1 **Supplemental Figure Legends:**

2 **Supplemental Figure 1. Mouse strain variability in hormone stimulated mammary ductal**

3 **side-branching.** Representative wholemount images of carmine-stained wild type (WT) mice

4 of different strains after treatment with E_2+P_4 for 48 hours. Scale bars represent 2mm;

5 Representative glands are shown from four individual mice per mouse background.

6

7 **Supplemental Figure 2. Temporal induction of ductal side-branching by progesterone.**

8 Representative mammary gland wholemount images of carmine-stained wild type (WT) mice

9 after different hormone treatments. Scale bars represent 2mm; Representative glands are

10 shown from four individual mice per treatment.

11

12 **Supplemental Figure 3. Temporal induction of proliferation by progesterone.**

13 Representative Ki67 immunohistochemistry images of wild type (WT) mice after different

14 hormone treatments. Scale bars represent 50um; Representative images are shown from three

15 individual mice per treatment.

16

17 **Supplemental Figure 4. Quantitative Real-Time PCR timecourse of known PR-dependent**

18 **target genes**. Quantitative Real-Time PCR timecourse for *Wnt4*, *Rankl*, *Areg*, and *Ccnd1*.

19 Student's t Test, *P < 0.01, **P < 0.0001; Three wild type mice per pool, tested in triplicate per

20 hormone treatment group; Results are means ± SEM of three independent biological replicates.

21

22 **Supplemental Figure 5. Comparison of progesterone-regulated gene signatures in**

23 **mouse mammary gland with published prolactin gene signature.** (A) Proportional Venn

24 diagram representing P_4-regulated genes at 24 hours under our conditions (red) and PRL-

25 regulated genes during early pregnancy as described by Harris et al. (blue) (Harris et al., 2006).

26 (B) Quantitative Real-Time PCR of PRL-induced gene *Elf5*; Student's t Test, NS represents Not

27   Significant; Three mice per pool, tested in triplicate per hormone treatment group; Results are

28   means ± SEM of three independent biological replicates.

29

30   **Supplemental Figure 6. Validation of progesterone receptor ChIP-seq accuracy.** (A)

31   Proportional Venn diagram representing the intersection of PR binding sites identified in two

32   ChIP-seq replicates; Three mice per replicate. (B) Pearson correlation of the PR binding sites

33   of two ChIP-seq replicates (r = .91). (C) Conservation plot of mouse PR binding sites with high

34   conservation around peak centers compared to flanking regions.

35

36   **Supplemental Figure 7. Progesterone regulates distinct tissue selective functions.** (A)

37   Proportional Venn diagram representing PR binding regions within the mammary gland (25,598)

38   and PR binding regions within the uterus (18,433) (Rubel et al. 2012). There are a total of 5,227

39   overlapping binding sites corresponding with 3,675 unique genes. (B) Of these unique genes,

40   148 are transcriptionally regulated within the mammary gland and 408 are transcriptionally

41   regulated within the uterus.

42

43   **Supplemental Figure 8. Representative screen shots of ChIP-seq data showing gene**

44   **progesterone receptor recruitment at 6 hours after exposure to progesterone.** UCSC

45   Genome Browser screen shots representing PR binding peaks in relation to the TSS of (A)

46   *Eps8l1*, (B) *Itga3*, (C) *Vil2*, (D) *Bdnf*, (E) *Vav2*, (F) *Wnt4*, (G) *Rankl*, or (H) *Zbtb16*. Peak

47   locations relative to the TSS are listed below each screen shot and peak values are listed in red

