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CERTIFICATION

AOAC® *Performance Tested*[SM]

Certificate No.

041801

The AOAC Research Institute hereby certifies the test kit known as:

PolySkope One Multiplex Pathogen Detection Assay

manufactured by

PolySkope Labs
755 Research Pkwy
Oklahoma City, OK 73104
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 (February 13, 2020 – December 31, 2020). 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

February 13, 2020
Date

METHOD AUTHORS	SUBMITTING COMPANY
Paul Smith and Michael Centola	**PolySkope Labs**
	755 Research Pkwy
	Oklahoma City, OK 73104
	USA

KIT NAME(S)	CATALOG NUMBERS
PolySkope One Multiplex Pathogen Detection Assay	04.00MX.096.QS5.01.01

INDEPENDENT LABORATORY	AOAC EXPERTS AND PEER REVIEWERS
Q Laboratories, Inc.	Yvonne Salfinger[1], Michael Brodsky[2], Brad Stawick[3]
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APPLICABILITY OF METHOD

Target organisms – STEC [*E. coli* O157:H7 & Non-O157 STEC Big 6 (O26, O45, O103, O111, O121, O145), *Listeria monocytogenes*, *Salmonella* species**

Matrices – 25 g – fresh raw ground beef (73% lean), deli turkey, fresh baby spinach, stainless steel (4 x 4 in sponge)

Performance claims - Performance equivalent to that of the reference

REFERENCE METHODS

USDA/FSIS-MLG 5.09: *Detection, Isolation and Identification of Escherichia coli O157:H7 from Meat Products and Carcass and Environmental Sponges.* **January, 2015. (2)**
USDA/FSIS-MLG 5B.05: *Detection and Isolation of non-O157 Shiga Toxin-Producing Escherichia coli (STEC) from Meat Products and Carcass and Environmental Sponges.* **June, 2014.(3)**
USDA MLG 8.10: *Isolation and Identification of Listeria monocytogenes from Red Meat, Poultry, Ready-To-Eat Siluriformes (Fish) and Egg Products, and Environmental Sponges.* **January 2ndt, 2017.http://www.fsis.usda.gov/wps/wcm/connect/1710bee8-76b9-4e6c-92fc-fdc290dbfa92/MLG-8.pdf?MOD=AJPERES (4)**
USDA MLG 4.09: *Isolation and Identification of Salmonella from Meat, Poultry, Pasteurized Egg, and Siluriformes (Fish) Products and Carcass and Environmental Sponges.* **Updated: January 2017. https://www.fsis.usda.gov/wps/wcm/connect/700c05fe-06a2-492a-a6e1-3357f7701f52/MLG-4.pdf?MOD=AJPERES (5)**
ISO/ Technical Specification (ISO/TS) 13136: 2012: *Microbiology of food and animal feed – Real-time polymerase chain reaction (PCR)-BASED METHOD FOR THE DETECTION OF FOOD-BORNE PATHOGENS – Horizontal method for the detection of Shiga toxin-producing, Escherichia coli (STEC) and the determination of O157, O111, O26, O103 and O1045 serogroups.* **First Edition (2012-11-15) (6)**
FDA BAM Ch 10: *Detection and Enumeration of Listeria monocytogenes in Foods.* **February, 2013. http://www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/ucm071400.htm (7)**
FDA BAM Ch 5: *Salmonella.* **Revised: August 2016. https://www.fda.gov/food/foodscienceresearch/laboratorymethods/ucm070149.htm (8)**

ORIGINAL CERTIFICATION DATE	CERTIFICATION RENEWAL RECORD
April 03, 2018	**Renewed annually through December 2020**

METHOD MODIFICATION RECORD	SUMMARY OF MODIFICATION
1. **February 2020 Level 1**	1. **Editorial/clerical changes resulting from kit name change from "PolySkope 1.0 Multiplex Pathogen Detection Assay" to "PolySkope One Multiplex Pathogen Detection Assay".**

Under this AOAC® *Performance Tested*^{SM} License Number, 041801 this method is distributed by: NONE	Under this AOAC® *Performance Tested*^{SM} License Number, 041801 this method is distributed as: NONE

PRINCIPLE OF THE METHOD (1)

The Polyskope 1.0 Multiplex Pathogen Detection Assay is a test based on gene amplification and detection by real-time PCR. Ready-to-use PCR reagents contain oligonucleotides (primers and probes) specific to *E. coli* O157, non-O157 STEC, *Listeria monocytogenes,* and *Salmonella* species as well as DNA polymerase and nucleotides. PCR is a well-established technique used to rapidly generate profuse copies of target DNA. During the PCR reaction, cycles of heating and cooling promote DNA denaturation, followed by primers binding to specific target regions. The DNA polymerase then recognizes these primers and utilizes deoxynucleotide triphosphates (dNTPs) to extend the DNA, creating copies of the target DNA, called amplicons. Next, specific probes are used to detect the DNA during the amplification, by hybridizing to the amplicons. These probes are bound to a fluorophore which fluoresces only when hybridized to the correct target sequence. In the absence of target DNA, no fluorescence will be detected. As the amplicons increase with each round of amplification, fluorescence intensity also increases. At the annealing step of each PCR cycle, the detector measures this fluorescence and the associated software plots the fluorescence intensity versus number of cycles. This method allows a simple determination of the presence, or absence, of up to five targets in a single reaction.

An unrelated DNA "internal control" is included in the reaction mix. This control is amplified with a specific probe at the same time as the other probe target DNA sequences and detected by a specific fluorophore. It allows for the validation of any negative result. Polyskope 1.0 Multiplex Pathogen Detection assay is specifically designed to detect pathogenic bacteria capable of human infection. The oligonucleotides are targeted to specific pathogen-related genes that are present in these bacteria and distinguish them from closely related non-pathogenic bacteria.

