

The Use of Environmental DNA to Monitor the Presence of Feral Hog (*Sus Scrofa*) Populations Within Cattle Communities

Eva Morgan^{*1}, Jason Apple¹, Sejal Bakhati², Rudolf Bohm²

Texas A&M University – Kingsville

¹Department of Animal Science & Veterinary Technology, ²Department of Biological and Earth Sciences

Abstract

Feral swine populations pose severe risk of damage to agricultural commodities each year. Among this damage is the potential for disease transmission within rangeland cattle communities. Methods of monitoring and control are generally costly, time demanding, and labor intensive. The use of environmental DNA (eDNA) to monitor species populations is gaining popularity due to its ease and quick results but lacks research within nonaquatic species and the interaction between other biotic factors such as the presence of another animal's DNA within the sample. This study will determine how the presence of cattle manure will degrade feral swine eDNA in samples where both are present. In a lab setting, researchers mixed domestic hog and cattle manure at 5 different concentrations, including positive and negative controls, and tested each at 0 hours and every 14 days for a 42-day period. Researchers utilized conventional polymerase chain reaction (cPCR) testing to provide positive or negative results for the detection of hog DNA. Samples will mimic what may be taken in natural bodies of water, therefore providing results regarding when samples should be taken for the best possibility of successful monitoring. Preliminary results support that the only significant relationship is that between hog DNA concentration and the PCR response. This relationship indicates that when extended to a 56-day sample period, the concentration containing the lowest amount of swine DNA may be undetectable. The results of this experiment may be immediately applied to real life scenarios and benefit the future of feral hog monitoring.

Introduction

Research that focuses on the effects of cattle's presence on accurate testing for feral swine populations will be vital for producers looking for cost effective and timely testing. The goal of this study is to thoroughly investigate using eDNA as a monitoring tool for feral swine in cattle communities by monitoring feral swine eDNA degradation when cattle manure is present. Samples with varied concentrations will be in a controlled environment and tested at different intervals throughout each trial.

Methods

- To best mimic a natural aqueous solution, water from a stock tank used for small ruminants was collected and tested to ensure no contamination.
- The cattle and hog manure were mixed into five 20 g concentrations. Concentrations were numbered for labeling purposes.
- Researchers homogenized samples and began DNA extraction directly without any preservation method, using the DNeasy mericon Food Kit Standard Protocol (2g) (Qiagen). Researchers used *Sus Scrofa* primers (Integrated DNA Technologies) previously successful in similar published research (Williams et al., 2017). Each cPCR reaction was a 25 µl reaction.
- The thermal cycling program optimized for sample amplification involved 30s at 94°C; 34 cycles of 30s at 94°C, 30s at 55°C, 1 min at 68°C; and the final extension time of 5 min at 68°C.
- Samples were then run in an agarose gel electrophoresis for two hours and viewed under UV light.

