



Salmonella Quantification (SalQuant®) with Hygiena's BAX® System for Poultry Crops and Lungs

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BAX® System Q7

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INTRODUCTION:

Salmonella is well established in poultry and continues to be a food safety concern for the industry. The organism can be introduced to live poultry through several routes including direct ingestion, contact and even inhalation of infectious particles that can spread throughout many internal organs (1, 2). To prevent further dissemination during slaughter and processing, the pathogen status of the flock should be determined prior to transport. Quantitative data for on-farm sampling can provide the most accurate and reliable information for this decision-making.

PURPOSE:

1. Develop a linear equation for poultry crops and lungs.
2. Verify the BAX® System Real-Time PCR assay for *Salmonella* quantification (SalQuant®) for crops and lungs.

REGISTERED TRADEMARKS:

BAX® is a registered trademark of Hygiena for its line of equipment, reagents and software used to analyze samples for microbial contamination. SalQuant® is a registered trademark of Hygiena.

METHOD:

SAMPLE PREPARATION

Primary enrichments were created for crops and lungs using different volumes of BPW. From this homogenate, 30 mL test portions were aliquoted for inoculation.

INOCULATION

For each matrix, fifteen (15) samples were inoculated with 0.00 – 4.00 Log CFU/mL of a cold stressed *Salmonella* Typhimurium to create three biological replicates per Log level. One sample was left uninoculated for a negative control.

ENRICHMENT

Immediately following inoculation, samples were combined with equal volumes of pre-warmed (45 °C) BAX MP media with Quant™ Solution, incubated at 42 °C for 4-24 hours, and tested by real-time PCR in quintuplet.

At the same time, a 3-tube x 5-dilution MPN was conducted for each inoculation level following the USDA FSIS Appendix 2.05.

RESULTS:

The 6-hour enrichment produced the best linear fit equation for both matrices (Figures 1 & 2).

Crops

- R² of 0.91
- Log RMSE of 0.35

Lungs

- R² of 0.94
- Log RMSE of 0.28

When compared to MPN, real-time PCR results displayed no statistical difference (Figures 3 & 4).

