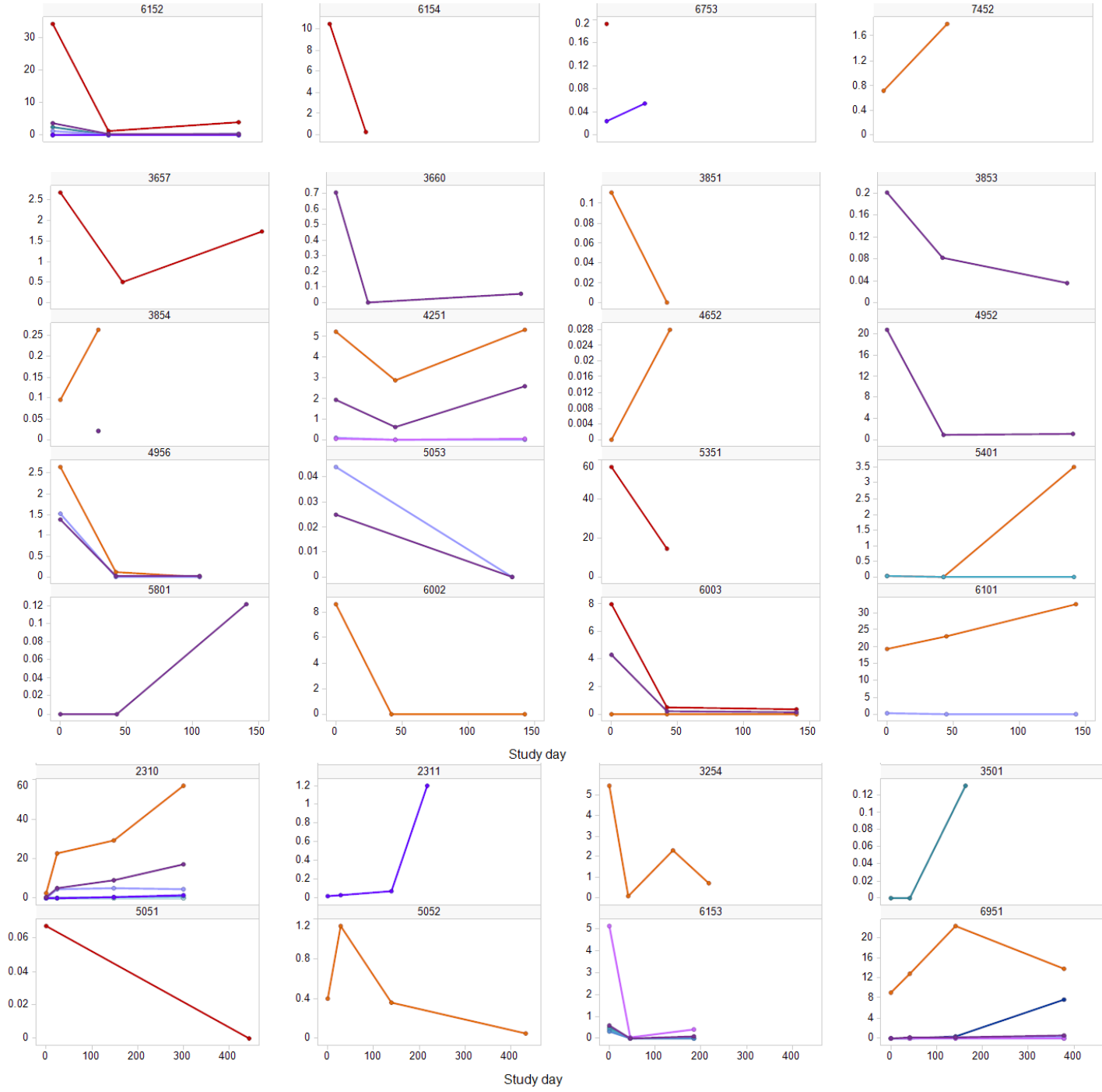
Supplementary Figure 4 Kaplan-Meier plots of progression-free survival in part 1 patients. PFS of the fulvestrant control arm (PL, blue) vs fulvestrant + pictilicib (0941, red) for patients with wildtype (WT, dashed line) or mutant (solid line) *ESR1* (A) and *PIK3CA* (B). Hazard ratios are relative to PL/WT for both plots.

