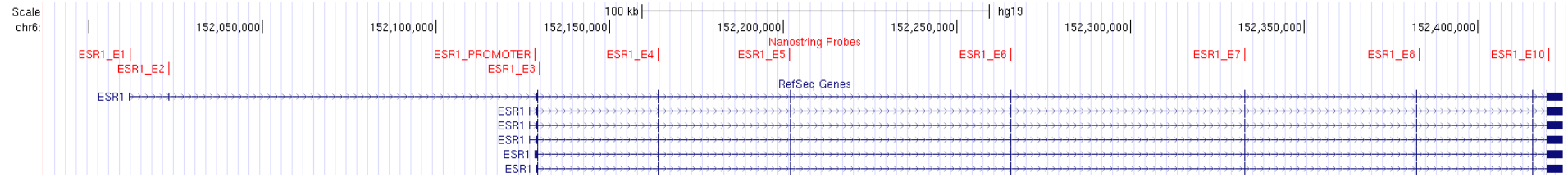
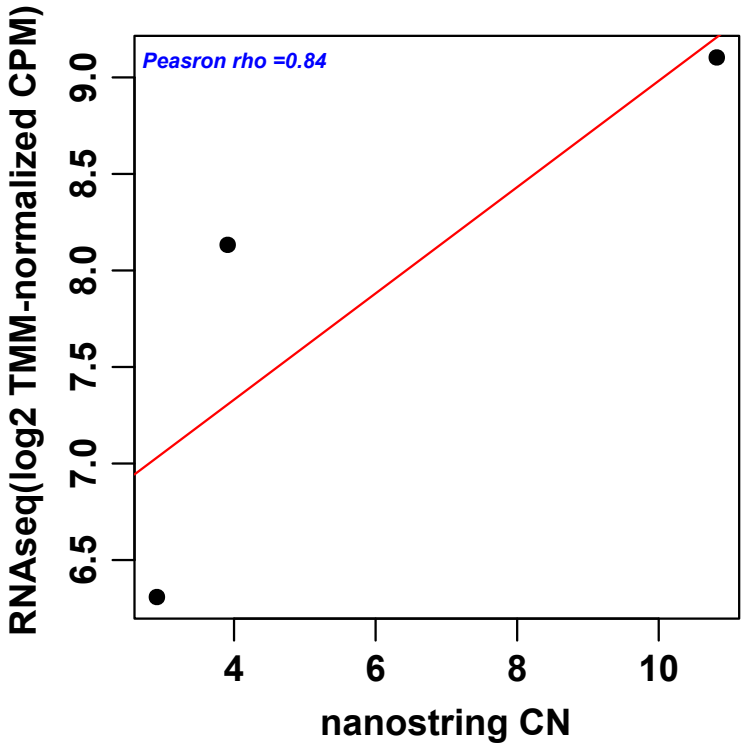

Scale
chr6:
100 kb
hg19
152,050,000
152,100,000
152,150,000
152,200,000
152,250,000
152,300,000
152,350,000
152,400,000
Nanostring Probes
ESR1_E1
ESR1_E2
ESR1_PROMOTER
ESR1_E3
ESR1_E4
ESR1_E5
ESR1_E6
ESR1_E7
ESR1_E8
ESR1_E10
RefSeq Genes
ESR1
ESR1
ESR1
ESR1
ESR1
ESR1

Figure S1


Peasron rho =0.84
RNAseq(log2 TMM-normalized CPM)
6.5
7.0
7.5
8.0
8.5
9.0
4
6
8
10
nanostring CN

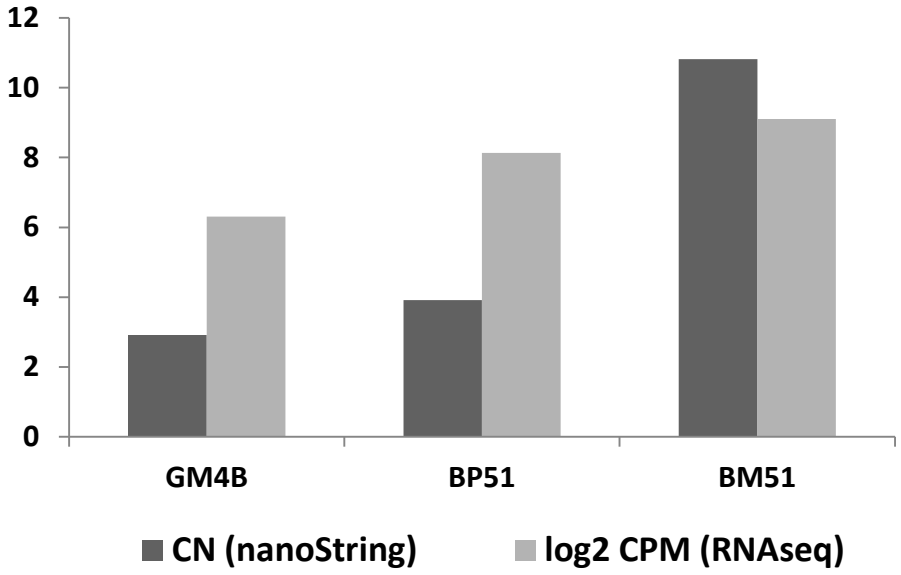

12
10
8
6
4
2
0
GM4B
BP51
BM51
CN (nanoString)
log2 CPM (RNAseq)

