# CERTIFICATION

## AOAC® *Performance Tested*SM

Certificate No.

# 100201

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

**BAX® System PCR Assay for *Salmonella* 2**
**BAX® System X5 PCR Assay for *Salmonella***

manufactured by

**Hygiena**
**2 Boulden Circle**
**New Castle, DE 19720**
**USA**

This method has been evaluated in the AOAC® *Performance Tested Methods*SM Program and found to perform as stated by the manufacturer contingent to the comments contained in the manuscript. This certificate means that an AOAC® Certification Mark License Agreement has been executed which authorizes the manufacturer to display the AOAC *Performance Tested*SM certification mark along with the statement - "THIS METHOD'S PERFORMANCE WAS REVIEWED BY AOAC RESEARCH INSTITUTE AND WAS FOUND TO PERFORM TO THE MANUFACTURER'S SPECIFICATIONS" - on the above-mentioned method for a period of one calendar year from the date of this certificate (December 13, 2021 – December 31, 2022). Renewal may be granted at the end of one year under the rules stated in the licensing agreement.

Scott Coates

Scott Coates, Senior Director
Signature for AOAC Research Institute

December 13, 2021
Date

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**MODIFICATION NOVEMBER 2008 – F. Morgan Wallace, Siqun Wang, George Tice, Bridget Andaloro, Jack Janes, Viviana Fino, and Eugene Davis**
**MOIDIFICATION APRIL 2009 – Morgan Wallace**
**MODIFICATION JUNE 2009 – Linda X. Peng, George Tice, Morgan Wallace, Bridget Andaloro, Dawn Fallon, Lois Fleck, and Dan Delduco**
**MODIFICATION OCTOBER 2010 – Frank R. Burns, Andrew Famum, George Tice, Bridget Andaloro, Eugene Davis, Jeff Rohrbeck, and Morgan Wallace**
**MODIFICATION NOVEMBER 2015 – Jefff Rohrbeck, Alain Minelli, Eugene Davis, Gongbo Wang, Lois Fleck, Dawn Fallon, Steve Hoeizer, and Morgan Wallace**

**SUBMITTING COMPANY**
**Qualicon, Inc.**
**3531 Silverside Road**
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**Wilmington, DE 19810**

**CURRENT SPONSOR**
**Hygiena**
**2 Boulden Circle**
**New Castle, DE 19720**
**USA**

**METHOD NAMES**
BAX® System PCR Assay for *Salmonella* 2
BAX® System X5 PCR Assay for *Salmonella*
Formerly known as:
DuPont™ BAX® System PCR Assay for *Salmonella* 2
DuPont™ BAX® System X5 PCR Assay for *Salmonella*
RETIRED: BAX® System PCR Assay for *Salmonella* formerly known as DuPont™ BAX® System PCR Assay for *Salmonella*

**CATALOG NUMBERS**
BAX® System Salmonella 2 Kit KIT2011 (D14368501)
BAX® System X5 Assay KIT2025 (D15407187)
MP Media MED2003 (D12404925)

RETIRED: BAX® System Salmonella 1 Kit KIT2012 (D11000133)

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Original Validation: Campden & Chorleywood Food Research Association
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[4] Consultant, Loganville, Georgia, USA
[5] Modifications: November 2008, April 2009, June 2009
[6] Modifications: October 2010, November 2015

**APPLICABILITY OF METHOD**
**Target organism – *Salmonella***

**Matrixes – Original Validation - (25 g) black pepper, custard, 2% milk, chilled ready meal, chipped ham, chocolate, cooked chicken, raw ground chicken, hot dogs, nonfat dry milk, orange juice, peanut butter, alfalfa sprouts, cooked fish, prawns, macaroni, pizza dough, frozen peas, dry pet food, soy protein flour, ground beef, spinach, lettuce, liquid egg, cream cheese, peanut butter (ERV 2009)**
**(65 g) beef trim**
**(4 x 4 in sponge) concrete, stainless steel, ceramic tile, plastic, epoxy material affixed to a support matrix**

**Performance claims – Method performs equivalent to reference methods.**

**REFERENCE METHODS**
**USDA FSIS, Office of Public Health and Science. Microbiology Laboratory Guidebook [Internet]. Washington: The Dept; c1998 [rev 2001 January 10; cited 2002 August 27]. Chapter 4, Isolation and Identification of *Salmonella* from Meat, Poultry, and Egg Products [about 14 screens] (6)**

**US FDA, CFSAN. Bacteriological Analytical Manual Online [Internet]. Washington: The Admin; c1998 [rev 2001 October; cited 2002 August 27]. Chapter 5, *Salmonella* [about 12 screens] (7)**

| ORIGINAL CERTIFICATION DATE | CERTIFICATION RENEWAL RECORD |
|---|---|
| November 2002 | Renewed annually through December 2022. |

| METHOD MODIFICATION RECORD | SUMMARY OF MODIFICATION |
|---|---|
| 1. 2006 | 1. Addition of Q7 BAX® Instrument. |
| 2. November 2008 | 2. Additional of Environmental sponges using BAX® and BAX® Q7. |
| 3. April 2009 | 3. Matrix Extension to include Peanut Butter. |
| 4. June 2009 | 4. Screening of beef and produce using BAX® System PCR Assay for *E. coli* O157:H7 MP enrichment media and protocol. |
| 5. October 2010 | 5. Addition of hot start functionality. |
| 6. September 2013 | 6. Inclusion of DuPont™ Thermal Block for automated sample lysis. |
| 7. November 2015 | 7. Certification of BAX® X5 instrument. |
| 8. March 2017 Level 1 | 8. Name change from DuPont Nutrition & Health to Qualicon Diagnostics LLC., a Hygiena company. |
| 9. January 2018 Level 1 | 9. Editorial updates to Inserts, manuals, and labels to Hygiena. |
| 10. May 2019 Level 1 | 10. Editorial updates to inserts and corporate address. |
| 11. December 2019 Level 1 | 11. Editorial/clerical changes. |
| 12. December 2021 Level 1 | 12. Editorial and retired BAX® System Salmonella 1 Kit KIT2012 (D11000133). |

| Under this AOAC® *Performance Tested*SM License Number, 100201 this method is distributed by: | Under this AOAC® *Performance Tested*SM License Number, 100201 this method is distributed as: |
|---|---|
| NONE | NONE |

**PRINIPLE OF THE METHOD (1)**

The BAX® System uses the Polymerase Chain Reaction (PCR) to amplify a specific fragment of bacterial DNA, which is stable and unaffected by growth environment. The fragment is a genetic sequence that is unique to *Salmonella*, thus providing a highly reliable indicator that the organism is present. The automated BAX System then uses fluorescent detection [2] to analyze PCR product for positive or negative results. (The non-automated BAX system, which uses gel-detection methods, has already received AOAC-RI Performance Tested certification [3].)

PCR offers the potential for rapid and definitive detection of *Salmonella* and has been demonstrated to be effective for other organisms [5,6]. In order to realize this potential, however, some of the drawbacks of PCR must be overcome. The first of these is the complexity of the PCR procedure itself. The second is the potential for false positives due to contamination with one or more molecules of amplified PCR product in the laboratory. This most often happens during the many pipetting steps associated with traditional PCR. The BAX System with automated detection alleviates these drawbacks by combining the reagents needed for the PCR process into a stable, dry, manufactured tablet already packaged inside the PCR tubes. After amplification, these tubes remain sealed for the detection phase, thus significantly reducing the potential for contamination.

The BAX System combines primers, polymerase and nucleotides needed for PCR into a single tablet. The specificity of a PCR assay is determined by the DNA sequences of the primers employed. The basis for primer design for the BAX® System assay is a collection of 1572 strains of *Salmonella*, which have been analyzed by ribotyping [5]. This large, well-characterized culture collection has facilitated the design of primers that can detect all strains of *Salmonella*, while remaining highly specific for *Salmonella* and no other genus.

The product of the PCR reaction is automatically analyzed by fluorescent detection. Each PCR tablet contains a fluorescent dye, which binds with double-stranded DNA and emits a signal in response to excitation light. During the automated detection phase, the temperature of the samples is slowly increased to denature the DNA, which releases the dye causing a drop in emission signal. The temperature of denaturation and the magnitude of fluorescent signal change are measured, and the BAX® system software analyzes those data to determine a positive or negative result.

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

The method comparison studies, both internal and external, support equivalence of the BAX system method to the reference method for detection of Salmonella in the 19 foods analyzed from the same original pre-enrichment. All chi-square values were below the required level of 3.84 indicating no statistically significant difference at $P \leq 0.05$.

The details of the performance indicator calculations are shown below:

Accuracy is the fraction of confirmed positive samples that are correctly identified by the detection method. In this study, confirmation is the reference method.
Method positives /confirmed positives = 527/530 = 99.4%
False negative rate is calculated as BAX (-), Ref (+) samples / Total Ref (+) samples = 7/530 = 1.3 %
False positive rate is calculated as BAX (+), Ref (-) samples / Total Ref (-) samples = 4/245 = 1.6 %
Sensitivity is calculated as 1 – false negative rate = 100 – 1.3 = 98.7 %.
Specificity is calculated as 1 – false positive rate = 100 – 1.6 = 98.3 %.

Alfalfa sprouts were analyzed using a Z-test for Differences in Means of Two Populations [13] with a 5% significance level. The results indicate no significant difference in recovery rate.

The independent lab method comparison was done for ground beef and liquid egg. Again there was no significant difference by chi-square analysis indicating equivalent detection sensitivity and specificity.

The inclusivity/exclusivity study showed 100% agreement with expected results for the 194 Salmonella strains and the 35 non-Salmonella strains.

The ruggedness study demonstrated that the BAX® system was not sensitive to changes in lysis temperature, lysis sample volume, nor PCR sample volume. Each of the variations tested was beyond the tolerances indicated for the assay, i.e., +/- 1°C for the heaters and +/-10% for the volumes.

**Table 3 . Method performances (1)**

| | | | Samples Positive | | | | | Sensitivity rate [c] | | Incidence of false negatives among total positive samples, % [d] | | Specificity rate [e] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Total | BAX system | | Ref. [a] | | | | | | |
| Sample | Level | MPN/g | | Pres. [a] | Conf. [a] | | Chi sq [b] | BAX® | Ref. | BAX | Ref. | BAX |
| Alfalfa sprouts | Low | 1.08 | 20 | 15 | 15 | 4 | 0 | 100 | 26.7 | 0 | 73.3 | 100 |
| | High | 5.25 | 20 | 18 | 18 | 20 | 0.5 | 90 | 100 | 10 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Ground Beef | Low | 1.08 | 20 | 18 | 18 | 19 | 0 | 94.7 | 100 | 5.3 | 0 | 100 |
| | High | 11.5 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Cooked chicken | Low | 1.08 | 20 | 13 | 13 | 13 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 2.3 | 20 | 17 | 17 | 17 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Custard | Low | 6 | 20 | 14 | 14 | 15 | 0 | 93.3 | 100 | 6.7 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Chilled ready meal | Low | 6 | 20 | 18 | 18 | 18 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 23.75 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Chocolate | Low | 0.575 | 20 | 7 | 7 | 8 | 0 | 87.5 | 100 | 12.5 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Liquid eggs | Low | 1.075 | 20 | 18 | 18 | 18 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Cooked fish | Low | 2.3 | 20 | 13 | 13 | 13 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 27.5 | 20 | 17 | 17 | 17 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Frankfurters | Low | 2.3 | 20 | 10 | 10 | 10 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 0.375 | 20 | 18 | 18 | 18 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Chipped ham | Low | 2.3 | 20 | 12 | 12 | 12 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 2.3 | 20 | 17 | 17 | 17 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | 0 | – | – | – | – | – |

**Table 3. Method performances (cont'd.) (1)**

| | | | Samples Positive | | | | | Sensitivity rate [c] | | Incidence of false negatives among total positive samples, % [d] | | Specificity rate [e] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Total | BAX system | | Ref. [a] | | | | | | |
| Sample | Level | MPN/g | | Pres.[a] | Conf. [a] | | Chi sq [b] | BAX® | Ref. | BAX | Ref. | BAX |
| Elbow macaroni | Low | 1.08 | 20 | 11 | 11 | 12 | 0 | 91.7 | 100 | 8.3 | 0 | 100 |
| | High | 11.5 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | 0 | – | – | – | – | – |
| Liquid milk | Low | 1.08 | 20 | 9 | 9 | 9 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 10.75 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Nonfat dry milk | Low | 1.08 | 20 | 15 | 15 | 16 | 0 | 93.8 | 100 | 6.2 | 0 | 100 |
| | High | 2.33 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Orange juice | Low | 6 | 20 | 18 | 18 | 19 | 0 | 94.7 | 100 | 5.3 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Peanut butter | Low | 1.88 | 20 | 17 | 17 | 17 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 11.5 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Frozen peas | Low | 0.53 | 20 | 5 | 5 | 5 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 0.98 | 20 | 12 | 12 | 13 | 0 | 92.3 | 100 | 7.7 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Black pepper | Low | 6 | 20 | 19 | 19 | 19 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 10.75 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | 0 | – | – | – | – | – |
| Dry pet food | Low | 0.575 | 20 | 13 | 13 | 13 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 2.325 | 20 | 20 | 20 | 20 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Pizza dough | Low | 0.95 | 20 | 13 | 13 | 11 | 0 | 100 | 84.6 | 0 | 15.4 | 100 |
| | High | 23.25 | 20 | 20 | 20 | 18 | 0 | 100 | 90.0 | 0 | 10.0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |
| Seafood - prawns | Low | 0.375 | 20 | 5 | 5 | 5 | 0 | 100 | 100 | 0 | 0 | 100 |
| | High | 1.075 | 20 | 15 | 15 | 15 | 0 | 100 | 100 | 0 | 0 | 100 |
| | Control | < 0.003 | 0 | 0 | 0 | 0 | – | – | – | – | – | – |

[a]Pres. = presumptive, Conf. = confirmed

[a]Chi Sq = Chi square as defined by MCNemar – $(|a-b|-1)^2/(a+b)$ where a = samples positive by BAX and negative by the reference method, and b = samples negative by BAX and positive by the reference method. A chi square value greater than 3.84 indicates significance at $P<0.05$.

