# CERTIFICATION

## AOAC Research Institute

## *Performance Tested Methods*[SM]

Certificate No.

**072302**

**The AOAC Research Institute hereby certifies the method known as:**

**Thermo Scientific™ SureCount™ Salmonella species, Typhimurium and Enteritidis Multiplex PCR Kit and SureCount ™ MPN**

manufactured by

**Oxoid Ltd part of Thermo Fisher Scientific**
**Wade Rd**
**Basingstoke, Hampshire**
**RG24 8PW, UK**

This method has been evaluated in the AOAC Research Institute *Performance Tested Methods*[SM] Program and found to perform as stated in the applicability of the method. This certificate indicates an AOAC Research Institute Certification Mark License Agreement has been executed which authorizes the manufacturer to display the AOAC Research Institute *Performance Tested Methods*[SM] certification mark on the above-mentioned method for the period below. Renewal may be granted by the Expiration Date under the rules stated in the licensing agreement.

Bradley A. Stawick, Senior Director
Signature for AOAC Research Institute

| | |
|---|---|
| Issue Date | January 10, 2024 |
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**SUBMITTING COMPANY**
**Oxoid Ltd. part of Thermo Fisher Scientific**
**Wade Rd**
**Basingstoke, Hampshire**
**RG24 8PW, UK**

**METHOD NAMES**
Thermo Scientific™ SureCount™ Salmonella species, Typhimurium and Enteritidis Multiplex PCR Kit and SureCount™ MPN

**CATALOG NUMBER**
A56848

**INDEPENDENT LABORATORY**
Q Laboratories, Inc.
1400 Harrison Avenue
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**APPLICABILITY OF METHOD**
**Target Organism – *Salmonella* species, *Salmonella* ser. Typhimurium, and *Salmonella* ser. Enteritidis**

**Matrixes – (MLG 4.11 and MPN) – fresh raw ground turkey (325 g), chicken carcass rinse, fresh raw ground beef (325 g), fresh raw ground pork (325)**

**Performance claims – Comparable performance of the SureCount Salmonella Multiplex PCR and the SureCount MPN method, including an alternative confirmation procedure, to the U.S. Department of Agriculture Food Safety and Inspection Service *Microbiology Laboratory Guidebook* (MLG) Chapter 4.11 Isolation and Identification of *Salmonella* from Meat, Poultry, Pasteurized Egg, and Siluriformes (Fish) Products and Carcass and Environmental Sponges reference culture methods (2), following the procedure outlined in MLG Appendix 2.05 Most Probable Number Procedure and Tables (MPN) (3).**

**ORIGINAL CERTIFICATION DATE**
**July 27, 2023**

**CERTIFICATION RENEWAL RECORD**
**Renewal annually through December 2024.**

**METHOD MODIFICATION RECORD**
1. **July 2023 Level 3**
2. **January 2024 Level 1**

**SUMMARY OF MODIFICATION**
1. **Method modification of the PTM 081701 Thermo Scientific™ RapidFinder™ for *Salmonella* species, Typhimurium and Enteritidis Multiplex PCR Kit for (1) quantitative analysis of *Salmonella* spp., *S.* Enteritidis and *S.* Typhimurium in fresh raw ground turkey, fresh raw ground beef, fresh raw ground pork, and chicken carcass rinses including alternative confirmation procedure and (2) certification of PTM 072302 Thermo Scientific™ SureCount™ Salmonella species, Typhimurium and Enteritidis Multiplex PCR Kit and SureCount™ MPN.**
2. **Editorial/clerical changes.**

**Under this AOAC *Performance Tested Methods[SM]* License Number, 072301 this method is distributed by:**
**NONE**

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**NONE**

**PRINCIPLE OF THE METHOD (1)**

The current PTM validated RapidFinder Salmonella Multiplex PCR Kit is a real-time PCR test for the qualitative detection and differentiation of Salmonella species (SPP), S. Typhimurium (STY) and S. Enteritidis (SEN) from food and environmental samples (4). The modification described in this report details the addition of the quantitative workflow (SureCount Salmonella Multiplex), capable of quantifying target organisms that are present at a minimum of 1 colony forming unit (cfu)/ test portion based on the amount of cellular DNA present. A test portion is a quantity of matrix removed from the original laboratory sample. (e.g., 325 g ground turkey is the test portion removed from a larger bulk sample of ground turkey). The test portion homogenate refers to the 30 mL aliquot removed after mixing the 325 g test portion of ground meat/poultry sample types with the specified amount of enrichment media. The test portion rinsate refers to the 30 mL aliquot removed from the poultry carcass/parts rinses.

This report also details the extension to apply the SureCount MPN method to the U.S. Department of Agriculture Food Safety and Inspection Service *Microbiology Laboratory Guidebook* (MLG) Chapter 4.11 (2) and the (MLG) Appendix 2.05 (3) reference methods. The assay can be performed on the 3 tube, 5 dilution Buffered Peptone Water set-up to determine presence or absence of *Salmonella* within each tube, which can then be used to enumerate the test portion utilizing MLG Appendix 2.05. This negates the need for further selective broth sub-enrichments, biochemical and antigen identification saving approximately 48 hours of labor.

The SureCount Salmonella Multiplex PCR kit is supplied containing all necessary reagents to conduct the test portion homogenate/rinsate lysis, including Lysis Tubes and lyophilized PCR pellets, containing all necessary PCR reagents (target-specific primers, dye-labelled probes, and PCR master mix components) to easily conduct the PCR analysis of the test portion homogenate/rinsate. PCR probes are short oligonucleotides with a quencher molecule at one end that, when not bound to target DNA, greatly reduces fluorescence from the dye label at the opposite end of the probe molecule. The oligonucleotides target unique DNA sequences found only in *Salmonella*. If STY and/or SEN and/or SPP are present, the target DNA sequences will be amplified and the increasing fluorescent signal generated will be detected by the Applied Biosystems™ QuantStudio 5™ Food Safety Real-Time PCR Instrument and interpreted by the Applied Biosystems™ RapidFinder™ Analysis software v2.0 or higher.

In addition to detection of any target DNA, the SureCount PCR pellets contain probes, primers, and DNA templates for an internal positive control (IPC). During PCR cycling, the IPC template is amplified whether any target DNA is present or not. The probe used for the IPC is labelled with a different colored fluorescent dye to the probes used within the assay to detect target DNA, and so can be detected by the QuantStudio 5 PCR Instrument through a separate dye channel. Following a successful PCR run, the instrument will detect amplification of the IPC DNA sequence. If no target DNA is detected by the assay, the presence of the IPC amplification curve confirms that the PCR process has occurred successfully.

The PCR probes used in the SureCount Salmonella Multiplex PCR Kit are based on Applied Biosystems™ TaqMan™ chemistry. Results from this assay system are achieved in approximately 50 minutes after the prepared test portion homogenate/rinsate is loaded into the QuantStudio 5 PCR instrument and is displayed on the attached PC screen. Simple positive or negative symbols are directly presented in the software together with a separate quantitative result for each target in the assay, with no need of setting up a calibration curve manually. The SPP target includes the quantification value for all *Salmonella* serovars present in the test portion homogenate/rinsate, including SEN and STY. To calculate the quantity of SPP excluding STY and SEN, the following formula can be used [SPP= SPP-(STY+SEN)].