Supplementary Figure 5

Inpatient patterns of mutant allele frequency changes over time



Mutation Allele Frequency



Supplemental Figure 5 Absolute *ESR1* and *PIK3CA* mutant allele frequency changes in 60 patients with at least one mutation detected in at least one time point.

ESR1

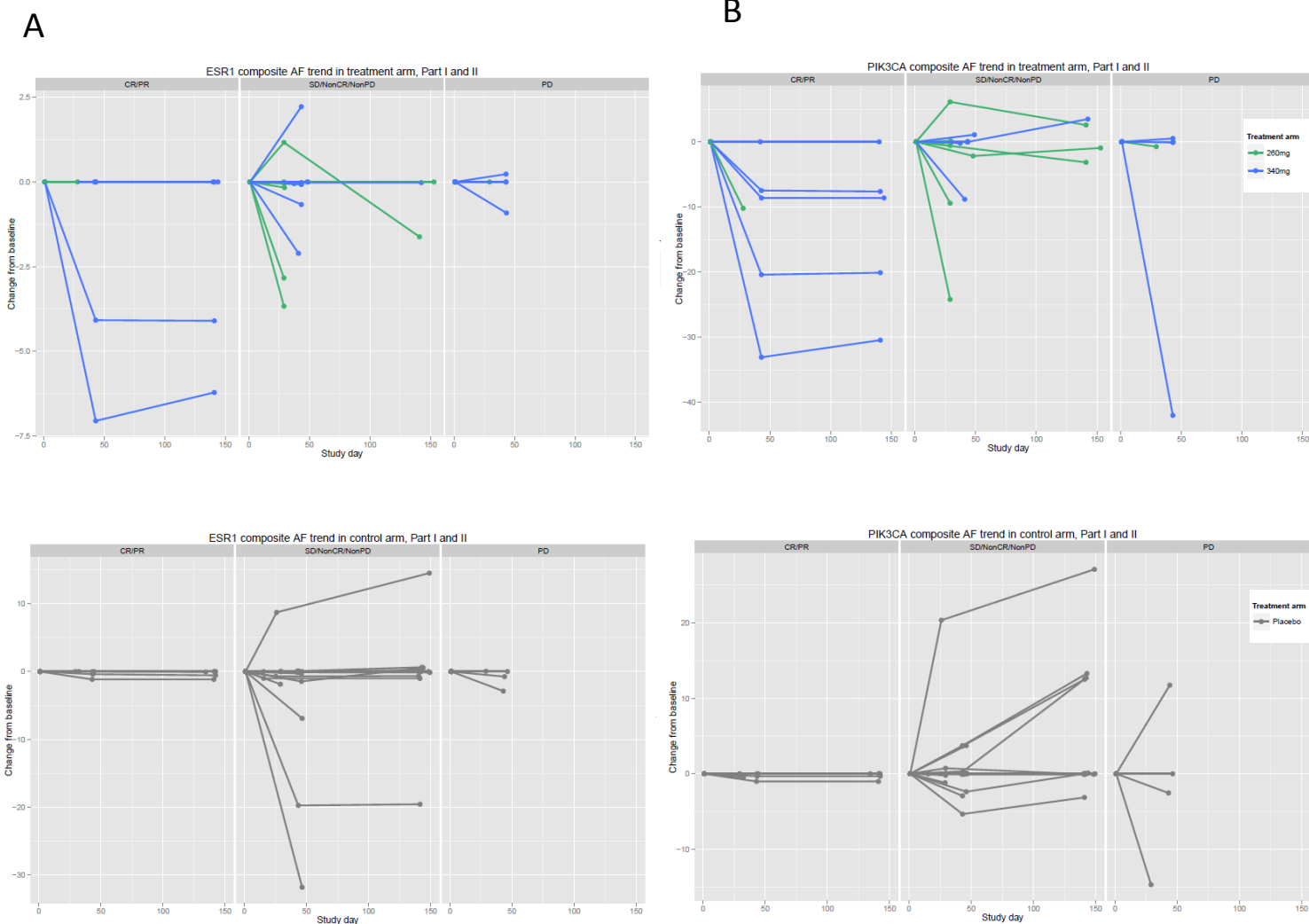
- E380Q
- S463P
- P535H
- L536Q
- L536R
- L536H
- L536P
- Y537N
- Y537S
- Y537C
- D538G

