

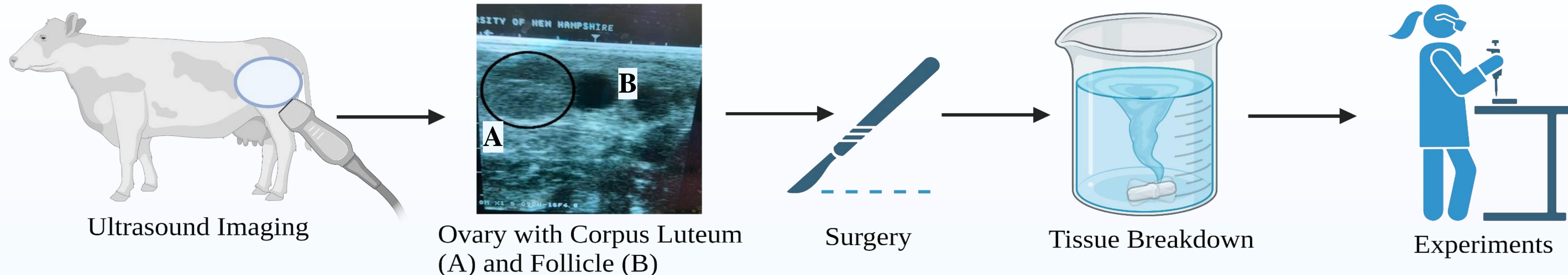


Dairy Barn to Lab Bench: Imaging Ovaries to Analyzing Cell Function



¹Quinn Poole, ²Dean Elder, ¹Paul Tsang

¹Department of Molecular, Cellular, and Biomedical Sciences, and ²Animal Resource Office, University of New Hampshire