[c]Sensitivity rate is defined as the total number of positive test samples among the confirmed positive samples for a method divided by the total number of confirmed positive samples by either method.

[d]Incidence of false negatives = 100 – sensitivity rate.

[e]Specificity rate is defined as the number of negative samples by the BAX method divided by the number of confirmed negative samples by both BAX and reference method.

**Table 4. BAX System Inclusivity (1)**

| Strain number | Salmonella serovar | O-Group | Source | BAX result | Strain number | Salmonella serovar | O-Group | Source | BAX result |
|---|---|---|---|---|---|---|---|---|---|
| 919 | *S. paratyphi A* | A | unknown | + | 1331 | *S. berta* | D1 | sausages | + |
| 1548 | *S. abony* | B | unknown | + | 1641 | *S. durban* | D1 | feces | + |
| 1335 | *S. agona* | B | chicken | + | 1236 | *S. enteritidis* | D1 | duck | + |
| 1360 | *S. agona* | B | chicken | + | 741 | *S. gallinarum* | D1 | unknown | + |
| 1531 | *S. altendorf* | B | unknown | + | 1691 | *S. inverness* | D1 | feces | + |
| 1553 | *S. ball* | B | unknown | + | 1701 | *S. miami* | D1 | unknown | + |
| 1338 | *S. brandenburg* | B | milk | + | 966 | *S. napoli* | D1 | unknown | + |
| 1435 | *S. brandenburg* | B | unknown | + | 1476 | *S. napoli* | D1 | unknown | + |
| 964 | *S. bredeney* | B | raw chicken | + | 1482 | *S. pullorum* | D1 | chicken liver | + |
| 1356 | *S. bredeney* | B | pork | + | 1507 | *S. pullorum* | D1 | unknown | + |
| 1238 | *S. heidelberg* | B | chicken | + | 4029 | *S. dublin* | D1 | unknown | + |
| 1244 | *S. heidelberg* | B | unknown | + | 3792 | *S. enteritidis* | D1 | chicken | + |
| 1655 | *S. reading* | B | unknown | + | 3794 | *S. enteritidis* | D1 | chicken | + |
| 1330 | *S. saintpaul* | B | bean sprouts | + | 4013 | *S. enteritidis* | D1 | chicken | + |
| 1372 | *S. saintpaul* | B | milk powder | + | 4020 | *S. enteritidis* | D1 | chicken | + |
| 1478 | *S. saintpaul* | B | unknown | + | 4022 | *S. enteritidis* | D1 | mayonaise | + |
| 1237 | *S. typhimurium* | B | raw egg | + | 2294 | *S. pullorum* | D1 | unknown | + |
| 1467 | *S. typhimurium* | B | unknown | + | 1558 | *S. canastel* | D1 | feed | + |
| 4084 | *S. africana* | B | unknown | + | 1552 | *S. alabama* | D2 | unknown | + |
| 3920 | *S. agona* | B | environmental | + | 1530 | *S. amager* | E1 | unknown | + |
| 4033 | *S. brandenburg* | B | unknown | + | 1649 | *S. lexington* | E1 | unknown | + |
| 2316 | *S. bredeney* | B | coconut | + | 1652 | *S. london* | E1 | unknown | + |
| 3461 | *S. heidelberg* | B | egg yolk | + | 1705 | *S. muenster* | E1 | unknown | + |
| 3868 | *S. heidelberg* | B | poultry feed | + | 3851 | *S. anatum* | E1 | chicken | + |
| 3871 | *S. heidelberg* | B | chicken | + | 3907 | *S. anatum* | E1 | poultry feed | + |
| 4038 | *S. heidelberg* | B | chicken | + | 3919 | *S. anatum* | E1 | chicken | + |
| 3852 | *S. indiana* | B | poultry feed | + | 3922 | *S. anatum* | E1 | environmental | + |
| 3898 | *S. neumuenster* | B | poultry feed | + | 4025 | *S. anatum* | E1 | chicken | + |
| 2645 | *S. reading* | B | turkey intestine | + | 2189 | *S. give* | E1 | unknown | + |
| 2637 | *S. swarzengrund* | B | chicken | + | 2353 | *S. kristianstad* | E1 | unknown | + |
| 2641 | *S. swarzengrund* | B | chicken | + | 3883 | *S. lexington* | E1 | poultry feed | + |
| 2647 | *S. swarzengrund* | B | llama | + | 2676 | *S. orion* | E1 | chicken feed | + |
| 3537 | *S. typhimurium* | B | unknown | + | 4012 | *S. orion* | E1 | chicken | + |
| 3854 | *S. typhimurium* | B | poultry feed | + | 1491 | *S. weltevreden* | E1 | prawns | + |
| 3858 | *S. typhimurium* | B | environmental | + | 1248 | *S. anatum* | E2 | unknown | + |
| 4017 | *S. typhimurium* | B | unknown | + | 1332 | *S. anatum* | E2 | shrimp | + |
| 1526 | *S. austin* | C1 | unknown | + | 1365 | *S. anatum* | E2 | paprika | + |
| 1510 | *S. bareilly* | C1 | unknown | + | 1366 | *S. anatum* | E2 | chicken | + |
| 2181 | *S. bareilly* | C1 | unknown | + | 1085 | *S. binza* | E2 | dried spice | + |
| 1329 | *S. branderup* | C1 | dried egg | + | 1707 | *S. newbrunswick* | E2 | unknown | + |
| 1628 | *S. colorado* | C1 | unknown | + | 3903 | *S. binza* | E2 | poultry feed | + |
| 1235 | *S. infantis* | C1 | avian meal | + | 2349 | *S. drypool* | E2 | unknown | + |
| 1255 | *S. montevideo* | C1 | egg | + | 4610 | *S. newbrunswick* | E2 | cereal | + |
| 1260 | *S. montevideo* | C1 | animal feed | + | 6177 | *S. arkansas* | E3 | chicken giblets | + |
| 1267 | *S. montevideo* | C1 | chicken | + | 2639 | *S. thomasville* | E3 | turkey intestine | + |
| 1274 | *S. montevideo* | C1 | animal feed | + | 3924 | *S. thomasville* | E3 | poultry feed | + |
| 1492 | *S. montevideo* | C1 | unknown | + | 740 | *S. senftenberg* | E4 | unknown | + |
| 1710 | *S. oranienburg* | C1 | unknown | + | 3882 | *S. broughton* | E4 | poultry feed | + |
| 1336 | *S. thompson* | C1 | chicken | + | 1624 | *S. chandans* | F | unknown | + |
| 3458 | *S. braenderup* | C1 | egg albumin | + | 1712 | *S. pretoria* | F | pig | + |
| 3003 | *S. choleraesuis* | C1 | unknown | + | 2220 | *S. senftenberg* | F | unknown | + |
| 3984 | *S. choleraesuis* | C1 | gallbladder | + | 2340 | *S. senftenberg* | F | coconut | + |
| 3985 | *S. choleraesuis* | C1 | unknown | + | 1562 | *S. montgomery* | F | unknown | + |
| 3986 | *S. choleraesuis* | C1 | unknown | + | 1489 | *S. poona* | G1 | clinical isolate | + |
| 3987 | *S. choleraesuis* | C1 | unknown | + | 1632 | *S. cubana* | G2 | chicken | + |
| 3869 | *S. infantis* | C1 | poultry feed | + | 1251 | *S. kedougou* | G2 | turkey | + |
| 3902 | *S. infantis* | C1 | poultry feed | + | 1703 | *S. mississippi* | G2 | feces | + |
| 4028 | *S. infantis* | C1 | liquid egg | + | 2245 | *S. havana* | G2 | pancake | + |
| 2263 | *S. lille* | C1 | pancake | + | 2271 | *S. havana* | G2 | pet food | + |
| 3155 | *S. lille* | C1 | environmental | + | 3876 | *S. havana* | G2 | poultry feed | + |
| 4036 | *S. livingstone* | C1 | chicken | + | 4039 | *S. kedougou* | G2 | chicken | + |
| 3870 | *S. mbandaka* | C1 | poultry feed | + | 1509 | *S. bovismorbidificans* | H | unknown | + |
| 3878 | *S. mbandaka* | C1 | poultry feed | + | 1621 | *S. carrau* | H | unknown | + |
| 6206 | *S. mbandaka* | C1 | chicken giblets | + | 1686 | *S. fayed* | H | unknown | + |
| 3760 | *S. montevideo* | C1 | unknown | + | 1620 | *S. carmel* | J | unknown | + |
| 4027 | *S. montevideo* | C1 | chicken | + | 1622 | *S. cerro* | K | unknown | + |
| 3892 | *S. ohio* | C1 | poultry feed | + | 3459 | *S. cerro* | K | whole egg | + |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| 3863 | *S. othmarschen* | C1 | environmental | + | 3460 | *S. cerro* | K | whole egg | + |
| 3874 | *S. othmarschen* | C1 | poultry feed | + | 3926 | *S. cerro* | K | poultry feed | + |
| 3536 | *S. tennessee* | C1 | unknown | + | 1557 | *S. chicago* | M | unknown | + |
| 6687 | *S. tennessee* | C1 | sesame seeds | + | 6696 | *S. cotham* | M | unknown | + |
| 3866 | *S. thompson* | C1 | environmental | + | 1469 | *S. ealing* | O | dried milk | + |
| 738 | *S. virchow* | C1 | unknown | + | 1543 | *S. adelaide* | O | unknown | + |
| 1257 | *S. virchow* | C1 | turkey | + | 3884 | *S. adelaide* | O | poultry feed | + |
| 1551 | *S. aequatoria* | C2 | unknown | + | 3885 | *S. adelaide* | O | poultry feed | + |
| 1521 | *S. amersfoort* | C2 | unknown | + | 1684 | *S. emmastad* | P | unknown | + |
| 1343 | *S. blockley* | C2 | environment | + | 1429 | *S. anfo* | Q | box meat | + |
| 1344 | *S. blockley* | C2 | chicken | + | 1623 | *S. champaign* | Q | chicken liver | + |
| 1424 | *S. manchester* | C2 | yeast | + | 2313 | *S. wandsworth* | Q | unknown | + |
| 1653 | *S. manhatan* | C2 | unknown | + | 1695 | *S. johannesburg* | R | unknown | + |
| 1261 | *S. newport* | C2 | duck | + | 1608 | *S. seminole* | R | snake feces | + |
| 1262 | *S. newport* | C2 | cotton seed | + | 2346 | *S. vietnam* | S | unknown | + |
| 1481 | *S. newport* | C2 | raw burger | + | 1523 | *S. berkeley* | U | diseased turkey | + |
| 3916 | *S. hadar* | C2 | chicken | + | 1680 | *S. dugbe* | W | unknown | + |
| 3917 | *S. hadar* | C2 | chicken | + | 1569 | *S.sp.* | ND | unknown | + |
| 3918 | *S. hadar* | C2 | chicken | + | 1568 | *S. arizona* | ND | unknown | + |
| 2673 | *S. manhattan* | C2 | avian | + | 1525 | *S. betioky* | ND | unknown | + |
| 4014 | *S. newport* | C2 | chicken | + | 1554 | *S. branalia* | ND | unknown | + |
| 6174 | *S. newport* | C2 | chicken giblets | + | 1535 | *S. brookfield* | ND | frog | + |
| 1231 | *S. hadar* | C3 | turkey | + | 1775 | *S. sp.* | ND | unknown | + |
| 3157 | *S. corvalis* | C3 | environmental | + | 1616 | *S. houten* | ND | bird feces | + |
| 3915 | *S. haardt* | C3 | chicken | + | 1566 | *S. sp.* | ND | rattlesnake skin | + |
| 3921 | *S. haardt* | C3 | environmental | + | 2376 | *S. sculcoates* | ND | unknown | + |
| 2195 | *S. kentucky* | C3 | unknown | + | 2379 | *S. sp.* | ND | unknown | + |
| 6250 | *S. santiago* | C3 | dried onion | + | 2380 | *S. sp.* | ND | unknown | + |
| 6253 | *S. santiago* | C3 | dried onion | + | 3804 | *S. sp.* | ND | unknown | + |
| 2229 | *S. theilalle* | C4 | unknown | + | 3805 | *S. sp.* | ND | unknown | + |