PIK3CA

- E542K
- E545K
- Q546K
- H1047R
- H1047L

Supplementary Figure 6

Mutant allele frequency changes and tumor response



Supplementary Figure 6 (A) ESR1 and (B) PIK3CA ctDNA mutation AF changes while on part 1 (green) or part 2 (blue) pictilisib + fulvestrant or placebo + fulvestrant (grey) treatment arms. Patients are binned according to their best clinical response. Each line represents one patient with the absolute change of allele frequency relative to baseline (study day 0) plotted for 2 or 3 time-points. The mutation allele frequency sum is plotted for patients with multiple ESR1 or PIK3CA mutations.

Supplementary Table 1: Baseline ctDNA mutation prevalence in molecular subsets

Part 1 Randomized GDC-0941 and Placebo GDC-0941 Patients	
	<i>ESR1</i> Mutation Prevalence n / total (%)
<i>PIK3CA</i> Status, ctDNA	
Wild-type	23 / 91 (25.3%)
Mutation	34 / 62 (54.8%)
<i>PIK3CA</i> Status, tissue	
Wild-type	23 / 76 (30.3%)
Mutation	28 / 63 (44.4%)
Luminal Status	
Luminal A	41 / 99 (41.4%)
Luminal B	14 / 44 (31.8%)
Other	2 / 10 (20%)
Progesterone Status	
Positive	42 / 109 (38.5%)
Negative	10 / 30 (33.3%)
Part 2 Randomized GDC-0941 and Placebo GDC-0941 Patients	
	<i>ESR1</i> Mutation Prevalence n / total (%)
<i>PIK3CA</i> Status, tissue	
Wild-type	0 / 0
Mutation	24 / 54 (44.4%)

Supplementary Table 2: BEAMing assay performance for *PIK3CA* from Part I samples

<i>PIK3CA</i> Plasma				
		Mutant	Wild-type	
<i>PIK3CA</i> Tissue	Mutant	48	14	Sensitivity 77%
	Wild-type	5	71	Specificity 93%
Concordance 86%				

N=142 matched tissue and plasma

4 pairs excluded due to different *PIK3CA* mutations detected in plasma vs tissue (2) or differences in assay coverage (2)

Supplementary Table 3: *ESR1* mutations detected in tissue

<i>ESR1</i> Tissue status	<i>ESR1</i> ctDNA status
Y537N (5.3%) E380Q (0.029%) L536Q (23.7%)	E380Q mutant (3.278%) Y537N mutant (0.088%) L536Q mutant (0.443%)
Y537C (1.9%) E380Q (6.7%)	Y537C mutant (5.129%) D538G mutant (0.576%) E380Q mutant (0.489%) S463P mutant (0.431%) L536H mutant (0.314%)
D538G (20.1%)	D538G mutant (1.678%) Y537C mutant (1.336%) E380Q mutant (0.072%)
D538G (26.7%)	D538G mutant (1.678%) Y537N mutant (0.19%)
D538G (0.49%)	D538G mutant (5.225%) E380Q mutant (0.022%)
P535H (5.7%)	P535H mutant (1.781%) Y537C mutant (0.465%)
D538G (45%)	D538G mutant (0.794%)
E380Q (0.025%)	E380Q mutant (0.024%)
E380Q (0.59%)	E380Q mutant (0.032%)
E380Q (18.1%)	E380Q mutant (0.064%)
Y537S (17.5%)	Y537S mutant (2.812%)
E380Q (0.1%)	Wild-type
Y537C (0.15%)	Wild-type
Y537N (20.1%)	Wild-type
Y537C (0.17%)	Wild-type

Supplementary Table 4: *ESR1* mutation status concordance between tissue and plasma

		<i>ESR1</i> Plasma			Concordance
		Mutant	Wild-type		
<i>ESR1</i> Tissue	Overall	Mutant	11	4	31%
	N=98	Wild-type	64	19	
	Primary	Mutant	0	0	5%
	N=42	Wild-type	40	2	
	Metastatic	Mutant	11	4	47%
	N=47	Wild-type	21	11	
	Tissue collected post AI	Mutant	9	3	57%
	N=21	Wild-type	6	3	
	Tissue collected pre AI	Mutant	2	1	23%
	N=77	Wild-type	58	16	