Background & Methods

Results

Summary & Future Work



Figure 1. Portable Ultrasound

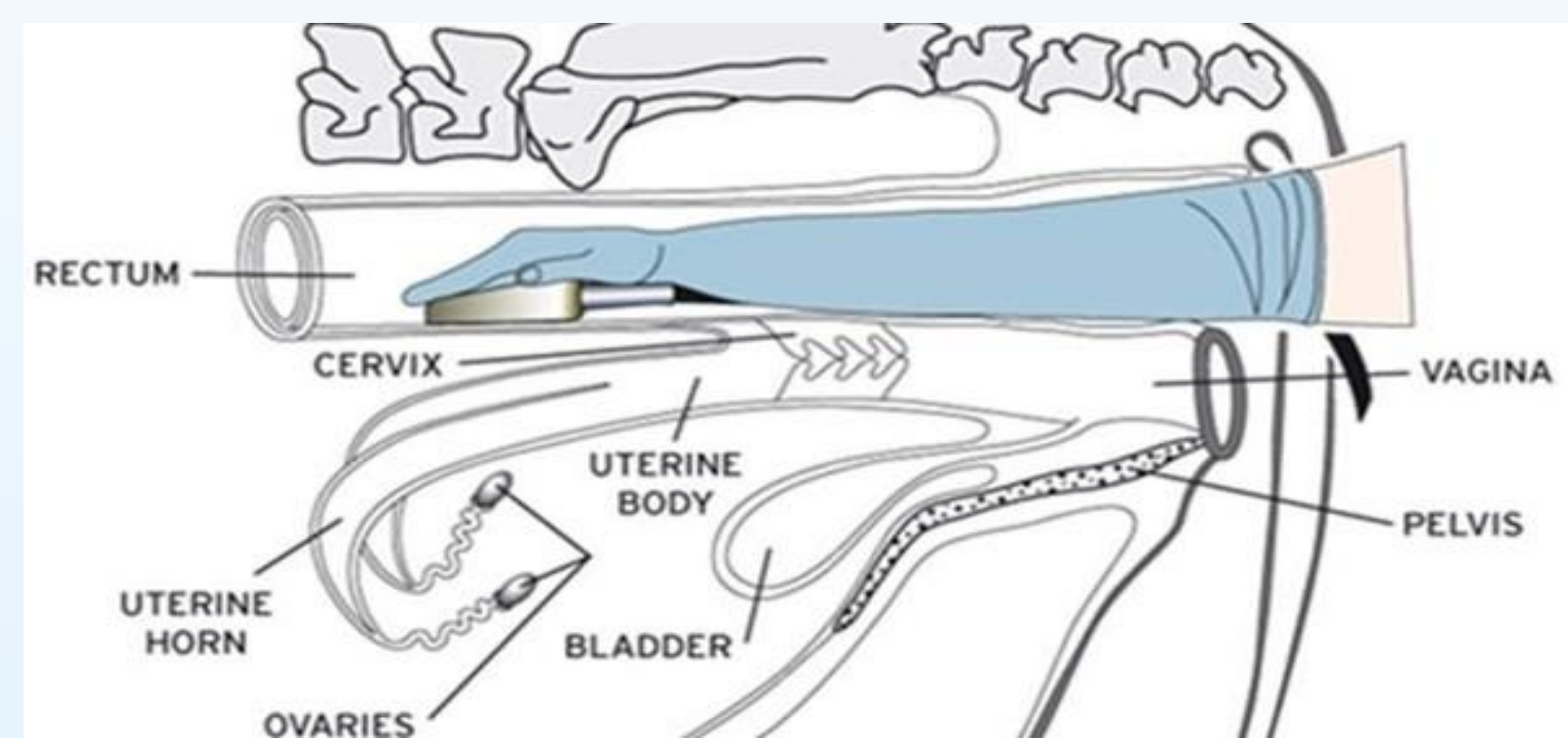


Figure 2. Ultrasound Imaging Technique

Objective: Use ultrasound imaging to identify and localize the corpus luteum to facilitate surgical removal for cell culture experiments

Ultrasound Imaging (Figs 1-4)

- Transducer emits sound waves from crystals and produces black-grey-white images based on density.
- Solid structures (bones) are white while fluid is black.

What is the Corpus Luteum?

- Corpus luteum produces progesterone and regulates fertility in animals.

Surgical Removal of the Corpus Luteum and Isolation of Cells in the Lab

- Enzymes were used to break down tissue and isolate cells from the corpus luteum.

Same-day Incubation of Isolated Luteal Cells

- Cells were incubated for 2 hours and treated with or without fetal bovine serum (FBS).
- Total RNA was extracted from cells and prepared for polymerase chain reaction (PCR).