The quantitative result provides an estimate of the true starting contamination level. The value given represents cfu/volume of liquid removed from the original test portion homogenate/rinsate. This value can either be used as provided (per 30 mL test portion homogenate/rinsate) or converted to cfu/mass or volume of original test portion (multiplication factor table detailed in the user guide) (5). The PCR amplification plots are easily accessible for review. All results interpreted by the RapidFinder Analysis software can be stored, printed, or downloaded by the user, as required.

**DISCUSSION OF THE VALIDATION STUDY (1)**

*Inclusivity/Exclusivity*

The inclusivity/exclusivity study correctly identified and excluded all isolates tested, respectively except for two inclusivity isolates of *Salmonella* Typhi. Both isolates were detected in mTSB and BPW without novobiocin, but the isolates were not detected in BPW with novobiocin. *S*. Typhi is documented for exhibiting sensitivity to novobiocin (6), and in combination with the enrichment length, this is likely why this species was not detected in BPW with novobiocin. However, prevalence of *S*. Typhi in poultry and meat samples is typically low (16), and as it was detected successfully in mTSB and BPW minus novobiocin this is not a concerning performance indicator. Several other isolates also exhibited moderate sensitivity to the Novobiocin addition, and inoculation levels had to be adjusted accordingly as outlined in Table 1.

Other than the two isolates mentioned, there was no difference in performance between either BPW with novobiocin or mTSB, and no performance differences between the isolates run at the method developer site or independent laboratory, highlighting the specificity and accuracy of the method.

*SureCount Salmonella Multiplex Matrix Study*

The SureCount Salmonella Multiplex method successfully enumerated all target organisms in all matrixes. Mean differences between the SureCount Salmonella Multiplex method results and the MLG 2.05/4.11 reference method results were <0.5 $\log_{10}$ in all cases except fresh raw ground turkey medium level for *S*. Typhimurium (0.821 $\log_{10}$), chicken carcass rinse high level for *S*. Hadar (0.543 $\log_{10}$), and raw fresh ground pork medium level for *S*. Enteritidis (0.559 $\log_{10}$). In each of these cases, the results from the SureCount method were closer to the inoculation level than the reference method. The inoculation level for *S*. Typhimurium medium level in fresh raw ground turkey was 30.8 cfu/g (1.49 $\log_{10}$), and the SureCount result was 1.411 $\log_{10}$, while the reference method result was 0.590 $\log_{10}$. The inoculation level for *S*. Hadar high level in chicken carcass rinse was 1,170 cfu/mL (3.07 $\log_{10}$), and the SureCount result was 2.381 $\log_{10}$, while the reference method result was 1.838 $\log_{10}$. The inoculation level for *S*. Enteritidis medium level in fresh raw ground pork and 69 cfu/g (1.84 $\log_{10}$), and the SureCount result 2.016 $\log_{10}$, while the reference method result was 1.457 $\log_{10}$. In addition, the $s_r$ were consistently lower for the SureCount method, 0.140, 0.144, and 0.119 respectively, while the $s_r$ for the reference method were particularly high in these three cases, 0.997, 1.124, and 0.622 respectively. It is important to note that the fresh raw ground turkey was co-inoculated *S*. Typhimurium and *S*. Enteritidis at the medium contamination level, and the chicken carcass rinse was co-inoculated with *S*. Hadar and *S*. Typhimurium at the high level. The variation and underestimation of the reference method is likely due to the challenge associated with cultural confirmation of multiple serovars on one plate. The MLG 4.11 is reliant on the isolation of colonies on selective plates and positive O group confirmations to confirm co-infection, which are particularly challenging in a co-infected sample given the contamination level (overgrowth of one strain) as well as competition of background flora. Chicken carcass rinses in particular are well documented for having a diverse and abundant microbiome which would have added further complication to isolation and confirmation given high levels of interspecific competition (6-10).

There was one outlier for the raw ground beef matrix at the low contamination level for the reference method, but no obvious explanation for what had caused this, so this outlier was not removed from the dataset during analysis. The most probable cause for this is a handling error due to the number of test portions being processed.

The SureCount Salmonella Multiplex method reliably estimated more consistent cfu/mL results per dataset than the MPN method, demonstrating both the accuracy and sensitivity of the method as all estimations were closer to the inoculation level than the reference method was. This is of particular importance given certain *Salmonella* spp. cases and antibiotic resistance are increasing annually. Specifically, the contaminating organism in this case, *Salmonella* Infantis, has seen a recent rise in antibiotic resistance (11, 12), thus the ability to detect and enumerate reliably is of marked importance in ensuring contaminated stock is not released but also that stock is not falsely retained.

The performance of the raw ground turkey matrix at both the method developer and independent laboratory demonstrated similar performance with differences of means within 0.5 log and 90% confidence intervals that were either within -0.5 to 0.5 or marginally outside. The data show that the SureCount Salmonella Multiplex method is a consistent and reliable method for estimating target *Salmonella* contamination levels compared to the reference method.

*SureCount MPN Matrix Study*

The SureCount MPN method demonstrated comparable or equivalent performance to the reference method for all matrixes. The data presented suggests that application of the SureCount MPN method is a suitable alternative for MPN enumeration compared to the full MLG 4.11 workflow.

Mean differences between the SureCount Salmonella MPN method results and the MLG 2.05/4.11 reference method results were <0.5 $\log_{10}$ in all cases except fresh raw ground turkey medium level for *S*. Typhimurium (0.832 $\log_{10}$), and chicken carcass rinse high level for *S*. Hadar (1.096 $\log_{10}$), In each of these cases, the results from the

SureCount MPN method were closer to the inoculation level than the reference method. The inoculation level for *S.* Typhimurium medium level in fresh raw ground turkey was 30.8 cfu/g (1.49 $\log_{10}$), and the SureCount result was 1.422 $\log_{10}$, while the reference method result was 0.590 $\log_{10}$. The inoculation level for *S.* Hadar high level in chicken carcass rinse was 1,170 cfu/mL (3.07 $\log_{10}$), and the SureCount result was 2.934 $\log_{10}$, while the reference method result was 1.838 $\log_{10}$. In addition, the $s_r$ were consistently lower for the SureCount method, 0.271 and 0.373 respectively, while the $s_r$ for the reference method were particularly high in both cases, 0.997 and 1.124, respectively. It is important to note that the fresh raw ground turkey was co-inoculated *S.* Typhimurium and *S.* Enteritidis at the medium contamination level, and the chicken carcass rinse was co-inoculated with *S.* Hadar and *S.* Typhimurium at the high level.

In addition, there was one outlier for raw ground beef from the reference method dataset. This outlier significantly impacted analysis, however there was no explainable cause for this, and so data has been presented both with and without the outlier. The most probable explanation for this is a handling error due to the number of test portions being processed.