Supplementary Table 5: Demographic and Baseline Characteristics by *ESR1* Status; Part 1 Randomized GDC-0941 and Placebo GDC-0941 Patients

	ESR1 Wild Type N=96	ESR1 Mutant N=57	All Patients N=153
ESR1 Mutations			
n	96	57	153
0	96 (100.0%)	0	96 (62.7%)
1	0	34 (59.6%)	34 (22.2%)
2	0	13 (22.8%)	13 (8.5%)
3	0	7 (12.3%)	7 (4.6%)
4	0	1 (1.8%)	1 (0.7%)
5	0	2 (3.5%)	2 (1.3%)
Age			
n	96	57	153
Mean (SD)	62.0 (10.3)	59.7 (10.3)	61.1 (10.4)
Median	62.5	60.0	62.0
Min - Max	40 - 90	36 - 82	36 - 90
Age group (yr)			
n	96	57	153
< 65	58 (60.4%)	40 (70.2%)	98 (64.1%)
>= 65	38 (39.6%)	17 (29.8%)	55 (35.9%)
Baseline Weight (kg)			
n	95	56	151
Mean (SD)	69.5 (14.9)	71.8 (16.2)	70.3 (15.4)
Median	68.0	68.1	68.0
Min - Max	38 - 119	39 - 115	38 - 119
Race			
n	96	57	153
Asian	7 (7.3%)	5 (8.8%)	12 (7.8%)
Black or African American	1 (1.0%)	1 (1.8%)	2 (1.3%)
White	85 (88.5%)	49 (86.0%)	134 (87.6%)
Other	3 (3.1%)	1 (1.8%)	4 (2.6%)
Multiple	0	1 (1.8%)	1 (0.7%)
Region			
n	96	57	153
Asia	17 (17.7%)	14 (24.6%)	31 (20.3%)
Europe	47 (49.0%)	22 (38.6%)	69 (45.1%)
North America	27 (28.1%)	19 (33.3%)	46 (30.1%)
Other	5 (5.2%)	2 (3.5%)	7 (4.6%)

US				
	n	96	57	153
	US	27 (28.1%)	19 (33.3%)	46 (30.1%)
	ex	69 (71.9%)	38 (66.7%)	107 (69.9%)
Baseline ECOG Score				
	n	95	57	152
	0	66 (69.5%)	33 (57.9%)	99 (65.1%)
	1	29 (30.5%)	24 (42.1%)	53 (34.9%)
Primary/Secondary Resistance (IVRS)				
	n	96	57	153
	Primary Resistance	46 (47.9%)	30 (52.6%)	76 (49.7%)
	Secondary Resistance	50 (52.1%)	27 (47.4%)	77 (50.3%)
Resistance to Prior AI (BOLERO2)				
	n	96	57	153
	Yes	22 (22.9%)	9 (15.8%)	31 (20.3%)
	No	74 (77.1%)	48 (84.2%)	122 (79.7%)
Measurable Disease (IVRS)				
	n	96	57	153
	Measurable	58 (60.4%)	28 (49.1%)	86 (56.2%)
	Non-Measurable	38 (39.6%)	29 (50.9%)	67 (43.8%)
Measurable Disease (Derived)				
	n	96	57	153
	Measurable	53 (55.2%)	33 (57.9%)	86 (56.2%)
	Non-Measurable	43 (44.8%)	24 (42.1%)	67 (43.8%)
ER Status				
	n	96	57	153
	Positive	93 (96.9%)	55 (96.5%)	148 (96.7%)
	Negative	1 (1.0%)	0	1 (0.7%)
	Status Unknown	2 (2.1%)	2 (3.5%)	4 (2.6%)
Her2				
	n	96	57	153
	Equivocal	0	1 (1.8%)	1 (0.7%)
	Negative	92 (95.8%)	52 (91.2%)	144 (94.1%)
	Positive	2 (2.1%)	1 (1.8%)	3 (2.0%)
	Unknown	2 (2.1%)	3 (5.3%)	5 (3.3%)
PR Status				
	n	96	57	153
	Positive	67 (69.8%)	42 (73.7%)	109 (71.2%)
	Negative	20 (20.8%)	10 (17.5%)	30 (19.6%)
	Status Unknown	9 (9.4%)	5 (8.8%)	14 (9.2%)
PIK3CA Mutation Status				
	n	96	57	153