**Table 5. BAX System exclusivity (1)**

| Strain DD# | Strain ID | Source | BAX result | Strain DD# | Strain ID | Source | BAX result |
|---|---|---|---|---|---|---|---|
| 715 | *Bacillus cereus* | unknown | - | 6247 | *Klebsiella spp.* | avian | - |
| 6712 | *Citrobacter freundii* | vegetables | - | 659 | *Lactococcus lactis* | unknown | - |
| 8990 | *Citrobacter freundii* | soil | - | 6121 | *Proteus mirabilis* | avian | - |
| 2560 | *Citrobacter freundii* | throat | - | 2357 | *Proteus mirabilis* | poultry | - |
| 3097 | *Citrobacter freundii* | Cake mix | - | 6921 | *Proteus mirabilis* | chicken entrails | - |
| 1074 | *Enterobacter cloacae* | skim milk powder | - | 960 | *Providencia alcalifaciens* | prawns | - |
| 3108 | *Enterobacter cloacae* | cereal | - | 295 | *Pseudomonas aeruginosa* | unknown | - |
| 3112 | *Enterobacter cloacae* | unknown | - | 9022 | *Pseudomonas aeruginosa* | soil | - |
| 1458 | *Escherichia coli* | human bloody diarrhea | - | 569 | *Pseudomonas fluorescens* | pre-filter tank | - |
| 2443 | *Escherichia coli* | unknown | - | 7083 | *Serratia marcesens* | unknown | - |
| 2513 | *Escherichia coli* | unknown | - | 700 | *Shigella sonnei* | unknown | - |
| 3785 | *Escherichia coli* | unknown | - | 6723 | *Shigella sonnei* | sandwich | - |
| 3786 | *Escherichia coli* | meninges - baby | - | 6832 | *Shigella sonnei* | unknown | - |
| 3789 | *Escherichia coli* | unknown | - | 1396 | *Staphylococcus aureus* | margarine | - |
| 3790 | *Escherichia coli* | unknown | - | 765 | *Staphylococcus warneri* | chicken | - |
| 6719 | *Escherichia hermannii* | sesame seeds | - | 3990 | *Stomatococcus mucilaginosus* | pharynx | - |
| 6588 | *Hafnia alvei* | raw vegetables | - | 7120 | *Yersinia enterocolitica* | unknown | - |
| 6708 | *Hafnia alvei* | vegetables | - | | | | |

**DISSCUSSION OF MODIFICATION APPROVED 2006 (8)**

The data in these studies, within their statistical uncertainty, support the product claims and demonstrate that the BAX ® system *Salmonella* pcr test kit when used with the Q7 instrument is comparable to standard reference methods for detecting *Salmonella* in a variety of foods. All chi-square values were below the required level of 3.84 indicating no statistically significant difference at $P \leq 0.05$. In addition, the assay demonstrated excellent inclusivity and exclusivity with all *Salmonella* isolates screened testing positive and all non-*Salmonella* isolates testing negative.

| Table 1. Inclusivity test panel for BAX Salmonella test kit using the Q7 Instrument (8) | | | | |
|---|---|---|---|---|
| DD# | Other Designation (Non-Qualicon Origin) | Serotype | Source (where available) | Result |
| 586 | | *Salmonella* Typhimurium | Chicken hearts and livers | **POS** |
| 706 | ATCC 13076 | *Salmonella* Enteritidis | Human clinical | **POS** |
| 707 | ATCC 6962 | *Salmonella* Newport | Human clinical fatal | **POS** |
| 725 | ATCC 13314 | *Salmonella* Arizonae | NCTC | **POS** |
| 917 | ATCC 13312 | *Salmonella* Choleraesuis | NCTC | **POS** |
| 1084 | | *Salmonella* Typhimurium | Unknown | **POS** |
| 1261 | | *Salmonella* Newport | Duck | **POS** |
| 1329 | | *Salmonella* Branderup | Dried egg | **POS** |
| 1330 | | *Salmonella* Saintpaul | Bean sprouts | **POS** |
| 1331 | | *Salmonella* Berta | Sausages | **POS** |
| 1332 | | *Salmonella* Anatum | Shrimp | **POS** |
| 1335 | | *Salmonella* Agona | Chicken | **POS** |
| 1336 | | *Salmonella* Thompson | Chicken | **POS** |
| 1338 | | *Salmonella* Brandenburg | Milk | **POS** |
| 1343 | | *Salmonella* Blockley | Environment | **POS** |
| 1344 | | *Salmonella* Blockley | Chicken | **POS** |
| 1356 | | *Salmonella* Bredeney | Pork | **POS** |
| 1360 | | *Salmonella* Agona | Chicken | **POS** |
| 1365 | | *Salmonella* Anatum | Paprika | **POS** |
| 1366 | | *Salmonella* Anatum | Chicken | **POS** |
| 1372 | | *Salmonella* Saintpaul | Milk powder | **POS** |
| 1424 | | *Salmonella* Manchester | Yeast | **POS** |
| 1429 | | *Salmonella* Anfo | Box meat | **POS** |
| 1435 | | *Salmonella* Brandenburg | Unknown | **POS** |
| 1483 | CRA 728[1] | *Salmonella* Hadar | Unknown | **POS** |
| 1492 | CRA 72[1] | *Salmonella* Montevideo | Unknown | **POS** |
| 1662 | | *Salmonella* Tranoroa | Unknown | **POS** |
| 1711 | | *Salmonella* Pomona | Turkey intestine | **POS** |
| 1773 | ATCC 43975 | *Salmonella* Brookfield | Unknown | **POS** |
| 1777 | ATCC 43972 | *Salmonella* Salamae | ATCC | **POS** |
| 2181 | | *Salmonella* Bareilly | Unknown | **POS** |
| 2274 | | *Salmonella* Anatum | ATCC | **POS** |
| 2317 | | *Salmonella* Othmarschen | Unknown | **POS** |
| 2341 | | *Salmonella* Barry | Unknown | **POS** |
| 2380 | | *Salmonella* Sya | Unknown | **POS** |
| 2628 | ATCC 9263 | *Salmonella* Kentucky | ATCC | **POS** |
| 2791 | | *Salmonella* Binza | Chicken | **POS** |
| 2824 | | *Salmonella* Cerro | Chicken | **POS** |
| 2992 | | *Salmonella* Lille | Unknown | **POS** |
| 3017 | | *Salmonella* Dublin | Unknown | **POS** |
| 3019 | | *Salmonella* Dublin | Unknown | **POS** |
| 3096 | | *Salmonella* Tennessee | Unknown | **POS** |
| 3984 | ATCC 8759 | *Salmonella* Choleraesuis | Gall bladder | **POS** |
| 3988 | ATCC 13428 | *Salmonella* Choleraesuis | ATCC | **POS** |
| 4022 | | *Salmonella* Enteritidis | Mayonnaise | **POS** |
| 4102 | | *Salmonella* Species | Nuts | **POS** |
| 5533 | | *Salmonella* Infantis | Thyme | **POS** |
| 7005 | | *Salmonella* Dublin | CMCC | **POS** |
| 8034 | | *Salmonella* Species | Raw chicken | **POS** |
| 8047 | | *Salmonella* Cubana | Oats | **POS** |

[1] Campden & Chorleywood Food Research Association Culture Collection (CRA)

| Table 2. Exclusivity test panel for BAX® *Salmonella* test kit using the Q7 Instrument (8) | | | | |
|---|---|---|---|---|
| DD# | Other Designation (Non-Qualicon Origin) | Serotype | Source (where available) | Result |
| 295 | | *Pseudomonas aeruginosa* | Unknown | **NEG** |
| 569 | ATCC 13525 | *Pseudomonas fluorescens* | Pre-filter tank | **NEG** |
| 715 | ATCC 14579 | *Bacillus cereus* | Unknown | **NEG** |
| 765 | | *Staphylococcus warneri* | Chicken | **NEG** |
| 846 | ATCC 29907 | *Escherichia blattae* | Insect | **NEG** |
| 960 | | *Providencia alcalifaciens* | Prawns | **NEG** |
| 1074 | | *Enterobacter cloacae* | Skim milk powder | **NEG** |
| 1396 | | *Staphylococcus aureus* | Margarine | **NEG** |
| 2357 | | *Proteus mirabilis* | Poultry | **NEG** |
| 2443 | | *E.coli O157 :H19* | PSU Reference Lab | **NEG** |
| 2560 | ATCC 25408 | *Citrobacter diversus* | Throat Swab | **NEG** |
| 3097 | | *Citrobacter freundii* | Cake mix | **NEG** |
| 3790 | | *Escherichia coli* | Unknown | **NEG** |
| 3990 | ATCC 25296 | *Stomatococcus mucilaginosus* | Pharynx | **NEG** |
| 6121 | | *Proteus mirabilis* | Avian | **NEG** |
| 6588 | | *Hafnia alvei* | Raw vegetables | **NEG** |
| 6719 | | *Escherichia hermanii* | Sesame seeds | **NEG** |
| 6832 | | *Shigella sonnei* | Unknown | **NEG** |
| 7083 | | *Serratia marcesens* | Unknown | **NEG** |
| 7120 | | *Yersinia enterocolitica* | Unknown | **NEG** |

| Table 3. Results of method comparison on 3 food matrixes (8) | | | | | |
|---|---|---|---|---|---|
| **Frankfurters** | | BAX pos | BAX neg | Total | |
| MPN = 3.8 / 25g | MLG pos | 18 | 0 | 18 | |
| APC = TFTC | MLG neg | 0 | 2 | 2 | Chi-square = 0.00 |
| Est. APC[1] = 0 | Total | 18 | 2 | 20 | Sensitivity = 100% |
| Negative Controls | MLG pos | 0 | 0 | 0 | Specificity = 100% |
| | MLG neg | 0 | 5 | 5 | False positive = 0% |
| | Total | 0 | 5 | 5 | False negative = 0% |
| **Raw Ground Chicken** | | BAX pos | BAX neg | Total | Chi-square = 0.00 |
| MPN = 1.1 / 25g | MLG pos | 10 | 0 | 10 | Sensitivity = 100% |
| APC[2] = 2.5 X $10^6$ cfu/g | MLG neg | 0 | 10 | 10 | Specificity = 100% |
| APC[3] = 1.8 X $10^7$ cfu/g | Total | 10 | 10 | 20 | False positive = 0% |
| | | | | | False negative = 0% |
| **Orange Juice** | | BAX pos | BAX neg | Total | |
| MPN = 0.38 / 25 ml | BAM pos | 11 | 0 | 11 | |
| APC = TFTC | BAM neg | 0 | 9 | 9 | Chi-square = 0.00 |
| Est. APC = 15 cfu/ml | Total | 11 | 9 | 20 | Sensitivity = 100% |
| Negative Controls | MLG pos | 0 | 0 | 0 | Specificity = 100% |
| | MLG neg | 0 | 5 | 5 | False positive = 0% |
| | Total | 0 | 5 | 5 | False negative = 0% |

[1] Est. APC – Results reported are from aerobic plate count plates that have too few colonies to accurately count from the most concentrated dilution (the 1:10 initial dilution in Preenrichment broth).
[2] At prescreening for *Salmonella*.
[3] At start of experiment.

**DISCUSSION OF MODIFICATION APPROVED JUNE 2008 (9)**

For laboratory contaminated samples, all LB enrichments that would ultimately be confirmed as containing *Salmonella* through the reference method protocol were positive by the BAX test method (tables 3a and 3c). No statistically significant differences were obtained when comparing the BAX ® test kit and the FDA-BAM reference method for any of the studies performed except for the naturally contaminated LB protocol with the standard *Salmonella* re-growth procedure. This protocol performed significantly differently (with a chi-square value of > 3.84 indicative of significance at the 95% confidence level) than the reference method. A modified re-growth protocol in which 1 ml of the LB enrichment is added to 9 ml of BHI and the re-growth time is extended from 3 to 5 hours was found to improve the method performance to give statistical, though not absolute, equivalence. In order to achieve absolute, as opposed to statistical, equivalence BPW enrichment with the standard re-growth protocol was used to support *Salmonella* growth to BAX detectable levels in these high background microflora sample types (Tables 3b and 3d). Studies using BPW and LB for the detection of *Salmonella* from artificially contaminated "clean" surfaces, however, indicated better performance with LB (data not shown but available upon request). Qualicon thus recommends that in testing from the less microbiologically complex finished product side of manufacturing facilities (surfaces similar to the artificially contaminated samples), LB enrichment should be used for best results, but when testing for the presence of *Salmonella* from concrete surfaces with heavy levels of competing microbial flora from the raw ingredients side of a manufacturing facility, BPW should be used. Environmental sponges collected from surface types other than concrete and containing high levels of non-*Salmonella* competing microbial flora would likely behave in a similar fashion; however, validation studies should be performed by end-users of this test kit on their particular surface type(s) other than concrete to determine whether the conclusions of this study are applicable to their matrixes.

