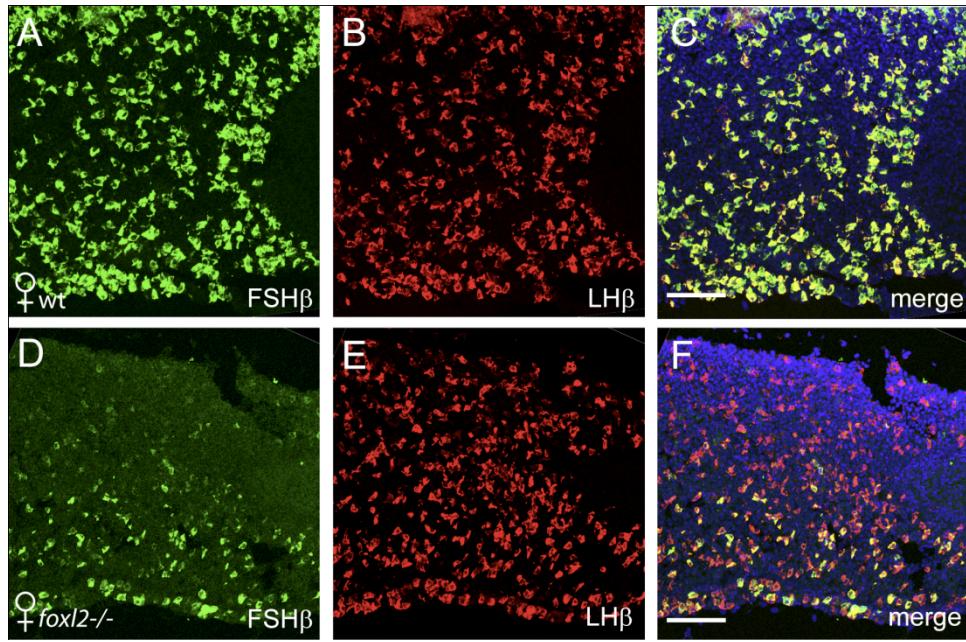


## Supplemental Figure S1



**Supplemental Fig. S1.** Expression of FSH $\beta$  in the *Foxl2* mutant female pituitary.

Confocal micrographs of pituitary sections from 3-week old female wildtype (A, B, C) or *FoxL2* mutant (D, E, F) animals stained for FSH $\beta$  (A, D), LH $\beta$  (B, E) or the merged images (C, F). In the wildtype, FSH $\beta$  positive cells (A) are found throughout the anterior lobe. In the *FoxL2* mutant animals, the number of FSH $\beta$  positive cells (D) is dramatically reduced. In contrast to the *Foxl2* mutant male pituitary where the remaining FSH $\beta$  positive cells are clustered in the ventral medial quadrant, the remaining FSH $\beta$  cells in the *Foxl2* mutant female are found spread out over the ventral half of the anterior lobe. Staining for LH $\beta$  (B, E) reveals gonadotropes throughout the anterior lobe of both the wildtype (B) and *FoxL2* mutant (E), indicating that gonadotropes are appropriately specified in the *FoxL2* mutant. (C, F) The merged images show that whereas the coincidence of FSH $\beta$  and LH $\beta$  is complete in the wildtype, in the *FoxL2* mutant, most gonadotropes fail to express FSH $\beta$ . Scale bar = 100 $\mu$ m.

**Supplemental Table S1.** Summary of cell count data for pituitaries of wildtype (wt) or *Foxl2* mutant (*Foxl2*-/-) male mice.

1	2	3	4	5	6	7	8
Pituitary Hormone	wt number	<i>Foxl2</i> -/- number	wt cell number/mm <sup>2</sup>	wt St. Err.	<i>Foxl2</i> -/- cell number/mm <sup>2</sup>	<i>Foxl2</i> -/- St. Err.	p value
<b>αGSU</b>	2 (2,0)	3 (3,0)	6500	2488	7716	2081	n.s.
<b>FSHβ</b>	4 (3,1)	5 (4,1)	3383	492	1083	221	0.003
<b>LHβ</b>	4 (2,2)	3 (2,1)	4676	1136	5938	1554	n.s.
<b>TSHβ</b>	3 (2,1)	2 (1,1)	3346	926	3130	1860	n.s.
<b>PRL</b>	2 (2,0)	2 (2,0)	4246	452	4759	289	n.s.
<b>GH</b>	2 (2,0)	2 (2,0)	5547	451	7851	522	0.05
<b>ACTH</b>	4 (4,0)	3 (3,0)	2796	501	4733	196	0.01

Column 1 lists the pituitary hormones evaluated by immunohistochemistry. Columns 2 and 3 list the total number of wildtype or *Foxl2* mutant animals used for cell counting; the number in parentheses indicate numbers of 3-week or 5-week old animals (3-week, 5-week). Columns 4 and 5 show the calculated numbers and the standard errors, respectively, of cells per square millimeter of pituitary sections (20 microns) of the wildtype animals. Columns 6 and 7 show the calculated numbers and the standard errors, respectively, of cells per square millimeter of the sections from *Foxl2* mutant animals. Column 8 shows the calculated p value to determine statistical significance (n.s.=not significant, p>0.05).

**Supplemental Table S2.** List of primers for qRT-PCR analyses.

	Forward Primers	Reverse Primers
<i>Fshb</i>	5'-gtgcggctactgtacact	5'-caggcaatcttacggctcg
<i>Lhb</i>	5'-ctcagccagtgtcacctac	5'-gaaaggagactatgggtctac
<i>Cga</i>	5'-tccctcaaaaagtccagagc	5'-gaagagaatgaagaatgcaggaa
<i>Tshb</i>	5'-aagagctgggttgtcaaa	5'-acaagcaagagcaaaaagcac
<i>Gh</i>	5'-gcttggcaatggctacaga	5'-gaaaaagcaactgcctcctg
<i>Prl</i>	5'-tgtccccagcagtccatt	5'-cagcaacaggaggagtgtcc
<i>Pomc</i>	5'-cagtgccaggacccacc	5'-cagcgagaggtcgagttg
<i>Gnhr</i>	5'-agcaacagcaagctgaaca	5'-ccagagccgtctgcttagta
<i>Fst</i>	5'-cctatgagggaaagtgtatcacaa	5'-tggaatcccataggcatttt
<i>Acvr1b</i>	5'-agagggtggggaccaaac	5'-tgcttcatgttattgtctcg
<i>Acvr2a</i>	5'-ccctcctgtacttgttctactca	5'-gcaatgggtcaaccctagt
<i>Acvr2b</i>	5'-tggctgtcggttgagc	5'-ggccatgtaccgtctggt
<i>Smad2</i>	5'-aggacggtagatgagcttgag	5'-gtccccaaatttcagagcaa
<i>Smad3</i>	5'-tcaagaagacggggcagtt	5'-agccgaccatccagtgac
<i>Gapdh</i>	5'-atggtaaggtcggtgtga	5'-aatctccacttgccactgc