Positive	35 (36.5%)	28 (49.1%)	63 (41.2%)
Wild Type	53 (55.2%)	23 (40.4%)	76 (49.7%)
Unknown	8 (8.3%)	6 (10.5%)	14 (9.2%)
Number of Previous Therapies in Metastatic Setting			
n	96	57	153
0	27 (28.1%)	13 (22.8%)	40 (26.1%)
1	38 (39.6%)	28 (49.1%)	66 (43.1%)
2	23 (24.0%)	12 (21.1%)	35 (22.9%)
3+	8 (8.3%)	4 (7.0%)	12 (7.8%)
Bone-only Disease			
n	96	57	153
Yes	24 (25.0%)	11 (19.3%)	35 (22.9%)
No	72 (75.0%)	46 (80.7%)	118 (77.1%)
Baseline Tumor Burden (Continuous)			
n	96	57	153
Mean (SD)	23.66 (26.07)	27.34 (33.73)	25.03 (29.11)
Median	16.30	20.00	17.00
Min – Max	0.0 – 87.0	0.0 – 184.1	0.0 – 184.1
Baseline Tumor Burden (Categorical)			
n	96	57	153
>= Median	48 (50.0%)	30 (52.6%)	78 (51.0%)
< Median	48 (50.0%)	27 (47.4%)	75 (49.0%)
Number of Metastatic Sites			
n	96	57	153
0	5 (5.2%)	1 (1.8%)	6 (3.9%)
1	37 (38.5%)	13 (22.8%)	50 (32.7%)
2	30 (31.3%)	24 (42.1%)	54 (35.3%)
3+	24 (25.0%)	19 (33.3%)	43 (28.1%)
Setting of Last CA Therapy			
n	96	57	153
Adjuvant	22 (22.9%)	12 (21.1%)	34 (22.2%)
Metastatic	72 (75.0%)	45 (78.9%)	117 (76.5%)
Other	2 (2.1%)	0	2 (1.3%)
Previous Chemotherapy			
n	96	57	153
No	33 (34.4%)	18 (31.6%)	51 (33.3%)
Yes – Adjuvant/Neo-Adjuvant	43 (44.8%)	28 (49.1%)	71 (46.4%)
Yes – Metastatic	20 (20.8%)	11 (19.3%)	31 (20.3%)
Number of Previous Therapies			
n	96	57	153
1	4 (4.2%)	1 (1.8%)	5 (3.3%)
2	13 (13.5%)	8 (14.0%)	21 (13.7%)

3+	79 (82.3%)	48 (84.2%)	127 (83.0%)
Prior Tamoxifen Use			
n	96	57	153
Yes	52 (54.2%)	26 (45.6%)	78 (51.0%)
No	44 (45.8%)	31 (54.4%)	75 (49.0%)
Number of Prior Tamoxifen Regimens			
n	96	57	153
0	44 (45.8%)	31 (54.4%)	75 (49.0%)
1	51 (53.1%)	26 (45.6%)	77 (50.3%)
2	1 (1.0%)	0	1 (0.7%)
Prior AI Use			
n	96	57	153
Yes	96 (100.0%)	57 (100.0%)	153 (100.0%)
Number of Prior AI Regimens			
n	96	57	153
1	71 (74.0%)	47 (82.5%)	118 (77.1%)
2	24 (25.0%)	10 (17.5%)	34 (22.2%)
3	1 (1.0%)	0	1 (0.7%)
Prior Endocrine Therapy Use			
n	96	57	153
Yes	96 (100.0%)	57 (100.0%)	153 (100.0%)
Number of Prior Endocrine Therapy Regimens			
n	96	57	153
1	35 (36.5%)	27 (47.4%)	62 (40.5%)
2	45 (46.9%)	24 (42.1%)	69 (45.1%)
3	14 (14.6%)	6 (10.5%)	20 (13.1%)
4	2 (2.1%)	0	2 (1.3%)