**Table 3a. Method performance for the detection of *Salmonella* species from environmental surfaces by the BAX ® System. Artificially inoculated surfaces. BAX testing was performed from the same LB enrichments used for subsequent culture confirmation as per the FDA-BAM. (9)**

| Surface | Level (cfu applied per analytical unit) | Total Samples (each treatment) | BAX Presumptive (# positive) | BAX Confirmed (# positive) | Reference Method (# positive) | Sensitivity %[1] | Specificity %[2] | False Negative %[3] | False Positive %[4] | $X^2$ Value [5] |
|---|---|---|---|---|---|---|---|---|---|---|
| Ceramic Tile | 114 | 20 | 15 | 15 | 15 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Plastic | 162 | 20 | 13 | 13 | 13 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Stainless Steel | 50 | 20 | 13 | 13 | 13 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Stainless Steel (Independent Laboratory) | 2.7 | 20 | 8 | 7 | 7 | 100 | 92.3 | 0 | 7.7 | 0.0 |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Epoxy Coated Flooring | 836 | 20 | 10 | 10 | 10 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |

[1] Sensitivity is calculated as 100% – false negative rate = 100%
[2] Specificity is calculated as 100% – false positive rate = 100%
[3] False negative rate is calculated as BAX (-) Ref (+) BAX enrichment samples / Tot Ref (+) samples
[4] False positive rate is calculated as BAX (+) Ref (-) / Tot Ref (-) samples
[5] McNemar Chi-Square test statistic calculated for paired enrichments

**Table 3b. Method performance for the detection of *Salmonella* species from environmental surfaces by the BAX System. Naturally contaminated surfaces. (9)**

| Surface | Re-growth Scheme | Total Samples (each treatment) | BAX Presumptive (# positive) | BAX Confirmed (# positive) | Reference Method (# positive) | Sensitivity % | Specificity % | False Negative % | False Positive % | $X^2$ Value [1,2] |
|---|---|---|---|---|---|---|---|---|---|---|
| Concrete (LB Enrichment) | 10 ul in 500 ul 3 hr | 20 | 6 | 4 | 10 | 40 | 80 | 60 | 20 | 1.1 |
| Concrete (LB Enrichment) | 1 ml in 9 ml 5 hr | 20 | 9 | 8 | 10 | 80 | 90 | 20 | 10 | 0.0 |
| Concrete (BPW Enrichment) | 10 ul in 500 ul 3 hr | 20 | 10 | 10 | 10 | 100 | 100 | 0 | 0 | 0.0 |

[1] McNemar Chi-Square test statistic calculated for paired enrichments (used for calculating data from single enrichments using LB)
[2] Mantel-Haenszel Chi-Square test statistic (used for calculating significance of results for adjacently collected sponges enriched in BPW for the test method)

**Table 3c. Method performance for the detection of *Salmonella* species from environmental surfaces by the BAX Q7 System. Artificially inoculated surfaces. BAX testing was performed from the same LB enrichments used for subsequent culture confirmation as per the FDA-BAM. (9)**

| Surface | Level (cfu applied per analytical unit) | Total Samples (each treatment) | BAX Presumptive (# positive) | BAX Confirmed (# positive) | Reference Method (# positive) | Sensitivity % | Specificity % | False Negative % | False Positive % | $X^2$ Value |
|---|---|---|---|---|---|---|---|---|---|---|
| Ceramic Tile | 114 | 20 | 15 | 15 | 15 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Plastic | 162 | 20 | 13 | 13 | 13 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Stainless Steel | 50 | 20 | 13 | 13 | 13 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Stainless Steel (Independent Laboratory) | 2.7 | 20 | 7 | 7 | 7 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |
| Epoxy Coated Flooring | 836 | 20 | 10 | 10 | 10 | 100 | 100 | 0 | 0 | - |
| | 0 | 5 | 0 | 0 | 0 | | 100 | 0 | 0 | |

**Table 3d. Method performance for the detection of *Salmonella* species from environmental surfaces by the BAX Q7 ® System. Naturally contaminated surfaces. (9)**

| Surface | Re-growth Scheme | Total Samples (each treatment) | BAX Presumptive (# positive) | BAX Confirmed (# positive) | Reference Method (# positive) | Sensitivity % | Specificity % | False Negative % | False Positive % | $X^2$ Value |
|---|---|---|---|---|---|---|---|---|---|---|
| Concrete (LB Enrichment) | 10 ul in 500 ul 3 hr | 20 | 7 | 5 | 10 | 40 | 80 | 60 | 20 | 0.6 |
| Concrete (LB Enrichment) | 1 ml in 9 ml 5 hr | 20 | 9 | 8 | 10 | 80 | 90 | 20 | 10 | 0.0 |
| Concrete (BPW Enrichment) | 10 ul in 500 ul 3 hr | 20 | 10 | 10 | 10 | 100 | 100 | 0 | 0 | 0.0 |

**DISCUSSION OF MODIFICATION APPROVED APRIL 2009 (10)**

In this three-laboratory collaborative study, results obtained were consistent across the participating sites. Each site also demonstrated identical results between platforms (BAX and BAX Q7 instruments). In the 135 samples assayed, one false negative result was found at one of the participating laboratories. It is not known whether this result is due to a candidate method related issue (*Salmonella* cell density reached a high enough level to make the transfer into the BAM secondary enrichments, but did not reach a level high enough to trigger a positive result by PCR or a failure to transfer from the correct primary enrichment to the BHI re-growth tube), or due to a contamination event in the reference method (though this seems unlikely since the discordant sample demonstrated positive results from both the TT and RV secondary enrichment broths). Since the theoretical limit of detection of the reference method from the primary LB enrichment is on the order of 1 cfu/ml (since 1 ml is transferred to the TT secondary enrichment broth tube) and the theoretical limit of detection of the BAX method from the primary LB enrichment is on the order of $10^3$ cfu/ml, it may be possible that the one discordant sample fell into this category. Another possibility is that the sample preparation may have been responsible. Since the sample matrix of peanut butter is very hydrophobic, if there is a less than complete homogenization of the sample in the enrichment broth, then the *Salmonella* cell(s) may not be exposed to the media for some period of the enrichment. Sample preparation is thus a critical step in the detection of *Salmonella* from this matrix (much more so than for matrixes that easily go into solution or are only susceptible to surface contamination) by this candidate method, and presumably by any other rapid method. Overall, though, the results of this study demonstrate that the BAX System method is comparable to the reference culture method for the detection of *Salmonella* from peanut butter even when the viable cell count approaches 1 cfu per analytical portion.

**Table 1. Summary Data Table – Qualicon, BAX Instrument (10)**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 20 | 20 | 20 | 20 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 20 | 2 | 2 | 2 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Control | - | 5 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 2. Summary Data Table – Q Laboratories, BAX Instrument**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 20 | 20 | 20 | 20 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 20 | 5 | 4 | 5 | 0.80 | 1.00 | 1.00 | 1.00 | 0 |
| Control | - | 5 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 3. Summary Data Table – retch laboratories, BAX Instrument**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 20 | 18 | 18 | 18 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 20 | 4 | 4 | 4 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Control | - | 5 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 4. Summary Data Table – Aggregate Data, BAX Instrument (10)**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 60 | 58 | 58 | 58 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 60 | 11 | 10 | 11 | 0.91 | 1.00 | 1.00 | 1.00 | 0 |
| Control | - | 15 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 5. Summary Data Table – Qualicon, BAX Q7 Instrument (10)**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 20 | 20 | 20 | 20 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 20 | 2 | 2 | 2 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Control | - | 5 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 6. Summary Data Table – Q Laboratories, BAX Q7 Instrument (10)**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 20 | 20 | 20 | 20 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 20 | 5 | 4 | 5 | 0.80 | 1.00 | 1.00 | 1.00 | 0 |
| Control | - | 5 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 7. Summary Data Table – retch laboratories, BAX Q7 Instrument (10)**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 20 | 18 | 18 | 18 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 20 | 4 | 4 | 4 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Control | - | 5 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**Table 8. Summary Data Table – Aggregate Data, BAX Q7 Instrument (10)**

| Level | MPN per 25 g | Replicates per method | RM Pos. | CM Pres. Pos. | CM Conf. Pos. | CM Sens. | CM Spec. | RM Sens. | RM Spec. | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|---|
| High | 11.5 | 60 | 58 | 58 | 58 | 1.00 | 1.00 | 1.00 | 1.00 | - |
| Low | 1.08 | 60 | 11 | 10 | 11 | 0.91 | 1.00 | 1.00 | 1.00 | 0 |
| Control | - | 15 | 0 | 0 | 0 | - | 1.00 | - | 1.00 | - |

**DISCUSSION OF MODIFICATION APPROVED JUNE 2009 (11)**

The results in these studies, within their statistical uncertainty, support the product claims that the BAX® System is an effective method and comparable to standard reference methods for detecting *Salmonella* in beef and produce enriched in the BAX System *E. coli* O157:H7 MP media and for detecting *Salmonella* in soy protein flour when enriched in lactose broth as primary enrichment followed by 3-hour re-growth in BHI broth.

**Table 2. Internal study of spiked and unspiked spinach (25 g) samples tested with BAX system method and FDA- BAM reference culture methods. (0.016 MPN[a]/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specificity[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 8 hr | BAX Classic | 20 | 7/7 | 87.5 | 12.5 | 0/5 | 100 | 0 | 0.1 |
| | BAX Q7 | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| 10 hr | BAX Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| | BAX Q7 | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| 24 hr | BAX Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| | BAX Q7 | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| FDA-BAM | | 20 | 8 | -- | -- | 0/5 | -- | -- | -- |

[a] Most probable number of colony forming units per test portion.

[b] Presump Pos: Positive by BAX® System assay for BAX® enrichments. Confirmed: At least one confirmed E. coli O157:H7 isolate was obtained by culture.

[c] Sensitivity rate: 100 times the number of presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.

[d] False negative rate: 100 minus sensitivity rate.

[e] Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples.

[f] False positive rate: 100 minus specificity rate.

[g] Chi-square: Mantel-Haenszel chi square. *Chi-square value > 3.84 indicates significance at P < 0.05.

**Table 3. Internal study of spiked and unspiked iceberg lettuce (25 g) samples tested with BAX system method and FDA-BAM reference culture methods. (0.092 MPN[a]/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specificity[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 7.5 hr | BAX Classic | 20 | 6/6 | 75 | 25 | 0/5 | 100 | 0 | 1.63 |
| | BAX Q7 | 20 | 6/6 | 75 | 25 | 0/5 | 100 | 0 | 1.63 |
| 10 hr | BAX Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.39 |
| | BAX Q7 | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.39 |
| 24 hr | BAX Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.39 |
| | BAX Q7 | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.39 |
| FDA-BAM | | 20 | 10 | -- | -- | 0/5 | -- | -- | -- |

*[a] Most probable number of colony forming units per test portion.*

*[b] Presump Pos: Positive by BAX® System assay for BAX® enrichments. Confirmed: At least one confirmed E. coli O157:H7 isolate was obtained by culture.*

*[c] Sensitivity rate: 100 times the number of presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.*

*[d] False negative rate: 100 minus sensitivity rate.*

*[e] Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples.*

*[f] False positive rate: 100 minus specificity rate*

*[g] Chi-square: Mantel-Haenszel chi square. *Chi-square value > 3.84 indicates significance at P < 0.05.*

**Table 4. Internal study of spiked and un-spiked ground beef (25 g) samples tested with BAX system method and FDA-BAM reference culture methods. (0.02 MPN[a]/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specificity[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 8 hr | BAX Classic | 20 | 6/6 | 75 | 25 | 0/5 | 100 | 0 | 0.94 |
| | BAX Q7 | 20 | 5/5 | 62.5 | 37.5 | 0/5 | 100 | 0 | 1.71 |
| 10 hr | BAX Classic | 20 | 7/7 | 87.5 | 12.5 | 0/5 | 100 | 0 | 0.41 |
| | BAX Q7 | 20 | 7/7 | 87.5 | 12.5 | 0/5 | 100 | 0 | 0.41 |
| 12 hr | BAX Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.1 |
| | BAX Q7 | 20 | 7/7 | 87.5 | 12.5 | 0/5 | 100 | 0 | 0.41 |
| 24 hr | BAX Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.1 |
| | BAX Q7 | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0.1 |
| FDA-BAM | | 20 | 9 | -- | -- | 0/5 | -- | -- | -- |

[a] *Most probable number of colony forming units per test portion.*

[b] *Presump Pos: Positive by BAX® System assay for BAX® enrichments. Confirmed: At least one confirmed E. coli O157:H7 isolate was obtained by culture.*

[c] *Sensitivity rate: 100 times the number of presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.*

[d] *False negative rate: 100 minus sensitivity rate.*

[e] *Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples.*

[f] *False positive rate: 100 minus specificity rate*

[g] *Chi-square: Mantel-Haenszel chi square. *Chi-square value > 3.84 indicates significance at P < 0.05.*

**Table 5. Internal study of spiked and un-spiked beef trim (65 g) samples tested with BAX system method and FDA- BAM reference culture methods. (0.0092 MPN[a]/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specificity[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 8 hr | BAX Classic | 20 | 14/14 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 13/13 | 92.9 | 7.1 | 0/5 | 100 | 0 | 0 |
| 10 hr | BAX Classic | 20 | 14/14 | 100 | 7.1 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 13/13 | 92.9 | 14.3 | 0/5 | 100 | 0 | 0.1 |
| 12 hr | BAX Classic | 20 | 14/14 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 14/14 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| 24 hr | BAX Classic | 20 | 14/14 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 14/14 | 100 | 7.1 | 0/5 | 100 | 0 | 0 |
| FDA-BAM | | 20 | 13 | -- | -- | 0/5 | -- | -- | -- |

[a] *Most probable number of colony forming units per test portion.*

[b] *Presump Pos: Positive by BAX® System assay for BAX® enrichments. Confirmed: At least one confirmed E. coli O157:H7 isolate was obtained by culture.*

[c] *Sensitivity rate: 100 times the number of presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.*

[d] *False negative rate: 100 minus sensitivity rate.*

[e] *Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples.*

[f] *False positive rate: 100 minus specificity rate*

[g] *Chi-square: Mantel-Haenszel chi square. *Chi-square value > 3.84 indicates significance at P < 0.05.*

**Table 6. Internal study of spiked and un-spiked soy protein flour (25 g) samples tested with BAX system method and FDA-BAM reference culture methods. (0.02 MPN/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specificity[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 20 hr | BAX® Classic | 20 | 8/8 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| FDA-BAM | | 20 | 8 | -- | -- | 0/5 | -- | -- | -- |