The PolySkope 1.0 Multiplex Pathogen Detection method allows the simultaneous detection of *E. coli* O157 STEC, *E. coli* non-O157 STEC*, Salmonella* spp. and *Listeria monocytogenes* in select environmental samples and select food products enriched with Polyskope Multiplex Enrichment Media (PMEM). [10]

DISCUSSION OF THE VALIDATION STUDY (1)

The PolySkope 1.0 Multiplex Pathogen Detection Assay provides qualitative detection of virulence factors (*stx1, stx 2 and eae*) for *E. coli* O157 and non-*E. coli* O157 STEC (O26, O45, O103, O111, O121 and O145), *Listeria monocytogenes* and *Salmonella* spp. Because the PolySkope method utilizes a multiplex reaction, it has the ability to detect multiple common pathogens within a single reaction. This enables the user to save time and cost per test by only having to prepare a single enrichment, conduct a single lysis sample, and run a single PCR reaction. The software is simple and easy to navigate and allows the user to view Real-Time results. Each individual reaction taking place within a single sample can be interpreted throughout the entire run, including the final analysis. The software does not present the typical stop light result (Green - positive, red - negative), but requires interpretation of the results. An analysis of the curves and the Cq values by a trained analyst are required to obtain a final result.

In the inclusivity and exclusivity evaluations, all inclusivity organisms were correctly included and all exclusivity organisms were correctly excluded. In the method comparison study, the PolySkope 1.0 Multiplex Pathogen Detection Assay demonstrated no statistically significant differences between candidate and reference method results (dPOD$_C$), or between presumptive and confirmed results (dPOD$_{CP}$) for all target pathogens. During the robustness evaluation, the change to the operational parameters of the method proved the method is robust and had not negative impact on the testing. For the product stability evaluation, the test kit proved to be unaffected by the storage conditions and lot-to-lot variations.

No.	Species	Serotype	Source	Origin	Result	No.	Species	Serotype	Source	Origin	Result
1	*E. coli*	O26	ATCC BAA-1653	Stool	+	26	*E. coli*	O103	QL 15071-2	Meat Powder	+
2	*E. coli*	O26	MSU TW 07862	Calf, Cow	+	27	*E. coli*	O111:H12	MSU DEC 6A	Infant	+
3	*E. coli*	O26	MSU TW02295	Infant	+	28	*E. coli*	O111:H8	MSU DEC 6C	Human	+
4	*E. coli*	O26	MSU DEC 9F	Human	+	29	*E. coli*	O111	MSU DEC 8D	Infant	+
5	*E. coli*	O26	MSU TW04270	Human	+	30	*E. coli*	O111	MSU TW07926	Human	+
6	*E. coli*	O26	MSU TW04284	Child	+	31	*E. coli*	O111	MSU TW14960	Human	+
7	*E. coli*	O26	MSU TW08031	Human	+	32	*E. coli*	O111	MSU TW06296	Child	+
8	*E. coli*	O26	MSU TW07814	Human	+	33	*E. coli*	O111	MSU TW05614	Human	+
9	*E. coli*	O26	MSU TW00971	Human Feces	+	34	*E. coli*	O111	MSU TW00186	Human	+
10	*E. coli*	O26	MSU TW05992	Human	+	35	*E. coli*	O111	MSU TW01387	Human	+
11	*E. coli*	O45	MSU TW10121	Human	+	36	*E. coli*	O121	PSU 10.0709	Not Available	+
12	*E. coli*	O45	MSU TW14003	Human	+	37	*E. coli*	O121	PSU 5.0959	Not Available	+
13	*E. coli*	O45	MSU TW07947	Human	+	38	*E. coli*	O121	PSU 7.1686	Not Available	+
14	*E. coli*	O45	MSU DEC 11C	Human	+	39	*E. coli*	O121	PSU 7.1709	Not Available	+
15	*E. coli*	O45	PSU 1.2622	Not Available	+	40	*E. coli*	O121	PSU 7.1732	Not Available	+
16	*E. coli*	O45	PSU 1.2635	Not Available	+	41	*E. coli*	O121	MSU TW07931	Human	+
17	*E. coli*	O45	PSU 2.0164	Not Available	+	42	*E. coli*	O121	MSU TW07614	Human	+
18	*E. coli*	O45	PSU 11.1079	Not Available	+	43	*E. coli*	O121	MSU TW08023	Human	+
19	*E. coli*	O103	MSU TW09101	Human	+	44	*E. coli*	O145	QL 15071-1	Meat Powder	+
20	*E. coli*	O103	MSU TW07971	Human	+	45	*E. coli*	O145	PSU 7.1711	Not Available	+
21	*E. coli*	O103	MSU TW11239	Child	+	46	*E. coli*	O145	PSU 10.0707	Not Available	+
22	*E. coli*	O103	MSU TW07697	Human	+	47	*E. coli*	O145	MSU TW09153	Human	+
23	*E. coli*	O103	PSU 5.1658	Not Available	+	48	*E. coli*	O145	MSU TW07596	Human	+
24	*E. coli*	O103	PSU 7.1691	Not Available	+	49	*E. coli*	O145	MSU TW09356	Human	+
25	*E. coli*	O103	PSU 9.0036	Not Available	+	50	*E. coli*	O145	MSU TW01664	Human	+