Figure S2

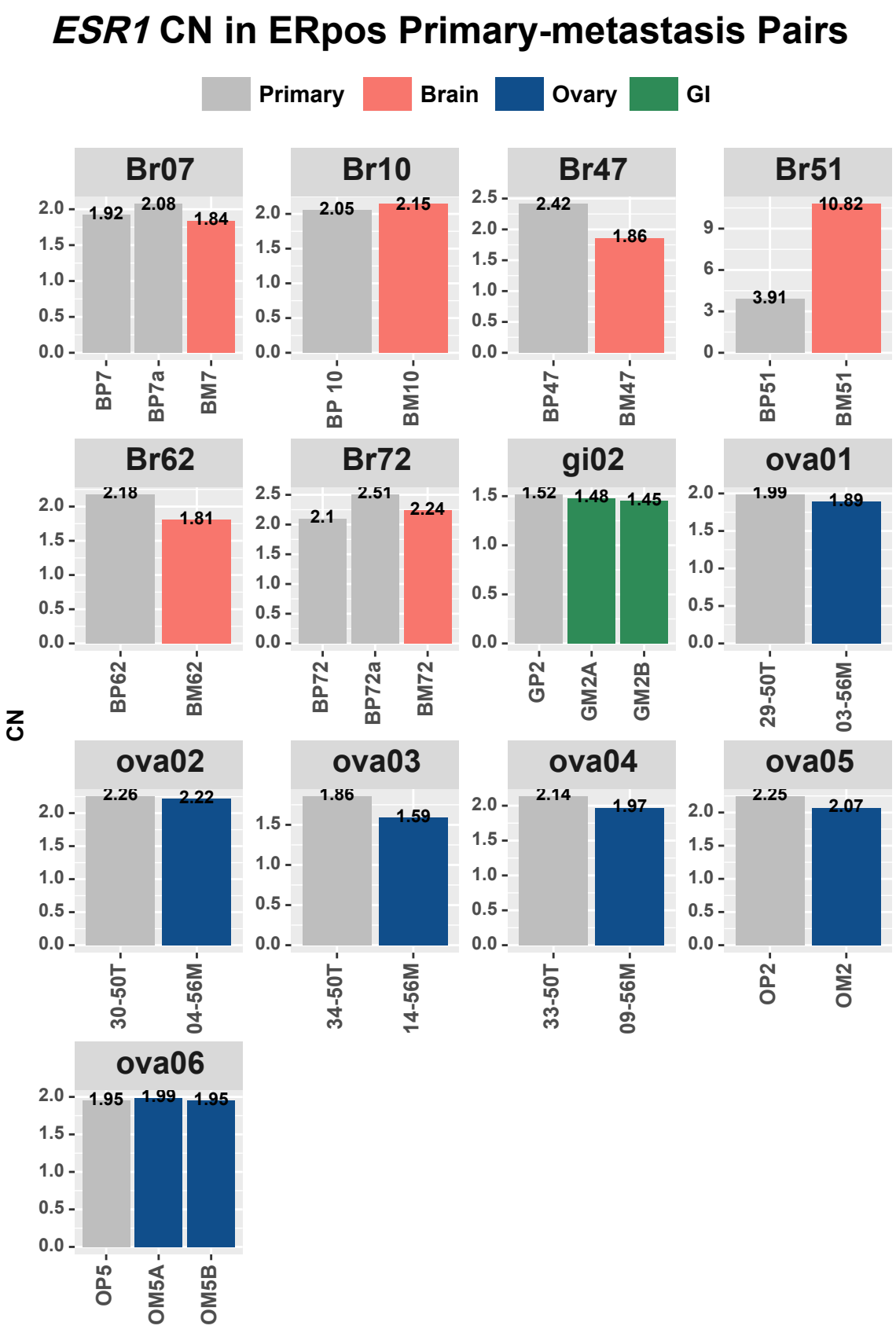
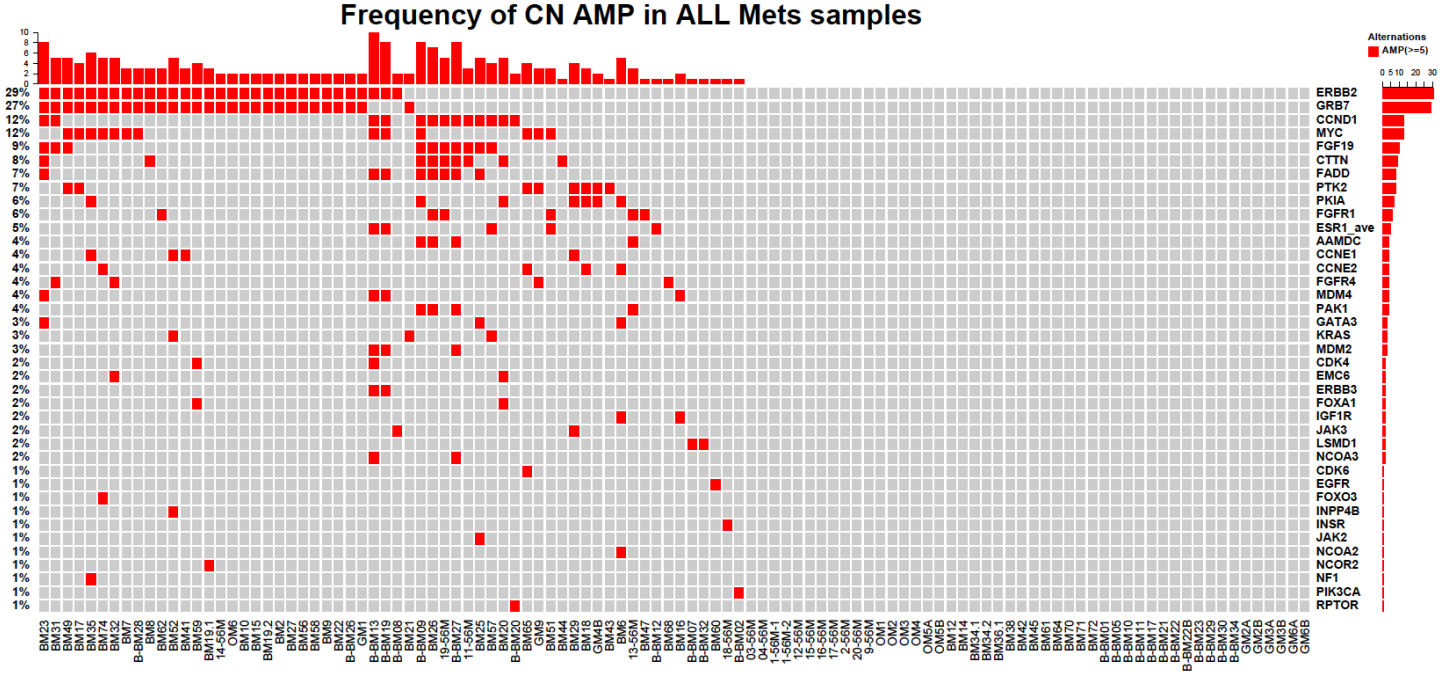