[a] *Most probable number of colony forming units per test portion.*

[b] *Presump Pos: Positive by BAX® System assay for BAX® enrichments. Confirmed: At least one confirmed E. coli O157:H7 isolate was obtained by culture.*

[c] *Sensitivity rate: 100 times the number of presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.*

[d] *False negative rate: 100 minus sensitivity rate.*

[e] *Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples.*

[f] *False positive rate: 100 minus specificity rate*

[g] *Chi-square: Mantel-Haenszel chi square. *Chi-square value > 3.84 indicates significance at P < 0.05.*

**Table 7. Independent laboratory study of spiked and un-spiked ground beef (25 g) samples tested with BAX system method and FDA-BAM reference culture methods. (0.024 MPN[a]/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specifi-city[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 8 hr | BAX Classic | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 12/12 | 92.3 | 7.7 | 0/5 | 100 | 0 | 0.43 |
| 10 hr | BAX Classic | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| 12 hr | BAX Classic | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| 24 hr | BAX Classic | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| | BAX Q7 | 20 | 13/13 | 100 | 0 | 0/5 | 100 | 0 | 0.11 |
| FDA-BAM | | 20 | 14 | -- | -- | 0/5 | -- | -- | -- |

[a] *Most probable number of colony forming units per test portion.*

[b] *Presump Pos: Positive by BAX® System assay for BAX® enrichments. Confirmed: At least one confirmed E. coli O157:H7 isolate was obtained by culture.*

[c] *Sensitivity rate: 100 times the number of presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.*

[d] *False negative rate: 100 minus sensitivity rate.*

[e] *Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples.*

[f] *False positive rate: 100 minus specificity rate*

[g] *Chi-square: Mantel-Haenszel chi square. *Chi-square value > 3.84 indicates significance at P < 0.05.*

**Table 12. Internal study of spiked and unspiked Romaine lettuce (25 g) samples tested with BAX system method and FDA-BAM reference culture methods. (0.0083 MPN[a]/g) (11)**

| Enrichment Time | Method | Total spiked | Presump.Pos /Confirmed[b] | Sensitivity[c] % | False Neg[d] % | Presump. Pos /Unspiked | Specificity[e] % | False Pos[f] % | Chi-square[g] |
|---|---|---|---|---|---|---|---|---|---|
| 8 hr | BAX Classic | 20 | 15/14 | 93.3 | 6.7 | 0/5 | 80 | 20 | 12.1 |
| | BAX Q7 | 20 | 15/14 | 93.3 | 6.7 | 0/5 | 80 | 20 | 12.1 |
| 22 hr | BAX Classic | 20 | 16/15 | 100 | 0 | 0/5 | 80 | 20 | 14.2 |
| | BAX Q7 | 20 | 16/15 | 100 | 0 | 0/5 | 80 | 20 | 14.2 |
| FDA-BAM | | 20 | 3 | -- | -- | 0/5 | -- | -- | -- |

**Table 8. *Salmonella* BAX® system Inclusivity**

| Strain No. | Serovar | O-Group | Source | Result | Strain No. | Serovar | O-Group | Source | Result |
|---|---|---|---|---|---|---|---|---|---|
| 1550 | *S*. Abaetetuba | F | unknown | Positive | 3915 | *S*. Haardt | C3 | Broiler breeders | Positive |
| 1548 | *S*. Abony | B | unknown | Positive | 1689 | *S*. Hartford | ND | unknown | Positive |
| 3885 | *S*. Adelaide | O | poultry feed | Positive | 13067 | *S*. Havana | G2 | Solae | Positive |
| 1551 | *S*. Aequatoria | C2 | Unkown | Positive | 12907 | *S*. Heidelberg | B | Tyson 03/2006 | Positive |
| 4084 | *S*. Africana | B | Unkown | Positive | 3852 | *S*. Indiana | B | poultry feed | Positive |
| 1552 | *S*. Alabama | D2 | unknown | Positive | 5533 | *S*. Infantis | C1 | Thyme | Positive |
| 1531 | *S*. Altendorf | B | unknown | Positive | 1693 | *S*. Javiana | ND | unknown | Positive |
| 1530 | *S*. Amager | E1 | unknown | Positive | 2628 | *S*. Kentucky | C3 | ATCC9263 | Positive |
| 1521 | *S*. Amersfoort | C2 | unknown | Positive | 2353 | *S*. Kristianstad | E1 | unknown | Positive |
| 1332 | *S*. Anatum | E2 | shrimp | Positive | 13068 | *S*. Lexington | E1 | Solae | Positive |
| 725 | *S*. Arizonae | C | ATCC13314 | Positive | 2263 | *S*. Lille | C1 | pancake | Positive |
| 1526 | *S*. Austin | C1 | unknown | Positive | 4036 | *S*. Livingstone | C1 | chicken | Positive |
| 1553 | *S*. Ball | B | unknown | Positive | 1653 | *S*. Manhattan | C2 | unknown | Positive |
| 1554 | *S*. Banalia | ND | unknown | Positive | 2309 | *S*. Maregrosso | ND | unknown | Positive |
| 1510 | *S*. Bareilly | C1 | unknown | Positive | 13069 | *S*. Mbandaka | C1 | Solae | Positive |
| 2341 | *S*. Barry | ND | unknown | Positive | 1703 | *S*. Mississippi | G2 | feces | Positive |
| 3185 | *S*. Bellevue | ND | Cocoa bean environment | Positive | 1492 | *S*. Montevideo | C1 | unknown | Positive |
| 1523 | *S*. Berkeley | U | diseased turkey | Positive | 3156 | *S*. Muenchen | C2 | Cocoa bean environment | Positive |
| 1525 | *S*. Betioky | ND | unknown | Positive | 2203 | *S*. Muenster | E1 | unknown | Positive |
| 1085 | *S*. Binza | E2 | dried spice | Positive | 966 | *S*. Napoli | D1 | unknown | Positive |
| 1343 | *S*. Blockley | C2 | environment | Positive | 3898 | *S*. Neumuenster | B | poultry feed | Positive |
| 2343 | *S*. Bockenheim | ND | unknown | Positive | 1707 | *S*. Newbrunswick | E2 | unknown | Positive |
| 1329 | *S*. Braenderup | C1 | dried egg | Positive | 707 | *S*. Newport | C2 | ATCC6962 | Positive |
| 1338 | *S*. Brandenburg | B | milk | Positive | 2735 | *S*. Ohio | C1 | Protein supplement for feed | Positive |
| 1356 | *S*. Bredeney | B | pork | Positive | 2676 | *S*. Orion | E1 | chicken feed | Positive |
| 1535 | *S*. Brookfield | ND | frog | Positive | 3863 | *S*. Othmarschen | C1 | environmental | Positive |
| 3882 | *S*. Broughton | E4 | poultry feed | Positive | 918 | *S*. Paratyphi A | A | ATCC9150 | Positive |
| 1558 | *S*. Canastel | D1 | feed | Positive | 1711 | *S*. Pomona | M | Turkey intestine in 1941 | Positive |
| 1620 | *S*. Carmel | J | unknown | Positive | 1712 | *S*. Pretoria | F | pig | Positive |
| 1621 | *S*. Carrau | H | unknown | Positive | 1482 | *S*. Pullorum | D1 | chicken liver | Positive |
| 2629 | *S*. Cerro | K | ATCC10723 | Positive | 1655 | *S*. Reading | B | unknown | Positive |
| 1615 | *S*. Chameleon | ND | Lizard liver | Positive | 1372 | *S*. Saintpaul | B | milk powder | Positive |
| 1623 | *S*. Champaign | Q | chicken liver | Positive | 6250 | *S*. Santiago | C3 | dried onion | Positive |
| 1625 | *S*. Chester | ND | unknown | Positive | 1608 | *S*. Seminole | R | snake feces | Positive |
| 1557 | *S*. Chicago | M | unknown | Positive | 13168 | *S*. Senftenberg | F | environmental sponge | Positive |
| 13167 | *S*. Chingola | ND | environmental sponge | Positive | 1566 | *S*. species | ND | rattlesnake skin | Positive |
| 917 | *S*. Choleraesuis | C1 | ATCC13312 | Positive | 6177 | *S*. species | ND | Chicken giblets | Positive |
| 1628 | *S*. Colorado | C1 | unknown | Positive | 6735 | *S*. species | ND | Sesame seeds | Positive |
| 3157 | *S*. Corvallis | C3 | environmental | Positive | 1590 | *S*. species | ND | Snake regurgitate | Positive |
| 2870 | *S*. Corvallis | ND | Cocoa bean environment | Positive | 739 | *S*. Stanley | ND | unknown | Positive |
| 1632 | *S*. Cubana | G2 | chicken | Positive | 2637 | *S*. Swarzengrund | B | chicken | Positive |
| 1675 | *S*. Daressalaam | ND | #N/A | Positive | 3536 | *S*. Tennessee | C1 | unknown | Positive |
| 13169 | *S*. Desau | ND | environmental sponge | Positive | 2639 | *S*. Thomasville | E3 | turkey intestine | Positive |
| 2349 | *S*. Drypool | E2 | unknown | Positive | 1336 | *S*. Thompson | C1 | chicken | Positive |
| 3017 | *S*. Dublin | D1 | unknown | Positive | 1613 | *S*. Tulndorp | ND | Zoo animal liver | Positive |
| 1680 | *S*. Dugbe | W | unknown | Positive | 584 | *S*. Typhi | D | ATCC6539 | Positive |
| 1641 | *S*. Durban | D1 | feces | Positive | 586 | *S*. Typhimurium | B | ATCC14028, Animal tissue | Positive |
| 1469 | *S*. Ealing | O | dried milk | Positive | 13081 | *S*. Virchow | C1 | Basil | Positive |
| 1684 | *S*. Emmastad | P | unknown | Positive | 1614 | *S*. Volksdorf | ND | Iguana bladder | Positive |
| 1773 | *S*. Enterica | ND | unknown | Positive | 2313 | *S*. Wandsworth | Q | unknown | Positive |
| 706 | *S*. Enteritidis | D1 | ATCC13076 | Positive | 1609 | *S*. Wassennaar | ND | Iguana swab | Positive |
| 1686 | *S*. Fayed | H | unknown | Positive | 1560 | *S*. Westpark | ND | Tortoise intestine | Positive |

**DISSCUSSION OF MODIFCATION APPROVED OCTOBER 2010 (12)**

Tables 1a - 1d (Modification results October 2010 below) show that the BAX system using the *Salmonella* 2 test kit performed in a statistically indistinguishable manner to the reference methods for detecting *Salmonella* in frankfurters, raw ground beef, cream cheese and orange juice analyzed from the same pre-enrichment. Additionally, results from samples tested from enrichments conducted using the BAX MP test media were statistically equivalent to the samples enriched using the reference method. All chi-square values were below the level of 3.84, indicating no statistically significant difference at $P < 0.05$. As shown in Tables 3 and 4, inclusivity and exclusivity of the modified test kit testing 352 inclusivity and 46 exclusivity strains gave correct results.

Within the statistical constraints of this study, the BAX system *Salmonella* 2 test kit supports the performance claims of the BAX system PCR assay for the detection of *Salmonella* that were validated in previous AOAC studies (≥ 98% sensitivity/specificity) when compared to the appropriate reference method.

**Table 1a. Results of Hot Dogs Strain *Salmonella enterica* Serotype Agona DD1333 (12)**

| Method | MPN Per 25g[a] | Enrichment Method (Med | Total spi | Presump.Pos /Confirmed | Sensitivity[b] | False Neg [c] % | Presump. Pos /Unspiked | Specificity[d] % | False Pos[e] % | Chi-squa |
|---|---|---|---|---|---|---|---|---|---|---|
| 22 hr BAX | 0.67 | BAX MP | 20 | 6/6 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| 22 hr BAX | 0.67 | BPW | 20 | 6/6 | 100 | 0 | 0/5 | 100 | 0 | - |
| Reference cult | 0.67 | BPW | 20 | 6 | - | - | - | - | - | |

**Table 1b. Results of Ground Beef Strain *Salmonella enterica* Serotype Heidelberg Agona DD13017 (12)**

| Method | MPN Per 25g[a] | Enrichment Method (Med | Total spi | Presump.Pos /Confirmed | Sensitivity[b] | False Neg [c] % | Presump. Pos /Unspiked | Specificity[d] % | False Pos[e] % | Chi-squa |
|---|---|---|---|---|---|---|---|---|---|---|
| 22 hr BAX | 0.79 | BAX MP | 20 | 12/12 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| 22 hr BAX | 0.79 | BPW | 20 | 12/12 | 100 | 0 | 0/5 | 100 | 0 | - |
| Reference cult | 0.79 | BPW | 20 | 12 | - | - | - | - | - | |

**Table 1c. Results of Orange Juice Strain *Salmonella enterica* Serotype Worthington DD403 (12)**

| Method | MPN Per 25g[a] | Enrichment Method (Med | Total spi | Presump.Pos /Confirmed | Sensitivity[b] | False Neg [c] % | Presump. Pos /Unspiked | Specificity[d] % | False Pos[e] % | Chi-squa |
|---|---|---|---|---|---|---|---|---|---|---|
| 22 hr BAX | 0.80 | BAX MP | 20 | 7/7 | 100 | 0 | 0/5 | 100 | 0 | 2.1 |
| 22 hr BAX | 0.80 | UPB | 20 | 3/3 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| Reference cult | 0.80 | UPB | 20 | 3 | - | - | - | - | - | - |

