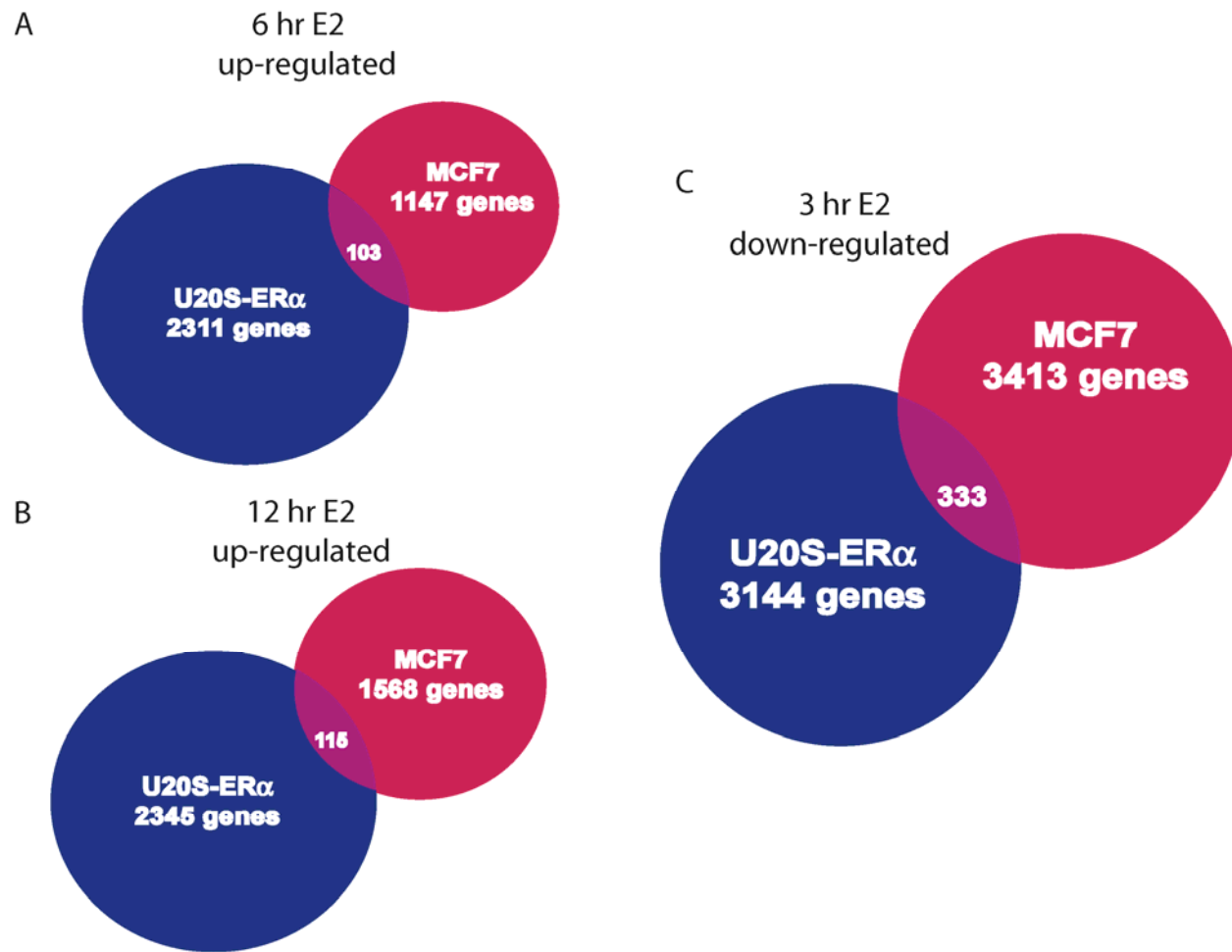


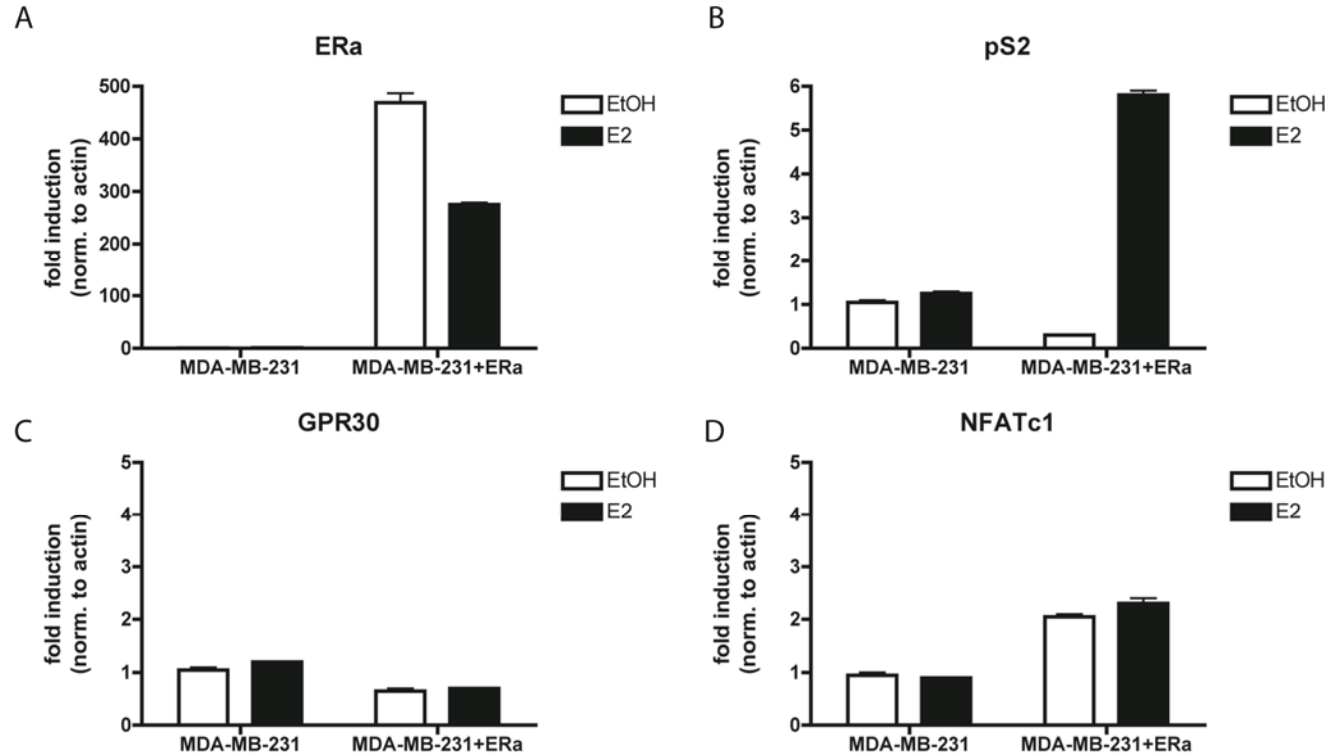
# Supplemental 1



Venn diagrams showing the number of genes up-regulated by 10 nM E2 for A, 6 hours, or B, 12 hours or C, down-regulated by E2 at 3 hours in MCF7 and U2OS-ER $\alpha$  cells.