48   above each peak. Red boxes represent peaks that were validated by ChIP-qPCR.
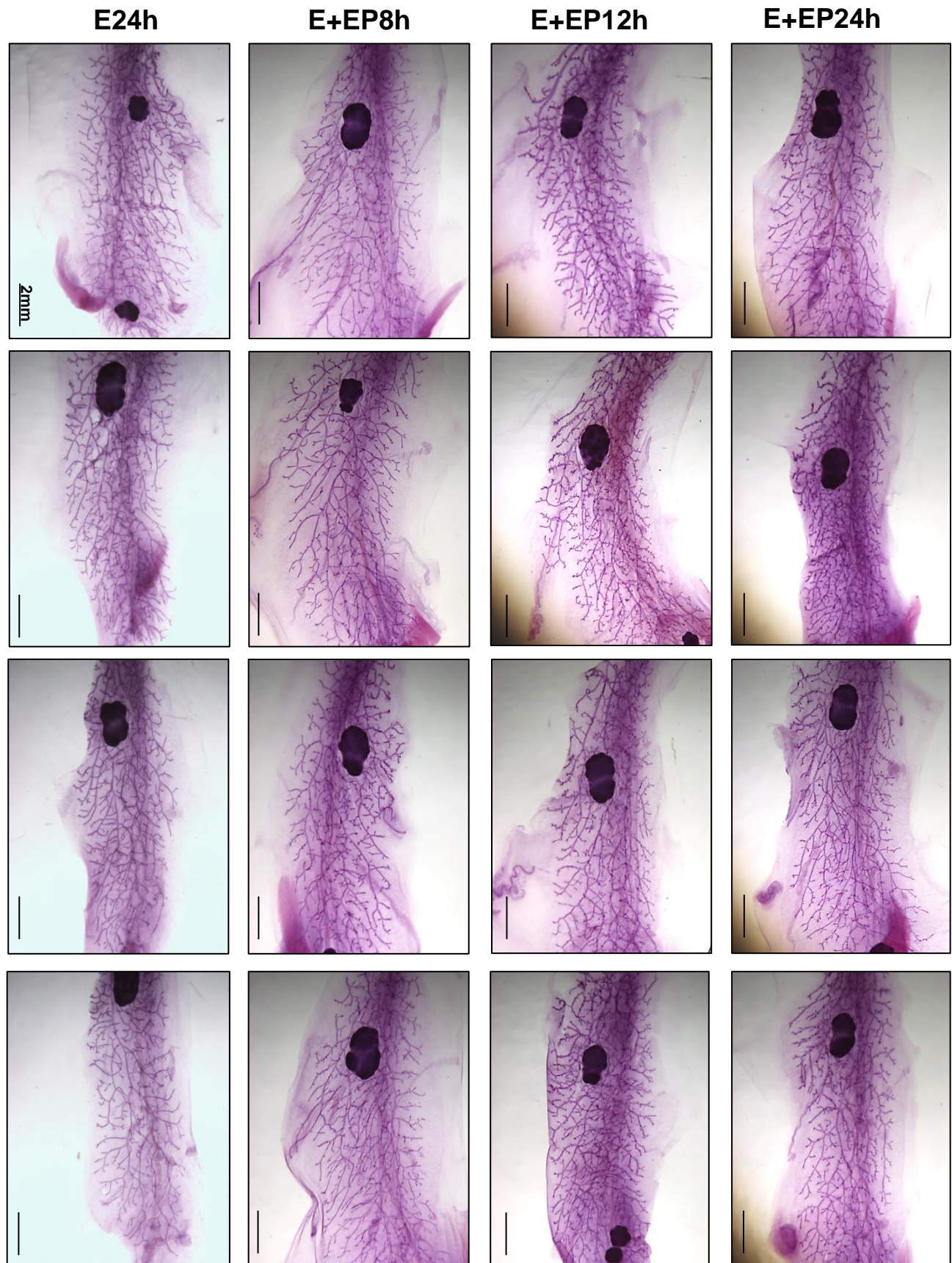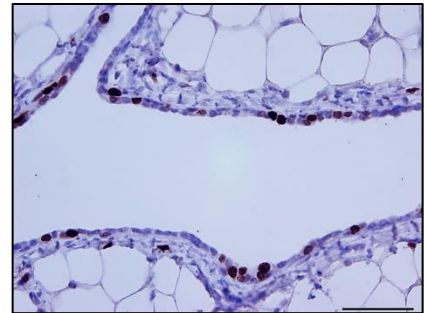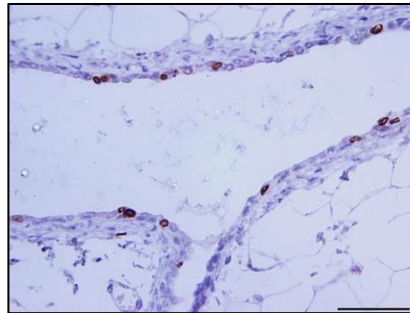
SUPPLEMENTAL FIGURE 1