**Table 1d. Results of Cream Cheese Strain *Salmonella enterica* Serotype Typhimurium DD586 (12)**

| Method | MPN Per 25g[a] | Enrichment Method (Med | Total spi | Presump.Pos /Confirmed | Sensitivity[b] | False Neg [c] % | Presump. Pos /Unspiked | Specificity[d] % | False Pos[e] % | Chi-square[f, 2] |
|---|---|---|---|---|---|---|---|---|---|---|
| 22 hr BAX | 0.67 | BAX MP | 20 | 7/7 | 100 | 0 | 0/5 | 100 | 0 | 0.46 |
| 22 hr BAX | 0.67 | BPW | 20 | 5/5 | 100 | 0 | 0/5 | 100 | 0 | 0 |
| Reference cult | 0.67 | BPW | 20 | 5 | - | - | - | - | - | - |

[a] *Most probable number of colony forming units per test portion.* ***MPN was performed on the day of testing using the reference method.*** *MPN values were calculated using the tables found in the FDA-BAM. Spike level was determined by performing a standard plate count on the incident cultures which were diluted for spiking each matrix on the day of introduction to the master sample.*

[b] *Sensitivity rate: 100 times the number of true presumptive positive results divided by total true positive results confirmed from enrichment of spiked samples.*

[c] *False negative rate: 100 minus sensitivity rate.*

[d] *Specificity rate: 100 times the number of assay-negative results divided by total number of true negative results, including unspiked samples. Specificity is determined by the USDA presumptive result compared with the actual culture confirmation.*

[e] *False positive rate: 100 minus specificity rate*

[f] *Mantel Haenszel Chi-square for unpaired samples. McNemar's Chi-square for paired samples.*

**Chi-square value > 3.84 indicates significance at $P < 0.05$.*

**Table 2. Inclusivity of the BAX ® *Salmonella* 2 Test Kit (12)**

| DuPont ID Number | Genus and Serotype | Isolate Source | Serogroup | BAX ® *Salmonella* 2 Result |
|---|---|---|---|---|
| 1550 | *Salmonella* Abaetetuba | | F | POS |
| 2166 | *Salmonella* Abaetetuba | | F | POS |
| 1547 | *Salmonella* Aberdeen | | F | POS |
| 1548 | *Salmonella* Abony | | B | POS |
| 1543 | *Salmonella* Adelaide | | O | POS |
| 1551 | *Salmonella* Aequatoria | | C1 | POS |
| 4084 | *Salmonella* Africana | | B | POS |
| 3218 | *Salmonella* Agama | Cocoa bean environment | B | POS |
| 1335 | *Salmonella* Agona | Chicken | B | POS |
| 1352 | *Salmonella* Agona | Cotton seeds | B | POS |

| | | | | |
|---|---|---|---|---|
| 1552 | *Salmonella* Alabama | | D1 | POS |
| 1556 | *Salmonella* Alachua | Soil, abbatoir | O | POS |
| 2966 | *Salmonella* Albany | | C3 | POS |
| 1531 | *Salmonella* Altendorf | | B | POS |
| 1530 | *Salmonella* Amager | | E1 | POS |
| 3432 | *Salmonella* Amager | | E1 | POS |
| 1521 | *Salmonella* Amersfoort | | C1 | POS |
| 7072 | *Salmonella* Amsterdam | | E1 | POS |
| 1332 | *Salmonella* Anatum | Shrimp | E1 | POS |
| 1334 | *Salmonella* Anatum | Egg | E1 | POS |
| 2274 | *Salmonella* Anatum | | E1 | POS |
| 725 | *Salmonella* Arizonae | | | POS |
| 726 | *Salmonella* Arizonae | | | POS |
| 2980 | *Salmonella* Arkansas | | B | POS |
| 2981 | *Salmonella* Arkansas | | B | POS |
| 1527 | *Salmonella* Atlanta | | G | POS |
| 1526 | *Salmonella* Austin | | C1 | POS |
| 1553 | *Salmonella* Ball | | B | POS |
| 1554 | *Salmonella* Banalia | | C2 | POS |
| 1510 | *Salmonella* Bareilly | | C1 | POS |
| 2172 | *Salmonella* Bareilly | | C1 | POS |
| 2341 | *Salmonella* Barry | | O54 | POS |
| 3185 | *Salmonella* Bellevue | Cocoa bean environment | C3 | POS |
| 1523 | *Salmonella* Berkeley | Diseased turkey | U | POS |
| 1606 | *Salmonella* Bern | Opossum | R | POS |
| 1331 | *Salmonella* Berta | Sausages | D1 | POS |
| 2795 | *Salmonella* Berta | Chicken intestine | D1 | POS |
| 1525 | *Salmonella* Betioky | | O59 | POS |
| 1085 | *Salmonella* Binza | Dried spice | E2 | POS |
| 2786 | *Salmonella* Binza | Chicken | E2 | POS |
| 1343 | *Salmonella* Blockley | Environment | C2 | POS |
| 2343 | *Salmonella* Bockenheim | | | POS |
| 1509 | *Salmonella* Bovismorbificans | | C2 | POS |
| 1329 | *Salmonella* Braenderup | Dried egg | C1 | POS |
| 1337 | *Salmonella* Braenderup | Chicken | C1 | POS |
| 1555 | *Salmonella* Brancaster | | B | POS |
| 1338 | *Salmonella* Brandenburg | Milk | B | POS |
| 964 | *Salmonella* Bredeney | Fresh chicken | B | POS |
| 1356 | *Salmonella* Bredeney | Pork | B | POS |
| 1535 | *Salmonella* Brookfield | Frog | O66 | POS |
| 3882 | *Salmonella* Broughton | Poultry feed | E4 | POS |
| 1668 | *Salmonella* California | | B | POS |
| 2178 | *Salmonella* California | | B | POS |
| 1558 | *Salmonella* Canastel | Feed | D1 | POS |
| 1620 | *Salmonella* Carmel | | O17 | POS |
| 1621 | *Salmonella* Carrau | | H | POS |
| 2629 | *Salmonella* Cerro | | K | POS |
| 2813 | *Salmonella* Cerro | Chicken chilled water tank | K | POS |
| 1615 | *Salmonella* Chameleon | Lizard liver | O16 | POS |
| 1623 | *Salmonella* Champaign | Liver of hen | Q | POS |
| 2180 | *Salmonella* Champaign | | Q | POS |
| 1624 | *Salmonella* Chandans | | F | POS |
| 3153 | *Salmonella* Chandans | Cocoa bean environment | F | POS |
| 1625 | *Salmonella* Chester | | B | POS |
| 1557 | *Salmonella* Chicago | | M | POS |
| 917 | *Salmonella* Choleraesuis | | UNK | POS |
| 3984 | *Salmonella* Choleraesuis paratyphi B | Gallbladder | B | POS |
| 3988 | *Salmonella* Choleraesuis paratyphi C | | C1 | POS |
| 1665 | *Salmonella* Colombo | | P | POS |
| 1628 | *Salmonella* Colorado | | C1 | POS |
| 2870 | *Salmonella* Corvallis | Cocoa bean environment | C3 | POS |
| 3157 | *Salmonella* Corvallis | Cocoa bean environment | C3 | POS |
| 3217 | *Salmonella* Cotham | Cocoa bean environment | O28 | POS |
| 6966 | *Salmonella* Cotham | | O28 | POS |
| 1632 | *Salmonella* Cubana | Chicks | G2 | POS |
| 1675 | *Salmonella* Daressalaam | | | POS |
| 1635 | *Salmonella* Daytona | | C1 | POS |
| 1638 | *Salmonella* Derby | | B | POS |
| 2186 | *Salmonella* Drypool | | O15 | POS |
| 2349 | *Salmonella* Drypool | | O15 | POS |

| | | | | |
|---|---|---|---|---|
| 3015 | *Salmonella* Dublin | | D1 | POS |
| 3017 | *Salmonella* Dublin | | D1 | POS |
| 3019 | *Salmonella* Dublin | | D1 | POS |
| 7005 | *Salmonella* Dublin | | D1 | POS |
| 1680 | *Salmonella* Dugbe | | W | POS |
| 1641 | *Salmonella* Durban | Faeces | D1 | POS |
| 2869 | *Salmonella* Durham | Cocoa bean environment | G2 | POS |
| 3187 | *Salmonella* Durham | Cocoa bean environment | G2 | POS |
| 1469 | *Salmonella* Ealing | Dried baby milk | O | POS |
| 1644 | *Salmonella* Ealing | Dried baby milk (1985-1986) | O | POS |
| 1684 | *Salmonella* Emmastad | | P | POS |
| 1773 | *Salmonella* enterica unk serotype | | | POS |
| 1775 | *Salmonella* enterica Typhimurium | | B | POS |
| 1777 | *Salmonella* enterica Dar-es-salaam | | | POS |
| 13035 | *Salmonella* enterica Choleraesuis | | | POS |
| 13036 | *Salmonella* enterica Typhimurium | | B | POS |
| 706 | *Salmonella* Enteritidis | | D1 | POS |
| 737 | *Salmonella* Enteritidis | | D1 | POS |
| 4022 | *Salmonella* Enteritidis | Mayonnaise | D1 | POS |
| 1686 | *Salmonella* Fayed | | C2 | POS |
| 1687 | *Salmonella* Ferlac | Ceylonese dessicated coconut | H | POS |
| 5908 | *Salmonella* Ferlac | | H | POS |
| 741 | *Salmonella* Gallinarum | | D1 | POS |
| 2350 | *Salmonella* Gallinarum | | D1 | POS |
| 2189 | *Salmonella* Give | | E1 | POS |
| 3915 | *Salmonella* Haardt | Broiler breeders | C3 | POS |
| 12967 | *Salmonella* Haardt | Poultry | C3 | POS |
| 12968 | *Salmonella*Haardt | Poultry | C3 | POS |
| 12969 | *Salmonella* Haardt | Poultry | C3 | POS |
| 12985 | *Salmonella* Haardt | Poultry | C3 | POS |
| 3917 | *Salmonella* Hadar | Broilers | C2 | POS |
| 3918 | *Salmonella* Hadar | Broilers | C2 | POS |
| 1689 | *Salmonella* Hartford | | C1 | POS |
| 2290 | *Salmonella* Hartford | Cheesecake, Dover | C1 | POS |
| 2245 | *Salmonella* Havana | Pancake | G2 | POS |
| 13067 | *Salmonella* Havana | Soy Plant Environmental | G2 | POS |
| 6667 | *Salmonella* Heidelberg | | B | POS |
| 12907 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12908 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12909 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12910 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12911 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12913 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12919 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12920 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12922 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12928 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12929 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12931 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12932 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12933 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12935 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12936 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12937 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12945 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12947 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12952 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12953 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12954 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12958 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12959 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12995 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12996 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12998 | *Salmonella* Heidelberg | Poultry | B | POS |
| 12999 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13000 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13001 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13004 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13014 | *Salmonella* Heidelberg | Poultry | B | POS |

| | | | | |
|---|---|---|---|---|
| 13017 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13018 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13019 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13020 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13021 | *Salmonella* Heidelberg | Poultry | B | POS |
| 13022 | *Salmonella* Heidelberg | Poultry | B | POS |
| 1616 | *Salmonella* Houten | Imported bird feces | O43 | POS |
| 3699 | *Salmonella* Hvittingfoss | Herbs or spices | I | POS |
| 1480 | *Salmonella* Indiana | Turkey | B | POS |
| 3852 | *Salmonella* Indiana | Poultry feed | B | POS |
| 7011 | *Salmonella* Indiana | | B | POS |
| 5533 | *Salmonella* infantis | Thyme | C1 | POS |
| 7111 | *Salmonella* Infantis | | C1 | POS |
| 1693 | *Salmonella* Javiana | | D1 | POS |
| 1695 | *Salmonella* Johannesburg | | R | POS |
| 3043 | *Salmonella* Jjohannesburg | | R | POS |
| 1251 | *Salmonella* Kedougou | Turkey | G2 | POS |
| 2628 | *Salmonella* Kentucky | | C3 | POS |
| 12912 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12914 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12915 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12916 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12917 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12918 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12921 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12924 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12925 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12926 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12927 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12941 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12943 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12946 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12948 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12949 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12950 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12951 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12955 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12956 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12957 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12981 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12989 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12990 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12993 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 12997 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13002 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13003 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13006 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13007 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13008 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13009 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13010 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13012 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13013 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13015 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 13016 | *Salmonella* Kentucky | Poultry | C3 | POS |
| 2196 | *Salmonella* Kiambu | | B | POS |
| 2312 | *Salmonella* Kottbus | | C2 | POS |
| 3038 | *Salmonella* Krefeld | | E4 | POS |
| 2353 | *Salmonella* Kristianstad | | E1 | POS |
| 7061 | *Salmonella* Kubacha | | B | POS |
| 7062 | *Salmonella* Kubacha | | B | POS |
| 2199 | *Salmonella* Lexington | | E1 | POS |
| 13068 | *Salmonella* Lexington | Soy Plant Environmental | E1 | POS |
| 2263 | *Salmonella* Lille | Pancake | C1 | POS |
| 2868 | *Salmonella* Lille | Cocoa bean environment | C1 | POS |
| 2992 | *Salmonella* Lille | | C1 | POS |
| 1650 | *Salmonella* Livingstone | Faeces | C1 | POS |
| 4036 | *Salmonella* Livingstone | Chicken | C1 | POS |
| 1652 | *Salmonella* London | | E1 | POS |
| 1698 | *Salmonella* Madelia | Liver of hen | H | POS |