Results

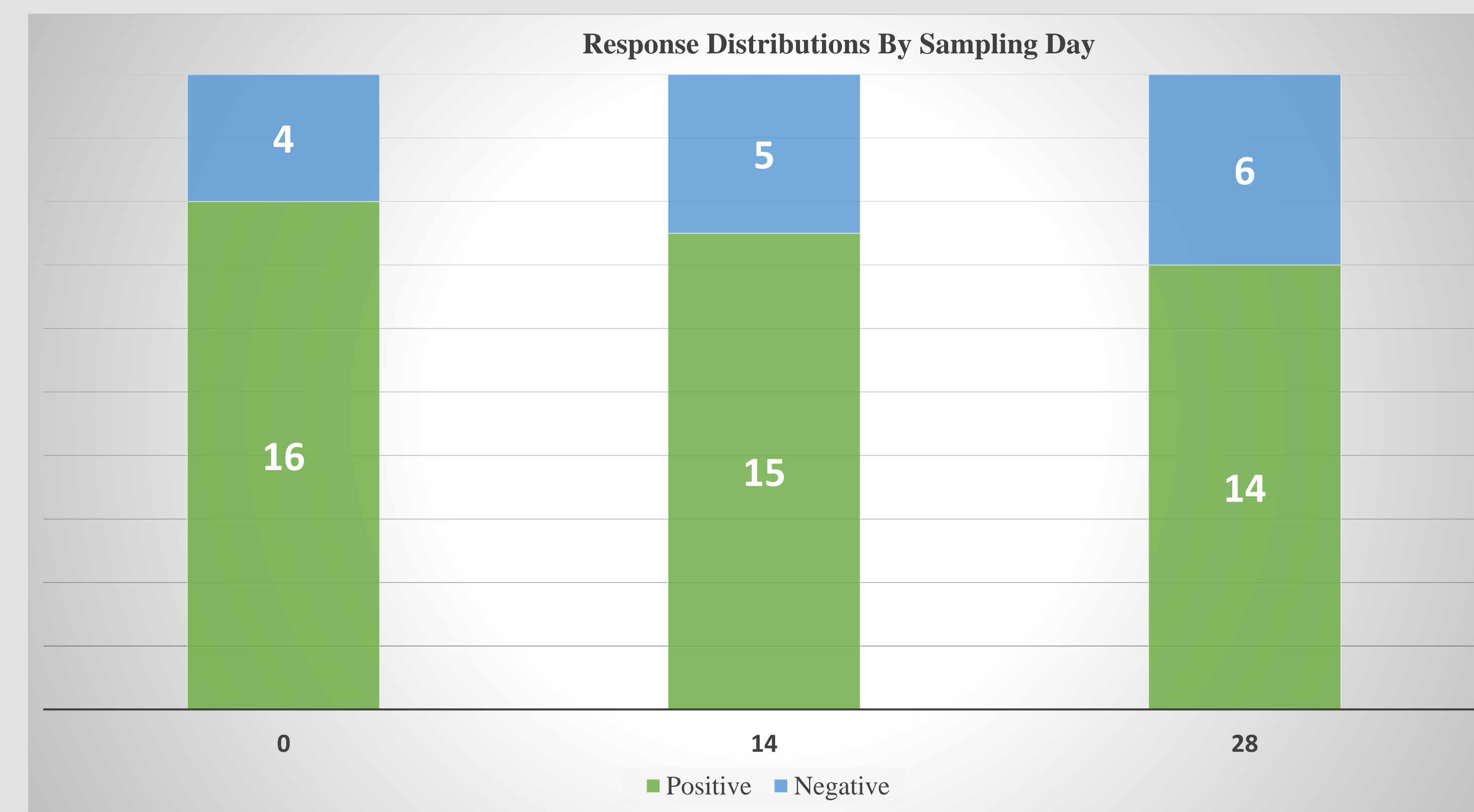