C57BL/6	129SvEv	BALB/c

SUPPLEMENTAL FIGURE 2

| E24h | E+EP8h | E+EP12h | E+EP24h |

2mm
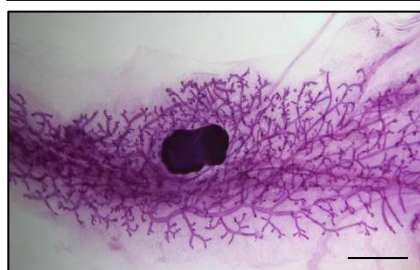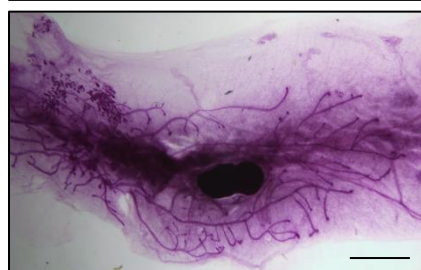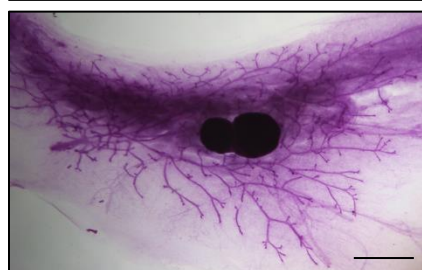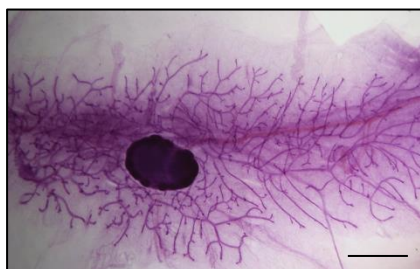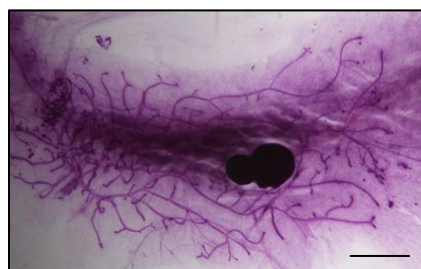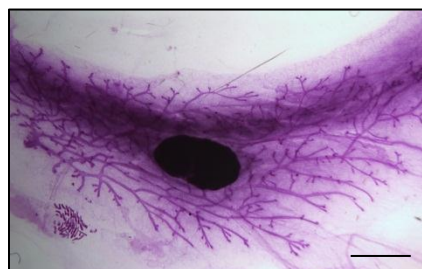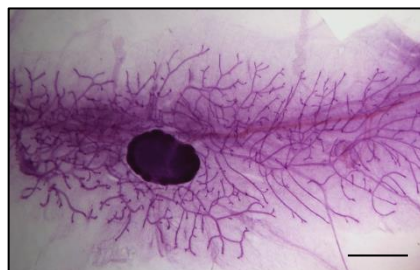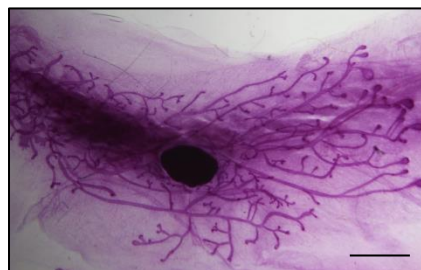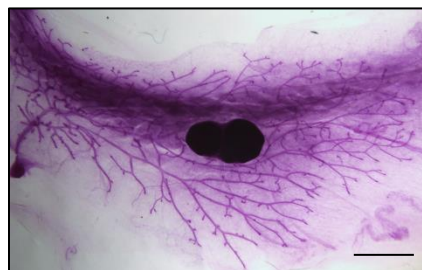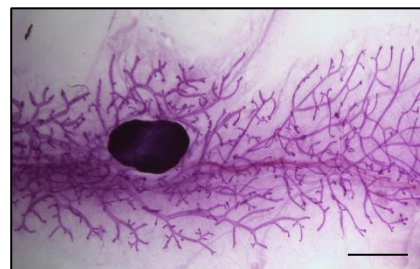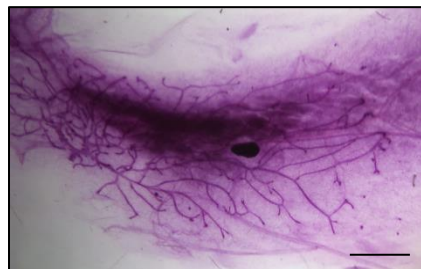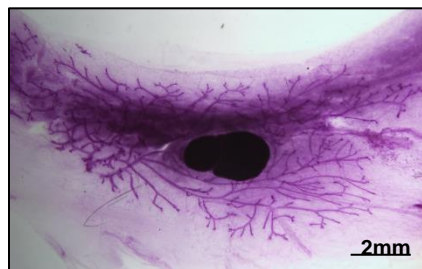
SUPPLEMENTAL FIGURE 3

