

AOAC-RI PTM and NF VALIDATION of the Thermo Scientific SureTect Salmonella species PCR Assay using the QuantStudio 5 Food Safety PCR Instrument

Jessica Williams¹, Ana-Maria Leonte¹, Katharine Evans¹, Annette Hughes¹, Charlotte Cooper¹, Maryse Rannou², Muriel Bernard² Ben Bastin³. ¹Thermo Fisher Scientific, Microbiology Basingstoke, UK, ²ADRIA Développement, Quimper, France, ³Q Laboratories Inc., Ohio, US.

INTRODUCTION

Studies were performed to extend the current AOAC-RI Performance tested methodSM (PTM) and NF VALIDATIONTM by AFNOR Certification claims for the Thermo ScientificTM SureTectTM Salmonella species PCR Assay (candidate method) to include the use of the Applied BiosystemsTM QuantStudioTM 5 Real-Time Food Safety PCR Instrument with associated Applied BiosystemsTM RapidFinderTM Analysis software (figure 1).

Figure 1. Thermo ScientificTM SureTectTM Real-Time PCR System

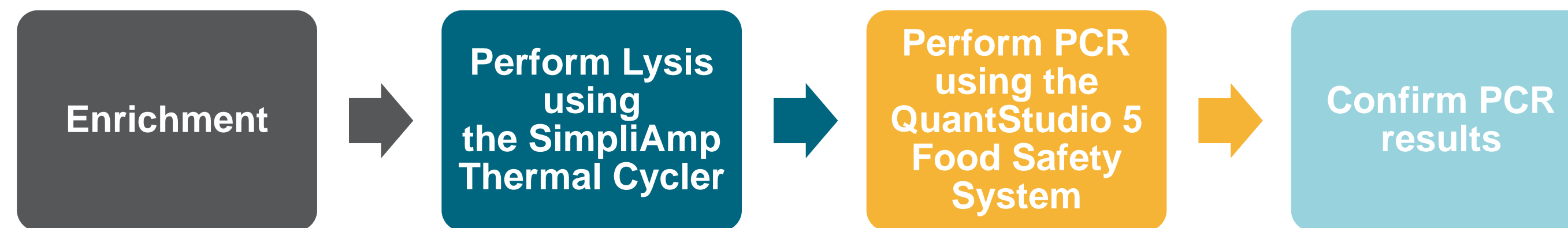


Left to right:
Applied BiosystemsTM SimpliAmpTM Thermal Cycler
Applied BiosystemsTM QuantStudioTM 5 Food Safety Real-Time PCR Instrument
Laptop with Applied BiosystemsTM RapidFinderTM Analysis Software
Thermo ScientificTM SureTectTM PCR Assays

MATERIALS AND METHODS

- The AOAC-RI PTM method modification study was conducted in comparison to ISO 6579-1:2017¹.
- The NF VALIDATION extension study was conducted in comparison to ISO 6579-1:2017 in accordance with ISO 16140-2:2016.
- The candidate method workflow is summarized in figure 2.

Figure 2. Thermo Scientific SureTect Salmonella species PCR Assay Workflow Summary



RESULTS

NF VALIDATION

Table 1: Sensitivity, Relative Trueness and False Positive Ratio of the Candidate method

Category	Sensitivity of the alternative method %	Sensitivity of the reference method %	Relative trueness %	False positive ratio %
A:	88.4	88.8	88.8	2.6
B:	88.5	88.1	88.4	3.5
C:	88.4	88.4	88.6	2.2
D:	88.5	87.7	88.2	3.1

A = Total of Dairy enriched with Thermo ScientificTM Buffered Peptone Water (BPW) and novobiocin, Raw beef 9hr and all other products
B = Total of Dairy enriched with Thermo ScientificTM One Broth Salmonella (OBS), Raw beef 9hr and all other products
C = Total of Dairy enriched with BPW and novobiocin, Raw beef 24hr and all other products
D = Total of Dairy enriched with OBS, Raw beef 24hr and all other products

The results in table 1 show that the alternative method is better or equivalent in performance to the ISO 6579-1:2017 reference method. The studies performed as part of the NF VALIDATION met the requirements of ISO 16140-2:2016.

AOAC PTM Validation

Table 2: POD Analysis Summary of the Candidate Method

Matrix	Spike level	N	Reference method positives	Alternative method positives	dPOD ^a	95% Confidence interval ^b
All food matrices ^c	n/a	20	0	0	0	-0.16, 0.16
	Low	80	46	38	-0.1	-0.25, 0.25
	High	20	16	15	-0.05	-0.30, 0.21
All surface matrices ^d	n/a	15	0	0	0	-0.20, 0.20
	Low	40	10	10	0	-0.19, 0.19
	High	10	8	8	0	-0.34, 0.34

^aDifference in POD between the alternative and reference methods
^bIf the 95% CI does not contain a zero the results are statistically significant at the 5% level
^cRaw ground beef (9 hr and 24 hr protocols), Skimmed milk powder, Lettuce
^dPlastic surface swabs (1x1") and sponges (4x4")

The results in table 2 show no statistically significant differences between the candidate method and the reference method. Inclusivity and exclusivity testing demonstrated that the candidate method successfully detected all target *Salmonella* species isolates and correctly excluded all non-target isolates

CONCLUSION

Superior Salmonella detection

- Detects Salmonella species in a broad range of food and environmental surfaces
- Superior or equivalent performance to the reference method.

AOAC and AFNOR Validated

- Data satisfied the acceptability criteria of AOAC PTM and NF VALIDATION by AFNOR Certification

Improved workflow using the QuantStudio 5 Food Safety System

- The QuantStudio 5 Food Safety System uses a 6-channel, 96-well cloud-enabled platform
- Suitable for running PCR solutions for food pathogen and authenticity testing
- Instrumentation offers touch screen technology along with intuitive software

REFERENCES

- ISO 6579-1:2017 Microbiology of the food chain -- Horizontal method for the detection, enumeration and serotyping of Salmonella -- Part 1: Detection of Salmonella spp
- 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

TRADEMARK STATEMENT

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LT2486A
September 2019

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