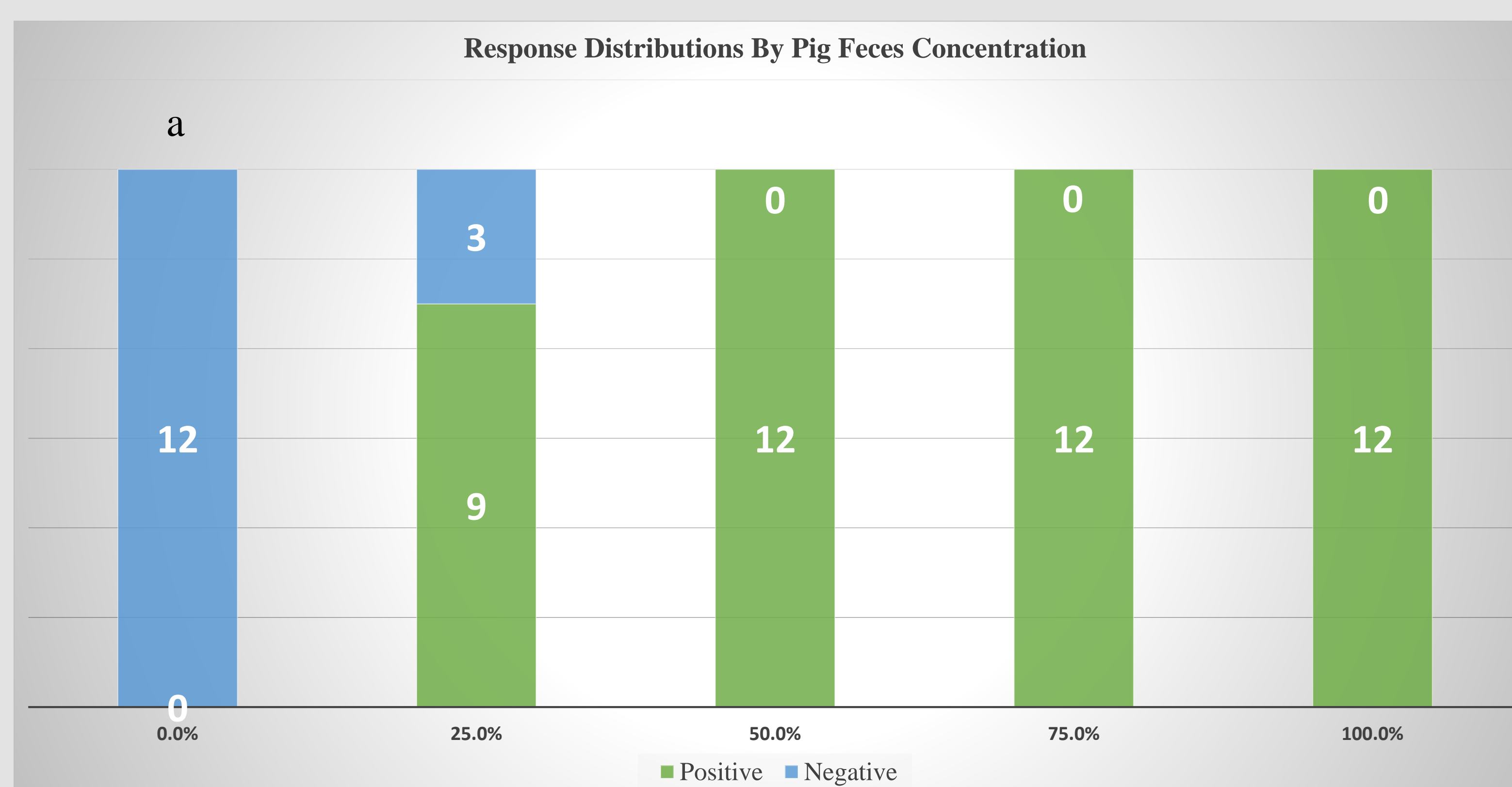