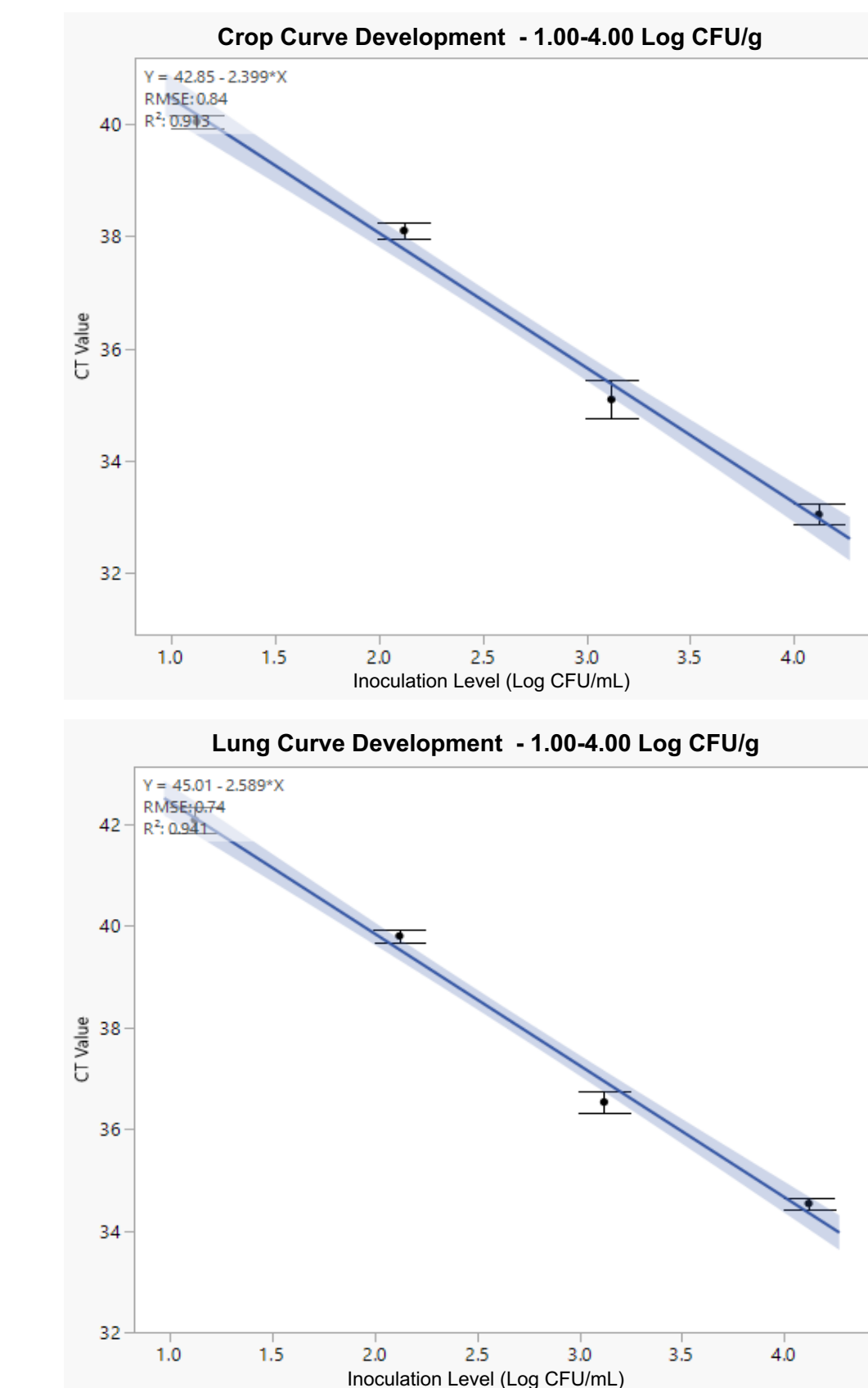


Figure 1 & 2. Mean (*Salmonella* Ct) and *Salmonella* Ct vs. Inoculated Log CFU/mL for Crops (Top) and Lungs (Bottom)

SIGNIFICANCE:

These studies demonstrate accurate and rapid quantification of *Salmonella* with an enumerable range of 1.00 to 4.00 Log CFU/g from poultry crops and lungs using the BAX System.

Processors that utilize these protocols for organs can apply SalQuant as an analytical tool to identify positive flocks and improve interventions during grow out to reduce incoming levels of *Salmonella* to the plant.



FIGURES – MPN VERIFICATION:

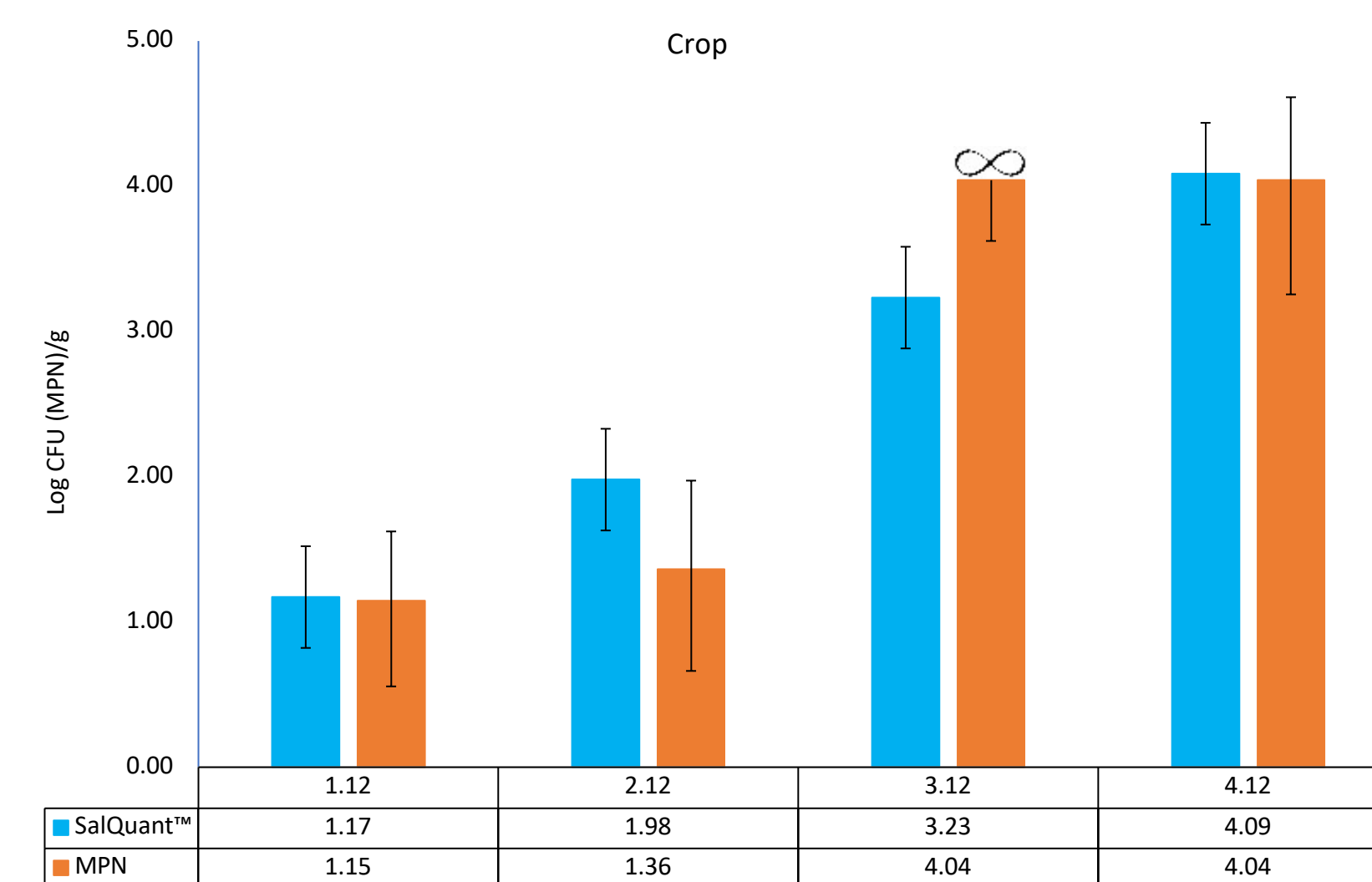


Figure 3 (Left). MPN and SalQuant comparison per inoculation level at 6 hours of enrichment for crops