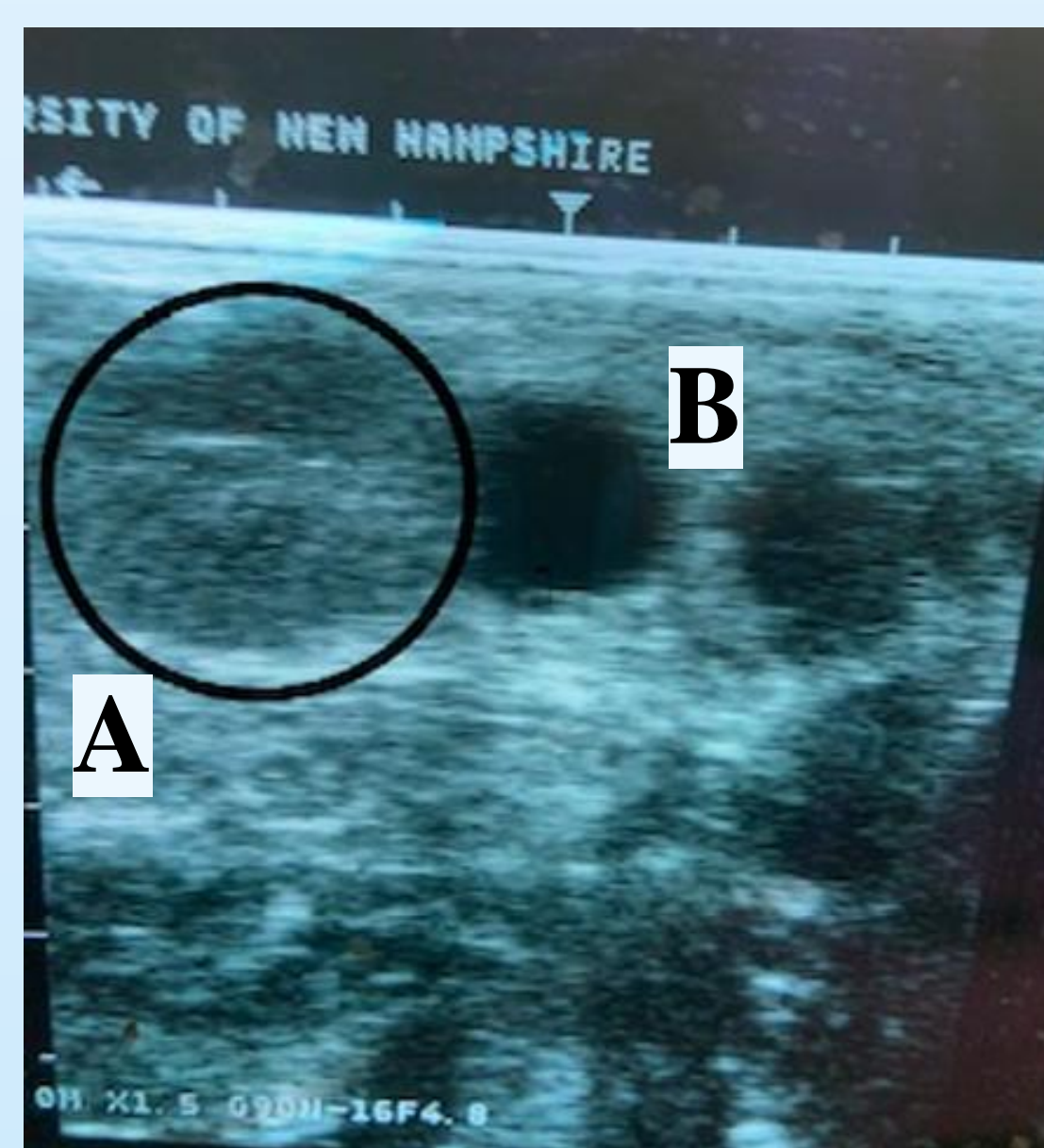


Figure 3. Ultrasound image of ovary, showing corpus luteum (A) and follicle (B)