The cultural confirmation of the MPN method is particularly challenging when matrixes are co-inoculated. For example, the chicken carcass rinse matrix; *S.* Typhimurium has a well-documented rapid growth rate and has also been shown to suppress the growth of *S.* Hadar through intraspecific competition (13, 14). This likely occurred in this case and led to poor growth of *S.* Hadar on agar plates resulting in the low MPN count. However, as PCR technology negates this issue, the SureCount MPN method was able to enumerate at the correct contamination level compared to the MPN method, leading to the large confidence interval for the individual *S.* Hadar analysis in favor of the candidate method.

The performance of the ground turkey matrix at both the method developer and independent laboratory demonstrated comparable performance with differences of means within 0.5 log and 90% confidence intervals that were either within -0.5 to 0.5 or marginally outside. The data show that the SureCount Salmonella MPN method is a shorter and reliable method for estimating target *Salmonella* contamination levels compared to the full MPN reference method.

**Table 1. Inclusivity results of the Thermo Scientific SureCount Salmonella species, Typhimurium and Enteritidis Multiplex PCR Kit (1)**

| No. | *Salmonella* serotype | Group/ comments | Source | Origin | BPW + Novobiocin Results | | | mTSB Results | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Spp[c] | SE[d] | ST[e] | Spp | SE | ST |
| 1 | *Salmonella* Uphill | II 42:b:e,n,x,$z_{15}$ | RDCC[a] 1726 | unknown[b] | + | - | - | + | - | - |
| 2 | *Salmonella* Donna | II 53:$z_4$,$z_{24}$:- | RDCC 2355 | unknown | + | - | - | + | - | - |
| 3 | *Salmonella* Locarno | II 57:$z_{29}$:$z_{42}$ | RDCC 2830 | unknown | + | - | - | + | - | - |
| 4 | *Salmonella* Tranaroa | II 55:k:$z_{39}$ | NCTC[f] 10252 | unknown | + | - | - | + | - | - |
| 5 | *Salmonella salamae* | II 58:l,z13,z28:z6 | TCC[g] 2599 | unknown | + | - | - | + | - | - |
| 6 | *Salmonella arizoniae* | IIIa 51:z4,z23:- | NCTC 8297 | unknown | + | - | - | + | - | - |
| 7 | *Salmonella arizoniae* | S 24, 223:- | TCC 2389 | unknown | + | - | - | + | - | - |
| 8 | *Salmonella arizoniae* | IIIa 41:z4,z23:- | TCC 2608 | unknown | + | - | - | + | - | - |
| 9 | *Salmonella arizoniae* | IIIa 40:z4,z23:- | TCC 2609 | unknown | + | - | - | + | - | - |
| 10 | *Salmonella arizoniae* | IIIa 48:g,z51:- | TCC 2610 | unknown | + | - | - | + | - | - |
| 11 | *Salmonella diarizonae* | 61:k:1,5,7 | TCC 2386 | Clinical | + | - | - | + | - | - |
| 12 | *Salmonella diarizonae* | 38:l,v:z53 | TCC 2388 | unknown | + | - | - | + | - | - |
| 13 | *Salmonella diarizonae* | IIIb 60:r:e,n,x,z15 | TCC 2616 | unknown | + | - | - | + | - | - |
| 14 | *Salmonella diarizonae* | IIIb 48:i:z | TCC 2617 | unknown | + | - | - | + | - | - |
| 15 | *Salmonella diarizonae* | IIIb 61:k:1,5,(7) | TCC 2618 | unknown | + | - | - | + | - | - |
| 16 | *Salmonella houtenae* | 51:z4,z23:- | RDCC 3732 | unknown | + | - | - | + | - | - |
| 17 | *Salmonella houtenae* | IV 50:g,z51:- | TCC 2624 | unknown | + | - | - | + | - | - |
| 18 | *Salmonella houtenae* | IV 48:g,z51:- | TCC 2625 | unknown | + | - | - | + | - | - |
| 19 | *Salmonella houtenae* | IV 44:z4,z23:- | TCC 2626 | unknown | + | - | - | + | - | - |
| 20 | *Salmonella houtenae* | IV 45:g,z51:- | TCC 2627 | unknown | + | - | - | + | - | - |
| 21 | *Salmonella indica* | 45:a:e,n,x | OCC[h] 2360 | unknown | + | - | - | + | - | - |
| 22 | *Salmonella indica* | VI 11:a:1,5 | OCC 2643 | unknown | + | - | - | + | - | - |
| 23 | *Salmonella indica* | VI 6,14,25:z10:1,(2),7 | TCC 2640 | unknown | + | - | - | + | - | - |
| 24 | *Salmonella indica* | VI 11:b:1,7 | TCC 2641 | unknown | + | - | - | + | - | - |
| 25 | *Salmonella indica* | VI 6,7:z41:1,7 | TCC 2642 | unknown | + | - | - | + | - | - |
| 26 | *Salmonella bongori* | unknown | NCTC 2215[i] | unknown | + | - | - | + | - | - |
| 27 | *Salmonella bongori* | 66:z41:- | NCTC 12419 | unknown | + | - | - | + | - | - |
| 28 | *Salmonella bongori* | 66:z41:- | NCTC 10946 | Frog | + | - | - | + | - | - |
| 29 | *Salmonella* Stanley | Group O:4 (B) | RDCC 3407 | unknown | + | - | - | + | - | - |
| 30 | *Salmonella* Abony | Group O:4 (B) | RDCC 5096 | unknown | + | - | - | + | - | - |
| 31 | *Salmonella* Saintpaul | Group O:4 (B) | RDCC 2150 | unknown | + | - | - | + | - | - |
| 32 | *Salmonella* Heidelberg | Group O:4 (B) | RDCC 3377 | unknown | + | - | - | + | - | - |
| 33 | *Salmonella* Agona | Group O:4 (B) | RDCC 2358 | unknown | + | - | - | + | - | - |
| 34 | *Salmonella* Brandenburg | Group O:4 (B) | RDCC 2839 | Clinical | + | - | - | + | - | - |
| 35 | *Salmonella* Indiana | Group O:4 (B) | RDCC 2840 | Turkey | + | - | - | + | - | - |
| 36 | *Salmonella* Abortus-equi | Group O:4 (B) | OCC 2296 | unknown | + | - | - | + | - | - |
| 37 | *Salmonella* Pomona | Group O:4 (B) | NCTC 6589 | Intestine of turkey | + | - | - | + | - | - |
| 38 | *Salmonella* Schwarzengrund | Group O:4 (B) | TCC 3017 | unknown | + | - | - | + | - | - |
| 39 | *Salmonella* Stanleyville | Group O:4 (B) | TCC3018 | unknown | + | - | - | + | - | - |
| 40 | *Salmonella* Sandiego | Group O:4 (B) | TCC 3019 | unknown | + | - | - | + | - | - |
| 41 | *Salmonella* Bredeney | Group O:4 (B) | RDCC 3381 | unknown | + | - | - | + | - | - |
| 42 | *Salmonella* Java | Group O:4 (B) | RDCC 30151 | unknown | + | - | - | + | - | - |
| 43 | *Salmonella* Paratyphi B | Group O:4 (B) | RDCC 30020 | unknown | + | - | - | + | - | - |
| 44 | *Salmonella* Dublin | Group O:9 ($D_1$) | RDCC 3399 | unknown | + | - | - | + | - | - |
| 45 | *Salmonella* Rostock | Group O:9 ($D_1$) | NCTC 3747 | unknown | + | - | - | + | - | - |
| 46 | *Salmonella* Gallinarum | Group O:9 ($D_1$) | RDCC 1774[j] | unknown | + | - | - | + | - | - |
| 47 | *Salmonella* Alabama | Group O:9 ($D_1$) | NCTC 9868 | unknown | + | - | - | + | - | - |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 48 | *Salmonella* Miami | Group O:9 ($D_1$) | TCC 3024 | unknown | + | - | - | + | - | - |
| 49 | *Salmonella* Lomalinda | Group O:9 ($D_1$) | TCC 3025 | Clinical | + | - | - | + | - | - |
| 50 | *Salmonella* Israel | Group O:9 ($D_1$) | TCC 3026 | unknown | + | - | - | + | - | - |
| 51 | *Salmonella* Portland | Group O:9 ($D_1$) | TCC 3027 | unknown | + | - | - | + | - | - |
| 52 | *Salmonella* Sendai | Group O:9 ($D_1$) | TCC 3028 | unknown | + | - | - | + | - | - |
| 53 | *Salmonella* Napoli | Group O:9 ($D_1$) | RDCC 2129 | Clinical | + | - | - | + | - | - |
| 54 | *Salmonella* Gallinarum | Group O:9 ($D_1$) | RDCC 2138 | unknown | + | - | - | + | - | - |
| 55 | *Salmonella* Eastbourne | Group O:9 ($D_1$) | RDCC 2139 | unknown | + | - | - | + | - | - |
| 56 | *Salmonella* Javiana | Group O:9 ($D_1$) | RDCC 2146 | unknown | + | - | - | + | - | - |
| 57 | *Salmonella* Berta | Group O:9 ($D_1$) | RDCC 2360 | unknown | + | - | - | + | - | - |
| 58 | *Salmonella* Typhi | Group O:9 ($D_1$) | RDCC 30001[k] | unknown | -[k] | - | - | + | - | - |
| 59 | *Salmonella* Typhi | Group O:9 ($D_1$) | OCC 365[k] | unknown | -[k] | - | - | + | - | - |
| 60 | *Salmonella* Kiel | Group O:2 (A) | OCC 1654 | unknown | + | - | - | + | - | - |
| 61 | *Salmonella* Ohio | Group O:7 ($C_1$) | RDCC 2135 | Clinical | + | - | - | + | - | - |