ESR1 CN in ERpos Primary-metastasis Pairs
Primary
Brain
Ovary
GI
CN
Br07
1.92
2.08
1.84
BP7
BP7a
BM7
Br10
2.05
2.15
BP 10
BM10
Br47
2.42
1.86
BP47
BM47
Br51
3.91
10.82
BP51
BM51
Br62
2.18
1.81
BP62
BM62
Br72
2.1
2.51
2.24
BP72
BP72a
BM72
gi02
1.52
1.48
1.45
GP2
GM2A
GM2B
ova01
1.99
1.89
29-50T
03-56M
ova02
2.26
2.22
30-50T
04-56M
ova03
1.86
1.59
34-50T
14-56M
ova04
2.14
1.97
33-50T
09-56M
ova05
2.25
2.07
OP2
OM2
ova06
1.95
1.99
1.95
OP5
OM5A
OM5B

Figure S3


Frequency of CN AMP in ALL Mets samples
Alternations
AMP(>=5)
ERBB2
GRB7
CCND1
MYC
FGF19
CTTN
FADD
PTK2
PKIA
FGFR1
ESR1_ave
AAMDC
CCNE1
CCNE2
FGFR4
MDM4
PAK1
GATA3
KRAS
MDM2
CDK4
EMC6
ERBB3
FOXA1
IGF1R
JAK3
LSMD1
NCOA3
CDK6
EGFR
FOXO3
INPP4B
INSR
JAK2
NCOA2
NCOR2
NF1
PIK3CA
RPTOR