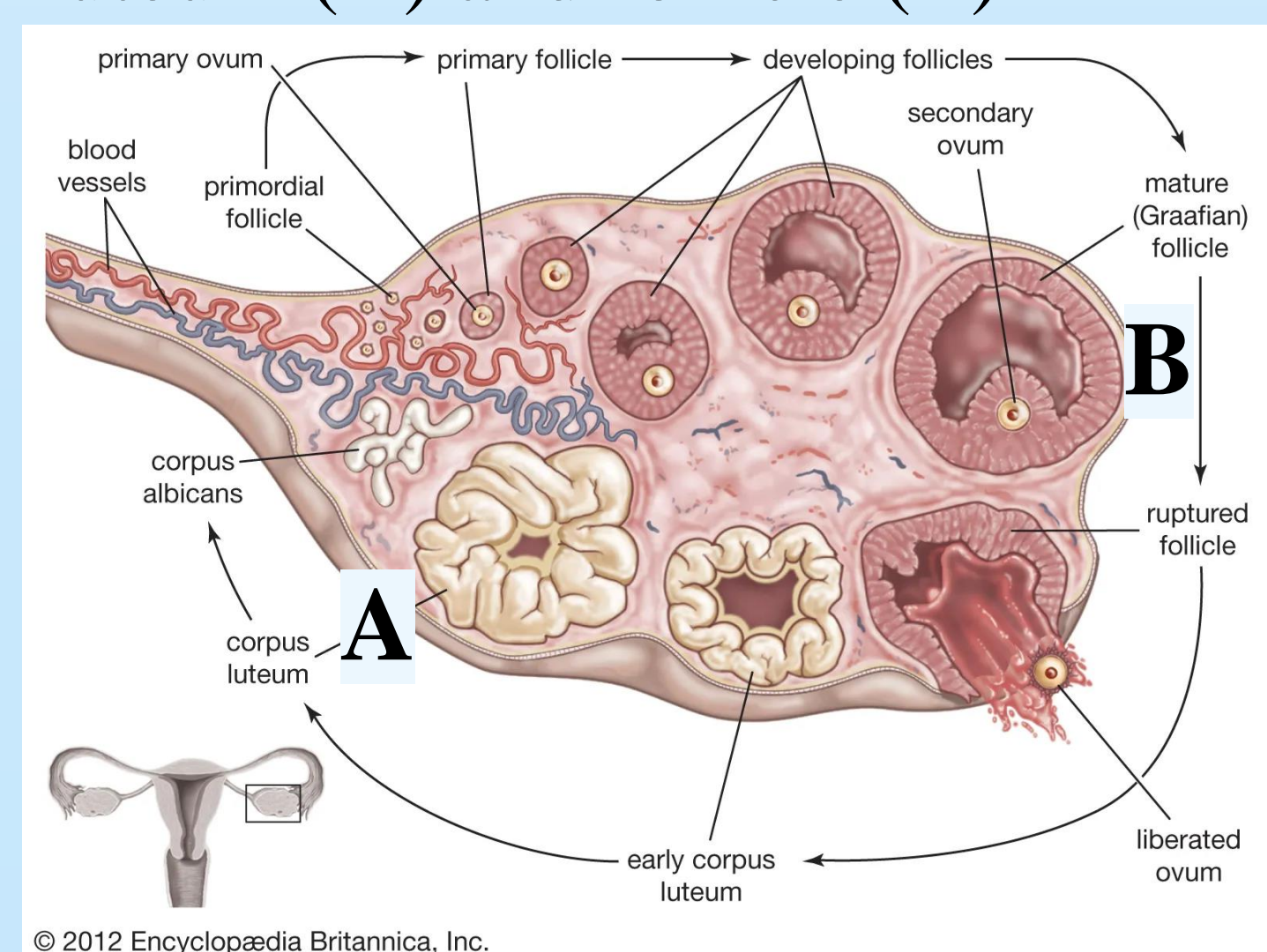


Figure 4. The Ovary

RNA Concentration vs. Cell Number

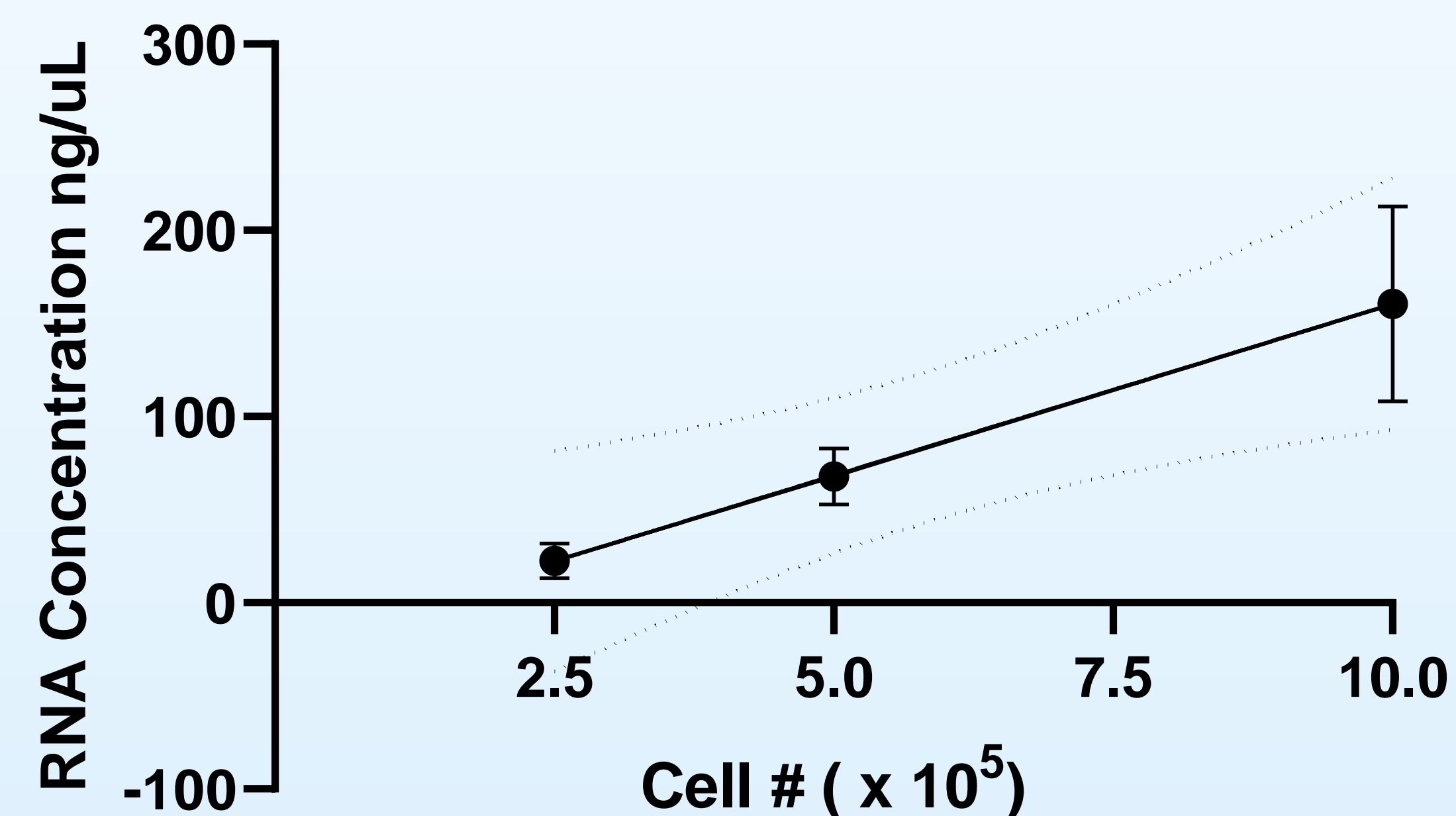


Figure 5. Relationship between cell number and RNA concentration. Isolated luteal cells (2.5 to 10.0 x 10⁵) were aliquoted into test tubes and incubated for 2 hours at 37°C (n=3). RNA was then extracted using RNeasy Plus kits (Qiagen).