| 62 | *Salmonella* Montevideo | Group O:7 ($C_1$) | RDCC 2359 | unknown | + | - | - | + | - | - |
| 63 | *Salmonella* Virchow | Group O:7 ($C_1$) | RDCC 2351 | unknown | + | - | - | + | - | - |
| 64 | *Salmonella* Infantis | Group O:7 ($C_1$) | RDCC 3400 | Clinical | + | - | - | + | - | - |
| 65 | *Salmonella* Bovis | Group O:8 ($C_2$-$C_3$) | RDCC 3402 | unknown | + | - | - | + | - | - |
| 66 | *Salmonella* Albany | Group O:8 ($C_2$-$C_3$) | RDCC 2125 | Clinical | + | - | - | + | - | - |
| 67 | *Salmonella* Bovis-Morbificans | Group O:8 ($C_2$-$C_3$) | OCC 2001 | unknown | + | - | - | + | - | - |
| 68 | *Salmonella* Kentucky | Group O:8 ($C_2$-$C_3$) | RDCC 3734 | unknown | + | - | - | + | - | - |
| 69 | *Salmonella* Newport | Group O:8 ($C_2$-$C_3$) | RDCC 2087 | unknown | + | - | - | + | - | - |
| 70 | *Salmonella* Muenchen | Group O:8 ($C_2$-$C_3$) | RDCC 2130 | unknown | + | - | - | + | - | - |
| 71 | *Salmonella* Hadar | Group O:8 ($C_2$-$C_3$) | RDCC 2131 | unknown | + | - | - | + | - | - |
| 72 | *Salmonella* Shanghai | Group O:3,10 ($E_1$) | RDCC 2132 | unknown | + | - | - | + | - | - |
| 73 | *Salmonella* Allerton | Group O:3,10 ($E_1$) | OCC 1623 | unknown | + | - | - | + | - | - |
| 74 | *Salmonella* Muenster | Group O:3,10 ($E_1$) | OCC 2683 | unknown | + | - | - | + | - | - |
| 75 | *Salmonella* Seftenburg | Group O:1,3,19 ($E_4$) | RDCC 2210 | Feces | + | - | - | + | - | - |
| 76 | *Salmonella* Krefeld | Group O:1,3,19 ($E_4$) | OCC 1655 | unknown | + | - | - | + | - | - |
| 77 | *Salmonella* Aberdeen | Group O:11 (F) | OCC 1612 | unknown | + | - | - | + | - | - |
| 78 | *Salmonella* Rubislaw | Group O:11 (F) | RDCC 3739 | unknown | + | - | - | + | - | - |
| 79 | *Salmonella* Poona | Group O:13 (G) | RDCC 3405 | Gastro-enteritis | + | - | - | + | - | - |
| 80 | *Salmonella* Ibidam | Group O:13 (G) | RDCC 2831 | unknown | + | - | - | + | - | - |
| 81 | *Salmonella* Madelia | Group O:6,14 (H) | RDCC 3736 | unknown | + | - | - | + | - | - |
| 82 | *Salmonella* Schalkwijk | Group O:6,14 (H) | OCC 2453 | unknown | + | - | - | + | - | - |
| 83 | *Salmonella* Saphra | Group O:16 (I) | OCC 2671 | unknown | + | - | - | + | - | - |
| 84 | *Salmonella* Huttingfoss | Group O:16 (I) | RDCC 2362 | unknown | + | - | - | + | - | - |
| 85 | *Salmonella* Michigan | Group O:17 (J) | OCC 2673 | unknown | + | - | - | + | - | - |
| 86 | *Salmonella* Cerro | Group O:18 (K) | RDCC 2127 | unknown | + | - | - | + | - | - |
| 87 | *Salmonella* Brisbane | Group O:28 (M) | OCC 2679 | unknown | + | - | - | + | - | - |
| 88 | *Salmonella* Urbana | Group O:30 (N) | RDCC 2100 | unknown | + | - | - | + | - | - |
| 89 | *Salmonella* Matopeni | Group O:30 (N) | RDCC 2356 | unknown | + | - | - | + | - | - |
| 90 | *Salmonella* Adelaide | Group O:35 (O) | RDCC 2122 | unknown | + | - | - | + | - | - |
| 91 | *Salmonella* Alachua | Group O:35 (O) | OCC 2690 | unknown | + | - | - | + | - | - |
| 92 | *Salmonella* Inverness | Group O:38 (P) | RDCC 2120 | unknown | + | - | - | + | - | - |
| 93 | *Salmonella* Champaign | Group O:39 (Q) | RDCC 2118 | unknown | + | - | - | + | - | - |
| 94 | *Salmonella* Riogrande | Group O:40 (R) | RDCC 5372 | unknown | + | - | - | + | - | - |
| 95 | *Salmonella* Johannesburg | Group O:40 (R) | OCC 2685 | unknown | + | - | - | + | - | - |
| 96 | *Salmonella* Vietnam | Group O:41 (S) | OCC 2675 | unknown | + | - | - | + | - | - |
| 97 | *Salmonella* Gera | Group O:42 (T) | OCC 2677 | unknown | + | - | - | + | - | - |
| 98 | *Salmonella* Berkeley | Group O:43 (U) | RDCC 2937 | unknown | + | - | - | + | - | - |
| 99 | *Salmonella* Tornow | Group O:45 (W) | OCC 2676 | unknown | + | - | - | + | - | - |
| 100 | *Salmonella* Teshi | Group O:47 (X) | RDCC 1728 | unknown | + | - | - | + | - | - |
| 101 | *Salmonella* Paratyphi A | Group O:2 (A) | RDCC 30017 | unknown | + | - | - | + | - | - |
| 102 | *Salmonella* Paratyphi C | Group O:7 ($C_1$) | RDCC 30026 | unknown | + | - | - | + | - | - |
| 103 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | RDCC 2207 | unknown | + | + | - | + | + | - |
| 104 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | RDCC 3379 | unknown | + | + | - | + | + | - |
| 105 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | RDCC 3729 | unknown | + | + | - | + | + | - |
| 106 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | RDCC 3900 | unknown | + | + | - | + | + | - |
| 107 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | OCC 723 | unknown | + | + | - | + | + | - |
| 108 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 1637 | unknown | + | + | - | + | + | - |
| 109 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 1638 | unknown | + | + | - | + | + | - |
| 110 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 1639 | unknown | + | + | - | + | + | - |
| 111 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 1640 | unknown | + | + | - | + | + | - |
| 112 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 1986 | unknown | + | + | - | + | + | - |
| 113 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2424 | unknown | + | + | - | + | + | - |
| 114 | *Salmonella* Enteritidis | 9,12:g,m:- ($D_1$) | TCC 2425 | unknown | + | + | - | + | + | - |
| 115 | *Salmonella* Enteritidis | 9,12:g,m:- ($D_1$) | TCC 2426 | Guinea pig | + | + | - | + | + | - |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 116 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2443 | Raw almonds | + | + | - | + | + | - |
| 117 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2450 | gastroenteritis | + | + | - | + | + | - |
| 118 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2591 | unknown | + | + | - | + | + | - |
| 119 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2668 | unknown | + | + | - | + | + | - |
| 120 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2669 | unknown | + | + | - | + | + | - |
| 121 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 2670 | unknown | + | + | - | + | + | - |
| 122 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | RDCC 3372 | unknown | + | + | - | + | + | - |
| 123 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | RDCC 3378 | unknown | + | + | - | + | + | - |
| 124 | *Salmonella* Enteritidis var. Dansyz | 1,9,12:g,m:- ($D_1$) | RDCC 3383 | Gastroenteritis | + | + | - | + | + | - |
| 125 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | TCC 1584 | unknown | + | + | - | + | + | - |
| 126 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | ATCC[l] BAA 1587[n] | unknown | + | + | - | + | + | - |
| 127 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL[m] 10155.1[n] | unknown | + | + | - | + | + | - |
| 128 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 10170.1[n] | unknown | + | + | - | + | + | - |
| 129 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 14255.2[n] | unknown | + | + | - | + | + | - |
| 130 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.40[n] | unknown | + | + | - | + | + | - |
| 131 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.80[n] | unknown | + | + | - | + | + | - |
| 132 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.121[n] | unknown | + | + | - | + | + | - |
| 133 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.184[n] | unknown | + | + | - | + | + | - |