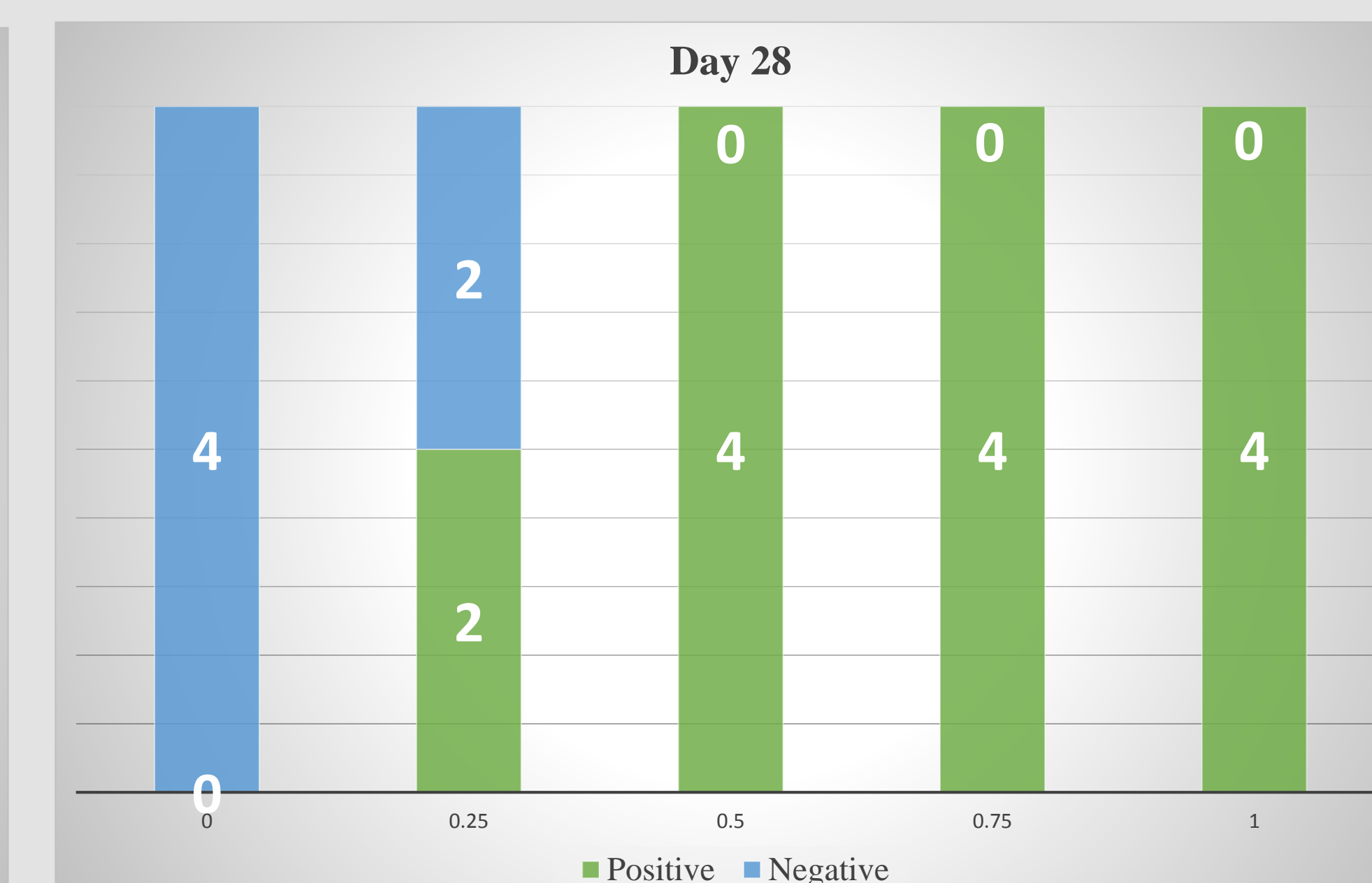
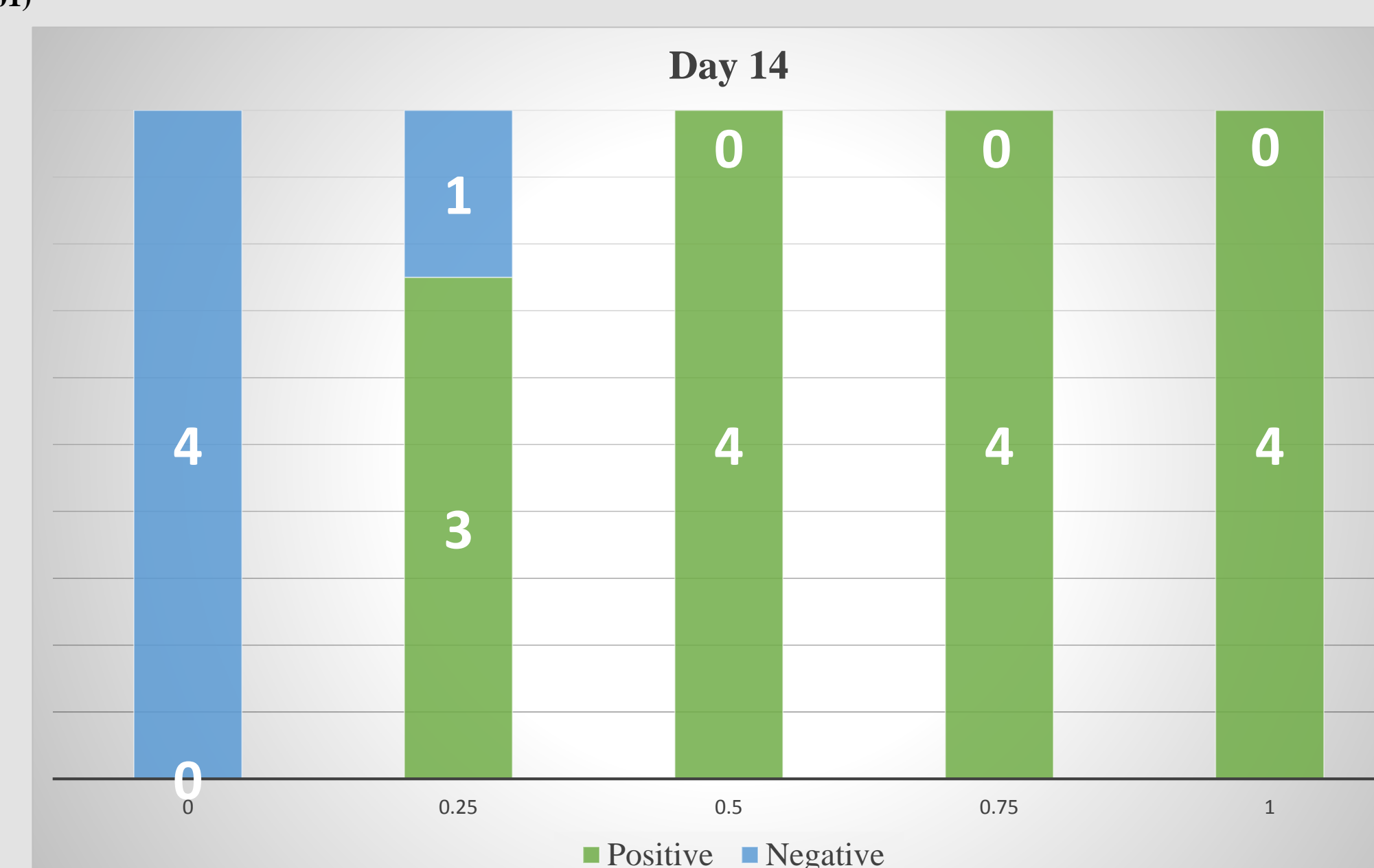
