

2. Primary enrichment

The dilution prepared in Half Fraser broth is incubated at $30^{\circ}\text{C} \pm 1^{\circ}\text{C}$ for 24-26 hours.

The sample from primary enrichment and secondary enrichment is then subcultured on HiCrome™ Listeria Ottaviani-Agosti Agar Base (M1540I) and on Listeria Oxford Medium Base (M1145) or Listeria Identification Agar Base (PALCAM) (M1064I). Incubate at $37 \pm 1^{\circ}\text{C}$ for 24 ± 2 hours. Additional incubation at $37 \pm 1^{\circ}\text{C}$ for 24 ± 2 hours is recommended for *Listeria* spp. other than *L.monocytogenes* for recovery of more species. (6,7)

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established hv efm oft should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets/

Limitations :

1. Presence of *L.monocytogenes* is often masked by other *Listeria* species like *L.inocua* and *L.ivanovii*.
2. Further subculture of organisms on selective media is required.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

bm u pouspm

bsbo

s bn up mmp ipnp o p t gs gmp o p e s

pmp s boe mbs u pg s bs e n e n

m ps t ou mmp pmp s e m bs tpm u po

5 H D F W L R Q

5 H D F W L R Q R I Z Y D T X H R X V V R O X W L R Q D W f & S + “

S +

& X O W X U D O U H V S R Q V H

Organism	Inoculum (CFU)	Growth	Esculin Hydrolysis	Recovery on M1540I*	Colour of colony on M1540*
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Productivity

Cultural characteristics observed on addition of FD141 after an incubation at $30 \pm 1^{\circ}\text{C}$ for 25 ± 1 hour. Further subculture is carried out on M1540I at $37 \pm 1^{\circ}\text{C}$ for 48 ± 4 hours.

<i>Listeria monocytogenes</i> 1/2a ATCC 35152 (00109*) +	50-100	good-luxuriant	positive reaction, blackening of medium	>10 colonies	Blue green colonies w/ opaque halo
<i>Escherichia coli</i> ATCC 25922 (00013*) +	$\geq 10^4$				
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	$\geq 10^4$				
<i>Listeria monocytogenes</i> 1/2a ATCC 35152 (00109*) +	50-100	good-luxuriant	positive reaction, blackening of medium	>10 colonies	Blue green colonies w/ opaque halo
<i>Escherichia coli</i> ATCC 8739 (00012*) +	$\geq 10^4$				
<i>Enterococcus faecalis</i> ATCC 19433 (00009*)	$\geq 10^4$				

Listeria monocytogenes 4b ATCC 13932 (00021*)+ <i>Escherichia coli</i> ATCC 8739 (00012*) + <i>Enterococcus faecalis</i> ATCC 19433 (00009*)	50-100 ≥10 ⁴ ≥10 ⁴	good-luxuriant	positive reaction, blackening of medium	>10 colonies	Blue green colonies w/ opaque halo
Listeria monocytogenes 4b ATCC 13932 (00021*)+ <i>Escherichia coli</i> ATCC 8739 (00012*) + <i>Enterococcus faecalis</i> ATCC 19433 (00009*)	50-100 ≥10 ⁴ ≥10 ⁴	good-luxuriant	positive reaction, blackening of medium	>10 colonies	Blue green colonies w/ opaque halo

Selectivity

Cultural characteristics observed on addition of FD141 after an incubation at 30 ± 1°C for 25 ± 1 hour. Further subculture is carried on Tryptone Soya Agar (M290) after an incubation at 37 ± 1°C for 48 ± 4 hours.

<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ⁴	inhibited	-	0
<i>Escherichia coli</i> ATCC 8739 (00012*)	≥10 ⁴	inhibited	-	0
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	≥10 ⁴	none-poor	-	<100 colonies
<i>Enterococcus faecalis</i> ATCC 19433 (00009*)	≥10 ⁴	none-poor	-	<100 colonies

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

Reference

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- Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
- Microbiology of the food chain — Horizontal method for the detection and enumeration of *Listeria monocytogenes* and of *Listeria* spp. - Part 1 , Detection method ; ISO 11290-1:2017
- Microbiology of the food chain — Horizontal method for the detection and enumeration of *Listeria monocytogenes* and of *Listeria* spp. - Part 2 , Detection method ; ISO 11290-2:2017

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 8. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
 9. Schuchat A. B., Swaminathan and C. V. Broome, Clin. Microbiol., Rev. 4 : 169-1

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