| 134 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.185[n] | unknown | + | + | - | + | + | - |
| 135 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.186[n] | unknown | + | + | - | + | + | - |
| 136 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.278[n] | unknown | + | + | - | + | + | - |
| 137 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.279[n] | unknown | + | + | - | + | + | - |
| 138 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 16078-2A.280[n] | unknown | + | + | - | + | + | - |
| 139 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 1698878.3[n] | unknown | + | + | - | + | + | - |
| 140 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 175599.1[n] | unknown | + | + | - | + | + | - |
| 141 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 182282[n] | unknown | + | + | - | + | + | - |
| 142 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 188498.1[n] | unknown | + | + | - | + | + | - |
| 143 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 191569.1[n] | unknown | + | + | - | + | + | - |
| 144 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | QL 194559.3[n] | unknown | + | + | - | + | + | - |
| 145 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | CCUG[o] 9563[n] | unknown | + | + | - | + | + | - |
| 146 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | CCUG 21288[n] | unknown | + | + | - | + | + | - |
| 147 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | CCUG 25340[n] | unknown | + | + | - | + | + | - |
| 148 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | CCUG 26522[n] | unknown | + | + | - | + | + | - |
| 149 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | CCUG 27004[n] | unknown | + | + | - | + | + | - |
| 150 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | CCUG 27021[n] | unknown | + | + | - | + | + | - |
| 151 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | FSL[p] S5-415[n] | unknown | + | + | - | + | + | - |
| 152 | *Salmonella* Enteritidis | 1,9,12:g,m:- ($D_1$) | FSL S5-483[n] | unknown | + | + | - | + | + | - |
| 153 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 962 | unknown | + | - | + | + | - | + |
| 154 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1793 | unknown | + | - | + | + | - | + |
| 155 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 2124 | unknown | + | - | + | + | - | + |
| 156 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 2836 | unknown | + | - | + | + | - | + |
| 157 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3380 | unknown | + | - | + | + | - | + |
| 158 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3384 | unknown | + | - | + | + | - | + |
| 159 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3740 | unknown | + | - | + | + | - | + |
| 160 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3741[q] | unknown | + | - | + | + | - | + |
| 161 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3897 | Tissue, animal | + | - | + | + | - | + |
| 162 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3920 | Dairy (Tiramisu) | + | - | + | + | - | + |
| 163 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3922 | Chocolate | + | - | + | + | - | + |
| 164 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 3924 | Cocoa beans | + | - | + | + | - | + |
| 165 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 4669 | unknown | + | - | + | + | - | + |
| 166 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1585 | unknown | + | - | + | + | - | + |
| 167 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1679 | unknown | + | - | + | + | - | + |
| 168 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1680 | unknown | + | - | + | + | - | + |
| 169 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1681 | Gastroenteritis | + | - | + | + | - | + |
| 170 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1683 | unknown | + | - | + | + | - | + |
| 171 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | RDCC 1684 | unknown | + | - | + | + | - | + |
| 172 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 1880 | unknown | + | - | + | + | - | + |
| 173 | *Salmonella* Typhimurium | 1,4,5,12:i:- (B) | TCC 2387 | unknown | + | - | + | + | - | + |
| 174 | *Salmonella* Typhimurium | 1, 4, 5, 12: - :- (B) | TCC 2390 | unknown | + | - | + | + | - | + |
| 175 | *Salmonella* Typhimurium | 1,4,5,12 i:1,2 (B) | ATCC 19585 | unknown | + | - | + | + | - | + |
| 176 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | ATCC BAA-1603 | Tomato | + | - | + | + | - | + |
| 177 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2461 | Feces | + | - | + | + | - | + |
| 178 | *Salmonella* Typhimurium | 4,5,12:I:1,2 (B) | TCC 2593 | unknown | + | - | + | + | - | + |
| 179 | *Salmonella* Typhimurium | DT104b | TCC 2645 | unknown | + | - | + | + | - | + |
| 180 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2646 | unknown | + | - | + | + | - | + |
| 181 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2647 | unknown | + | - | + | + | - | + |
| 182 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2648 | unknown | + | - | + | + | - | + |
| 183 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2649 | unknown | + | - | + | + | - | + |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 184 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2650 | unknown | + | - | + | + | - | + |
| 185 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2651 | unknown | + | - | + | + | - | + |
| 186 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2652 | unknown | + | - | + | + | - | + |
| 187 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2653 | unknown | + | - | + | + | - | + |
| 188 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2654 | unknown | + | - | + | + | - | + |
| 189 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2655 | unknown | + | - | + | + | - | + |
| 190 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2656 | unknown | + | - | + | + | - | + |
| 191 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2657 | unknown | + | - | + | + | - | + |
| 192 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 2658 | unknown | + | - | + | + | - | + |
| 193 | *Salmonella* Typhimurium/ DT104 | 1,4,5,12:i:1,2 (B) | TCC 2659 | unknown | + | - | + | + | - | + |
| 194 | *Salmonella* Typhimurium/ DT104 | 1,4,5,12:i:1,2 (B) | TCC 2660 | unknown | + | - | + | + | - | + |
| 195 | *Salmonella* Typhimurium/ DT104 | 1,4,5,12:i:1,2 (B) | TCC 2661 | unknown | + | - | + | + | - | + |
| 196 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | TCC 1586 | unknown | + | - | + | + | - | + |
| 197 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | QL 11007-2[n] | unknown | + | - | + | + | - | + |
| 198 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | QL 11414-2[n] | unknown | + | - | + | + | - | + |
| 199 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | QL 16078-2A.110[n] | unknown | + | - | + | + | - | + |
| 200 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | QL 16078-2A.112[n] | unknown | + | - | + | + | - | + |
| 201 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | QL 16078-2A.1[n] | unknown | + | - | + | + | - | + |
| 202 | *Salmonella* Typhimurium | 1,4,5,12:i:1,2 (B) | QL 16078-2A.108[n] | unknown | + | - | + | + | - | + |