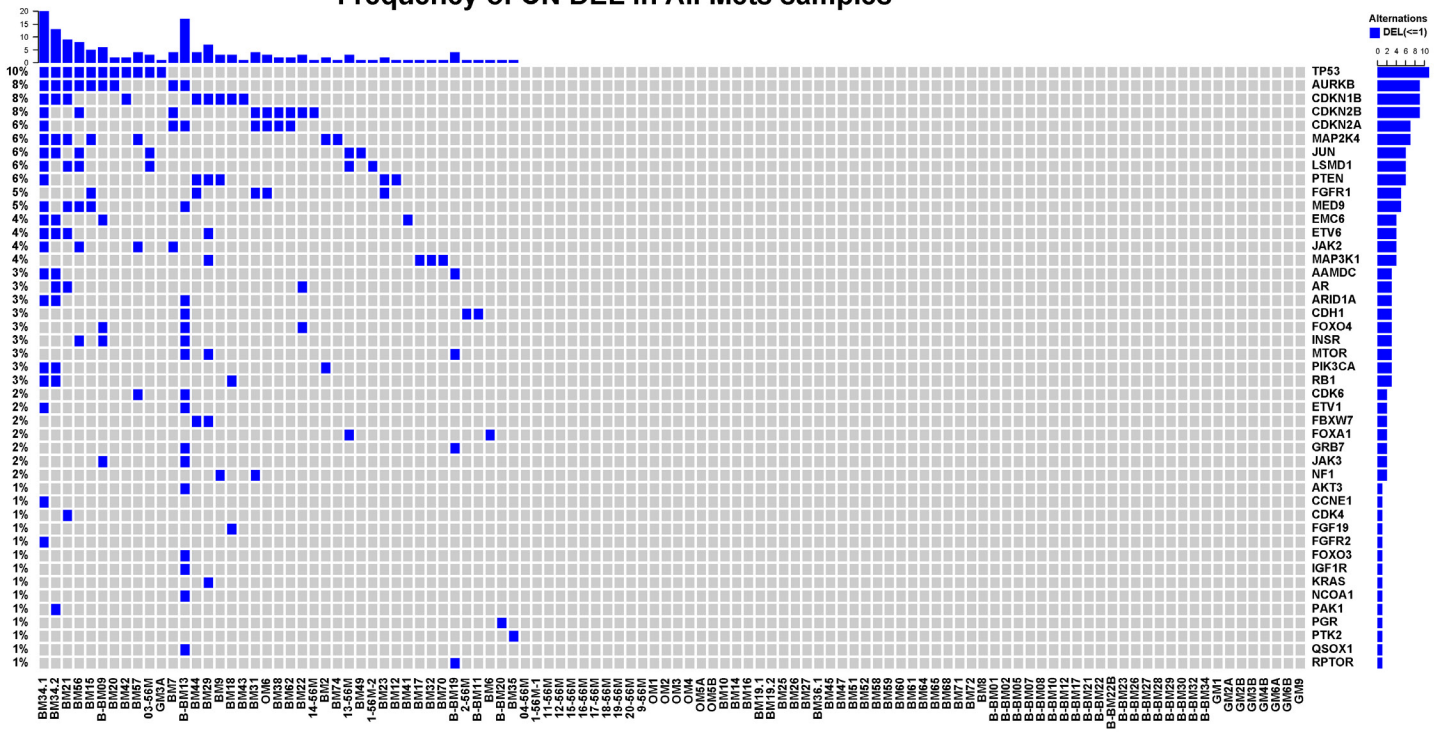

Frequency of CN DEL in All Mets samples
Alternations
DEL(<=1)
TP53
AURKB
CDKN1B
CDKN2B
CDKN2A
MAP2K4
JUN
LSMD1
PTEN
FGFR1
MED9
EMC6
ETV6
JAK2
MAP3K1
AAMDC
AR
ARID1A
CDH1
FOXO4
INSR
MTOR
PIK3CA
RB1
CDK6
ETV1
FBXW7
FOXA1
GRB7
JAK3
NF1
AKT3
CCNE1
CDK4
FGF19
FGFR2
FOXO3
IGF1R
KRAS
NCOA1
PAK1
PGR
PTK2
QSOX1
RPTOR

