

# AOAC-RI PTM and NF Validation of the Thermo Scientific *Listeria monocytogenes* PCR Assay using the QuantStudio 5 PCR Food Safety Instrument

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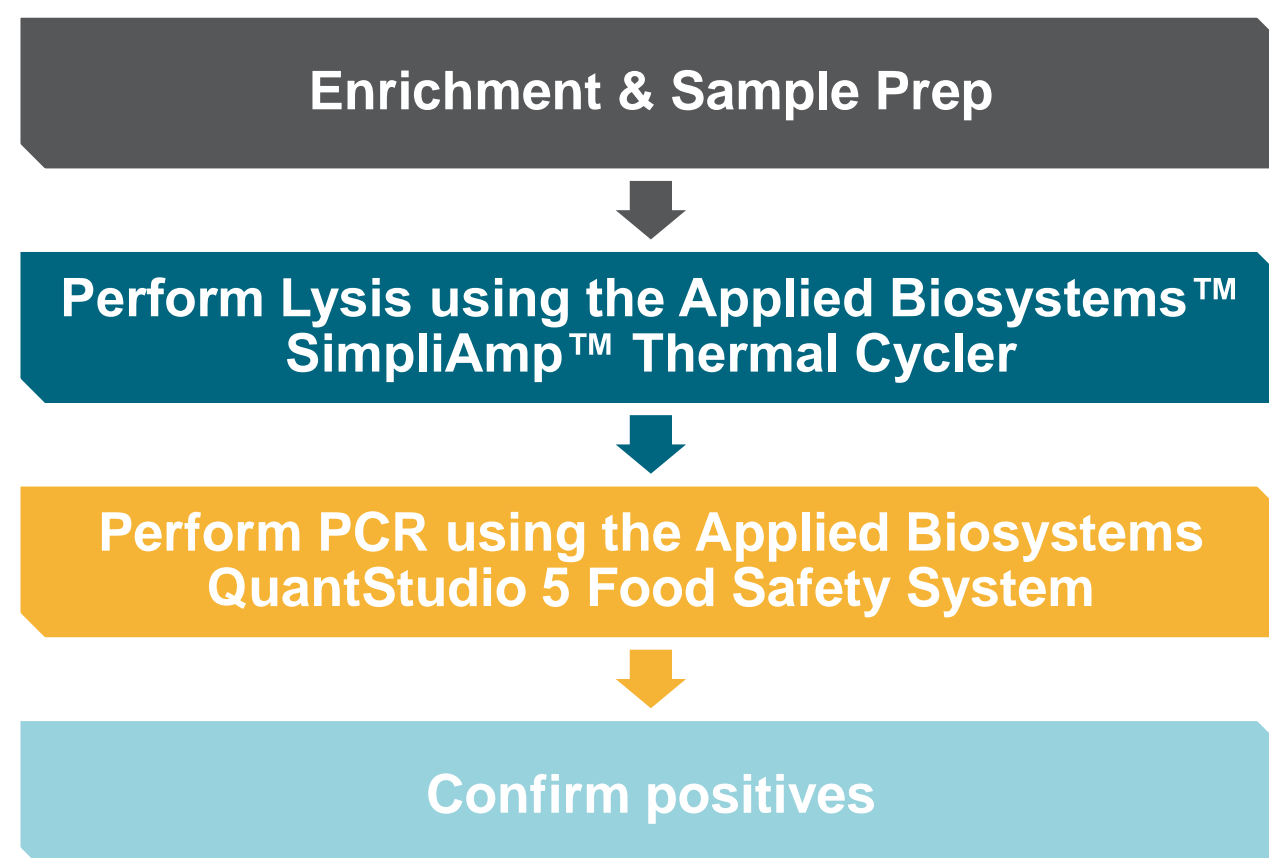
## INTRODUCTION

Studies were performed to extend the current AOAC-RI *Performance Tested Method*<sup>SM</sup> (PTM) and NF VALIDATION<sup>TM</sup> by AFNOR Certification claims for the Thermo Scientific<sup>TM</sup> SureTect<sup>TM</sup> *Listeria monocytogenes* PCR (candidate method) to include the use of the Applied Biosystems<sup>TM</sup> QuantStudio<sup>TM</sup> 5 Real-Time Food Safety PCR Instrument with associated Applied Biosystems<sup>TM</sup> RapidFinder<sup>TM</sup> Analysis software.

## MATERIALS AND METHODS

For the extension of the AOAC-RI PTM validation method modification studies were conducted in comparison to ISO 11290-1:1996<sup>1</sup>. For the NF VALIDATION by AFNOR Certification extension studies were conducted in comparison to ISO 11290-1:1996 in accordance with ISO 16140-2:2016<sup>2</sup>.

**Figure 1: Workflow for the SureTect *Listeria monocytogenes* PCR Assay**



## RESULTS

### AOAC PTM Validation

- The Probability of Detection (POD) analysis (table 1) demonstrated no statistically significant differences between the candidate methods and the reference method.
- Inclusivity and exclusivity testing demonstrated that the candidate methods successfully detected all target *L. monocytogenes* isolates and excluded all non-target isolates.

**Table 1. POD analysis of the SureTect *Listeria monocytogenes* PCR assay and the reference method**

Matrix type	Spike level	Number tested	Reference method positives	Candidate method positives <sup>a</sup>	dPOD <sup>b</sup>	95% CI <sup>c</sup>
All food matrices <sup>d</sup>	n/a	20	0	0	0.00	-0.16, 0.16
	Low	80	46	38	-0.10	-0.25, 0.05
	High	20	16	15	-0.05	-0.30, 0.21
All surface matrices <sup>e</sup>	n/a	15	0	0	0.00	-0.20, 0.20
	Low	40	10	10	0.00	-0.19, 0.19
	High	10	8	8	0.00	-0.34, 0.34

<sup>a</sup>Difference in POD between the candidate and reference methods  
<sup>b</sup>If the 95% confidence interval does not contain a zero the results are statistically significant at the 5% level  
<sup>c</sup>Raw ground beef (9 hr and 24 hr protocols), skimmed milk powder, lettuce  
<sup>d</sup>Plastic surface swabs (1x1") and sponges (4x4")

### AFNOR Validation

- Data was analyzed in accordance with ISO 16410-2:2016 (table 2).
- The candidate methods gave equivalent or improved performance to the reference method.

**Table 2: Sensitivity, relative trueness and false positive ratio of the candidate methods**

Number of samples	Sensitivity of the Candidate method	Sensitivity of the Reference method	Relative trueness	False positive ratio
387	80.2 %	77.8 %	81.9 %	2.7 %

**Figure 1. Applied Biosystems QuantStudio 5 Food Safety Real-Time PCR Instrument**



## CONCLUSION

The AOAC-RI PTM and NF VALIDATION studies demonstrated that the SureTect *Listeria monocytogenes* assay is a suitable method for the detection of *Listeria monocytogenes* from a variety of food and environmental surface samples when using the QuantStudio 5 Food Safety System (Figure 1).

## REFERENCES

- ISO 11290-1:1996, including Amendment 1:2004 'Microbiology of the food chain -- Horizontal method for the detection and enumeration of *Listeria monocytogenes* and of *Listeria spp.* -- Part 1: Detection method'
- ISO 16140-2:2016 Microbiology of the food chain -- Method validation -- Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method

## TRADEMARKS/ LICENSING

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