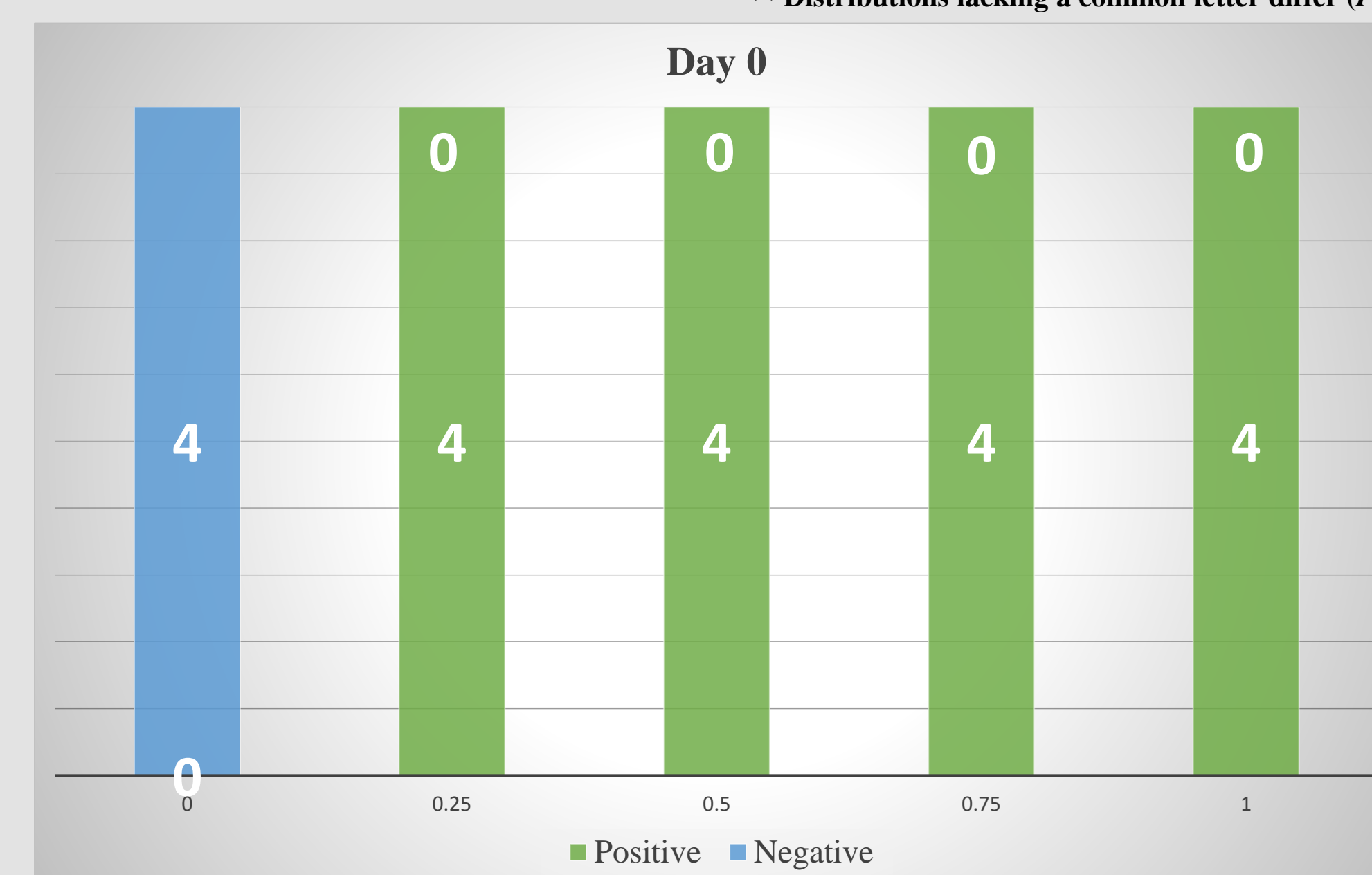