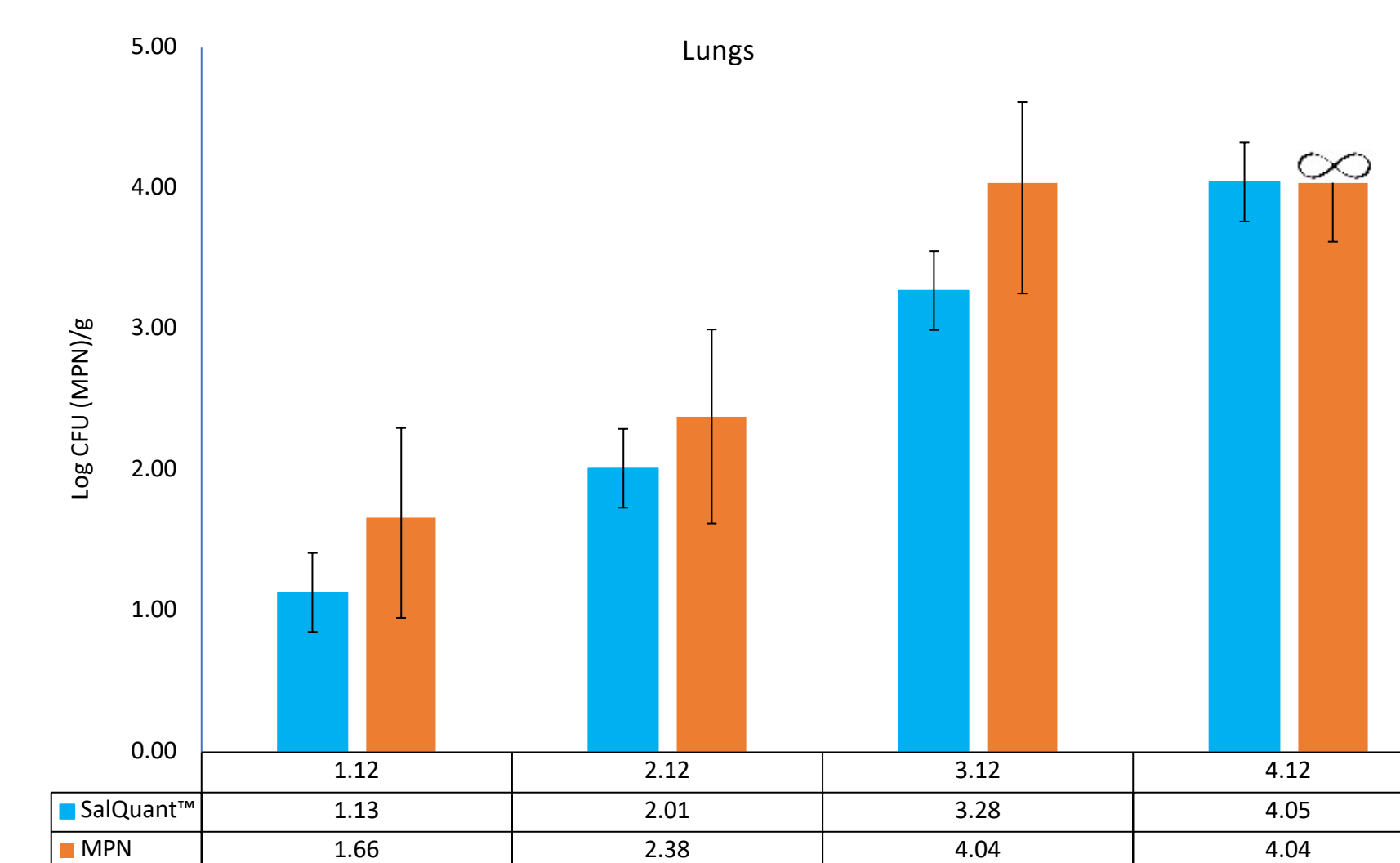


Figure 4 (Right). MPN and SalQuant comparison per inoculation level at 6 hours of enrichment for lungs

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1. Kallapura, G., Hernandez-Velasco, X., Pumford, N. R., Bielke, L. R., Hargis, B. M., and Tellez, G. (2104). Evaluation of respiratory route as a viable portal of entry for *Salmonella* in poultry. *Veterinary Medicine* (Auckland, N.Z.) 5, 59-73.
2. Nayak, R., O'Bryan, C., Kenney, P. B., Crandall, P. G., Ricke, S. 2012. Pre-and post-harvest intervention strategies for controlling *Salmonella* contamination in broiler production. *Salmonella: Classification, Genetics and Disease Outbreaks*. 1-38.