Figure S4

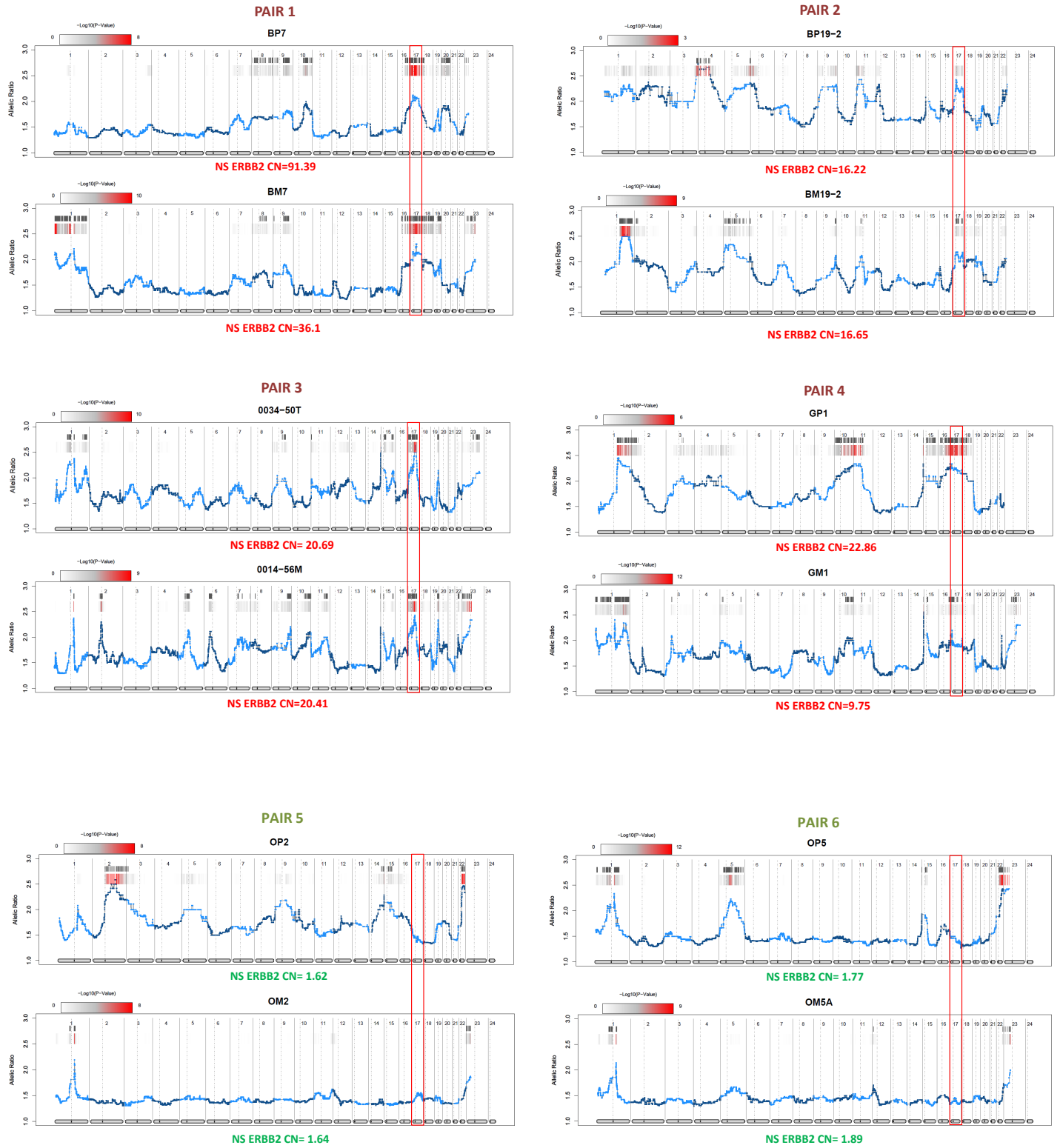

PAIR 1
BP7
NS ERBB2 CN=91.39
BM7
NS ERBB2 CN=36.1
PAIR 2
BP19−2
NS ERBB2 CN=16.22
BM19−2
NS ERBB2 CN=16.65
PAIR 3
0034−50T
NS ERBB2 CN= 20.69
0014−56M
NS ERBB2 CN=20.41
PAIR 4
GP1
NS ERBB2 CN=22.86
GM1
NS ERBB2 CN=9.75
PAIR 5
OP2
NS ERBB2 CN= 1.62
OM2
NS ERBB2 CN= 1.64
PAIR 6
OP5
NS ERBB2 CN= 1.77
OM5A
NS ERBB2 CN= 1.89

