

# Thermo Scientific SureTest Salmonella Species PCR Assay granted AOAC First Action Official Methods (OMA)

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## INTRODUCTION

*Salmonella* is a major food-borne pathogen and is frequently associated with illness in the US and around the world (1). Detecting *Salmonella* from food and environmental samples is crucial to preventing infection and illness. The Thermo Scientific™ SureTest™ Salmonella species PCR Assay is a Real-Time PCR test for the detection of *Salmonella* from one of the broadest ranges of foods and select environmental surfaces (Figure 1):

- Method scope covers 32 matrices.
- Includes challenging to test high-risk products.
- First to offer a validated method for large sample sizes e.g., raw beef meats, produce, cocoa and chocolate.

In this study, the accuracy, reliability and reproducibility of the SureTest Salmonella Species PCR method was demonstrated.

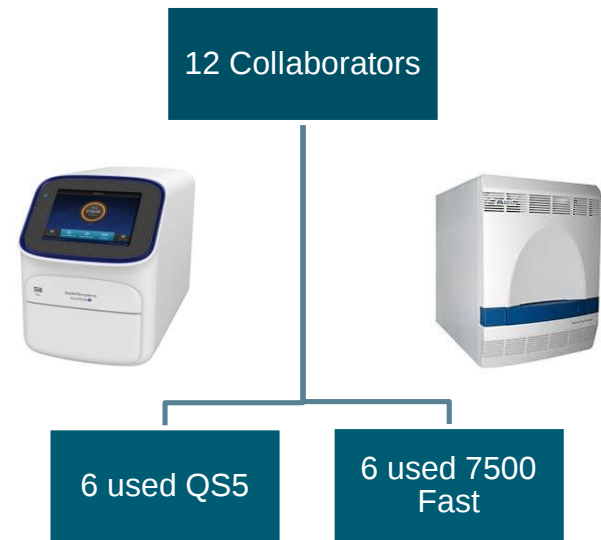
## METHODS

The unpaired collaborative study compared the SureTest Salmonella Species PCR method to the method outlined in FDA/BAM Chapter 5<sup>2</sup>. The study was run according to the guidelines in AOAC *Appendix J*<sup>3</sup>. The matrix selected was cocoa powder with a sample size of 375 g. This is a challenging matrix to test due to its large sample size, low moisture, and the presence of polyphenols, which can inhibit PCR.

For each method, collaborators tested 12 uninoculated portions (0 CFU/portion), 12 low inoculation portions (0.2-2 CFU/portion), and 12 high inoculation portions (2-5 CFU/portion). Therefore, each collaborator tested a total of 72 portions. The spiking organism was *Salmonella* Kentucky. The enrichment for the candidate method used a 1-in-10 dilution with Buffered Peptone Water. The candidate enrichments were analysed by PCR using the SureTest Salmonella Species PCR Assay on the Applied Biosystems™ QuantStudio™ 5 (QS5) or the Applied Biosystem™ 7500 Fast PCR instruments (see Figure 2) and streaked onto Thermo Scientific™ *Brilliance*™ Salmonella Agar. Presumptive purple-coloured colonies were confirmed using the candidate method confirmation procedure (Figure 3). Candidate enrichments were also tested using the FDA/BAM detection and confirmation procedures.

Twelve (12) collaborators submitted valid data. Out of the 12 collaborators, 11 were from eight laboratories across the US and one was from a laboratory in France, Europe.