Table 2: Inclusivity Results for *E. coli* O157 (1)											
No.	Species	Serotype	Source	Origin	Result	No.	Species	Serotype	Source	Origin	Result
1	*E. coli*	O157	MSU[1] TW00116	Human	+	26	*E. coli*	O157	MSU DEC3B	Human	+
2	*E. coli*	O157	MSU TW00975	Human	+	27	*E. coli*	O157	MSU DEC3C	Human	+
3	*E. coli*	O157	MSU TW02302	Hamburger	+	28	*E. coli*	O157	MSU DEC3D	Human	+
4	*E. coli*	O157	MSU TW04863	Human	+	29	*E. coli*	O157	MSU DEC3E	Human	+
5	*E. coli*	O157	MSU TW05356	Human	+	30	*E. coli*	O157	MSU DEC4A	Human	+
6	*E. coli*	O157	MSU TW07587	Human	+	31	*E. coli*	O157	MSU DEC4B	Human	+
7	*E. coli*	O157	ATCC[2] BAA-460	Human Feces	+	32	*E. coli*	O157	MSU DEC4C	Buffalo	+
8	*E. coli*	O157	NCTC[3] 12900	Not Available	+	33	*E. coli*	O157	MSU DEC4D	Cow, Calf	+
9	*E. coli*	O157	NCTC 13125	Human Stool	+	34	*E. coli*	O157	MSU DEC4E	Human	+
10	*E. coli*	O157	NCTC 13126	Not Available	+	35	*E. coli*	O157	QL[4] 164673	Beef Trim	+
11	*E. coli*	O157	NCTC 13127	Not Available	+	36	*E. coli*	O157	QL 2-202	Meat	+
12	*E. coli*	O157	NCTC 13128	Not Available	+	37	*E. coli*	O157	QL 2-203	Meat	+
13	*E. coli*	O157	ATCC 35150	Human Feces	+	38	*E. coli*	O157	QL 2-204	Meat	+
14	*E. coli*	O157	ATCC 43888	Human Feces	+	39	*E. coli*	O157	QL 2-205	Meat	+
15	*E. coli*	O157	ATCC 43889	Human Feces	+	40	*E. coli*	O157	QL 2-206	Meat	+
16	*E. coli*	O157	ATCC 43890	Human Feces	+	41	*E. coli*	O157	QL 2-207	Meat	+
17	*E. coli*	O157	ATCC 43894	Human Feces	+	42	*E. coli*	O157	QL 2-214	Meat	+
18	*E. coli*	O157	ATCC 43895	Raw Hamburger	+	43	*E. coli*	O157	QL 2-701	Beef	+
19	*E. coli*	O157	ATCC 51657	Clinical Isolate	+	44	*E. coli*	O157	QL 2-704	Beef	+
20	*E. coli*	O157	ATCC 51658	Clinical Isolate	+	45	*E. coli*	O157	QL 2-705	Beef	+
21	*E. coli*	O157	ATCC 51659	Clinical Isolate	+	46	*E. coli*	O157	QL 2-706	Beef	+
22	*E. coli*	O157	ATCC 700531	Clinical Isolate	+	47	*E. coli*	O157	QL 2-707	Beef	+
23	*E. coli*	O157	ATCC 700599	Salami	+	48	*E. coli*	O157	QL 2-708	Beef	+
24	*E. coli*	O157	ATCC 700927	Not Available	+	49	*E. coli*	O157	QL 2-710	Beef	+
25	*E. coli*	O157	MSU DEC3A	Human	+	50	*E. coli*	O157	QL 14077.1	Meat	+

1. MSU – Michigan State University Culture Collection, 2. ATCC – American Type Culture Collection, 3. NCTC – National Culture Type Collection, 4. QL – Q Laboratories Culture Collection

Table 3: Inclusivity Results for *Salmonella* (1)

No.	Organism	Source	Origin	Result	No.	Organism	Source	Origin	Result
1	*Salmonella* bongori	NCTC[1] 12419	Not Available	+	26	*Salmonella* Berta	UPENN STS 13	Not Available	+
2	*Salmonella* bongori	ATCC[2] 43975	Not Available	+	27	*Salmonella* Binza	UPENN STS 14	Not Available	+
3	*Salmonella* bongori	NCTC 10946	Amphibian, Frog	+	28	*Salmonella* Bovis-Morbificans	UPENN STS 16	Not Available	+
4	*Salmonella* Artis	ATCC 700149	Not Available	+	29	*Salmonella* Brandenburg	UPENN STS 18	Not Available	+
5	*Salmonella* Salamae	QL[3] 02415	Clinical Isolate	+	30	*Salmonella* Bredeney	NCTC 5731	Not Available	+
6	*Salmonella* Basel	ATCC 700151	Not Available	+	31	*Salmonella* California	NCTC 6018	Not Available	+
7	*Salmonella* Arizonae	ATCC 13314	Not Available	+	32	*Salmonella* Cerro	UPENN STS 22	Not Available	+
8	*Salmonella* Arizonae	ATCC BAA-1577	Not Available	+	33	*Salmonella* Choleraesuis	ATCC 10708	Not Available	+
9	*Salmonella* Arizonae	QL 11007-4	Veterinary	+	34	*Salmonella* Choleraesuis var Kunzendorf	ATCC 12011	Not Available	+
10	*Salmonella*. Diarizonae	ATCC BAA-1579	Not Available	+	35	*Salmonella* Cubana	UPENN STS 24	Not Available	+
11	*Salmonella* Diarizonae	ATCC BAA-216	Human Blood	+	36	*Salmonella* Derby	NCTC 5721	Not Available	+
12	*Salmonella*. Diarizonae	ATCC BAA-639	Human Feces	+	37	*Salmonella* Drypool	UPENN STS 26	Not Available	+
13	*Salmonella* Abaetetuba	ATCC 35640	Creek Water	+	38	*Salmonella* Dublin	UPENN STS 27	Not Available	+
14	*Salmonella* Abortusequi	FDA[4] 9842	Not Available	+	39	*Salmonella* Eastbourne	FDA 4017H	Not Available	+
15	*Salmonella* Abortusovis	NCTC10241	Not Available	+	40	*Salmonella* Enteritidis	ATCC 13076	Not Available	+
16	*Salmonella* Abony	NCTC 6017	Not Available	+	41	*Salmonella* Galiema	QL 024.2	Clinical Isolate	+
17	*Salmonella* Adelaide	UPENN[5] STS 2	Not Available	+	42	*Salmonella* Give	UPENN STS 42	Not Available	+
18	*Salmonella* Agona	ATCC 51957	Not Available	+	43	*Salmonella* Haardt	UPENN STS 44	Not Available	+
19	*Salmonella* Agama	UPENN STS 3	Not Available	+	44	*Salmonella* Hadar	ATCC 51956	Not Available	+
20	*Salmonella* Agoueve	UPENN STS 5	Not Available	+	45	*Salmonella* Havana	UPENN STS 47	Not Available	+
21	*Salmonella* Alachua	UPENN STS 6	Not Available	+	46	*Salmonella* Heidelberg	ATCC 8326	Not Available	+
22	*Salmonella* Albany	UPENN STS 7	Not Available	+	47	*Salmonella* Illinois	ATCC 11646	Not Available	+
23	*Salmonella* Anatum	ATCC 9270	Pork Liver	+	48	*Salmonella* Indiana	NCTC 11304	Turkey	+
24	*Salmonella* Arkansas	UPENN STS 11	Not Available	+	49	*Salmonella* Infantis	ATCC 51741	Pasta	+
25	*Salmonella* Bareilly	FDA 1206H	Not Available	+	50	*Salmonella* Javiana	ATCC 10721	Not Available	+