Figure S5

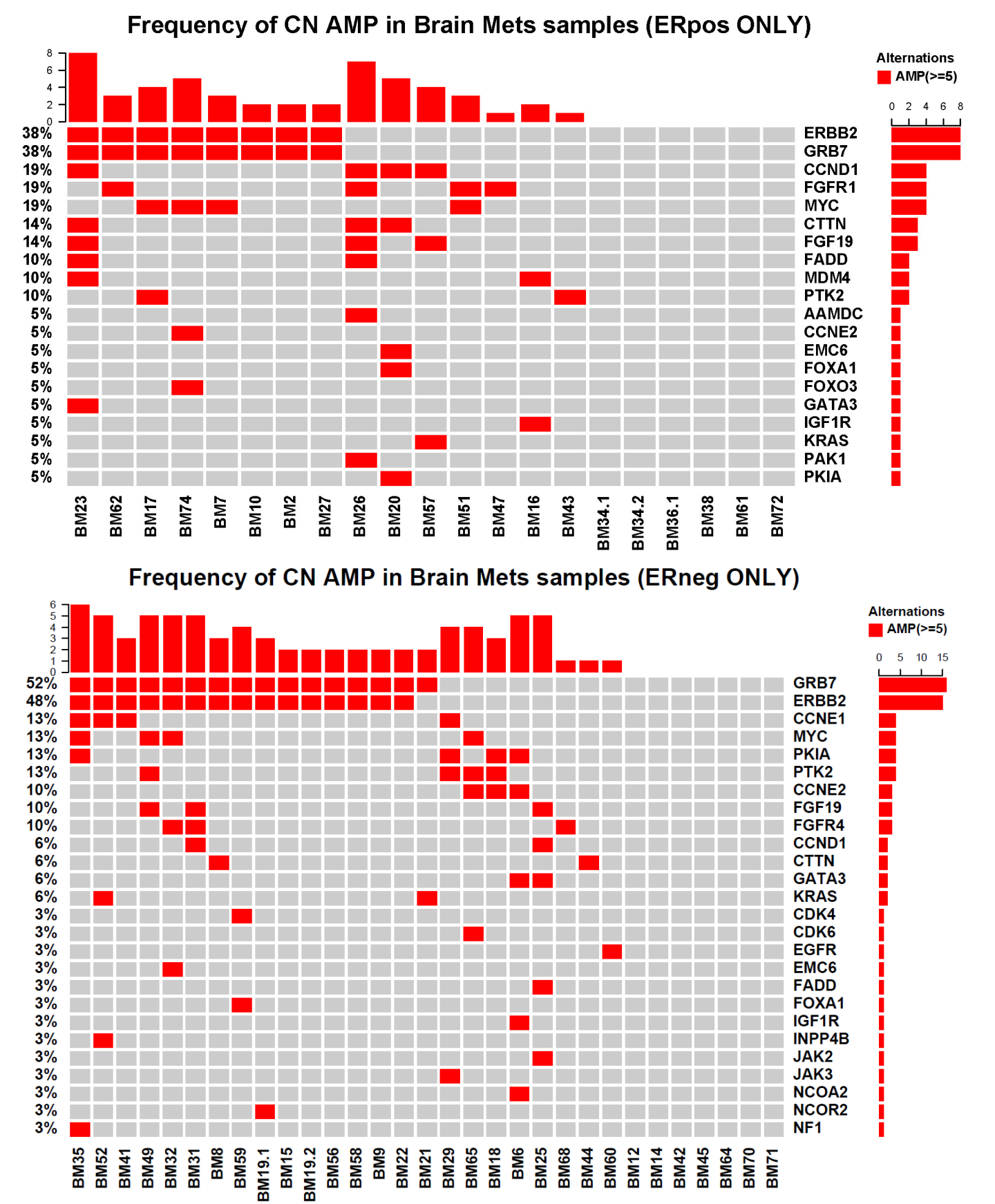
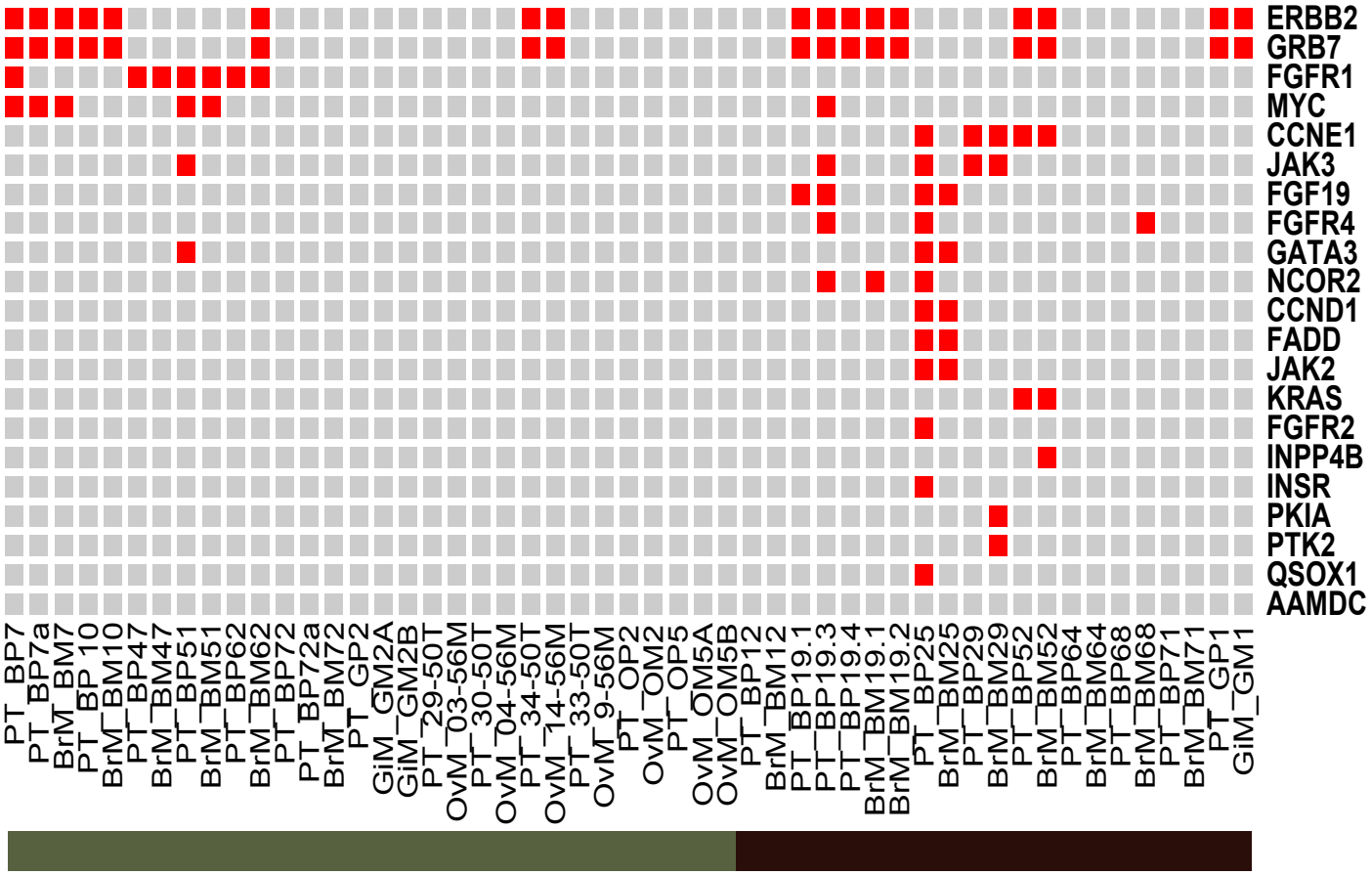