Figure 2. PCR Instruments used in Collaborative study



## RESULTS

Figure 4. Cocoa Powder Collaborative study dLPOD: Candidate vs. Reference

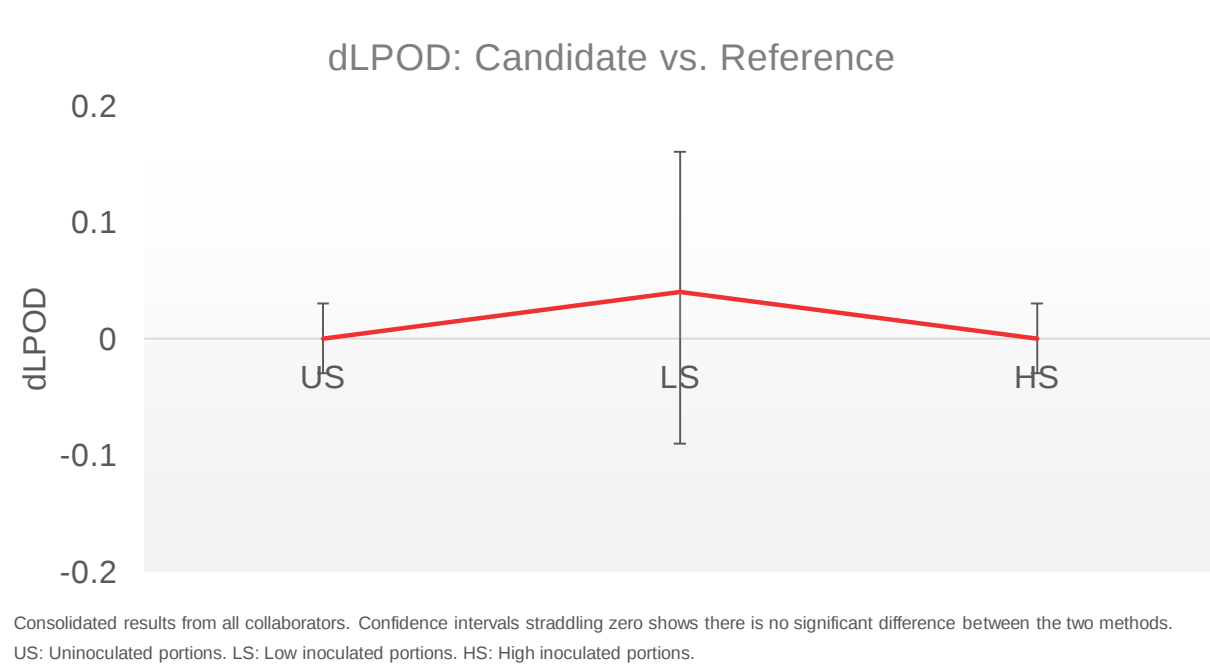


Figure 5. Cocoa Powder Collaborative study dLPOD: Candidate Presumptive vs. Candidate Confirmed

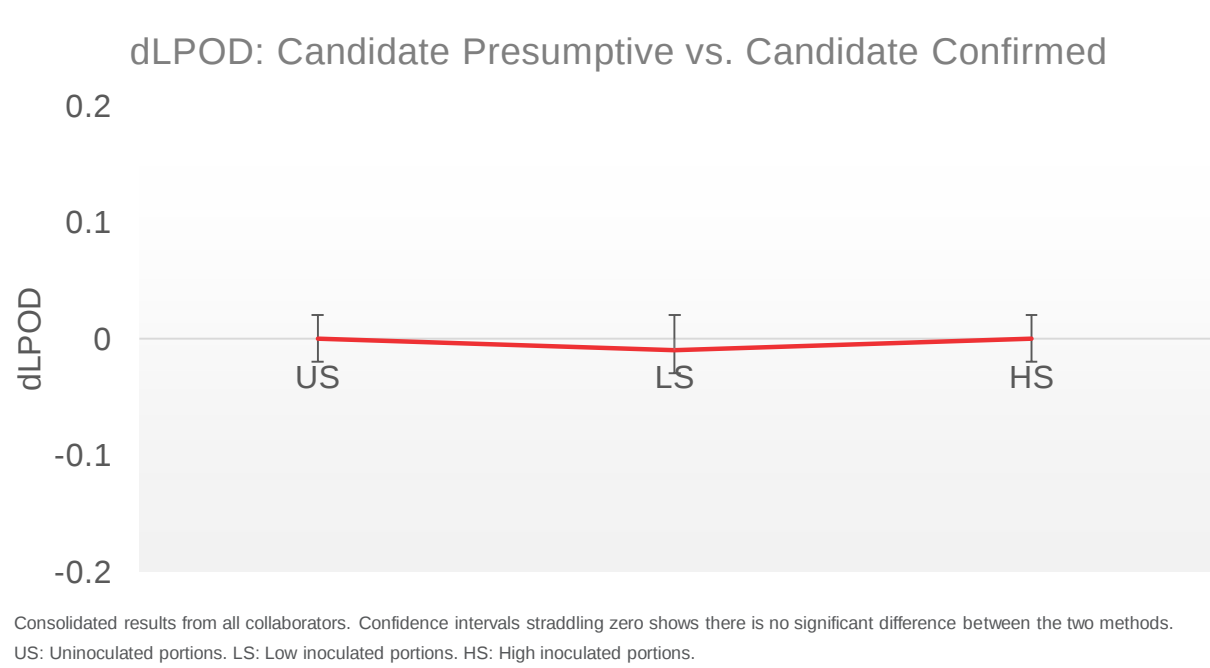
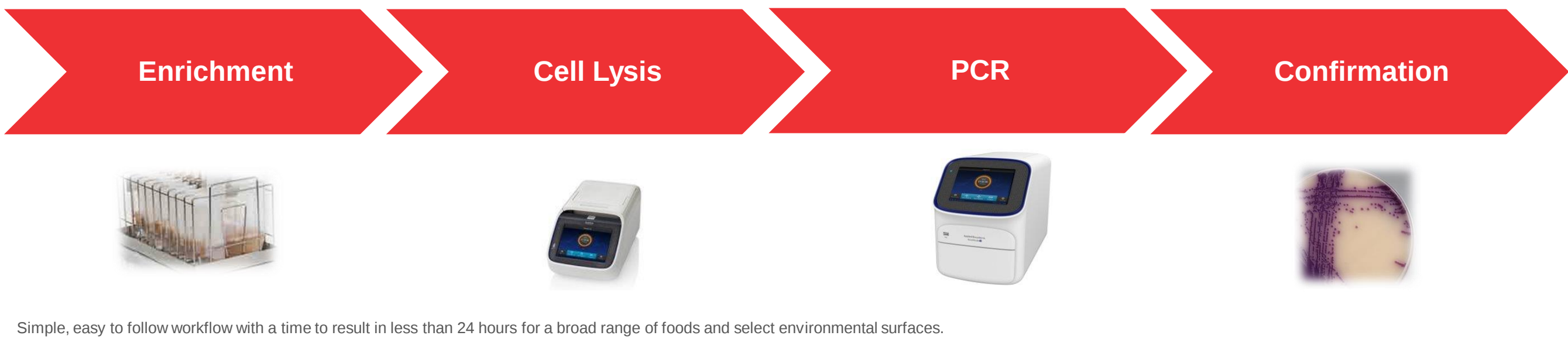