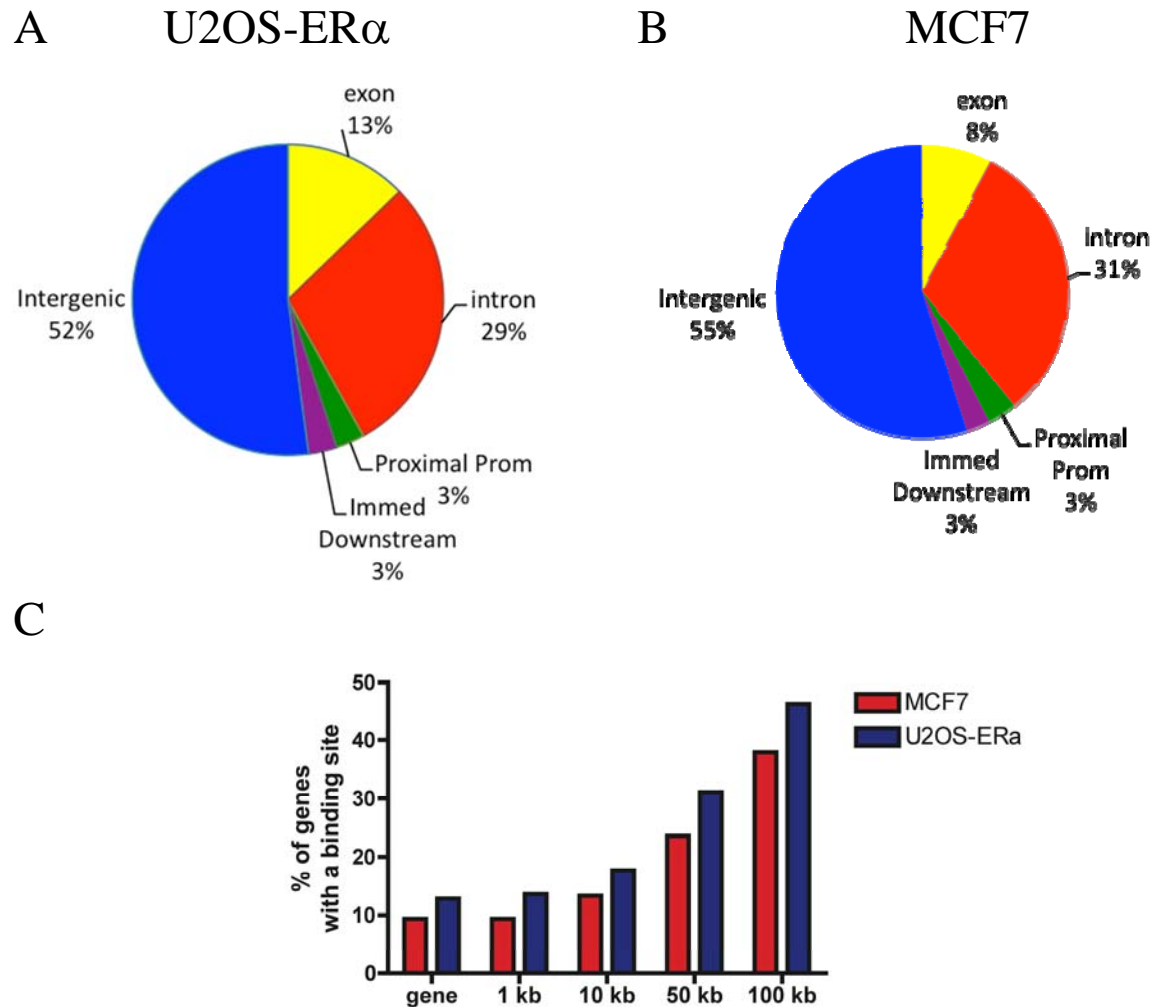
2345 genes

## Supplemental 2



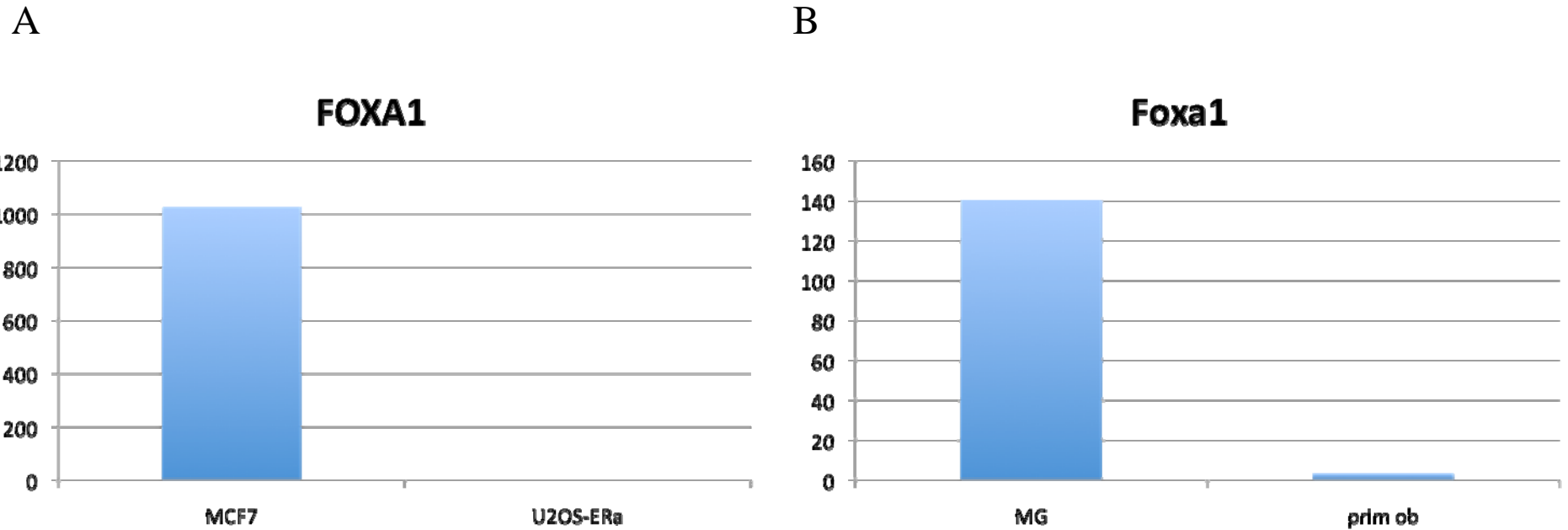
Over-expression of ER $\alpha$  does not regulate osteoblast-specific genes in MDA-MB-231 cells. MDA-MB-231 or MDA-MB-231 cells stably transfected with ER $\alpha$  were treated for 3 hours with vehicle (EtOH) or 10 nM E2. RNA was obtained and quantitative PCR was performed with primers for A. ER $\alpha$ , B. pS2, C. GPR30 and D. NFATc1.