Table 1. $\chi^2 = 48.0 (P < 0.0001)$
*a,b,c Distributions lacking a common letter differ ($P < 0.01$)

Table 2. $\chi^2 = 0.53 (P = 0.7659)$



Tables 3a,b,c. $\chi^2 = 2.67 (P = 0.2636)$

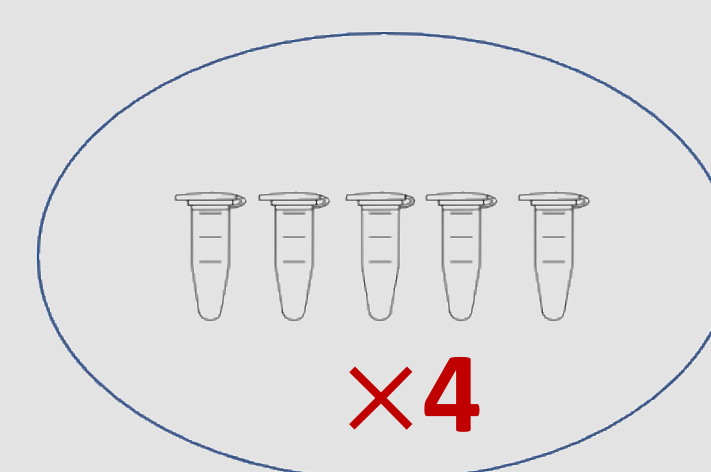
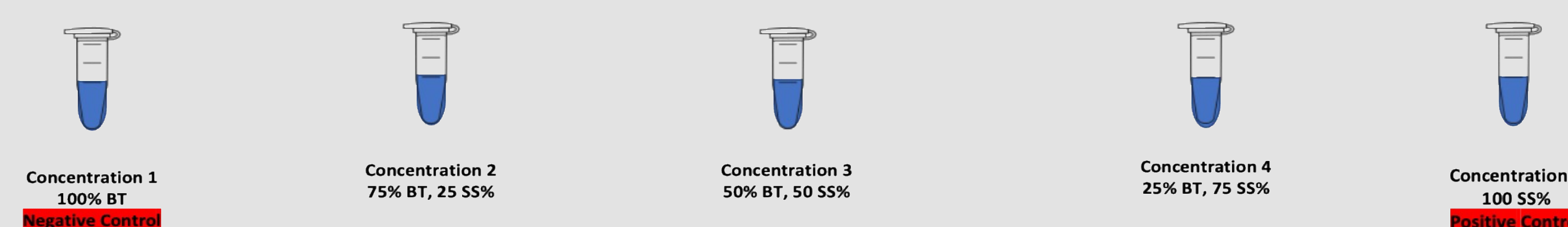