1. NCTC – National Culture Type Collection, 2. ATCC – American Type Culture Collection, 3. QL – Q Laboratories Culture Collection, 4. FDA – US Food and Drug Administration Culture Collection, 5. UPENN – University of Pennsylvania Culture Collection

No.	Organism	Source	Origin	Result	No.	Organism	Source	Origin	Result
51	*Salmonella* Jerusalem	QL[1] 024.12	Dry Dog Food	+	76	*Salmonella* Paratyphi A	ATCC 9150	Not Available	+
52	*Salmonella* Johannesburg	UPENN[2] STS 56	Not Available	+	77	*Salmonella* Paratyphi B	ATCC 10719	Not Available	+
53	*Salmonella* Kahla	ATCC[3] 17980	Human Feces	+	78	*Salmonella* Paratyphi C	ATCC 13428	Not Available	+
54	*Salmonella* Kaitaan	QL 024.7	Clinical Isolate	+	79	*Salmonella* Pomona	ATCC 10729	Clinical Isolate	+
55	*Salmonella* Kentucky	ATCC 9263	Not Available	+	80	*Salmonella* Poona	NCTC 4840	Infant	+
56	*Salmonella* Krefeld	UPENN STS 58	Not Available	+	81	*Salmonella* Preston	QL 024.16	Clinical Isolate	+
57	*Salmonella* Indica	ATCC BAA-1578	Unknown, India	+	82	*Salmonella* Pullorum	ATCC 13036	Egg	+
58	*Salmonella* Ferlac	ATCC 43976	Not Available	+	83	*Salmonella* Rubislaw	UPENN STS 92	Not Available	+
59	*Salmonella* Ferlac	NCTC[4] 10458	Desiccated Coconut	+	84	*Salmonella* Saintpaul	ATCC 9712	Cystitis	+
60	*Salmonella* Lille	UPENN STS 59	Not Available	+	85	*Salmonella* San-Diego	UPENN STS 94	Not Available	+
61	*Salmonella* Livingstone	UPENN STS 63	Not Available	+	86	*Salmonella* Schalkwijk	QL 024.10	Clinical Isolate	+
62	*Salmonella* London	UPENN STS 64	Not Available	+	87	*Salmonella* Schwarzengrund	UPENN STS 95	Not Available	+
63	*Salmonella* Manhattan	UPENN STS 65	Not Available	+	88	*Salmonella* Senftenberg	ATCC 43845	Not Available	+
64	*Salmonella* Mbankaka	FDA[5] 37N	Not Available	+	89	*Salmonella* Stanley	ATCC 7308	Not Available	+
65	*Salmonella* Menden	ATCC 15992	Human Feces	+	90	*Salmonella* Tallahassee	ATCC 12002	Not Available	+
66	*Salmonella* Meleagridis	QL 12074-1	Not Available	+	91	*Salmonella* Tennessee	QL 024.6	Clinical Isolate	+
67	*Salmonella* Menhaden	QL 024.20	Clinical Isolate	+	92	*Salmonella* Thompson	FDA 2051H	Not Available	+
68	*Salmonella* Montevideo	ATCC 8387	Not Available	+	93	*Salmonella* Typhi	ATCC 6539	Not Available	+
69	*Salmonella* Muenchen	ATCC BAA-1594	Roma Tomatoes	+	94	*Salmonella* Typhimurium	ATCC 14028	Animal Tissue	+
70	*Salmonella* Neasden	QL 024.4	Clinical Isolate	+	95	*Salmonella* Utrect	NCTC 10077	Not Available	+
71	*Salmonella* Newington	QL 0248	Clinical Isolate	+	96	*Salmonella* Urbana	UPENN STS 110	Not Available	+
72	*Salmonella* Newport	ATCC 6962	Unknown, England	+	97	*Salmonella* Vellore	ATCC 15611	Rectal Swab	+
73	*Salmonella* Ohio	UPENN STS 81	Unknown, Illinois Hospital	+	98	*Salmonella* Virchow	ATCC 51955	Not Available	+
74	*Salmonella* Oranienburg	ATCC 9239	Not Available	+	99	*Salmonella* Volta	QL 024.9	Clinical Isolate	+
75	*Salmonella* Orthmarshen	QL 024.13	Clinical Isolate	+	100	*Salmonella* Westhampton	QL 024.14	Clinical Isolate	+