E+EP12h	E+EP24h	E+EP48h
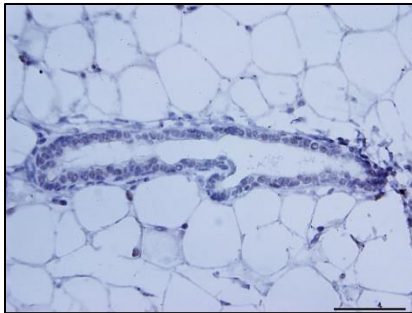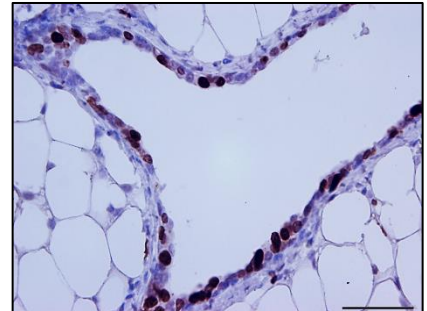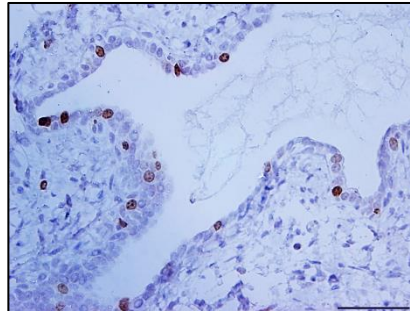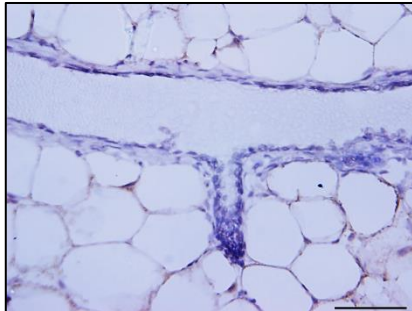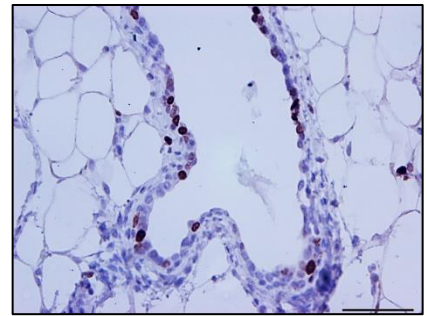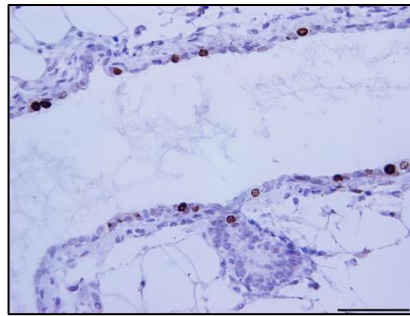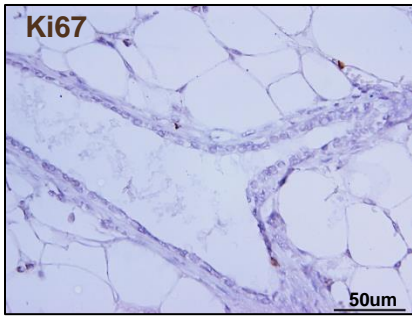
SUPPLEMENTAL FIGURE 4

SUPPLEMENTAL FIGURE 5

A

3%

■ Progesterone-regulated genes in the mammary gland 24h after P exposure (genes = 566)

■ Prolactin-induced genes in the mammary gland (genes = 199)

■ Overlap (genes = 26)