Frequency of CN AMP in Brain Mets samples (ERpos ONLY)
Alternations
AMP(>=5)
38% ERBB2
38% GRB7
19% CCND1
19% FGFR1
19% MYC
14% CTTN
14% FGF19
10% FADD
10% MDM4
10% PTK2
5% AAMDC
5% CCNE2
5% EMC6
5% FOXA1
5% FOXO3
5% GATA3
5% IGF1R
5% KRAS
5% PAK1
5% PKIA
BM23 BM62 BM17 BM74 BM7 BM10 BM2 BM27 BM26 BM20 BM57 BM51 BM47 BM16 BM43 BM34.1 BM34.2 BM36.1 BM38 BM61 BM72
Frequency of CN AMP in Brain Mets samples (ERneg ONLY)
Alternations
AMP(>=5)
52% GRB7
48% ERBB2
13% CCNE1
13% MYC
13% PKIA
13% PTK2
10% CCNE2
10% FGF19
10% FGFR4
6% CCND1
6% CTTN
6% GATA3
6% KRAS
3% CDK4
3% CDK6
3% EGFR
3% EMC6
3% FADD
3% FOXA1
3% IGF1R
3% INPP4B
3% JAK2
3% JAK3
3% NCOA2
3% NCOR2
3% NF1
BM35 BM52 BM41 BM49 BM32 BM31 BM8 BM59 BM19.1 BM15 BM19.2 BM56 BM58 BM9 BM22 BM21 BM29 BM65 BM18 BM6 BM25 BM68 BM44 BM60 BM12 BM14 BM42 BM45 BM64 BM70 BM71