[a]RDCC = Research and Development culture collection – Proprietary to Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK.
[b]Unknown = Origin of the strain is not listed or provided by the source.
[c]*Salmonella* species.
[d]*Salmonella* Enteritidis.
[e]*Salmonella* Typhimurium.
[f]NCTC = National Collection of Type Cultures, Health Protection Agency, London, UK.
[g]TCC = Trials Culture Collection Number-Proprietary to Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK.
[h]OCC = Oxoid Culture Collection-Proprietary to Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK.
[i]Starting cfu was 800 cfu/mL for BPW with Novobiocin.
[j]Starting cfu was 1100 cfu/10mL to be detected at 7 hours in BPW with Novobiocin and 110 cfu/10mL in mTSB.
[k]Positive in BPW without novobiocin and mTSB.
[l]ATCC = American Type Culture Collection, Manassas, Virginia, USA.
[m]QL = Q Laboratories LLC, Cincinnati, Ohio, USA.
[n]Tested at independent laboratory.
[o]CCUG = Culture Collection University of Gothenburg, Gothenburg, Sweden.
[p]FSL = Cornell University Food Safety Laboratory, Ithaca, NY.
[q]Required approximately 500 cfu/mL to be detected in BPW w/ 12 mg/L Novobiocin at 7 h.

**Table 2. Exclusivity results of the Thermo Scientific SureCount Salmonella species, Typhimurium and Enteritidis Quantitative Multiplex PCR Kit (1)**

| | | | | BPW + Novobiocin Result | | | mTSB Result | | |
|---|---|---|---|---|---|---|---|---|---|
| No. | Exclusivity strain | Source | Origin | Ssp[e] | SE[f] | ST[g] | Ssp | SE | ST |
| 1 | *Enterobacter faecalis* | TCC[a] 1388 | Unknown[b] | - | - | - | - | - | - |
| 2 | *Morganella morganii* | TCC 1431 | Clinical | - | - | - | - | - | - |
| 3 | *Proteus vulgaris* | TCC 1552 | Unknown | - | - | - | - | - | - |
| 4 | *Klebsiella aerogenes* | TCC 1804 | Unknown | - | - | - | - | - | - |
| 5 | *Escherichia coli* | TCC 1809 | Unknown | - | - | - | - | - | - |
| 6 | *Citrobacter youngae* | TCC 2043 | Unknown | - | - | - | - | - | - |
| 7 | *Escherichia hermanii* | TCC 2046 | Unknown | - | - | - | - | - | - |
| 8 | *Enterobacter amnigenus- Biogroup 1* | TCC 2198 | Clinical | - | - | - | - | - | - |
| 9 | *Providencia alcalifaciens* | TCC 2209 | Clinical | - | - | - | - | - | - |
| 10 | *Yersinia enterocolitica* | TCC 2215 | Frozen prawn | - | - | - | - | - | - |
| 11 | *Edwardsiella tarda* | RDCC 2027 | Unknown | - | - | - | - | - | - |
| 12 | *Klebsiella aerogenes* | ATCC[c] 35029[d] | Unknown | - | - | - | - | - | - |
| 13 | *Hafnia alvei* | ATCC 51815[d] | Milk | - | - | - | - | - | - |
| 14 | *Morganella morganii* | ATCC 25829[d] | Human | - | - | - | - | - | - |
| 15 | *Proteus vulgaris* | ATCC 6380[d] | Clinical | - | - | - | - | - | - |
| 16 | *Yersinia enterocolitica* | ATCC 49397[d] | Clinical | - | - | - | - | - | - |

[a]TCC = Trials Culture Collection Number-Proprietary to Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK.
[b]Unknown = Origin of the strain is not listed or provided by the source.
[c]ATCC = American Type Culture Collection, Manassas, Virginia, USA.
[d]Tested by independent laboratory.
[e]*Salmonella* species.
[f]*Salmonella* Enteritidis.
[g]*Salmonella* Typhimurium.