Table 3: Inclusivity Results for *Salmonella* (continued) (1)

1. QL – Q Laboratories Culture Collection, 2. UPENN – University of Pennsylvania Culture Collection, 3. ATCC – American Type Culture Collection, 4. NCTC – National Culture Type Collection, 5. FDA – US Food and Drug Administration Culture Collection

Table 4: Exclusivity Results for Gram Negative Organisms (1)

No	Organism	Source	Origin	Result	No	Organism	Source	Origin	Result
1	*Alcaligenes faecalis*	ATCC[1] 8750	Not Available	-	16	*Escherichia hermanii*	ATCC 33650	Mouse Brain	-
2	*Aeromonoas hydrophila*	ATCC 49140	Clinical Isolate	-	17	*Escherichia vulneris*	ATCC 29943	Human Wound	-
3	*Citrobacter braakii*	ATCC 43162	Clinical Isolate	-	18	*Hafnia alvei*	ATCC 51815	Milk	-
4	*Citrobacter farmeri*	ATCC 51633	Human Feces	-	19	*Haemophilus influenzae*	ATCC 19418	Not Available	-
5	*Cronobacter sakazakii*	QL[2] 17031.4	Infant Formula	-	20	*Klebsiella pneumoniae*	ATCC 4352	Cow Milk	-
6	*Edwardsiella tarda*	ATCC 15947	Human Feces	-	21	*Morganella morganii*	ATCC 25829	Human	-
7	*Enterobacter aerogenes*	ATCC 13048	Sputum	-	22	*Mycobacterium smegmatis*	ATCC 19420	Not Available	-
8	*Escherichia blattae*	ATCC 29907	Insect	-	23	*Pantoea agglomerans*	ATCC 19552	Sewage	-
9	*Escherichia coli* O55	MSU[3] DEC1A	Human Feces	-	24	*Proteus mirabilis*	ATCC 7002	Urine	-
10	*Escherichia coli* O113	NCTC[4] 9113	Not Available	-	25	*Providencia rettgeri*	ATCC 14505	Not Available	-
11	*Escherichia coli* O115	NCTC10444	Calf	-	26	*Pseudomonas aeruginosa*	ATCC 9027	Ear Infection	-
12	*Escherichia coli* O117	NCTC 9117	Not Available	-	27	*Rahnella aquatilis*	ATCC 55046	Soil	-
13	*Escherichia coli* O118	NCTC 9118	Not Available	-	28	*Serratia marcescens*	ATCC 13880	Human	-
14	*Escherichia coli* O163	NCTC 11021	Human Feces	-	29	*Shigella boydii*	ATCC 9290	Pork Liver	-
15	*Escherichia fergusonii*	ATCC 35469	Human Feces	-	30	*Vibrio vulnificus*	QL 02111-1A	Seafood Product	-

1. ATCC – American Type Culture Collection, 2. QL – Q Laboratories Culture Collection, 3. MSU – Michigan State University Culture Collection, 4. NCTC – National Culture Type Collection