# Supplemental 3



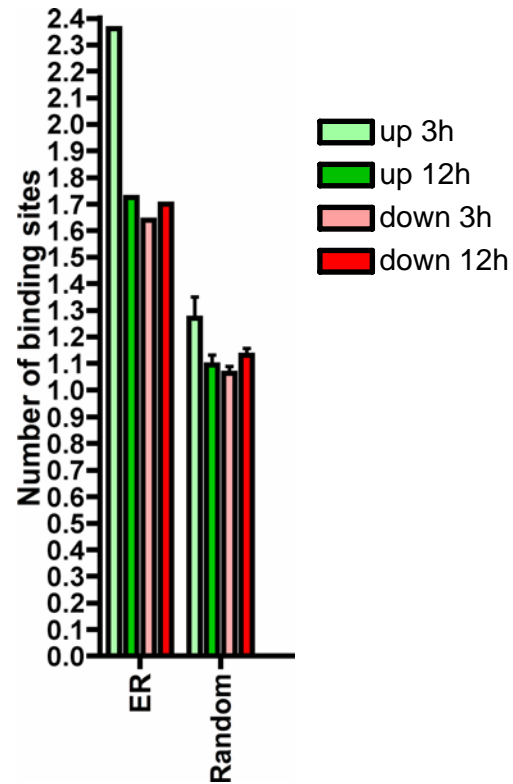
Supplemental 2: Location of ER $\alpha$  binding sites. A and B: Location of ER $\alpha$  binding sites in MCF7 and U2OS-ER $\alpha$  cells. Location analysis of binding sites was performed using CEAS (47). C: Up-regulated genes in U2OS-ER $\alpha$  cells or MCF7 cells were analyzed for the presence of binding sites within the gene or within the gene plus the indicated distance upstream and downstream of the gene.

# Supplemental 4



A. The basal levels of FOXA1 mRNA in MCF7 and U2OS-ER $\alpha$  cells as determined by qPCR. B. The basal levels of Foxa1 mRNA in mouse mammary gland (MG) and primary osteoblasts (prim ob) as determined by qPCR.

## Supplemental 5



Genes regulated by E2 at 3 hours have a higher number of ER $\alpha$  binding sites. Each of the genes up- or down-regulated at 3 or 12 hours by E2 in MCF7 cells was analyzed for the number of ER $\alpha$  binding sites in the gene and 30 kb upstream of the gene. A randomly generated set of binding sites was also compared to the regulated genes in MCF7 cells.

## Supp. Table 1

| Human mRNA primers    |                         |                           |
|-----------------------|-------------------------|---------------------------|
| SEQUENCE              | FORWARD PRIMER 5' TO 3' | REVERSE PRIMER 5' TO 3'   |
| <b>B-ACTIN</b>        | GGACTTCGAGCAAGAGATGG    | AGCACTGTGTTGGCGTACAG      |
| <b>GPR30</b>          | GCAATTGCACTCATGTGGAC    | TTCCGCACATGACAGGTTTA      |
| <b>FASL</b>           | GGCCCATTTAACAGGCAAGTC   | GGCCACCCTTCTTATACTTCAC    |
| <b>XBP1</b>           | GCGCCTCACGCACCT         | GCTGCTACTCTGTTTTTCAGTTTCC |
| <b>CTSD</b>           | GACCTGCCTCTCCACTTTGA    | CACTGCAAACCTGCTGGACAT     |
| <b>C-FOS</b>          | AGAATCCGAAGGGAAAAGGAA   | CTTCTCCTTCAGCAGGTTGG      |
| <b>NFATC1</b>         | AGAAAGCGAAGCCAGTACCA    | CGGTCTCACTAACGGGACAT      |
| <b>CDH26</b>          | CAAACAGGGACTTTCCAGAA    | AGTGTTTGGTGGCCTTCATC      |
| <b>NR5A2</b>          | CAGTGCTCCCCACTGAAAAT    | GCCAGGTTACAAATCGGCTA      |
| <b>SOX5</b>           | CAGCAGCTGGTGAGATTTGA    | AGTCACTTGGGAGGATGTGG      |
| <b>MYB</b>            | TACCCAACCTGTTACGCAGA    | CTTTCCACAGGATGCAGGTT      |
| <b>NR1P</b>           | TCGCACTCACCACAGAAAAC    | AGCCAAGCTCTTCTCCATGT      |
| <b>PAX7</b>           | CACTGTGACCGAAGCACTGT    | GTCAGGTTCCGACTCCACAT      |
| <b>MYT1L</b>          | CCCCCATTTGTTATAACAGC    | CCAACGTTAGATGAGCAGCA      |
| <b>ALKALINE PHOS.</b> | CCACGTCTTCACATTTGGTG    | AGACTGCGCCTGGTAGTTGT      |
| <b>FOXO1</b>          | AAGAGCGTGCCCTACTTCAA    | TTCTTCATTCTGCACACGA       |
| <b>FOXO4</b>          | ACGAGTGGATGGTCCGTACT    | GCCTCGTTGTGAACCTTGAT      |