**Table 4. Matrix study summary results: Thermo Scientific SureCount Salmonella species, Typhimurium, and Enteritidis Multiplex PCR kit vs MLG 2.05/4.11 reference method procedure for *Salmonella* (1)**

| | | | SureCount Multiplex | | MLG 4.11[d] | | | | 90% CI[g] | | 95% CI | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Matrix | Inoculating strain(s) | Cont. level[a] | $Log_{10}$ Mean[b] | $s_r$[c] | $Log_{10}$ Mean | $s_r$ | DOM[e] | SE[f] | LCL[h] | UCL[i] | LCL | UCL |
| Fresh Ground Beef 325 g | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Infantis (RDCC[j] 2133) | Low – including outlier[k] | 0.374 | 0.151 | 0.673 | 1.346 | -0.299 | 0.606 | -1.591 | 0.993 | -1.981 | 1.383 |
| | *S.* Infantis (RDCC 2133) | Low - no outlier | 0.374 | 0.151 | 0.080 | 0.281 | 0.293 | 0.156 | -0.039 | 0.626 | -0.139 | 0.726 |
| | *S.* Infantis (RDCC 2133) | Med | 1.389 | 0.216 | 1.253 | 0.301 | 0.137 | 0.166 | -0.177 | 0.451 | -0.255 | 0.529 |
| | *S.* Infantis (RDCC 2133) | High | 2.275 | 0.188 | 2.427 | 0.331 | -0.151 | 0.170 | -0.483 | 0.180 | -0.568 | 0.266 |
| Fresh Ground Turkey 325 g | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Enteritidis (RDCC 2207) | Low | 0.144 | 0.173 | 0.385 | 0.47 | -0.241 | 0.224 | -0.692 | 0.210 | -0.816 | 0.334 |
| | *S.* Enteritidis & *S.* Typhimurium (RDCC 2207 and RDCC 3897) | Med | 1.585 | 0.129 | 1.492 | 0.136 | 0.093 | 0.084 | -0.067 | 0.252 | -0.106 | 0.291 |
| | *S.* Enteritidis (RDCC 2207) | Med | 1.080 | 0.209 | 1.119 | 0.472 | -0.040 | 0.087 | -0.204 | 0.125 | -0.245 | 0.166 |
| | *S.* Typhimurium (RDCC 3897) | Med | 1.411 | 0.140 | 0.590 | 0.997 | 0.821 | 0.087 | 0.657 | 0.986 | 0.616 | 1.027 |
| | *S.* Enteritidis (RDCC 2207) | High | 2.381 | 0.195 | 2.156 | 0.232 | 0.225 | 0.135 | -0.032 | 0.481 | -0.095 | 0.545 |
| Chicken Carcass Rinse | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Typhimurium (RDCC 5586) | Low | 0.982 | 0.173 | 0.490 | 0.180 | 0.492 | 0.112 | 0.281 | 0.704 | 0.228 | 0.757 |
| | *S.* Typhimurium (RDCC 5586) | Med | 1.821 | 0.260 | 1.624 | 0.236 | 0.197 | 0.157 | -0.100 | 0.494 | -0.174 | 0.568 |
| | *S.* Typhimurium & *S.* Hadar (RDCC 5586 and RDCC 2131) | High | 2.580 | 0.109 | 2.936 | 0.376 | -0.356 | 0.175 | -0.729 | 0.017 | -0.842 | 0.130 |
| | *S.* Typhimurium (RDCC 5586) | High | 2.138 | 0.064 | 2.615 | 0.259 | -0.477 | 0.119 | -0.731 | -0.222 | -0.808 | -0.145 |
| | *S.* Hadar (RDCC 2131) | High | 2.381 | 0.144 | 1.838 | 1.124 | 0.543 | 0.507 | -0.537 | 1.623 | -0.864 | 1.949 |
| Fresh Ground Pork 325 g | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Enteritidis & S. Newport (RDCC 5793 and TCC[l] 1663) | Low | 1.105 | 0.243 | 1.518 | 0.335 | -0.413 | 0.185 | -0.763 | -0.062 | -0.850 | 0.025 |
| | *S.* Enteritidis (RDCC 5793) | Low | 1.101 | 0.246 | 0.733 | 0.758 | 0.368 | 0.087 | 0.203 | 0.533 | 0.162 | 0.573 |
| | S. Newport (TCC 1663) | Low | 1.105 | 0.243 | 1.043 | 0.126 | 0.062 | 0.087 | -0.103 | 0.227 | -0.144 | 0.268 |
| | *S.* Enteritidis (RDCC 5793) | Med[m] | 2.016 | 0.119 | 1.457 | 0.622 | 0.559 | 0.283 | -0.045 | 1.163 | -0.228 | 1.346 |
| | *S.* Enteritidis (RDCC 5793) | High | 3.083 | 0.114 | 2.662 | 0.390 | 0.421 | 0.182 | 0.034 | 0.809 | -0.084 | 0.926 |
| Fresh Ground Turkey 325 g (Independent Laboratory) | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Enteritidis (ATCC[n] 13076) | Low | 0.861 | 0.108 | 0.872 | 0.369 | -0.011 | 0.172 | -0.377 | 0.355 | -0.488 | 0.466 |
| | *S.* Enteritidis & *S.* Typhimurium (ATCC 13076 and ATCC 14028) | Med | 2.393 | 0.030 | 2.307 | 0.445 | 0.085 | 0.199 | -0.339 | 0.510 | -0.468 | 0.639 |
| | *S.* Enteritidis (ATCC 13076) | Med | 1.952 | 0.055 | 1.939 | 0.203 | 0.012 | 0.087 | -0.153 | 0.177 | -0.193 | 0.218 |
| | *S.* Typhimurium (ATCC 14028) | Med | 2.197 | 0.024 | 1.831 | 0.189 | 0.366 | 0.087 | 0.201 | 0.530 | 0.160 | 0.571 |
| | *S.* Enteritidis (ATCC 13076) | High | 2.905 | 0.115 | 2.682 | 0.235 | 0.223 | 0.117 | -0.013 | 0.459 | -0.078 | 0.524 |

[a]All matrixes were artificially contaminated. Non=non-inoculated.
[b]Mean of five replicate portions, after logarithmic transformation: Log10[cfu/g + (0.1) f], where f is the reported cfu/unit corresponding to the smallest reportable result and unit is the reported unit of measure.
[c]Repeatability standard deviation.
[d]USDA FSIS Microbiology Laboratory Guidebook (MLG) 2.05, Most Probable Number Procedure and Tables, and MLG 4.11, Isolation and Identification of *Salmonella* from Meat, Poultry, Pasteurized Egg, and Siluriformes (Fish) Products and Carcass and Environmental Sponges.
[e]Difference of means between the candidate and reference methods, analyzed using an unpaired statistical analysis.
[f]Standard error.
[g]Confidence interval.
[h]Lower confidence limit for difference of means.
[i]Upper confidence limit for difference of means.
[j]Research and Development Culture Collection - Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK
[k]Outlier from MPN reference method.
[l]Trials Culture Collection - Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK
[m]Candidate method analysed with inclusion of outlier.
[n]American Type Culture Collection - Manassas, Virginia, USA.

