Verification of Poultry Carcass Rinsates with the BAX® System Real-Time PCR Assay for Highly Pathogenic *Salmonella*

Introduction

Poultry is a significant reservoir for *Salmonella* contamination, which can persist on carcasses and be distributed throughout processing. Since rinsates reflect overall carcass contamination, even low levels of *Salmonella* indicate a potential risk for downstream product contamination. *Salmonella* serotypes vary in virulence and their potential to cause human illness; therefore, understanding both the presence and pathogenicity is increasingly important. Detection methods that differentiate levels of pathogenicity can provide poultry processors with actionable insights to prioritize risks, strengthen control strategies and support more informed risk-based food safety decisions.

Sample Preparation and Enrichment

Three *Salmonella* serotypes were selected based on relative pathogenicity: *Salmonella* Typhimurium (High), *Salmonella* Reading (Moderate) and *Salmonella* Kentucky (Low). Each strain was grown overnight at 35 °C in Brain Heart Infusion (BHI) broth. The following day, cultures were serially diluted (10-fold), plated and held at 4 °C until enumeration was complete.

Carcass rinses were collected from a commercial poultry processing facility and divided into 30 mL test portions. Each portion was inoculated with a single *Salmonella* culture at two levels of contamination: 6 low-level replicates expected to yield fractional positive results and 6 high-level replicates expected to yield all positive results. This was repeated for each serotype. Three uninoculated samples were included for negative controls. All samples were equilibrated at 4 °C for 48 hours to acclimate the target organism in the matrix.

Samples were then enriched with 30 mL of pre-warmed (35 °C) Buffered Peptone Water (BPW), homogenized and incubated at 35 °C for 18-24 hours. Sample aliquots were removed at 18, 20 and 24 hours and analyzed using the BAX® System method.

Method

BAX System Method
All samples were processed following the procedures described in the Product Instructions for the BAX System Real-Time PCR Assay for Highly Pathogenic *Salmonella* (KIT2048).

Reference Method
All samples were confirmed by culture, regardless of BAX System results, following the USDA FSIS MLG 4.15.

Results

At the low inoculation level, the BAX System Real-Time PCR assay for HPS returned positive results for *Salmonella* Typhimurium in 6/6 samples, *Salmonella* Reading in 4/6 samples and *Salmonella* Kentucky in 5/6 samples, for a total of 15/18 positive samples. At the high inoculation level, all 6 samples were positive for each of the three *Salmonella* serotypes for a total of 18/18 positive samples. The expected high, moderate and low (H, M, L) designations were correctly assigned. All uninoculated controls returned negative results. Compared to culture, there was 100% agreement with no false positives or false negatives.

Table 1. BAX System Method Presumptive vs. Confirmed Results

Poultry Rinsate – Highly Pathogenic *Salmonella*						
Inoculating Strain	**CFU/Test Portion**	**N**	**BAX System Presumptive**			**Confirmation**
			18 h	**20 h**	**24 h**	
N/A	Control	3	0	0	0	0
Salmonella Typhimurium	0.46	6	6 (H)	6 (H)	6 (H)	6 (H)
	2.75	6	6 (H)	6 (H)	6 (H)	6 (H)
Salmonella Reading	0.64	6	4 (M)	4 (M)	4 (M)	4 (M)
	1.10	6	6 (M)	6 (M)	6 (M)	6 (M)
Salmonella Kentucky	0.54	6	5 (L)	5 (L)	5 (L)	5 (L)
	3.30	6	6 (L)	6 (L)	6 (L)	6 (L)

H – High pathogenicity. Detection of 2 or more HPS targets
M – Moderate pathogenicity. Detection of Target B or Target X
L – Low pathogenicity. Detection of *Salmonella* spp. or Target A

Conclusions

This study demonstrates that the BAX System Real-Time PCR assay for Highly Pathogenic *Salmonella* provides reliable and accurate detection in poultry carcass rinses. The assay consistently detected low and high levels of *Salmonella* across serotypes representing high, moderate, and low pathogenicity. These results confirm that the BAX System is an effective approach for rapidly screening poultry rinsates, providing actionable insights into *Salmonella* presence and pathogenicity to support risk-based food safety programs and informed decision-making.

- **Enrich 30 mL poultry rinse with 30 mL of pre-warmed (35 °C) BPW, homogenize and incubate at 35 °C for 18-24 hours.**

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