



CERTIFICATION

AOAC[®] Performance TestedSM

Certificate No.

060402

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

Microgen Listeria-ID

manufactured by

Microgen Bioproducts Ltd.

Unit 1, Watchmoor Point

Camberley, Surrey GU15 3AD

United Kingdom

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 22, 2018 – December 31, 2018). Renewal may be granted at the end of one year under the rules stated in the licensing agreement.

Deborah McKenzie

Deborah McKenzie, Senior Director
Signature for AOAC Research Institute

February 22, 2018

Date

METHOD AUTHORS

ORIGINAL VALIDATION: Stuart Clark
 MODIFICATION FEBRUARY 2018: Microgen Bioproducts Ltd.

SUBMITTING COMPANY

Microgen Bioproducts Limited
 1 Admiralty Way
 Camberley, Surrey GU15 3DT
 United Kingdom

CURRENT LOCATION

Microgen Bioproducts Ltd.
 Unit 1, Watchmoor Point
 Camberley, Surrey GU15 3AD
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KIT NAME(S)

Microgen Listeria ID

CATALOG NUMBERS

MID-67

INDEPENDENT LABORATORY

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AOAC EXPERTS AND PEER REVIEWERS

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APPLICABILITY OF METHOD

Target organism – *Listeria species (monocytogenes, innocua, welshimeri, seeligeri, ivanovii, grayi)*

Matrices – Pure Culture

Performance claims - The Microgen Listeria ID product has been validated to:

Differentiate accurately between members of the genus *Listeria*
 Identify individual *Listeria* species from a single colony on a selective agar plate

Produce an accurate result within 18-24 hours

Match the performance of the FDA/BAM method

Match the performance of a leading competitor product which has AOAC performance tested status (API Listeria)

REFERENCE METHOD

Hitchins A.D. (1998) *Listeria monocytogenes* US FDA Bacteriological Analytical Manual 8th Edition, AOAC, Arlington, VA (3)

ORIGINAL CERTIFICATION DATE

September 27, 2004

CERTIFICATION RENEWAL RECORD

Renewed Annually through December 2018

METHOD MODIFICATION RECORD

1. February 2018 Level 2

SUMMARY OF MODIFICATION

1. Location change

Under this AOAC® Performance TestedSM License Number, 060402 this method is distributed by:
 NONE

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 NONE

PRINCIPLE OF THE METHOD (1)

The genus *Listeria* represents a group of species including *L. monocytogenes*, *L. innocua*, *L. welshimeri*, *L. seeligeri*, *L. ivanovii* and *L. grayi*. The product under test Microgen Listeria ID (MID 67) has been designed to enable laboratories to properly identify / differentiate these species when sampled from a single colony isolated on selective agar plates. The product utilizes the ability of each of these species to metabolise certain chemical substrates. The pattern of substrate metabolism produced in the MID 67 multiwell strip is characteristic for each species. The results from the multiwell plate are converted into four digit codes which are analysed by a dedicated computer software programme which calculates the percentage probability that the isolate being tested is one of the *Listeria* species. The most probable species is identified and this is taken as the result. If non-*Listeria* species colonies are introduced into the test system (which should not happen if the proper pre-testing has been correctly performed as per the manufacturers' insert) there are a number of tests included in the multiwell strip which should be metabolized by all *Listeria* species so non-*Listeria*s can be identified. Many of the substrates used in the MID 67 product are taken directly from the FDA BAM method including an in well haemolysis test to differentiate *L. monocytogenes* from *L. innocua*.

DISCUSSION OF THE VALIDATION STUDY (1)

The validation studies performed to assess the equivalence of the Microgen Listeria ID kit (Product Code MID 67) to the FDA / BAM approved *Listeria* Identification procedure have generated strong evidence that both methods agree with each other 100%. Furthermore, both methods successfully identified all of the individual *Listeria species* isolates and can discriminate between *Listeria species* and all of the non-*Listeria species* isolates even those which can be considered closely related.

Table 8: Summary of results of inclusivity study (1)

Organism	Microgen Listeria ID	Confirmed Status	Comments
<i>L. monocytogenes</i>	55	55	All 1/1
<i>L. innocua</i>	17	17	All 1/1
<i>L. seeligeri</i>	8	8	All 1/1
<i>L. welshimeri</i>	5	5	3 (1/1) / 2 (1/8)
<i>L. ivanovii</i>	3	3	All 1/3
<i>L. grayi</i>	3	3	All 1/2
<i>Other</i>	0	0	N/A
<i>Total</i>	91	91	

Table 9: Summary of Exclusivity study results (1)