Data Cutoff Date: 07MAR2014

Data Extract Date: 15SEP2014

Supplementary Table 6: Demographic and Baseline Characteristics
by *PIK3CA* Status; Part 1 Randomized GDC-0941 and Placebo
GDC-0941 Patients

	PIK3CA Wild Type N=94	PIK3CA Mutant N=62	All Patients N=156
ESR1 Mutations			
n	91	62	153
0	68 (74.7%)	28 (45.2%)	96 (62.7%)
1	13 (14.3%)	21 (33.9%)	34 (22.2%)
2	7 (7.7%)	6 (9.7%)	13 (8.5%)
3	1 (1.1%)	6 (9.7%)	7 (4.6%)
4	1 (1.1%)	0	1 (0.7%)
5	1 (1.1%)	1 (1.6%)	2 (1.3%)
Age			
n	94	62	156
Mean (SD)	62.0 (10.8)	59.4 (9.4)	61.0 (10.3)
Median	63.0	60.5	62.0
Min – Max	40 – 90	36 – 80	36 – 90
Age group (yr)			
n	94	62	156
< 65	57 (60.6%)	44 (71.0%)	101 (64.7%)
>= 65	37 (39.4%)	18 (29.0%)	55 (35.3%)
Baseline Weight (kg)			
n	92	62	154
Mean (SD)	71.6 (15.3)	68.1 (15.1)	70.2 (15.3)
Median	68.8	64.8	67.8
Min – Max	47 – 119	38 – 106	38 – 119
Race			
n	94	62	156
Asian	6 (6.4%)	7 (11.3%)	13 (8.3%)
Black or African American	0	2 (3.2%)	2 (1.3%)
White	85 (90.4%)	51 (82.3%)	136 (87.2%)
Other	2 (2.1%)	2 (3.2%)	4 (2.6%)
Multiple	1 (1.1%)	0	1 (0.6%)
Region			
n	94	62	156
Asia	16 (17.0%)	18 (29.0%)	34 (21.8%)
Europe	42 (44.7%)	27 (43.5%)	69 (44.2%)
North America	30 (31.9%)	16 (25.8%)	46 (29.5%)
Other	6 (6.4%)	1 (1.6%)	7 (4.5%)
US			
n	94	62	156
US	30 (31.9%)	16 (25.8%)	46 (29.5%)

ex	64 (68.1%)	46 (74.2%)	110 (70.5%)
Baseline ECOG Score			
n	93	62	155
0	61 (65.6%)	38 (61.3%)	99 (63.9%)
1	32 (34.4%)	24 (38.7%)	56 (36.1%)
Primary/Secondary Resistance (IVRS)			
n	94	62	156
Primary Resistance	43 (45.7%)	34 (54.8%)	77 (49.4%)
Secondary Resistance	51 (54.3%)	28 (45.2%)	79 (50.6%)
Resistance to Prior AI (BOLERO2)			
n	94	62	156
Yes	21 (22.3%)	11 (17.7%)	32 (20.5%)
No	73 (77.7%)	51 (82.3%)	124 (79.5%)
Measurable Disease (IVRS)			
n	94	62	156
Measurable	55 (58.5%)	32 (51.6%)	87 (55.8%)
Non-Measurable	39 (41.5%)	30 (48.4%)	69 (44.2%)
Measurable Disease (Derived)			
n	94	62	156
Measurable	54 (57.4%)	33 (53.2%)	87 (55.8%)
Non-Measurable	40 (42.6%)	29 (46.8%)	69 (44.2%)
ER Status			
n	94	62	156
Positive	93 (98.9%)	58 (93.5%)	151 (96.8%)
Negative	0	1 (1.6%)	1 (0.6%)
Status Unknown	1 (1.1%)	3 (4.8%)	4 (2.6%)
Her2			
n	94	62	156
Equivocal	1 (1.1%)	0	1 (0.6%)
Negative	88 (93.6%)	59 (95.2%)	147 (94.2%)
Positive	2 (2.1%)	1 (1.6%)	3 (1.9%)
Unknown	3 (3.2%)	2 (3.2%)	5 (3.2%)
PR Status			
n	94	62	156
Positive	64 (68.1%)	46 (74.2%)	110 (70.5%)
Negative	21 (22.3%)	10 (16.1%)	31 (19.9%)
Status Unknown	9 (9.6%)	6 (9.7%)	15 (9.6%)
Number of Previous Therapies in Metastatic Setting			
n	94	62	156
0	23 (24.5%)	18 (29.0%)	41 (26.3%)
1	42 (44.7%)	25 (40.3%)	67 (42.9%)
2	21 (22.3%)	15 (24.2%)	36 (23.1%)
3+	8 (8.5%)	4 (6.5%)	12 (7.7%)
Visceral Disease			
n	94	62	156