No.	Organism	Source	Origin	Result	No.	Organism	Source	Origin	Result
	Table 5: Inclusivity Results for *Listeria monocytogenes* (1)								
1	*L. monocytogenes* (1/2C)	CWD[1] 1553	Not Available	+	26	*L. monocytogenes* (N/A)	ATCC 19113	Not Available	+
2	*L. monocytogenes* (1/2A)	CWD 1554	Unknown, Carlisle, 1981	+	27	*L. monocytogenes* (4A)	ATCC 19114	Animal Tissue	+
3	*L. monocytogenes* (4B)	CWD 1563	Unknown, Lausanne ,1987	+	28	*L. monocytogenes* (4B)	ATCC 19115	Human	+
4	*L. monocytogenes* (4B)	CWD 1567	Unknown, Los Angeles, 1985	+	29	*L. monocytogenes* (4C)	ATCC 19116	Chicken	+
5	*L. monocytogenes* (4B)	CWD 1571	Not Available	+	30	*L. monocytogenes* (4E)	ATCC 19118	Chicken	+
6	*L. monocytogenes* (4B)	CWD 1590	Unknown, San Francisco	+	31	*L. monocytogenes* (N/A)	ATCC 49953	Goat, Belgium	+
7	*L. monocytogenes* (3B)	CWD 1600	Not Available	+	32	*L. monocytogenes* (1/2A)	ATCC 49594	Food, France	+
8	*L. monocytogenes* (1/2A)	CWD 1609	Turkey Factory	+	33	*L. monocytogenes* (3A)	ATCC 51782	Cheese	+
9	*L. monocytogenes* (1/2A)	CWD 1620	Turkey Factory	+	34	*L. monocytogenes* (N/A)	ATCC BAA-2658	Not Available	+
10	*L. monocytogenes* (1/2B)	CWD 1626	Turkey Franks	+	35	*L. monocytogenes* (N/A)	QL[5] 030911-10	Clinical	+
11	*L. monocytogenes* (1/2B)	CWD 1627	Mother/Baby	+	36	*L. monocytogenes* (4B)	CWD 1561	Placenta	+
12	*L. monocytogenes* (4D)	ATCC[2] 19117	Sheep	+	37	*L. monocytogenes* (1/2B)	CWD 1601	Unknown, Los Angeles	+
13	*L. monocytogenes* (1/2A)	ATCC 51772	Not Available	+	38	*L. monocytogenes* (1/2A)	CWD 1612	Turkey Factory	+
14	*L. monocytogenes* (4B)	ATCC 51778	Dairy Products	+	39	*L. monocytogenes* (1/A)	CWD 1613	Turkey Factory	+
15	*L. monocytogenes* (1/2B)	ATCC 51780	Cheese	+	40	*L. monocytogenes* (1/2A)	CWD 1614	Unknown, Oklahoma	+
16	*L. monocytogenes* (1/2B)	ATCC BAA-751	Not Available	+	41	*L. monocytogenes* (1/2A)	CWD 1618	Turkey Factory	+
17	*L. monocytogenes* (7)	NCTC[3] 10890	Human Feces	+	42	*L. monocytogenes* (1/2A)	CWD 1629	Turkey Franks	+
18	*L. monocytogenes* (4B)	FSL[4]-F6-367	Not Available	+	43	*L. monocytogenes* (1/2A)	CWD 1630	Turkey Factory	+
19	*L. monocytogenes* (4AB)	FSL J1-129	Not Available	+	44	*L. monocytogenes* (4B)	CWD 1574	Unknown, Halifax, 1983	+
20	*L. monocytogenes* (3C)	FSL J1-049	Not Available	+	45	*L. monocytogenes* (1/2B)	CWD 1584	Not Available	+
21	*L. monocytogenes* (1/2C)	ATCC 7644	Human	+	46	*L. monocytogenes* (3B)	CWD 1586	Not Available	+
22	*L. monocytogenes* (4B)	ATCC 13932	Child with Meningitis	+	47	*L. monocytogenes* (1/2B)	CWD 1588	Not Available	+
23	*L. monocytogenes* (1/2A)	ATCC 15313	Rabbit	+	48	*L. monocytogenes* (4B)	CWD 1596	Not Available	+
24	*L. monocytogenes* (1)	ATCC 19111	Poultry	+	49	*L. monocytogenes* (1/2B)	CWD 1597	Not Available	+
25	*L. monocytogenes* (2)	ATCC 19112	Spinal Fluid	+	50	*L. monocytogenes* (1/2A)	CWD 1611	Turkey Factory	+

1. CWD – University of Vermont Culture Collection, 2. ATCC – American Type Culture Collection, 3. NCTC – National Culture Type Collection, 4. FSL – Cornell University Culture Collection, 5. QL – Q Laboratories Culture Collection

Table 6: Exclusivity Results for *Listeria monocytogenes* (1)

No	Organism	Source	Origin	Result	No	Organism	Source	Origin	Result
1	*L. grayi*	ATCC[1] 19120	Animal Feces	-	16	*Enterococcus faecalis*	ATCC 19433	Not Available	-
2	*L. innocua*	ATCC 33090	Cow Brain	-	17	*Kurthia gibsonii*	ATCC 43195	Meat	-
3	*L. ivanovii*	ATCC 19119	Sheep	-	18	*Lactobacillus fermentum*	ATCC 9338	Not Available	-
4	*L. marthii*	ATCC BAA-1595	Soil	-	19	*Lactobacillus acidophilus*	ATCC 314	Not Available	-
5	*L. rocourtiae*	FSL[2] F6-0920	Not Available	-	20	*Lactobacillus plantarum*	ATCC 8014	Not Available	-
6	*L. welshimeri*	ATCC 35897	Not Available	-	21	*Lactococcus lactis*	ATCC 4797	Not Available	-
7	*L. seeligeri*	ATCC 35967	Soil	-	22	*Rhodococcus equi*	ATCC 6939	Not Available	-
8	*Aeromonas hydrophila*	ATCC 49140	Clinical Isolate	-	23	*Staphylococcus aureus*	ATCC 29213	Wound	-
9	*Bacillus cereus*	ATCC 6464	Soil	-	24	*Staphylococcus saprophyticus*	ATCC 15305	Urine	-
10	*Bacillus mycoides*	ATCC 6462	Soil	-	25	*Staphylococcus epidermidis*	ATCC 12228	Not Available	-
11	*Bacillus subtilis*	ATCC 27370	Not Available	-	26	*Staphylococcus haemolyticus*	ATCC 29970	Human Skin	-
12	*Bacillus lichenformis*	ATCC 12759	Plant	-	27	*Staphylococcus hominis*	ATCC 27844	Human Skin	-
13	*Brochothrix thermosphacta*	ATCC 11509	Animal Derived Foodstuff	-	28	*Staphylococcus warneri*	ATCC 29885	Not Available	-
14	*Enterobacter cloacae*	ATCC 23355	Not Available	-	29	*Streptococcus mutans*	ATCC 25175	Not Available	-
15	*Enterococcus durans*	ATCC 19432	Not Available	-	30	*Streptococcus pyogenes*	ATCC 19615	Pharynx of Child	-

1. ATCC – American Type Culture Collection, 2. FSL – Cornell University Culture Collection

Table 17: PolySkope 1.0 Multiplex Pathogen Detection Assay, Candidate vs. Reference – POD Results (1)