**Table 5. Matrix study summary results: Thermo Scientific SureCount MPN Method vs MLG 2.05/4.11 reference method procedure for *Salmonella* (1)**

| | | | SureCount MPN | | MLG 4.11[d] | | | | 90% CI[g] | | 95% CI | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Matrix | Inoculating strain(s) | Cont. level[a] | $Log_{10}$ Mean[b] | $s_r$[c] | $Log_{10}$ Mean | $s_r$ | DOM[e] | SE[f] | LCL[h] | UCL[i] | LCL | UCL |
| Fresh Ground Beef 325 g | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S. Infantis* (RDCC[j] 2133) | Low | 0.431 | 0.364 | 0.673 | 1.346 | -0.241 | 0.624 | -1.571 | 1.088 | -1.973 | 1.490 |
| | *S. Infantis* (RDCC 2133) | Low - no outlier | 0.431 | 0.364 | 0.080 | 0.281 | 0.351 | 0.215 | -0.067 | 0.768 | -0.175 | 0.877 |
| | *S. Infantis* (RDCC 2133) | Med | 1.134 | 0.165 | 1.253 | 0.301 | -0.119 | 0.153 | -0.417 | 0.179 | -0.495 | 0.256 |
| | *S. Infantis* (RDCC 2133) | High | 2.427 | 0.331 | 2.427 | 0.331 | 0.000 | 0.210 | -0.390 | 0.390 | -0.483 | 0.483 |
| Fresh Ground Turkey 325 g | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Enteritidis (RDCC 2207) | Low | 0.282 | 0.555 | 0.385 | 0.469 | -0.104 | 0.325 | -0.720 | 0.513 | -0.873 | 0.666 |
| | *S.* Enteritidis & *S.* Typhimurium (RDCC 2207 and RDCC 3897) | Med | 1.230 | 0.331 | 1.492 | 0.136 | -0.262 | 0.160 | -0.585 | 0.060 | -0.674 | 0.149 |
| | *S.* Enteritidis (RDCC 2207) | Med | 1.230 | 0.331 | 1.119 | 0.472 | 0.110 | 0.087 | -0.055 | 0.275 | -0.095 | 0.316 |
| | *S.* Typhimurium (RDCC 2207) | Med | 1.422 | 0.271 | 0.590 | 0.997 | 0.832 | 0.087 | 0.667 | 0.997 | 0.626 | 1.038 |
| | *S.* Enteritidis (RDCC 3897) | High | 2.446 | 0.420 | 2.156 | 0.232 | 0.290 | 0.215 | -0.127 | 0.707 | -0.235 | 0.815 |
| Chicken Carcass Rinse | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Typhimurium (RDCC 5586) | Low | 0.448 | 0.216 | 0.490 | 0.180 | -0.042 | 0.126 | -0.281 | 0.196 | -0.340 | 0.255 |
| | *S.* Typhimurium (RDCC 5586) | Med | 1.487 | 0.271 | 1.624 | 0.236 | -0.137 | 0.161 | -0.441 | 0.168 | -0.516 | 0.243 |
| | *S.* Typhimurium & *S. hadar* (RDCC 5586 and RDCC 2131) | High | 2.934 | 0.373 | 2.934 | 0.373 | 0.000 | 0.236 | -0.439 | 0.439 | -0.545 | 0.545 |
| | *S.* Typhimurium (RDCC 5586) | High | 2.465 | 0.392 | 2.615 | 0.259 | -0.149 | 0.210 | -0.558 | 0.259 | -0.664 | 0.365 |
| | *S.* Hadar (RDCC 2131) | High | 2.934 | 0.373 | 1.838 | 1.124 | 1.096 | 0.530 | -0.033 | 2.225 | -0.374 | 2.567 |
| Fresh Ground Pork 325 g | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Enteritidis & S. Newport (RDCC 5793 and TCC[k] 1663) | Low | 1.485 | 0.333 | 1.518 | 0.335 | -0.033 | 0.211 | -0.433 | 0.367 | -0.533 | 0.466 |
| | *S.* Enteritidis (RDCC 5793) | Low | 0.585 | 0.713 | 0.733 | 0.758 | -0.149 | 0.087 | -0.313 | 0.016 | -0.354 | 0.057 |
| | S. Newport (TCC 1663) | Low | 1.485 | 0.333 | 1.043 | 0.126 | 0.442 | 0.087 | 0.277 | 0.606 | 0.236 | 0.647 |
| | *S.* Enteritidis (RDCC 5793) | Med | 1.695 | 0.237 | 1.457 | 0.622 | 0.238 | 0.298 | -0.362 | 0.838 | -0.527 | 1.004 |
| | *S.* Enteritidis (RDCC 5793) | High | 2.480 | 0.539 | 2.660 | 0.388 | -0.180 | 0.297 | -0.743 | 0.383 | -0.883 | 0.522 |
| Fresh Ground Turkey 325 g (Independent Laboratory) | N/A | Non | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A | N/A |
| | *S.* Enteritidis (ATCC[l] 13076) | Low | 1.032 | 0.472 | 1.039 | 0.474 | -0.006 | 0.299 | -0.573 | 0.560 | -0.714 | 0.701 |
| | *S.* Enteritidis & *S.* Typhimurium (ATCC 13076 and ATCC 14028) | Med | 2.213 | 0.318 | 1.974 | 0.283 | 0.239 | 0.190 | -0.121 | 0.600 | -0.211 | 0.689 |
| | *S.* Enteritidis (ATCC 13076) | Med | 2.015 | 0.297 | 1.974 | 0.283 | 0.041 | 0.087 | -0.123 | 0.206 | -0.164 | 0.247 |
| | *S.* Typhimurium (ATCC 14028) | Med | 1.792 | 0.243 | 1.744 | 0.246 | 0.048 | 0.087 | -0.117 | 0.213 | -0.157 | 0.254 |
| | *S.* Enteritidis (ATCC 13076) | High | 2.966 | 0.169 | 2.966 | 0.169 | 0.000 | 0.107 | -0.199 | 0.199 | -0.247 | 0.247 |

[a]All matrixes were artificially contaminated. Non=non-inoculated.
[b]Mean of five replicate portions, after logarithmic transformation: Log10[cfu/g + (0.1) f], where f is the reported cfu/unit corresponding to the smallest reportable result and unit is the reported unit of measure.
[c]Repeatability standard deviation.
[d]USDA FSIS Microbiology Laboratory Guidebook (MLG) 2.05, Most Probable Number Procedure and Tables, and MLG 4.11, Isolation and Identification of *Salmonella* from Meat, Poultry, Pasteurized Egg, and Siluriformes (Fish) Products and Carcass and Environmental Sponges.
[e]Difference of means between the candidate and reference methods, analyzed using an unpaired statistical analysis.
[f]Standard error.
[g]Confidence interval.
[h]Lower confidence limit for difference of means.
[i]Upper confidence limit for difference of means.
[J]Research and Development Culture Collection - Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK
[K]Trials Culture Collection - Thermo Fisher Scientific, Microbiology Division, Basingstoke, UK
[L]American Type Culture Collection - Manassas, Virginia, USA

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