Yes	51 (54.3%)	31 (50.0%)	82 (52.6%)
No	43 (45.7%)	31 (50.0%)	74 (47.4%)
Resistance to Prior AI (Derived)			
n	94	62	156
Primary	35 (37.2%)	25 (40.3%)	60 (38.5%)
Secondary	58 (61.7%)	37 (59.7%)	95 (60.9%)
Unknown	1 (1.1%)	0	1 (0.6%)
Bone-only Disease			
n	94	62	156
Yes	18 (19.1%)	18 (29.0%)	36 (23.1%)
No	76 (80.9%)	44 (71.0%)	120 (76.9%)
Baseline Tumor Burden (Continuous)			
n	94	62	156
Mean (SD)	26.40 (30.82)	22.72 (26.37)	24.93 (29.10)
Median	18.50	15.00	17.00
Min – Max	0.0 – 184.1	0.0 – 97.0	0.0 – 184.1
Baseline Tumor Burden (Categorical)			
n	94	62	156
>= Median	50 (53.2%)	29 (46.8%)	79 (50.6%)
< Median	44 (46.8%)	33 (53.2%)	77 (49.4%)
Number of Metastatic Sites			
n	94	62	156
0	5 (5.3%)	1 (1.6%)	6 (3.8%)
1	30 (31.9%)	21 (33.9%)	51 (32.7%)
2	31 (33.0%)	24 (38.7%)	55 (35.3%)
3+	28 (29.8%)	16 (25.8%)	44 (28.2%)
Setting of Last CA Therapy			
n	94	62	156
Adjuvant	19 (20.2%)	16 (25.8%)	35 (22.4%)
Metastatic	74 (78.7%)	45 (72.6%)	119 (76.3%)
Other	1 (1.1%)	1 (1.6%)	2 (1.3%)
Previous Chemotherapy			
n	94	62	156
No	35 (37.2%)	18 (29.0%)	53 (34.0%)
Yes – Adjuvant/Neo-Adjuvant	39 (41.5%)	33 (53.2%)	72 (46.2%)
Yes – Metastatic	20 (21.3%)	11 (17.7%)	31 (19.9%)
Number of Previous Therapies			
n	94	62	156
1	5 (5.3%)	0	5 (3.2%)
2	13 (13.8%)	8 (12.9%)	21 (13.5%)
3+	76 (80.9%)	54 (87.1%)	130 (83.3%)
Prior Tamoxifen Use			
n	94	62	156
Yes	48 (51.1%)	32 (51.6%)	80 (51.3%)
No	46 (48.9%)	30 (48.4%)	76 (48.7%)

Number of Prior Tamoxifen Regimens

n	94	62	156
0	46 (48.9%)	30 (48.4%)	76 (48.7%)
1	47 (50.0%)	32 (51.6%)	79 (50.6%)
2	1 (1.1%)	0	1 (0.6%)

Prior AI Use

n	94	62	156
Yes	94 (100.0%)	62 (100.0%)	156 (100.0%)

Number of Prior AI Regimens

n	94	62	156
1	74 (78.7%)	46 (74.2%)	120 (76.9%)
2	19 (20.2%)	16 (25.8%)	35 (22.4%)
3	1 (1.1%)	0	1 (0.6%)

Prior Endocrine Therapy Use

n	94	62	156
Yes	94 (100.0%)	62 (100.0%)	156 (100.0%)