| | | | | |
|---|---|---|---|---|
| 2201 | *Salmonella* Madelia | | H | POS |
| 1424 | *Salmonella* Manchester | Autolysed yeast | C2 | POS |
| 1653 | *Salmonella* Manhattan | | C2 | POS |
| 2673 | *Salmonella* Manhattan | Avian | C2 | POS |
| 6729 | *Salmonella* Manila | Sesame seeds | E2 | POS |
| 2309 | *Salmonella* Maregrosso | | | POS |
| 2755 | *Salmonella* Mbandaka | Swine tissue composite | C1 | POS |
| 13069 | *Salmonella* Mbandaka | Soy Plant Environmental | C1 | POS |
| 1701 | *Salmonella* Miami | | D1 | POS |
| 2204 | *Salmonella* Minnesota | | L | POS |
| 1703 | *Salmonella* Mississippi | Faeces in 1942 | G2 | POS |
| 2205 | *Salmonella* Mississippi | | G2 | POS |
| 1255 | *Salmonella* Montevideo | Egg | C1 | POS |
| 1492 | *Salmonella* Montevideo | | C1 | POS |
| 13071 | *Salmonella* Montevideo | Soy Plant Environmental | C1 | POS |
| 1704 | *Salmonella* Muenchen | | C2 | POS |
| 3156 | *Salmonella* Muenchen | Cocoa bean environment | C2 | POS |
| 2748 | *Salmonella* Muenster | Chicken | E1 | POS |
| 966 | *Salmonella* Napoli | | D1 | POS |
| 1476 | *Salmonella* napoli | | D1 | POS |
| 3898 | *Salmonella* Neumuenster | Poultry feed | B | POS |
| 1707 | *Salmonella* Newbrunswick | | E1 | POS |
| 2283 | *Salmonella* Newbrunswick | Malted barley flour | E1 | POS |
| 707 | *Salmonella* Newport | Fatal case of food poisoning | C2 | POS |
| 1261 | *Salmonella* Newport | Duck | C2 | POS |
| 13079 | *Salmonella* Newport | Basil | C2 | POS |
| 2735 | *Salmonella* Ohio | Protein supplement for feed | C1 | POS |
| 1710 | *Salmonella* Oranienburg | | C1 | POS |
| 3863 | *Salmonella* Othmarschen | Poultry hatchery | C1 | POS |
| 1248 | *Salmonella* Panama | Pork sausages | D1 | POS |
| 918 | *Salmonella* Paratyphi A | | A | POS |
| 919 | *Salmonella* Paratyphi A | | A | POS |
| 1711 | *Salmonella* Pomona | Turkey intestine in 1941 | M | POS |
| 2215 | *Salmonella* Poona | | G1 | POS |
| 1712 | *Salmonella* Pretoria | Pig | F | POS |
| 1482 | *Salmonella* Pullorum | Chicks livers | D1 | POS |
| 1507 | *Salmonella* Pullorum | Chicks livers | D1 | POS |
| 1655 | *Salmonella* Reading | | B | POS |
| 4558 | *Salmonella* Redlands | | I | POS |
| 2289 | *Salmonella* Rubislaw | Barley malt berries | F | POS |
| 1372 | *Salmonella* Saintpaul | Milk powder | B | POS |
| 13080 | *Salmonella* Saintpaul | Basil | B | POS |
| 1657 | *Salmonella* Sandiego | | B | POS |
| 2218 | *Salmonella* Sandiego | | B | POS |
| 2935 | *Salmonella* Sandiego | | B | POS |
| 6250 | *Salmonella* Santiago | Dried onion | C3 | POS |
| 6586 | *Salmonella* Santiago | Bourgignon powder | C3 | POS |
| 2352 | *Salmonella* Saphra | | I | POS |
| 1658 | *Salmonella* Schwarzengrund | | B | POS |
| 2637 | *Salmonella* Schwarzengrund | Chicken | B | POS |
| 2641 | *Salmonella* Schwarzengrund | Chicken | B | POS |
| 3184 | *Salmonella* Sculcoates | Cocoa bean environment | I | POS |
| 1610 | *Salmonella* Seminole | Lizard coelomic fluid | O40 | POS |
| 12960 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12961 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12962 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12963 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12964 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12965 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12966 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12970 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12971 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12972 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12973 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12975 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12978 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12980 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12982 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12983 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12984 | *Salmonella* Senftenberg | Poultry | E4 | POS |

| | | | | |
|---|---|---|---|---|
| 12986 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12987 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 12988 | *Salmonella* Senftenberg | Poultry | E4 | POS |
| 13056 | *Salmonella* Senftenberg | Soy Plant Environmental | E4 | POS |
| 13057 | *Salmonella* Senftenberg | Soy Plant Environmental | E4 | POS |
| 13058 | *Salmonella* Senftenberg | Soy Plant Environmental | E4 | POS |
| 13059 | *Salmonella* Senftenberg | Soy Plant Environmental | E4 | POS |
| 13060 | *Salmonella* Senftenberg | Soy Plant Environmental | E4 | POS |
| 1659 | *Salmonella* Shangani | | E1 | POS |
| 2373 | *Salmonella* species | | | POS |
| 2376 | *Salmonella* species | | | POS |
| 2380 | *Salmonella* species | | | POS |
| 4102 | *Salmonella* species | Nuts | | POS |
| 6177 | *Salmonella* species | Chicken giblets | | POS |
| 6696 | *Salmonella* species | Chicken giblets | | POS |
| 6735 | *Salmonella* species | Sesame seeds | | POS |
| 8034 | *Salmonella* species | Raw chicken | | POS |
| 12241 | *Salmonella* species | Gum arabic | | POS |
| 13037 | *Salmonella* Species | Isolate from NYS Department of Ag and Markets | | POS |
| 13070 | *Salmonella* Species | Soy Plant Environmental | | POS |
| 13075 | *Salmonella* Species | celery seed | | POS |
| 739 | *Salmonella* Stanley | | B | POS |
| 1333 | *Salmonella* Stanley | Chicken | B | POS |
| 3194 | *Salmonella* Stanleyville | Cocoa bean environment | B | POS |
| 1660 | *Salmonella* Sundsvall | | H | POS |
| 2867 | *Salmonella* Sya | Cocoa bean environment | X | POS |
| 3186 | *Salmonella* Sya | Cocoa bean environment | X | POS |
| 1661 | *Salmonella* Tennessee | | C1 | POS |
| 3536 | *Salmonella* Tennessee | | C1 | POS |
| 13061 | *Salmonella* Tennessee | Soy Plant Environmental | C1 | POS |
| 13062 | *Salmonella* Tennessee | Soy Plant Environmental | C1 | POS |
| 13063 | *Salmonella* Tennessee | Soy Plant Environmental | C1 | POS |
| 13064 | *Salmonella* Tennessee | Soy Plant Environmental | C1 | POS |
| 13065 | *Salmonella* Tennessee | Soy Plant Environmental | C1 | POS |
| 13066 | *Salmonella* Tennessee | Soy Plant Environmental | C1 | POS |
| 2229 | *Salmonella* Theilalle | | | POS |
| 2639 | *Salmonella* Thomasville | Turkey intestine | E3 | POS |
| 3924 | *Salmonella* Thomasville | Poultry feed | E3 | POS |
| 1336 | *Salmonella* Thompson | Chicken | C1 | POS |
| 1339 | *Salmonella* Thompson | Egg | C1 | POS |
| 12904 | *Salmonella* Tranorora | 700148 | O55 | POS |
| 1613 | *Salmonella* Tuindorp | Zoo animal liver | O43 | POS |
| 584 | *Salmonella* Typhi | | D1 | POS |
| 585 | *Salmonella* Typhi | | D1 | POS |
| 586 | *Salmonella* Typhimurium | Animal tissue | B | POS |
| 1084 | *Salmonella* Typhimurium | | B | POS |
| 1467 | *Salmonella* Typhimurium | | B | POS |
| 13005 | *Salmonella* Typhimurium | Poultry | B | POS |
| 13011 | *Salmonella* Typhimurium | Poultry | B | POS |
| 2238 | *Salmonella* Urbana | | N | POS |
| 2239 | *Salmonella* Uzaramo | | H | POS |
| 2346 | *Salmonella* Vietnam | | S | POS |
| 738 | *Salmonella* Virchow | | C1 | POS |
| 13081 | *Salmonella* Virchow | Basil | C1 | POS |
| 1614 | *Salmonella* Volksdorf | Iguana bladder | O43 | POS |
| 2313 | *Salmonella* Wandsworth | | Q | POS |
| 1609 | *Salmonella* Wassennaar | Iguana swab | O50 | POS |
| 1714 | *Salmonella* Wassennaar | Human | O50 | POS |
| 4035 | *Salmonella* Waycross | | S | POS |
| 1491 | *Salmonella* Weltevreden | Prawns | E1 | POS |
| 1560 | *Salmonella* Westpark | Tortoise intestine | O3,10 | POS |
| 4043 | *Salmonella* Worthington | | G2 | POS |
| 1429 | *Salmonella* species | African box meat (1967) | | POS |

**Table 3. Exclusivity of the BAX *Salmonella* 2 Test Kit (12)**

| DuPont Strain ID Number | ATCC Strain Number | Genus and Species | Isolate Source | BAX *Salmonella* 2 Result |
|---|---|---|---|---|
| 373 | 13883 | *Klebsiella pneumoniae* | | NEG |
| 374 | 29906 | *Proteus mirabilis* | | NEG |
| 383 | 8090 | *Citrobacter freundii* | | NEG |
| 640 | 43889 | *Escherichia coli O157:H7* | HUS Case Stool | NEG |
| 641 | 43890 | *Escherichia coli O157:H7* | Human Feces | NEG |
| 657 | 11296 | *Klebsiella ozaenae* | | NEG |
| 658 | 13182 | *Klebsiella oxytoca* | Pharyngeal Tonsil | NEG |
| 2389 | 13337 | *Hafnia alvei* | | NEG |
| 2417 | | *Serratia liquefaciens* | Raw Mince | NEG |
| 2558 | 43864 | *Citrobacter freundii* | | NEG |
| 3064 | | *Morganella morganii* | Environmental Swab | NEG |
| 3982 | 27853 | *Pseudomonas aeruginosa* | Blood Culture | NEG |
| 5588 | | *Hafnia alvei* | Ground Beef | NEG |
| 6121 | | *Proteus mirabilis* | Herring Gull Cloacae | NEG |
| 13142 | | *Morganella morganii* | | NEG |
| 13147 | | *Providencia rettgeri* | | NEG |
| 13148 | | *Pseudomonas aeruginosa* | | NEG |
| 13186 | | *Enterobacter amnigenus* | | NEG |
| 13187 | | *Enterobacter amnigenus* | | NEG |
| ES9 | | *Enterobacter sakazakii* | | NEG |
| ES14 | | *Enterobacter sakazakii* | | NEG |
| ES53 | | *Enterobacter sakazakii* | | NEG |
| ES1 | | *Enterobacter sakazakii* | | NEG |
| ES20 | | *Enterobacter sakazakii* | | NEG |
| ES34 | | *Enterobacter sakazakii* | | NEG |
| ES35 | | *Enterobacter sakazakii* | | NEG |
| ES38 | | *Enterobacter sakazakii* | | NEG |
| 700 | | *Shigella sonnei* | | NEG |
| 1083 | | *Shigella flexneri* | | NEG |
| 702 | | *Shigella sonnei* | | NEG |
| 846 | 29907 | *Escherichia blattae* | Hindgut of Cockroach | NEG |
| 847 | 35469 | *Escherichia fergusonii* | Human Feces | NEG |
| 848 | 33650 | *Escherichia hermannii* | Human Toe | NEG |
| 849 | 21073 | *Escherichia intermedia* | | NEG |
| 850 | 33821 | *Escherichia vulneris* | Human Wound | NEG |
| 854 | 35539 | *Staphylococcus gallinarum* | Chicken Nares | NEG |
| 862 | 4698 | *Micrococcus luteus* | | NEG |
| 863 | 12600 | *Staphylococcus aureus* | Human Clinical | NEG |
| 864 | 14990 | *Staphylococcus epidermidis* | Nose | NEG |
| 3354 | | *Listeria welshimeri* | | NEG |
| 1309 | | *Listeria monocytogenes* | Soft Cheese | NEG |
| 1154 | | *Listeria innocua* | Pate | NEG |
| QC201 | 13048 | *Enterobacter aerogenes* | Sputum | NEG |
| QC203 | 51113 | *Citrobacter brakii* | Snake | NEG |
| QC204 | 700814 | *Bacillus pumilus* | | NEG |
| QC102 | 51740 | *Staphylococcus aureus* | Margarine | NEG |

**DISCUSSION OF MODIFICATION APPROVED NOVEMBER 2015 (13)**

The method comparison study demonstrated that the alternative method as performed using the X5 BAX instrument is equivalent to the USDA-FSIS reference method for ground beef. Data demonstrated positive results across a wide titer of target in both pure culture and in enriched food matrix demonstrating that the X5 instrument performs in a similar fashion to the currently validated instruments when using the *Salmonella* 2 assay. Additional studies showed broad inclusivity and the ability to discriminate against non-target species. Robustness studies indicated that several of the factors tested could have a negative impact on assay performance. These factors will be explicitly called out in the instrument user guide and/or end user training materials.