B

Elf5

NS
P = .083

NS
P = .026
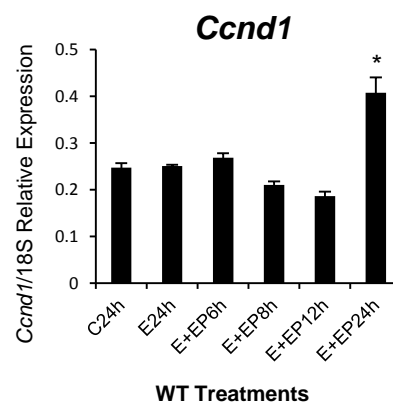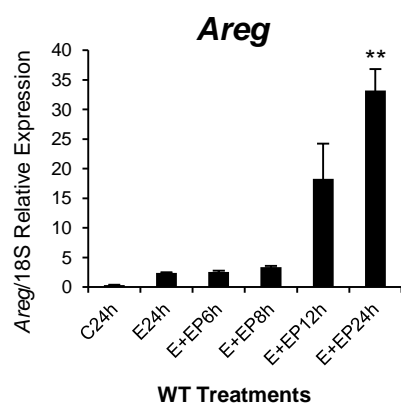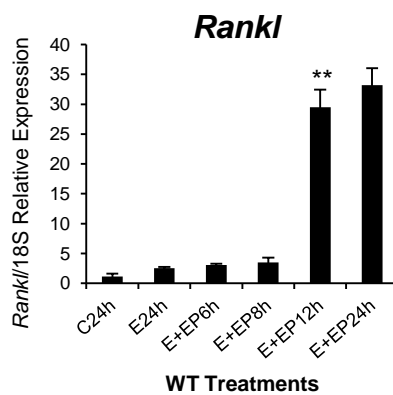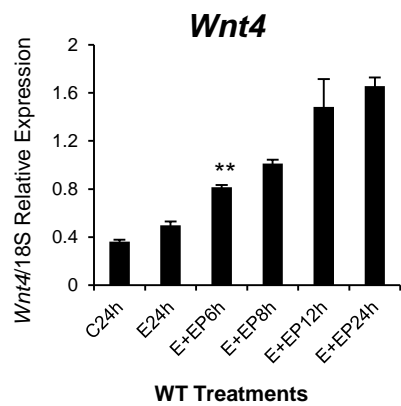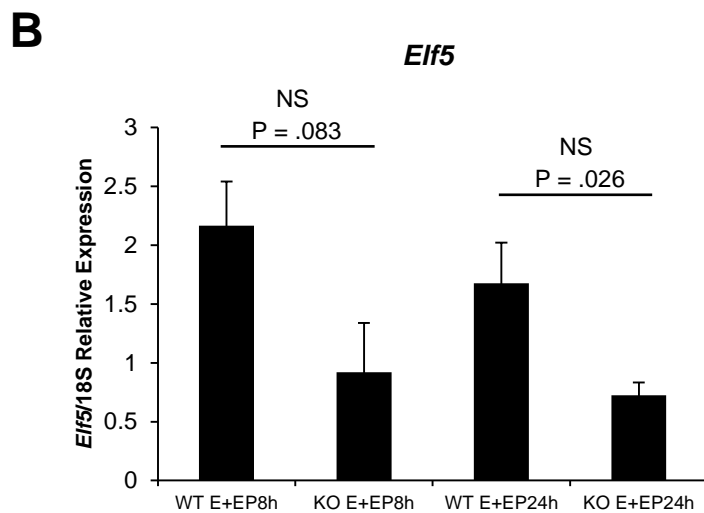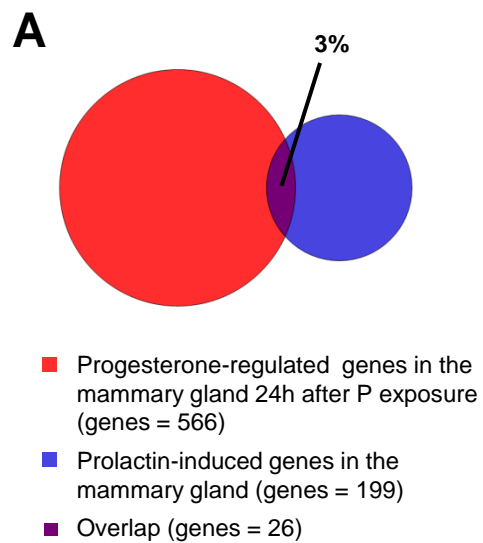
SUPPLEMENTAL FIGURE 6
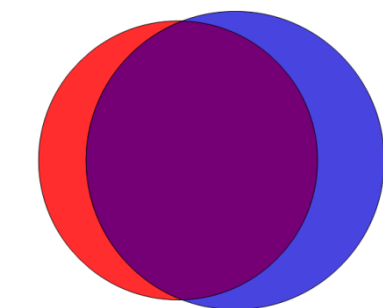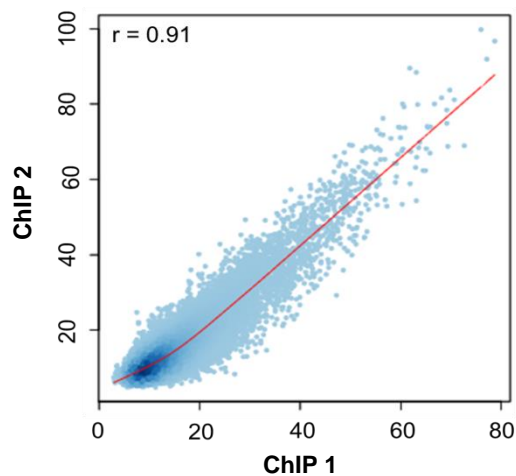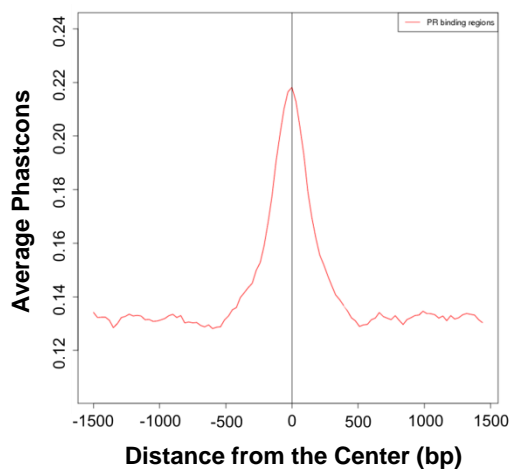
A