Figure 1. Concentrations used in experiment, for each sample set, each concentration was replicated four times and contained positive and negative controls to account for any mistakes made during the experiment.

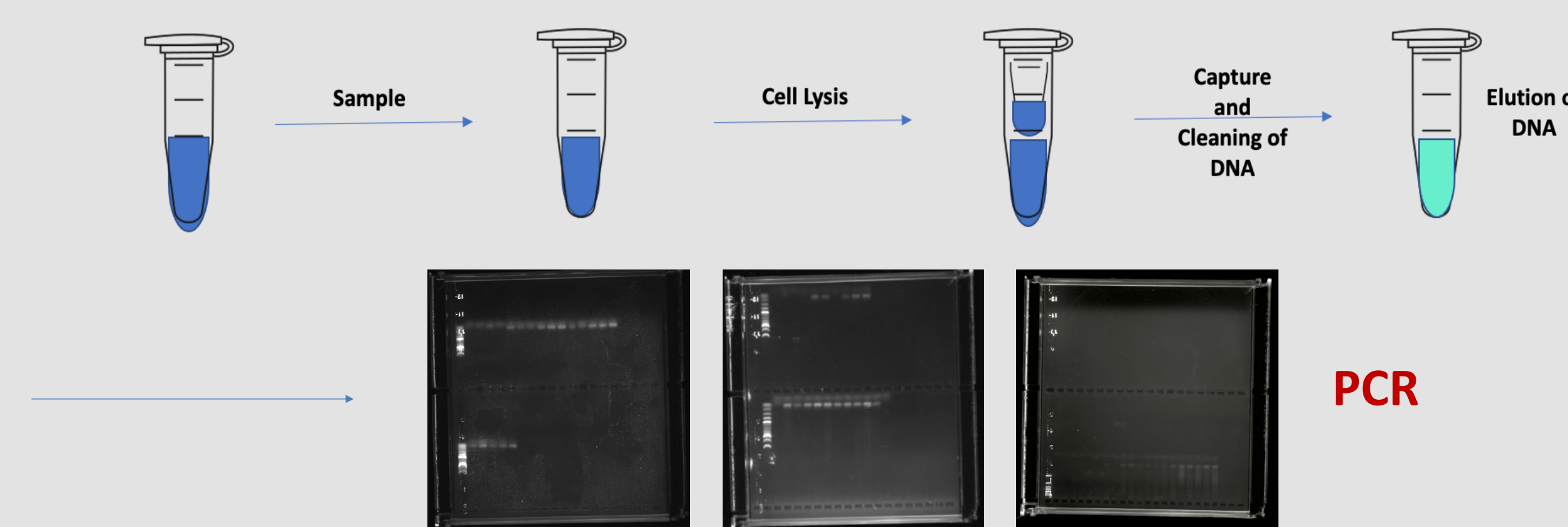


Figure 2. DNA Extraction protocol using the DNeasy mericon Food Kit Standard Protocol (2g) (Qiagen), PCR Sample Date A,B,C.

Conclusion

- Preliminary data indicates that it is highly possible that when extended to the next sample (56 days), the 25% swine DNA / 75% cattle DNA sample will be undetectable.
- The slow decrease in positive detection within the 25% swine DNA / 75% cattle DNA sample indicates that there may be a relationship between the degradation of the swine DNA and the presence of cattle DNA in eDNA sampling.
- With a preliminary experiment such as this, there must be field trials to corroborate results. The next phase of this experiment should include purposely introducing swine eDNA into artificial wallows or ponds on land occupied by cattle and continue similar forms of sampling and testing.

Acknowledgements

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References

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