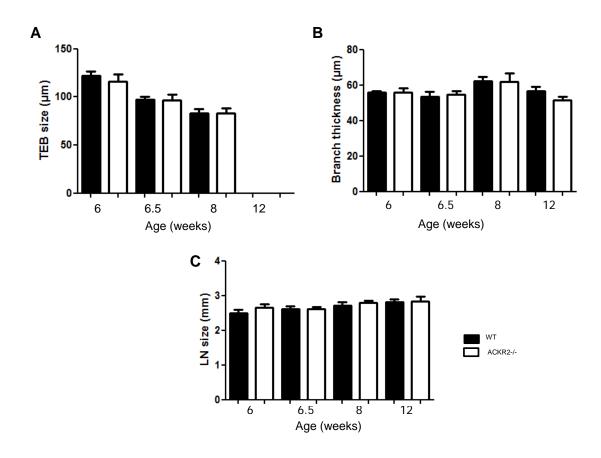
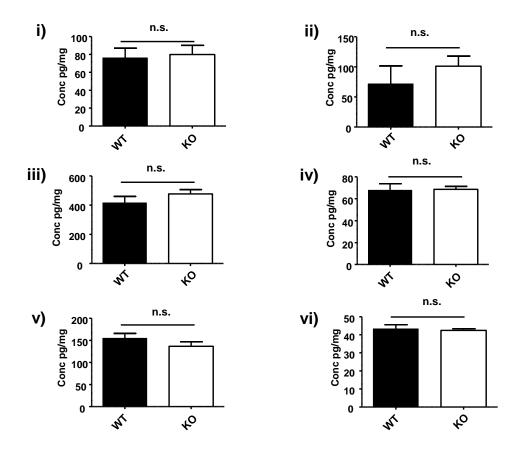
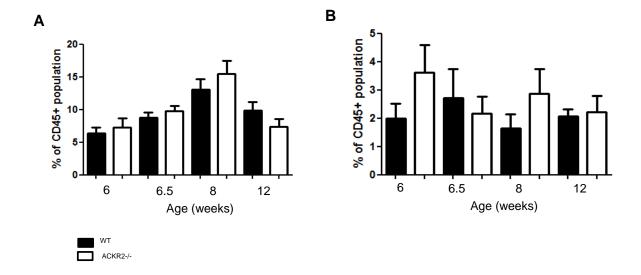
Supplemental Information



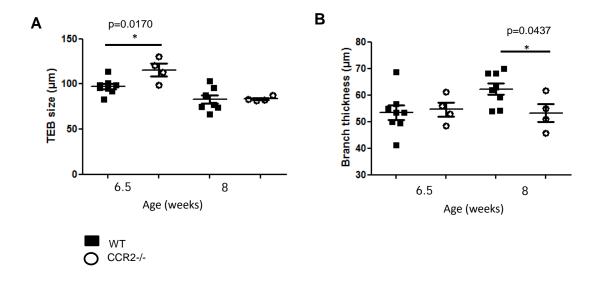
Supplemental Figure S1: ACKR2 does not control morphology of branches or TEBs within the developing mammary gland. Carmine alum whole-mounts were obtained from virgin WT and ACKR2-/- mammary glands at 6 (n=6), 6.5 (n=8), 8 (n=8) and 12 weeks old (n=9). a) The average width of all the TEBs from 3 F.O.V (5x) per gland was measured. b) Branch thickness was the average from 3 measurements from 3 F.O.V (5x) per gland, and c) the length of the inguinal LN was measured using ImageJ. Where there is no indication results were not statistically significant. Error bars represent S.E.M.



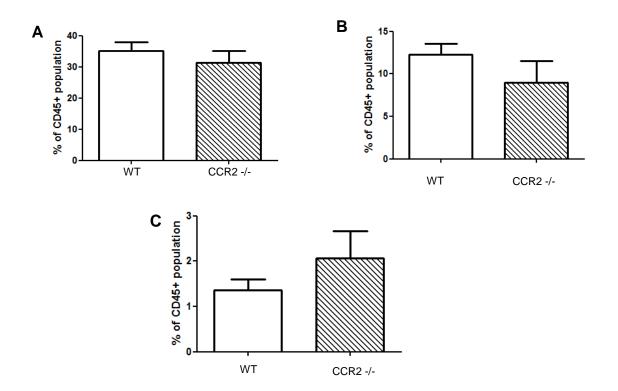
Supplemental Figure S2: Chemokine and cytokine expression in WT and ACKR2-/- mammary glands. Multiplexing was used to assess levels of i) CCL2; ii) CCL4; iii) CCL5; iv) CCL22; v) CXCL1 and vi) CSF-1. n=8 per group n.s. = not significant.



Supplemental Figure S3: ACKR2 does not control recruitment of a) eosinophils or b) mast cells to the developing mammary gland. FACS analysis of WT and ACKR2-/- deficient glands was carried out at 6, 6.5, 8 and 12 weeks. Results were combined from 2 independent experiments, n=8 to 11. Where there is no indication results were not statistically significant. Error bars represent S.E.M.



Supplemental Figure S4: CCR2-/- mice display mild alterations in branch morphology. Whole-mount images were analysed for a) TEB size and b) branch thickness at 6.5 and 8 weeks. WT data points are the same as those used in Supplementary Figure 1. Where there is no indication results were not statistically significant. Error bars represent S.E.M.



Supplemental Figure S5: CCR2 does not control recruitment of a) macrophages, b) eosinophils, or c) mast cells to the developing mammary gland at 8 weeks. Results were combined from 2 independent experiments, n=9 to 11. Where there is no indication results were not statistically significant. Error bars represent S.E.M.

Supplemental Table S1: Differential gene regulation in pubertal ACKR2 deficient mice

6.5 week Upregulated (>2 fold)	6.5 week Downregulated (>-2 fold)	8 week Upregulated (>2 fold)	8 week Downregulated (>-2 fold)
CCR5	CCR4	CCL1	CXCR5
CCL24	CXCR2	ACKR4	CCR1I1
CCL9	CXCL3	Cmtm6	CCR2
CCR1	IL-6	CCL22	CXCR2
CXCL4 (PF4)	Cmtm2a	Cmtm5	Cmtm2a
CXCL7 (PPBP)		TYMP	CCR8
IL-4			CCR7
CCL3			CCR4
XCR1			
CCR1I1			
CCR6			
CCR3			
CCR8	and different control of	tion = 40.05	

Red type denotes significantly different gene regulation, p<0.05