Number of Prior Endocrine Therapy Regimens

n	94	62	156
1	41 (43.6%)	21 (33.9%)	62 (39.7%)
2	38 (40.4%)	34 (54.8%)	72 (46.2%)
3	13 (13.8%)	7 (11.3%)	20 (12.8%)
4	2 (2.1%)	0	2 (1.3%)

Data Cutoff Date: 07MAR2014

Data Extract Date: 15SEP2014

Supplementary Table 7: Droplet digital PCR (ddPCR) assay sequences
for tissue assessment of *ESR1* mutation status

*ESR1*ddPCR

Assay	Forward Primer Sequence	Reverse Primer Sequence	WT Probe Sequence	Mutant Probe Sequence
E380Q	GGATTTGACCCTCCATGA	AGACCAATCATCAGGATCTC	ACATTCTAGAAGGTGGACC	ACATTGTAGAAGGTGGACC
P535H	ATCTGTACAGCATGAAGTG	GTCCAGCATCTCCAGC	TAGAGGGGCACCACG	TAGAGGTGCACCACGT
L536H	GTACAGCATGAAGTGCAA	GCGTCCAGCATCTCC	TCATAGAGGGGCACCA	TCATAGTGGGGCACCA
L536P	GTACAGCATGAAGTGCAA	GCGTCCAGCATCTCC	TCATAGAGGGGCACCA	TCATAGGGGGGCACC
L536Q	GTACAGCATGAAGTGCAA	GCGTCCAGCATCTCC	TGGTGCCCCTCTATGA	TGGTGCCCCAGTATGA
L536R	GTACAGCATGAAGTGCAA	GCGTCCAGCATCTCC	TGCCCCTCTATGACCTG	TGCCCCGCTATGACC
Y537C	GTACAGCATGAAGTGCAA	GGGCGTCCAGCATC	CCCCTCTATGACCTGCT	TGCCCCTCTGTGACC
Y537N	GTACAGCATGAAGTGCAA	GGCGTCCAGCATCTC	CCCCTCTATGACCTGC	AGGTCATTGAGGGGCA
Y537S	GTACAGCATGAAGTGCAA	GGGCGTCCAGCATC	AGCAGGTCATAGAGGGG	AGCAGGTCAGAGAGGG
D538G	TACAGCATGAAGTGCAAG	TGGGCGTCCAGCA	CCCCTCTATGACCTGCT	TCTATGGCCTGCTGCT

The BioRad QX200™ Droplet Digital PCR System is used for tissue DNA assessment of *ESR1* mutation status.

Supplementary Table 8: PAM50 signature gene set

<u>Gene</u>	<u>Accession Number</u>	<u>Gene</u>	<u>Accession Number</u>
ACTR3B	NM_001040135.1	KRT17	NM_000422.2
ANLN	NM_018685.2	KRT5	NM_000424.2
BAG1	NM_004323.3	MAPT	NM_016834.3
BCL2	NM_000657.2	MDM2	NM_006878.2
BIRC5	NM_001168.2	MELK	NM_014791.2
BLVRA	NM_000712.3	MIA	NM_006533.1
CCNB1	NM_031966.2	MKI67	NM_002417.2
CCNE1	NM_001238.1	MLPH	NM_024101.4
CDC20	NM_001255.2	MMP11	NM_005940.3
CDC6	NM_001254.3	MYBL2	NM_002466.2
CDH3	NM_001793.4	MYC	NM_002467.3
CENPF	NM_016343.3	NAT1	NM_001160174.1
CEP55	NM_018131.3	NDC80	NM_006101.2
CXXC5	NM_016463.7	NUF2	NM_031423.3
EGFR	NM_201282.1	ORC6	NM_014321.2
ERBB2	NM_004448.2	PGR	NM_000926.2
ESR1	NM_000125.2	PHGDH	NM_006623.2
EXO1	NM_003686.3	PTTG1	NM_004219.2
FGFR4	NM_002011.3	RRM2	NM_001034.1
FOXA1	NM_004496.2	SFRP1	NM_003012.3
FOXC1	NM_001453.1	SLC39A6	NM_012319.2
GPR160	NM_014373.2	TMEM45B	NM_138788.3
GRB7	NM_005310.2	TYMS	NM_001071.1
KIF2C	NM_006845.3	UBE2C	NM_181803.1
KRT14	NM_000526.4	UBE2T	NM_014176.3