Twin status of prepubertal ewe lambs may impact mammary gland growth and development: role of aromatase?

Author: Jaimie Watts

Major: Animal Science

Project Advisor: Kristy Daniels

Previous data from our lab suggests that presence of testosterone in the intrauterine environment due to the presence of a ram twin may latently affect mammary growth and composition in female co-twins. Ewe lambs that were gestated with males (FM) had mammary fat pads (MFP) that were almost 30% larger and contained more total lipid than their female-female twin (FF) counterparts. Testosterone is not implicated in mammary development but estrogen is. Aromatase is a rate-limiting enzyme involved in the conversion of testosterone to estrogen. Aromatase has been found at the gene and protein level in MFP of Holstein heifers, but to our knowledge, has not been studied in sheep. We hypothesized that aromatase mRNA and its mature protein product would be found in MFP of all lambs analyzed. We further hypothesized that aromatase mRNA and protein abundance would be different in FF twins compared to FM twins. The sample set used consisted of 27 ewe lambs born to first parity ewes (8 sets of FF twins (n=16) and 11 ewe lambs with male co-twins). Ewe lambs averaged 130 days of age and 46 kg at sample collection; these variables were unaffected by twin status ($P = 0.319$ and $P = 0.569$, respectively). Paraffin-embedded samples of mammary tissue were subjected to immunohistochemical staining for aromatase protein. Immunohistochemistry revealed staining patterns for aromatase that did not fully support our initial hypotheses. Aromatase protein was observed in MFP as we thought, but aromatase localization to mammary epithelial cells (MEC) exceeded that of cells which comprise MFP (mainly adipocytes and fibroblasts). For this reason, we investigated aromatase gene expression in mammary parenchyma only. Parenchyma is heterogeneous tissue but contains roughly 20% MEC. Results from quantitative reverse transcription-PCR showed no difference in relative abundance of aromatase due to twin-status ($P = 0.267$). Immunohistochemistry data have yet to be analyzed but similar results are expected. While our preliminary results do not fully support our main hypotheses, we were able to demonstrate aromatase at both the gene and protein levels in ewe lambs, which is a novel finding. This perhaps suggests that aromatase is important for prepubertal mammary growth, though not in the manner we originally hypothesized.