Matrix	Strain	MPN[a]/ Test Portion	N[b]	Candidate			Reference			dPOD$_C$[f]	95% CI[g]
				x[c]	POD$_C$[d]	95% CI	X	POD$_R$[e]	95% CI		
Fresh Raw Ground Beef	*E. coli* O157 ATCC 43895	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.44 (0.21, 0.76)	20	7	0.35	0.18, 0.57	6	0.30	0.15, 0.52	0.05	-0.23, 0.32
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	L. monocytogenes ATCC 7644	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.55 (0.29, 0.94)	20	7	0.35	0.18, 0.57	8	0.40	0.22, 0.61	-0.05	-0.32, 0.23
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	Salmonella Typhimurium ATCC 14028	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.35 (0.17, 0.62)	20	9	0.45	0.26, 0.66	5	0.25	0.11, 0.47	0.20	-0.09, 0.45
		2.29 (1.05, 5.02)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43

[a]MPN = Most Probable Number is calculated using the LCF MPN calculator provided by AOAC RI, with 95% confidence interval
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD$_C$ = Candidate method confirmed positive outcomes divided by the total number of trials
[e]POD$_R$ = Reference method confirmed positive outcomes divided by the total number of trials
[f]dPOD$_C$= Difference between the confirmed candidate method result and reference method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 18: PolySkope 1.0 Multiplex Pathogen Detection Assay, Candidate vs. Reference – POD Results (1)

Matrix	Strain	MPN[a]/ Test Portion	N[b]	Candidate			Reference			dPOD_C[f]	95% CI[g]
				x[c]	POD_C[d]	95% CI	X	POD_R[e]	95% CI		
Deli Turkey	E. coli O26 MSU TW00971	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.35 (0.14, 0.62)	20	7	0.35	0.18, 0.57	5	0.25	0.11, 0.47	0.10	-0.18, 0.36
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	L. monocytogenes ATCC 19115	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.55 (0.29, 0.93)	20	11	0.55	0.34, 0.74	8	0.40	0.22, 0.61	0.15	-0.15, 0.41
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	Salmonella Dublin ATCC 15480	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.76 (0.41, 1.27)	20	13	0.65	0.43, 0.82	10	0.50	0.30, 0.70	0.15	-0.15, 0.41
		4.38 (1.72, 11.15)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43

[a]MPN = Most Probable Number is calculated using the LCF MPN calculator provided by AOAC RI, with 95% confidence interval
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD_C = Candidate method confirmed positive outcomes divided by the total number of trials
[e]POD_R = Reference method confirmed positive outcomes divided by the total number of trials
[f]dPOD_C = Difference between the confirmed candidate method result and reference method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 19: PolySkope 1.0 Multiplex Pathogen Detection Assay, Candidate vs. Reference – POD Results (1)

Matrix	Strain	MPN[a]/ Test Portion	N[b]	Candidate			Reference			dPOD$_C$[f]	95% CI[g]
				x[c]	POD$_C$[d]	95% CI	X	POD$_R$[e]	95% CI		
Fresh Baby Spinach	*E. coli* O145 MSU TW09153	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.54 (0.29, 0.90)	20	9	0.45	0.26, 0.66	7	0.35	0.18, 0.57	0.10	-0.19, 0.37
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	L. monocytogenes ATCC BAA-2658	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.35 (0.14, 0.63)	20	7	0.35	0.18, 0.57	5	0.25	0.11, 0.47	0.10	-0.18, 0.36
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	Salmonella Enteritidis ATCC 13076	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		0.34 (0.14, 0.61)	20	7	0.35	0.18, 0.57	6	0.30	0.15, 0.52	0.05	-0.23, 0.32
		2.29 (1.05, 5.02)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43

[a]MPN = Most Probable Number is calculated using the LCF MPN calculator provided by AOAC RI, with 95% confidence interval
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD$_C$ = Candidate method confirmed positive outcomes divided by the total number of trials
[e]POD$_R$ = Reference method confirmed positive outcomes divided by the total number of trials
[f]dPOD$_C$= Difference between the confirmed candidate method result and reference method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 20: PolySkope 1.0 Multiplex Pathogen Detection Assay, Candidate vs. Reference – POD Results (1)

Matrix	Strain	CFU[a]/ Test Area	N[b]	Candidate			Reference			dPOD_C[f]	95% CI[g]
				x[c]	POD_C[d]	95% CI	X	POD_R[e]	95% CI		
Stainless Steel	E. coli O103 MSU TW08101	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		51	20	7	0.35	0.18, 0.57	6	0.30	0.15, 0.52	0.05	-0.23, 0.32
		440	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	L. monocytogenes ATCC 51780	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		64	20	4	0.20	0.08, 0.42	7	0.35	0.18, 0.57	-0.15	-0.40, 0.12
		650	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43
	Salmonella Kentucky ATCC 9263	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.43, 0.43
		45	20	5	0.25	0.11, 0.47	7	0.35	0.18, 0.57	-0.10	-0.36, 0.18
		580	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.43, 0.43

[a]CFU/Test Area = Results of the CFU/Test area were determined by plating the inoculum for each matrix in triplicate
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD_C = Candidate method confirmed positive outcomes divided by the total number of trials
[e]POD_R = Reference method confirmed positive outcomes divided by the total number of trials
[f]dPOD_C= Difference between the confirmed candidate method result and reference method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 21: PolySkope 1.0 Multiplex Pathogen Detection Assay, Presumptive vs. Confirmed – POD Results (1)											
Matrix	Strain	MPN[a]/ Test Portion	N[b]	Presumptive			Confirmed			dPOD$_{CP}$[f]	95% CI[g]
				x[c]	POD$_{CP}$[d]	95% CI	X	POD$_{CC}$[e]	95% CI		
Fresh Raw Ground Beef	E. coli O157 ATCC 43895	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.44 (0.21, 0.76)	20	7	0.35	0.18, 0.57	7	0.35	0.18, 0.57	0.00	-0.13, 0.13
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	L. monocytogenes ATCC 7644	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.55 (0.29, 0.94)	20	7	0.35	0.18, 0.57	7	0.35	0.18, 0.57	0.00	-0.13, 0.13
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	Salmonella Typhimurium ATCC 14028	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.35 (0.17, 0.62)	20	9	0.45	0.26, 0.66	9	0.45	0.26, 0.66	0.00	-0.13, 0.13
		2.29 (1.05, 5.02)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47