Replicate Overlap

- ChIP 1 only (locations = 6311)
- ChIP 2 only (locations = 10813)
- ChIP 1 and ChIP 2
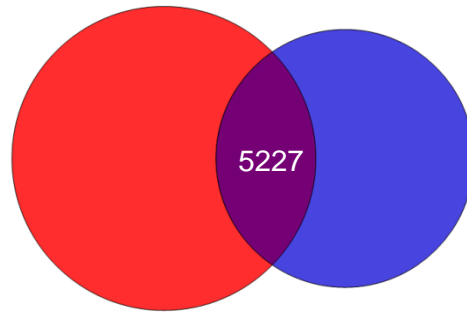 (shared locations = 25178)

B

Replicate Correlation

r = 0.91

ChIP 2

ChIP 1

C

Sequence Conservation

PR binding regions

Average Phastcons

Distance from the Center (bp)

SUPPLEMENTAL FIGURE 7

A
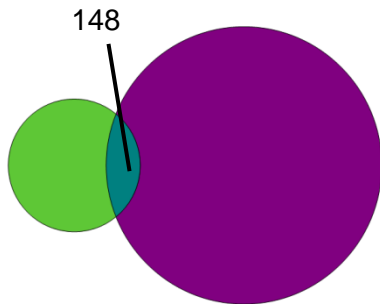
Mammary gland and uterus ChIP-seq overlap

5227

■ PR binding sites only in the mammary gland (locations = 20371)

■ PR binding sites only in the uterus (locations = 13206)

■ PR binding sites in the mammary gland and uterus (locations =5227)
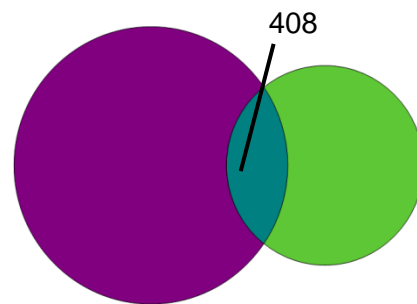
5227 binding locations = 3675 unique genes

B

Mammary Gland

148

■ P-regulated genes in the mammary gland (locations = 695)

■ PR binding sites in the mammary gland and uterus (locations = 3527)

■ P-regulated genes in the mammary gland that recruit PR (locations = 148)

Uterus

408

■ P-regulated genes in the uterus (locations = 1495)

■ PR binding sites in the mammary gland and uterus (locations = 3267)

■ P-regulated genes in the uterus that recruit PR (locations = 408)

SUPPLEMENTAL FIGURE 8