Figure 1. Broad range of food categories and select environmental surfaces

| Short protocols                                                                                                                                                                                                                      | Specific protocols and/or large sample sizes                                                                                                                                                                                                                                                                                                                          | Broad range of foods                                                                                                                                                             | Environmental surfaces                                                                                                                                                                                            |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
|  <ul style="list-style-type: none"><li>✓ Ground beef (25 g)</li><li>✓ Up to 375 g ground beef, beef trim</li><li>✓ Up to 375 g spinach</li></ul> |  <ul style="list-style-type: none"><li>✓ Sprouted seeds (25 g)</li><li>✓ Liquid whole egg (25 g)</li><li>✓ Non-fat dried milk powder (Up to 375 g)</li><li>✓ Chocolate (25 g)</li><li>✓ Up to 375 g cocoa powder, cocoa butter, cocoa liquor, dark chocolate (&gt;70%)</li></ul> |  <p>e.g., Raw meats, dairy, vegetables and fruits, ready-to-eat, ready-to-preheat, etc.</p> |  <ul style="list-style-type: none"><li>✓ Stainless steel (swab &amp; sponge)</li><li>✓ Plastic (swab &amp; sponge)</li></ul> |

Figure 3. SureTest Salmonella method workflow



## CONCLUSIONS

### Reproducibility

- Results between collaborators were consistent and there were no statistically significant differences between the two methods.

### Accuracy & Reliability

- The SureTest Samonella Species PCR method was statistically comparable to the FDA/BAM reference method for the detection of *Salmonella*.

### Broad Testing Scope

- The SureTest Salmonella Species PCR method has a broad range of foods claim against the ISO reference method<sup>4</sup>.
- To extend this claim to FDA, 11 AOAC matrix studies were run against the FDA/BAM reference method.

### Simple & Flexible Workflow

- Due to the COVID-19 pandemic, training had to be carried out remotely at a couple of the participating laboratories. No peculiar results were observed from these laboratories.
- The SureTest method offers extended incubation conditions, with a temperature range between 34 °C and 38 °C, and an 8 hours difference between the lower and upper incubation time limits.

## REFERENCES

1. Centers for Disease Control and Prevention, Salmonella <https://www.cdc.gov/salmonella/index.html>.
2. USFDA Bacteriological Analytical Manual (BAM), Chapter 5 – Salmonella
3. Official Methods of Analysis of AOAC INTERNATIONAL. (2019) 21st Ed., Appendix J: AOAC INTERNATIONAL Methods Committee Guidelines for Validation of Microbiological Methods for Food and Environmental Surfaces, AOAC INTERNATIONAL, Gaithersburg, MD, [http://www.eoma.aoac.org/app\\_j.pdf](http://www.eoma.aoac.org/app_j.pdf)
4. EN ISO 6579: Microbiology of the food chain — Horizontal method for the detection, enumeration and serotyping of Salmonella — Part 1: Detection of Salmonella spp.

## ACKNOWLEDGEMENTS

We would like to thank all the collaborators who made it possible for us to complete this study in difficult circumstances. Finally, a very special thank you to the team at Q Laboratories who coordinated the study.

## TRADEMARKS/LICENSING

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