**Table 1. Method results POD Food Matrixes – Candidate Method Compared to Culture Results from Candidate Enrichments (13)**

| Matrix | Strain | MPN[a]/test portion | N | Candidate Method Presumptive | | | Candidate Method Confirmed | | | dPOD$_{CP}$[e] | 95% CI[f] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | X[b] | POD$_{CP}$[c] | 95% CI | x | POD$_{CC}$[d] | 95% CI | | |
| Ground Beef BAX System X5 BPW | *S.* Typhimurium *DD586* | 5.0 (2.9, 8.8) | 5 | 5 | 1.0 | (0.57, 1.0) | 5 | 1.0 | (0.57, 1.0) | 0 | (-0.45, 0.45) |
| | | 0.5 (0.28, 0.88) | 20 | 6 | 0.30 | (0.14, 0.52) | 6 | 0.30 | (0.14, 0.52) | 0 | (-0.14, 0.14) |
| | | Negative Control | 5 | 0 | 0 | (0, 0.43) | 0 | 0 | (0, 0.43) | 0 | (-0.45, 0.45) |

**Table 2. Method results POD Food Matrixes – Candidate Method Compared to Culture Results from Candidate Enrichments**

| Matrix | Strain | MPN[a]/test portion | N | Candidate Method Presumptive | | | Candidate Method Confirmed | | | dPOD$_{CP}$[e] | 95% CI[f] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | X[b] | POD$_{CP}$[c] | 95% CI | x | POD$_{CC}$[d] | 95% CI | | |
| Ground Beef BAX System X5 mTSB | *S.* Typhimurium *DD586* | 5.0 (2.9, 8.8) | 5 | 5 | 1.0 | (0.57, 1.0) | 5 | 1.0 | (0.57, 1.0) | 0 | (-0.45, 0.45) |
| | | 0.5 (0.28, 0.88) | 20 | 5 | 0.25 | (0.11, 0.47) | 5 | 0.25 | (0.11, 0.47) | 0 | (-0.14, 0.14) |
| | | Negative Control | 5 | 0 | 0 | (0, 0.43) | 0 | 0 | (0, 0.43) | 0 | (-0.45, 0.45) |

**Table 3. Method results POD Food Matrixes – Candidate Method Compared to Reference Culture Method**

| Matrix | Strain | MPN[a]/test portion | N | Test Method | | | Reference Method | | | dPOD$_{C}$[e] | 95% CI[f] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | X[b] | POD$_{C}$[c] | 95% CI | x | POD$_{R}$[d] | 95% CI | | |
| Ground Beef Reference method mTSB v Candidate method BPW | *S.* Typhimurium *DD586* | 5.0 (2.9, 8.8) | 5 | 5 | 1.0 | (0.57, 1.0) | 5 | 1.0 | (0.57, 1.0) | 0 | (-0.43, 0.43) |
| | | 0.5 (0.28, 0.88) | 20 | 6 | 0.30 | (0.11, 0.46) | 5 | 0.25 | (0.14, 0.52) | 0.050 | (-0.22, 0.31) |
| | | Negative Control | 5 | 0 | 0 | (0, 0.43) | 0 | 0 | (0, 0.43) | 0 | (-0.43, 0.43) |
| Ground Beef Reference method mTSB v Candidate method mTSB | *S.* Typhimurium *DD586* | 5.0 (2.9, 8.8) | 5 | 5 | 1.0 | (0.57, 1.0) | 5 | 1.0 | (0.57, 1.0) | 0 | (-0.45, 0.45) |
| | | 0.5 (0.28, 0.88) | 20 | 5 | 0.25 | (0.11, 0.47) | 5 | 0.25 | (0.11, 0.47) | 0 | (-0.14, 0.14) |
| | | Negative Control | 5 | 0 | 0 | (0, 0.43) | 0 | 0 | (0, 0.43) | 0 | (-0.45, 0.45) |

[a]MPN = Most Probable Number based on the POD of reference method test portions using the AOAC MPN calculator (with 95% confidence interval).

[b]X = Number of positive test portions.

[c]POD$_C$ = Confirmed candidate method positive outcomes divided by the total number of trials.

[d]POD$_R$ = Confirmed reference method positive outcomes divided by the total number of trials.

[e]dPOD$_C$ = Difference between the candidate method and reference method POD values.

[f]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level.

**Table 4. Inclusivity Results for the BAX System X5 PCR Assay for *Salmonella* (13)**

| DuPont Strain ID | *Salmonella* Serotype and Serogroup | Strain Source | Result | DuPont Strain ID | *Salmonella* Serotype and Serogroup | Strain Source | Result |
|---|---|---|---|---|---|---|---|
| 584 | Typhi | Unknown | Pos | 1705 | Muenster | Unknown | Pos |
| 585 | Typhi | Unknown | Pos | 1712 | Pretoria | Pig | Pos |
| 586 | Typhimurium | Animal Tissue | Pos | 2189 | Give | Unknown | Pos |
| 707 | Newport | Fatal Case of Food Poisoning | Pos | 2263 | Lille | Pancake | Pos |
| 739 | Stanley | Unknown | Pos | 2283 | Newbrunswick | Malted Barley Flour | Pos |
| 741 | Gallinarum | Unknown | Pos | 2289 | Rubislaw | Barley Malt Berries | Pos |
| 919 | Paratyphi | Unknown | Pos | 2290 | Hartford | Cheesecake | Pos |
| 966 | Napoli | Unknown | U | 2313 | Wandsworth | Unknown | Pos |
| 1085 | Binza | Dried Spice | Pos | 2343 | Bockenheim | Unknown | Pos |
| 1248 | Panama | Pork Sausages | Pos | 2349 | Drypool | Unknown | Pos |
| 1251 | Kedougou | Turkey | Pos | 2353 | Kristianstad | Unknown | Pos |
| 1329 | Braenderup | Dried Egg | Pos | 2376 | Sculcoates | Unknown | Pos |
| 1332 | Anatum | Shrimp | Pos | 2637 | Schwarzengrund | Chicken | Pos |
| 1336 | Thompson | Chicken | Pos | 2639 | Thomasville | Turkey Intestine | Pos |
| 1352 | Agona | Cotton Seeds | Pos | 2735 | Ohio | Protein Supplement for Feed | Pos |
| 1356 | Bredeney | Pork | Pos | 2813 | Cerro | Chicken Chilled Water Tank | Pos |
| 1429 | Anfo | African Box Meat (1967) | Pos | 2867 | Sya | Cocoa Bean Environment | Pos |
| 1435 | Brandenburg | Unknown | Pos | 2870 | Corvallis | Cocoa Bean Environment | Pos |
| 1469 | Ealing | Dried Baby Milk | Pos | 3019 | Dublin | Unknown | Pos |
| 1482 | Pullorum | Chicks Livers | Pos | 3156 | Muenchen | Cocoa Bean Environment | Pos |
| 1509 | Bovismorbificans | Unknown | Pos | 3217 | Cotham | Cocoa Bean Environment | Pos |
| 1510 | Abaetetuba | Unknown | Pos | 3218 | Agama | Cocoa Bean Environment | Pos |
| 1521 | Bareilly | Unknown | Pos | 3699 | Hvittingfoss | Herbs or Spices | Pos |
| 1523 | Berkeley | Diseased Turkey | Pos | 3852 | Indiana | Poultry Feed | Pos |
| 1525 | Betioky | Unknown | Pos | 3863 | Oranienburg | Poultry Hatchery | Pos |
| 1526 | Austin | Unknown | Pos | 3882 | Broughton | Poultry Feed | Pos |
| 1530 | Altendorf | Unknown | Pos | 3898 | Thompson | Poultry Feed | Pos |
| 1535 | Brookfield | Frog | Pos | 3915 | Haardt | Broiler Breeders | Pos |
| 1543 | Adelaide | Unknown | Pos | 3984 | Java | Gallbladder | Pos |
| 1547 | Aberdeen | Unknown | Pos | 4022 | Enteritidis | Mayonnaise | Pos |
| 1548 | Abony | Unknown | Pos | 4036 | Livingstone | Chicken | Pos |
| 1552 | Alabama | Unknown | Pos | 4102 | Saintpaul | Nuts | Pos |
| 1553 | Ball | Unknown | Pos | 5533 | Infantis | Thyme | Pos |
| 1557 | Chicago | Unknown | Pos | 6177 | Arkansas | Chicken Giblets | Pos |
| 1568 | Arizonae | Turkey Egg | Pos | 6250 | Santiago | Dried Onion | Pos |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1585 | Arizonae | Turtle Soil | Pos | 6735 | Albany | Sesame Seeds | Pos |
| 1590 | *Salmonella* 3b | Snake Regurgitate | Pos | 7111 | Infantis | Unknown | Pos |
| 1597 | *Salmonella* 3b | Gila Monster Feces | Pos | 12907 | Heidelberg | Poultry | Pos |
| 1608 | Seminole | Snake Feces | Pos | 12914 | Kentucky | Poultry | Pos |
| 1609 | Wassennaar | Iguana Swab | Pos | 12960 | Senftenberg | Poultry | Pos |
| 1614 | Kralendyk | Iguana Bladder | Pos | 12968 | Blockley | Unknown | Pos |
| 1616 | Houten IV | Imported Bird Feces | Pos | 13005 | Typhimurium | Poultry | Pos |
| 1620 | Carmel | Unknown | Pos | 13035 | Choleraesuis | ATCC 10708 | Pos |
| 1621 | Carrau | Unknown | Pos | 13056 | Senftenberg | Soy Manufacturing | Pos |
| 1623 | Champaign | Liver of Hen | Pos | 13064 | Tennessee | Soy Manufacturing | Pos |
| 1638 | Derby | Unknown | Pos | 13066 | Tennessee | Soy Manufacturing | Pos |
| 1652 | London | Unknown | Pos | 13067 | Havana | Soy Manufacturing | Pos |
| 1653 | Yovokome | Unknown | Pos | 13068 | Lexington | Soy Manufacturing | Pos |
| 1657 | Reading | Unknown | Pos | 13069 | Mbandaka | Soy Manufacturing | Pos |
| 1668 | California | Unknown | Pos | 13071 | Montevideo | Soy Manufacturing | Pos |
| 1680 | Dugbe | Unknown | Pos | 13075 | Cubana | Celery Seed | Pos |
| 1684 | Emmastad | Unknown | Pos | 13079 | Newport | Basil | Pos |
| 1686 | Fayed | Unknown | Pos | 13081 | Virchow | Basil | Pos |
| 1687 | Ferlac | Ceylonese Dessicated Coconut | Pos | 13344 | Berta | Unknown | Pos |
| 1698 | Madelia | Liver of Hen | Pos | 13630 | Hadar | Unknown | Pos |
| 1703 | Mississippi | Faeces in 1942 | Pos | SAFE73 | *Salmonella I4,[5],12:i:-* | Unknown | Pos |

**Table 5. Exclusivity Results for the BAX System X5 PCR Assay for *Salmonella* (13)**

| DuPont Strain ID | Genus and Species | Result | DuPont Strain ID | Genus and Species | Result |
|---|---|---|---|---|---|
| 373 | *Klebsiella pneumoniae* | Neg | 2554 | *Enterococcus faecalis* | Neg |
| 375 | *Entrobacter cloacae* | Neg | 2558 | *Citrobacter freundii* | Neg |
| 383 | *Citrobacter freundii* | Neg | 2559 | *Citrobacter amalonaticus* | Neg |
| 569 | *Pseudomonas fluorescens* | Neg | 2560 | *Citrobacter koseri* | Neg |
| 572 | *Aeromonas hydrophila* | Neg | 2584 | *Enterobacter hormaechei* | Neg |
| 576 | *Pseudomonas mendocina* | Neg | 2586 | *Klebsiella planticola* | Neg |
| 577 | *Pseudomonas stutzeri* | Neg | 2604 | *Enterobacter amnigenus* | Neg |
| 592 | *Yersinia enterocolitica* | Neg | 2631 | *Vibrio fluvialis* | Neg |
| 610 | *Staphylococcus aureus* | Neg | 2632 | *Vibrio vulnificus* | Neg |
| 657 | *Klebsiella ozaenae* | Neg | 3097 | *Citrobacter freundii* | Neg |
| 659 | *Lactococcus lactis* | Neg | 3785 | *Escherichia coli* | Neg |
| 700 | *Shigella sonnei* | Neg | 3982 | *Pseudomonas aeruginosa* | Neg |
| 715 | *Bacillus cereus* | Neg | 5588 | *Hafnia alvei* | Neg |
| 1081 | *Shigella boydii* | Neg | 6121 | *Prot. Mirabilis* | Neg |
| 1082 | *Shigella dysenteriae* | Neg | 6523 | *Klebsiella oxytoca* | Neg |
| 1450 | *Escherichia coli* | Neg | 6719 | *Escherichia hermanni* | Neg |
| 1458 | *Escherichia coli* | Neg | 6832 | *Shigella sonnei* | Neg |
| 2357 | *Proteus mirabilis* | Neg | 7083 | *Serratia marcesens* | Neg |
| 2389 | *Hafnia alvei* | Neg | 8877 | *Xanthomonas maltophilia* | Neg |
| 2399 | *Yersinia aldovae* | Neg | 10006 | *Enterobacter sakazakii* | Neg |
| 2435 | *Escherichia coli* | Neg | 11232 | *Vibrio mimicus* | Neg |
| 2443 | *Escherichia coli* | Neg | 12720 | *Enterobacter sakazakii* | Neg |
| 2514 | *Escherichia coli* | Neg | 12760 | *Enterobacter cloacae* | Neg |
| 2552 | *Enterococcus faecium* | Neg | 13041 | *Escherichia coli* | Neg |

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