## Supp. Table 2

| Mouse mRNA primers    |                         |                         |
|-----------------------|-------------------------|-------------------------|
| SEQUENCE              | FORWARD PRIMER 5' TO 3' | REVERSE PRIMER 5' TO 3' |
| <b>C-FOS</b>          | CCGACTCCTTCTCCAGCAT     | TCACCGTGGGGATAAAGTTG    |
| <b>XBP1</b>           | TCAAATGTCCTTCCCCAGAG    | GGTCCCCACTGACAGAGAAA    |
| <b>NRIP</b>           | ACGACTTCCAGACCCACAAC    | CTCAGCAAGCGACTCAACAG    |
| <b>PAX7</b>           | GATCACCCCTCATCCAGTGCT   | GGTGTCTTGTCGGTTCAGGT    |
| <b>MYT1L</b>          | GGTGTGCAATCCTGTGTCAG    | GTGCCGCTGGGATATTCTTA    |
| <b>ALKALINE PHOS.</b> | GACGCAGAGTCCCTTCAGAC    | CACCCCTACTCCCCATACCT    |
| <b>FOXO1</b>          | GGGTCTGTCTCCCTTTCCTC    | CAACTGCCCATGATTCACAC    |
| <b>SOX5</b>           | AACAAGCACAGATCCCCATC    | TGTCCTCAGCCTGGATCTCT    |
| <b>FOXO4</b>          | CAAGAAGAAGCCGTCTGTCC    | CTGACGGTGCTAGCATTTGA    |
| <b>B-ACTIN</b>        | AGCCATGTACGTAGCCATCC    | CTCTCAGCTGTGGTGGTGAA    |

## Supp. Table 3

| Human ChIP primers |                         |                         |
|--------------------|-------------------------|-------------------------|
| SEQUENCE           | FORWARD PRIMER 5' TO 3' | REVERSE PRIMER 5' TO 3' |
| XPB1 PROMOTER      | CATAGCCACGGTCCTGAAAC    | CCACCACCATAGCTCCAGAC    |
| XPB1 ENH1          | ATACTTGGCAGCCTGTGACC    | GGTCCACAAAGCAGGAAAAA    |
| XPB1 ENH2          | TTGCTGTGCAAACAATAGCC    | GTCCAAGGGCACATTTCAT     |
| XPB1 ENH3          | AGGACTCCTTTGCGGGTAAT    | GTGAAAAATTTCGGTGGCATT   |
| PAX7 PROMOTER      | AGCAAGGAGCTCAGAGTTGG    | CAGGTTTCTCCTCCCCTTC     |
| PAX7 ENHANCER      | CAAAACGATCACTGCTCGAA    | ATGGGAGGAAGACCCTGAGT    |
| FASL PROMOTER      | TCCTGTAGCTGGGAGCAGTT    | AGAGCAAACCCCTGGAAGTT    |
| FASL ENHANCER 1    | CCACCAGGACCAGAATGTTT    | GGTTCCAGCTGACCAAATGT    |
| FASL ENHANCER 2    | GGCCTCCCAAAGGTATTAGC    | GTTGCTCAATGGCCAAGAAT    |
| ALPL 1             | GTGTGATCATTGCCCTCTT     | GCTGAGCTTTGTCTGGGAAC    |
| ALPL 2             | CACTGGGAGCCTAAGCAGAC    | CCTGAGGTCAGGTTGTTGT     |
| ALPL 3             | TCAACCTGTTTGTGGATGGA    | GACTCTCCCTAGGCCACACA    |
| B-ACTIN            | CAGTGCCTAGGTCACCCACT    | AGAAGTCGCAGGACCACACT    |
| HBB                | TGGTATGGGGCCAAGAGATA    | TAGATGGCTCTGCCCTGACT    |