No.	Listeria ID Result	Software Result	Probability	Organism Tested	Comment
1	4241	<i>L. ivanovii</i>	1/14,660,184	<i>B. cereus</i>	ARL Negative
2	4001	<i>L. ivanovii</i>	1/79,076,143	<i>B. cereus</i>	ARL Negative
3	4001	<i>L. ivanovii</i>	1/79,076,143	<i>B. mycoides</i>	ARL Negative
4	4001	<i>L. ivanovii</i>	1/79,076,143	<i>B. subtilis</i>	ARL Negative
5	6260	<i>L. grayi</i>	1/25,197,646	<i>Brochothrix</i>	ARL Negative
6	4240	<i>L. grayi</i>	1/815,539	<i>C. divergens</i>	ARL Negative
7	4260	<i>L. grayi</i>	1/814,723/887	<i>C. gallinarum</i>	ARL Negative
8	6244	<i>L. grayi</i>	1/58,853	<i>C. piscicola</i>	ARL Negative
9	3341	<i>L. ivanovii</i>	1/100,100,100	<i>C. freundii</i>	ARL Negative
10	5605	<i>L. ivanovii</i>	1/329	<i>E. aerogenes</i>	TRE Negative
11	0000	No. ID	N/A	<i>E. rhusiopathiae</i>	No ID
12	0000	No. ID	N/A	<i>Kurthia</i>	No ID
13	6060	<i>L. grayi</i>	1/100,100,100	<i>L. casei</i>	ARL Negative
14	6041	<i>L. ivanovii</i>	1/100,100,100	<i>L. lactis</i>	ARL Negative
15	6240	<i>L. grayi</i>	1/25,223	<i>L. plantarum</i>	ARL Negative
16	0000	No. ID	N/A	<i>Micrococcus sp.</i>	No ID
17	0000	No. ID	N/A	<i>P. freundenreichii</i>	No ID
18	0201	<i>L. ivanovii</i>	1/100,100,100	<i>P. mirabilis</i>	ESC Negative
19	1201	<i>L. ivanovii</i>	1/100,100,100	<i>P. vulgaris</i>	ESC Negative
20	0000	No. ID	N/A	<i>R. equi</i>	No ID
21	3340	<i>L. grayi</i>	1/100,100,100	<i>S. typhimurium</i>	ESC Negative
22	3240	<i>L. grayi</i>	1/100,100,100	<i>S. enteritidis</i>	ESC Negative
23	2041	<i>L. ivanovii</i>	1/100,100,100	<i>S. aureus</i>	ESC Negative
24	2041	<i>L. ivanovii</i>	1/100,100,100	<i>S. aureus</i>	ESC Negative
25	2240	<i>L. grayi</i>	1/25,197,646	<i>S. aureus</i>	ESC Negative
26	2041	<i>L. ivanovii</i>	1/100,100,100	<i>S. xylois</i>	ESC Negative
27	4041	<i>L. ivanovii</i>	1/9,773,456	<i>Streptococcus sp.</i>	ARL Negative
28	0230	<i>L. ivanovii</i>	1/100,100,100	<i>Streptococcus sp.</i>	ARL Negative
29	4241	<i>L. ivanovii</i>	1/14,660,184	<i>Streptococcus sp.</i>	ARL Negative
30	6260	<i>L. grayi</i>	1/25,197,646	<i>Streptococcus sp.</i>	ARL Negative
31	4240	<i>L. grayi</i>	1/815,539	<i>E. durans</i>	ARL Negative
32	0000	No. ID	N/A	<i>C. renale</i>	No ID

Table 11: Summary of Method Comparison Results (1)

Listeria Species	Sero-Type	ID Code	Microgen ID Result	Prob	FDA BAM Result
<i>L. monocytogenes</i>	1/2A	4547	<i>L. monocytogenes</i>	1/1	<i>L. monocytogenes</i>
<i>L. monocytogenes</i>	1/2B	4547	<i>L. monocytogenes</i>	1/1	<i>L. monocytogenes</i>
<i>L. monocytogenes</i>	1/2C	4547	<i>L. monocytogenes</i>	1/1	<i>L. monocytogenes</i>
<i>L. monocytogenes</i>	4B	4547	<i>L. monocytogenes</i>	1/1	<i>L. monocytogenes</i>
<i>L. monocytogenes</i>	4C	4547	<i>L. monocytogenes</i>	1/1	<i>L. monocytogenes</i>
<i>L. innocua</i>	6A	4546	<i>L. innocua</i>	1/1	<i>L. innocua</i>
<i>L. seeligeri</i>	1/2B	5445	<i>L. seeligeri</i>	1/1	<i>L. seeligeri</i>
<i>L. welshimeri</i>	6B	5566	<i>L. welshimeri</i>	1/1	<i>L. welshimeri</i>
<i>L. ivanovii</i>	N/A	5455	<i>L. ivanovii</i>	1/3	<i>L. ivanovii</i>
<i>L. grayi</i>	N/A	6642	<i>L. grayi</i>	1/2	<i>L. grayi</i>

Note: Probability = Prob.

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