Pilot Experiment

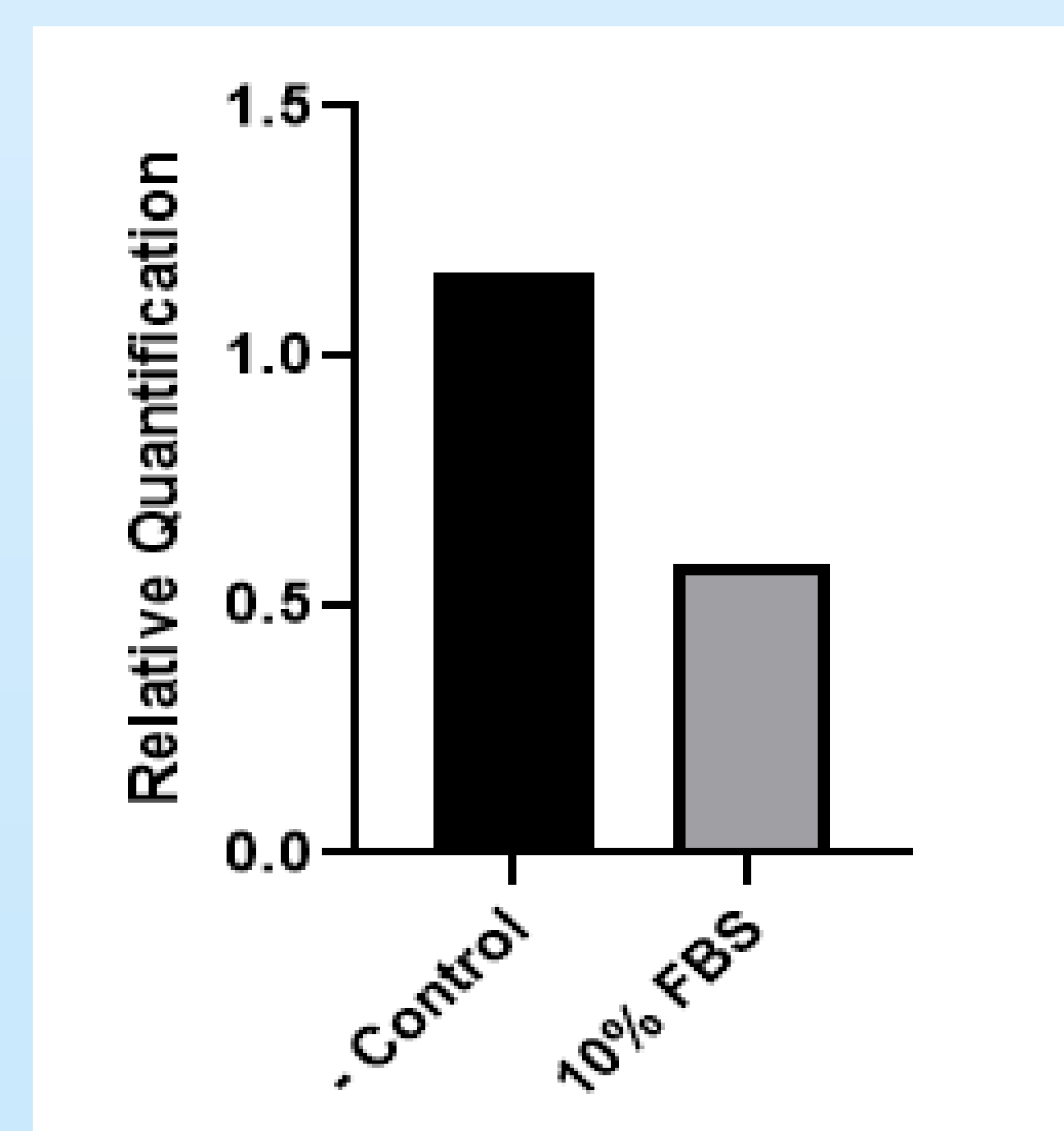


Figure 6. Regulation of cellular communication network factor 1 (CCN1), a builder of blood vessels, by FBS. Luteal cells (5.0 x 10⁵) were treated with 10% FBS for 2 hours. Then, RNA was extracted before performing quantitative PCR for CCN1 expression. Analysis was done using QuantStudio Cloud Connect Utility (ThermoFisher).

Summary:

- Validated the relationship between RNA concentration and cell number ($R^2 = 0.99$) in short term incubations of luteal cells isolated immediately after surgery.
- The pilot experiment suggested that FBS inhibited CCN1, which was the opposite of what we expected. This requires confirmation through more replicate experiments.

Future Work:

- Future analysis of progesterone concentrations in the medium will provide additional information about luteal cell function in short term incubations.

Acknowledgements

Thank you to Dr. Paul Tsang and Dr. Dean Elder for their mentorship, and to Donnelly Hutchings and Bryan Landry for teaching me laboratory techniques. A special thank you to Jon Whitehouse and his staff at the Fairchild Dairy Teaching and Research Center. All animal procedures were conducted in accordance with UNH IACUC approval (#201001) Supported by USDA NIFA Hatch Animal Health Project.

References

1. Battista PJ, Condon WA. Serotonin-induced stimulation of progesterone production by cow luteal cells in vitro. *Journal of Reproduction and Fertility*, 1986; 76(1):231-238. doi: 10.1530/jrf.0.0760231.
2. Berisha, B., and D. Schams. Ovarian function in ruminants. *Domestic Animal Endocrinology*, 2005; 29(2): 305-331, <https://doi.org/10.1016/j.domaniend.2005.02.035>.
3. Reeves, J.J., et al. Transrectal real-time ultrasound scanning of the cow reproductive tract. *Theriogenology*, 1984; 21(3): 485-494, [https://doi.org/10.1016/0093-691x\(84\)90410-2](https://doi.org/10.1016/0093-691x(84)90410-2).