Figure S6

Frequency of CN AMP in Paired samples



Frequency of CN DEL in Paired samples

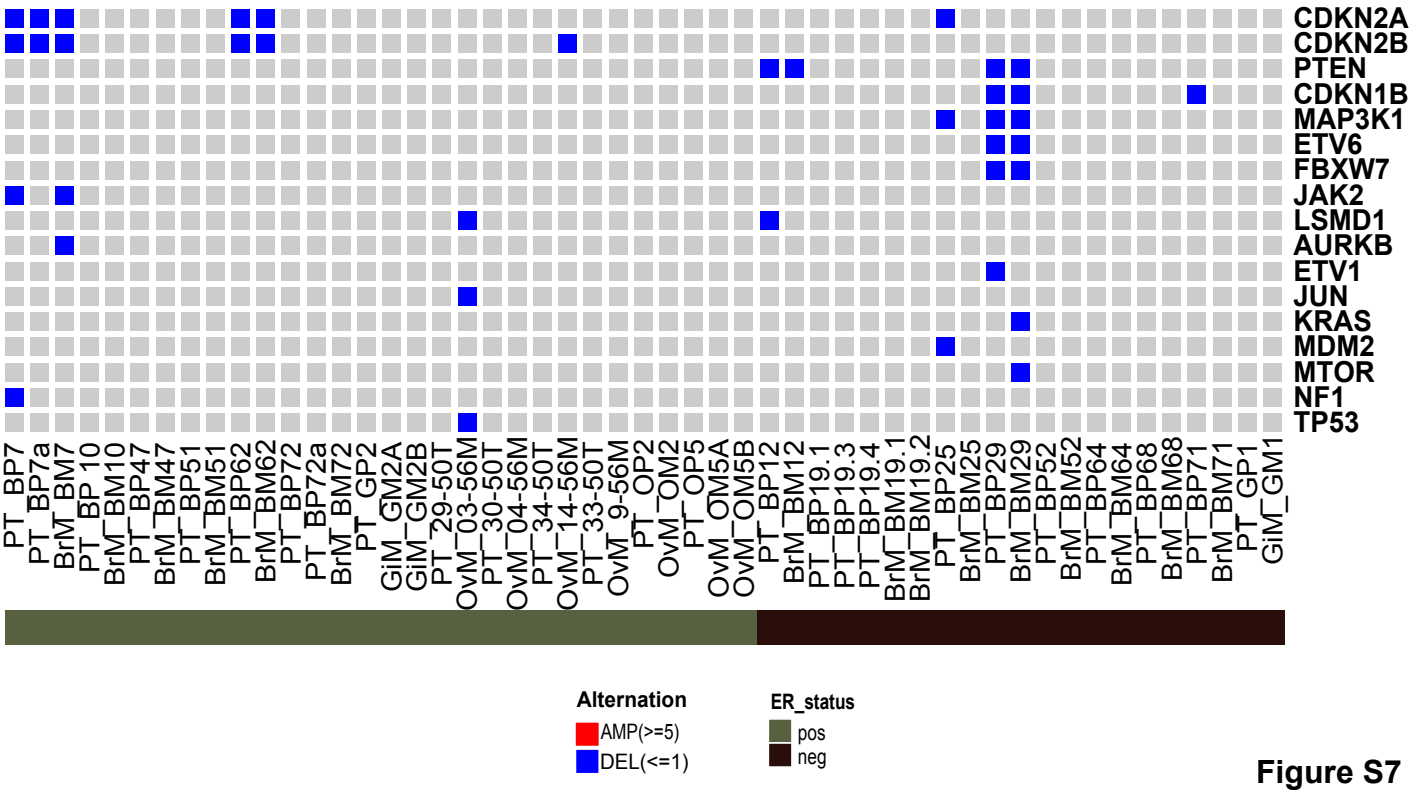


Figure S7

Supplementary Figures:

Figure S1: Genomic organization of *ESR1* nanoString probes

Genomic coordinates for nanoString probes comprehensively covering *ESR1* exons are highlighted in red. Different *ESR1* isoforms (RefSeq genes) are listed in blue for comparison. Exons are represented by blocks connected by horizontal lines representing introns. Thinner blocks represent untranslated regions (UTRs).

Figure S2: Correlation of *ESR1* amplification with mRNA expression

The data shows dose-dependent correlation between copy number (nanoString) and expression (RNAseq) for 3 amplified samples.

Figure S3: *ESR1* copy number status in ER+ primary-metastasis pairs

Each pair is plotted separately. Y-axis: CN= copy number call. Different colors represent different sites. Br=brain pair; ova= ovarian pair; gi= gi pair. Pair Br51 showed a clear increase in *ESR1* copy number for the brain metastasis.

Figure S4: Oncoprint visualization of copy number alterations in clinical samples

Copy number alterations by genes (rows) and samples (columns) show amplifications (top in red) and deletions (bottom in blue) frequency. Top and right-side bars show counts for alteration events within a sample and by gene, respectively.

Figure S5: eSNP-Karyotyping analysis of ERBB2 amplified samples

Shown are moving median plots for 6 paired tumors (4 *ERBB2* amplified + two unamplified). Color bars represent FDR-corrected p-value. Positions with $p < 0.01$ are marked by black line. Highlighted in red is the *ERBB2* region in chromosome 17. The left side of the figure shows the copy number calls by nanoString analysis.

Figure S6: Oncoprint visualization of copy number alterations in ER+ vs ER+ brain metastases

Copy number alterations by genes (rows) and samples (columns) show amplifications frequencies in ER+ (top) and ER- (bottom) samples. Top and right side bars show counts for alteration events within a sample and by gene, respectively. ER+ tumors showed enrichment for FGFR1 amplifications (Fisher's exact test p= 0.0221).

Figure S7: Oncoprint visualization of copy number alterations in paired primary-met samples

Copy number alterations by genes (rows) and samples (columns) show amplifications (amplifications) and deletions frequencies (bottom) in paired-samples (columns organized by pairs). Top and right-side bars in each plot show counts for alteration events within a sample and by gene, respectively. Pair BP-BM29 showed increase in *PKIA* and *PTK* CN, while pair BP-BM68 showed increase in *FGFR4* CN. PT: primary tumor; BrM: brain metastasis; GiM: GI metastasis; OvM: ovarian metastasis.