[a]MPN = Most Probable Number is calculated using the LCF MPN calculator provided by AOAC RI, with 95% confidence interval
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD$_{CP}$ = Candidate method presumptive positive outcomes divided by the total number of trials
[e]POD$_{CC}$ = Candidate method confirmed positive outcomes divided by the total number of trials
[f]dPOD$_{CP}$= Difference between the candidate method presumptive result and candidate method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 22: PolySkope 1.0 Multiplex Pathogen Detection Assay, Presumptive vs. Confirmed – POD Results (1)

Matrix	Strain	MPN[a]/ Test Portion	N[b]	Presumptive			Confirmed			dPOD$_{CP}$[f]	95% CI[g]
				x[c]	POD$_{CP}$[d]	95% CI	X	POD$_{CC}$[e]	95% CI		
Deli Turkey	E. coli O26 MSU TW00971	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.35 (0.14, 0.62)	20	7	0.35	0.18, 0.57	7	0.35	0.18, 0.57	0.00	-0.13, 0.13
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	L. monocytogenes ATCC 19115	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.55 (0.29, 0.93)	20	11	0.55	0.34, 0.74	11	0.55	0.34, 0.74	0.00	-0.13, 0.13
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	Salmonella Dublin ATCC 15480	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.76 (0.41, 1.27)	20	14	0.70	0.48, 0.85	13	0.65	0.43, 0.82	0.05	-0.11, 0.21
		4.38 (1.72, 11.15)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47

[a]MPN = Most Probable Number is calculated using the LCF MPN calculator provided by AOAC RI, with 95% confidence interval
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD$_{CP}$ = Candidate method presumptive positive outcomes divided by the total number of trials
[e]POD$_{CC}$ = Candidate method confirmed positive outcomes divided by the total number of trials
[f]dPOD$_{CP}$= Difference between the candidate method presumptive result and candidate method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 23: PolySkope 1.0 Multiplex Pathogen Detection Assay, Presumptive vs. Confirmed – POD Results (1)											
Matrix	Strain	MPN[a]/ Test Portion	N[b]	Presumptive			Confirmed			dPOD$_{CP}$[f]	95% CI[g]
				x[c]	POD$_{CP}$[d]	95% CI	X	POD$_{CC}$[e]	95% CI		
Fresh Baby Spinach	*E. coli* O145 MSU TW09153	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.54 (0.29, 0.90)	20	9	0.45	0.26, 0.66	9	0.45	0.26, 0.66	0.00	-0.13, 0.13
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	L. monocytogenes ATCC BAA-2658	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.35 (0.14, 0.63)	20	7	0.35	0.18, 0.57	7	0.35	0.18, 0.57	0.00	-0.13, 0.13
		3.01 (1.31, 6.89)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	Salmonella Enteritidis ATCC 13076	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		0.34 (0.14, 0.61)	20	7	0.35	0.18, 0.57	7	0.35	0.18, 0.57	0.00	-0.13, 0.13
		2.29 (1.05, 5.02)	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47

[a]MPN = Most Probable Number is calculated using the LCF MPN calculator provided by AOAC RI, with 95% confidence interval
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD$_{CP}$ = Candidate method presumptive positive outcomes divided by the total number of trials
[e]POD$_{CC}$ = Candidate method confirmed positive outcomes divided by the total number of trials
[f]dPOD$_{CP}$= Difference between the candidate method presumptive result and candidate method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

Table 24: PolySkope 1.0 Multiplex Pathogen Detection Assay, Presumptive vs. Confirmed – POD Results (1)

Matrix	Strain	CFU[a]/ Test Area	N[b]	Presumptive			Confirmed			dPOD$_{CP}$[f]	95% CI[g]
				x[c]	POD$_{CP}$[d]	95% CI	X	POD$_{CC}$[e]	95% CI		
Stainless Steel	E. coli O103 MSU TW08101	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		51	20	7	0.35	0.18, 0.57	7	0.35	0.18, 0.57	0.00	-0.13, 0.13
		440	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	L. monocytogenes ATCC 51780	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		64	20	4	0.20	0.08, 0.42	4	0.20	0.08, 0.42	0.00	-0.13, 0.13
		650	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47
	Salmonella Kentucky ATCC 9263	-	5	0	0.00	0.00, 0.43	0	0.00	0.00, 0.43	0.00	-0.47, 0.47
		45	20	5	0.25	0.11, 0.47	5	0.25	0.11, 0.47	0.00	-0.13, 0.13
		580	5	5	1.00	0.57, 1.00	5	1.00	0.57, 1.00	0.00	-0.47, 0.47

[a]CFU/Test Area = Results of the CFU/Test area were determined by plating the inoculum for each matrix in triplicate
[b]N = Number of test portions
[c]x = Number of positive test portions
[d]POD$_{CP}$ = Candidate method presumptive positive outcomes divided by the total number of trials
[e]POD$_{CC}$ = Candidate method confirmed positive outcomes divided by the total number of trials
[f]dPOD$_{CP}$= Difference between the candidate method presumptive result and candidate method confirmed result POD values
[g]95% CI = If the confidence interval of a dPOD does not contain zero, then the difference is statistically significant at the 5% level

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