

## Enterobacteria Enrichment Broth Mossel

LQ119

### Intended use

For the enrichment of bile tolerant organisms in accordance with harmonized methods of USP, EP, BP, JP & IP.

### Composition\*\*

Ingredients	Gms / Litre
Gelatin peptone #	10.000
Glucose monohydrate	5.000
Dehydrated bile ##	20.000
Disodium hydrogen phosphate, dihydrate	8.000
Potassium dihydrogen phosphate	2.000
Brilliant green	0.015
pH after heating ( at 25°C)	7.2±0.2

# Pancreatic digest of gelatin

## Dehydrated ox-bile

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Label the ready to use LQ119 bottle. Inoculate the sample and Incubate at specified temperature and time.

### Principle And Interpretation

EE Broth, Mossel, formulated by Mossel et. al.(5-9) is recommended as an enrichment medium for bile tolerant gram-negative bacteria in the biological examination of foods (5), animal feed stuffs (13) and is in accordance with the harmonized method of USP/EP/BP/JP & IP (12,2,1,4,11).

Gelatin peptone and glucose monohydrate allows the growth of most of the members of *Enterobacteriaceae*. Brilliant green and dehydrated bile, purified are the inhibitory agents for gram-positive bacteria. Phosphates act as a good buffering agent and neutralizes acids produced by lactose fermenters that otherwise would adversely affect the growth of the organism.

Lactose negative, anaerogenic lactose-positive or late lactose fermenting *Enterobacteriaceae* are often missed by the standard Coli-aerogenes test. To overcome this problem, lactose is replaced by glucose in this medium. Phosphates form the buffering system of the medium. The cells damaged while drying or low pH are resuscitated in well-aerated Tryptone Soya Broth (M011) for 2 hours at 25°C prior to enrichment in EE Broth. The resuscitation procedure is recommended for dried foods (8), animal feeds (9) and semi-preserved foods (10). EE Broth is an enrichment broth and a loopful of the enriched sample is then subcultured onto Violet Red Bile Glucose Agar (MH581) after an initial incubation at 30-35°C for 24 hours. Typical pink colonies from MH581 are subcultured for biochemical confirmation by oxidase and fermentation reactions (4).

### Type of specimen

Pharmaceutical samples

### Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per guidelines (12,2,1,4,11). After use, contaminated materials must be sterilized by autoclaving before discarding.

### 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 guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

## Limitations

1. Further isolation on selective media is required to be carried out for confirmation.

## 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.

## Quality Control

### Appearance

Sterile clear Enterobacteria Enrichment Broth Mossel in bottle

### Colour

Emerald green coloured solution.

### Quantity of medium

100ml of medium in bottle

### pH

7.00- 7.40

### Sterility test

Passes release criteria

### Cultural Response

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP & IP. Cultural response was observed after an incubation at 30-35°C for specified time.

Organism	Growth	Inoculum (CFU)	Acid
<b>Growth for &lt;=24 hours</b>			
<i>Escherichia coli</i> ATCC 8739 (00012*)	luxuriant	50 -100	positive reaction, yellow colour
<i>Pseudomonas aeruginosa</i> ATCC 9027 (00026*)	luxuriant	50 -100	positive reaction, yellow colour
<b>Inhibition after 48 hours</b>			
<i>Staphylococcus aureus</i> subsp.aureus ATCC 6538 (00032*)	inhibited	>=10 <sup>3</sup>	
<b>Additional Microbiological testing</b>			
Cultural characteristics observed after incubation at 30-35°C for 24-48 hours.			
<i>Salmonella Enteritidis</i> ATCC 13076 (00030*)	luxuriant	50 -100	variable reaction
# <i>Klebsiella aerogenes</i> ATCC 13048 (00175*)	luxuriant	50 -100	positive reaction, yellow colour
<i>Proteus mirabilis</i> ATCC 25933	luxuriant	50 -100	positive reaction, yellow colour
<i>Shigella boydii</i> ATCC 12030	luxuriant	50 -100	negative reaction, no colour change
<i>Escherichia coli</i> ATCC 25922 (00013*)	luxuriant	50 -100	positive reaction, yellow colour
<i>Escherichia coli</i> NCTC 9002	luxuriant	50 -100	positive reaction, yellow colour
<i>Pseudomonas aeruginosa</i> ATCC 27853 (00025*)	luxuriant	50 -100	positive reaction, yellow colour
<i>Staphylococcus aureus</i> subsp. aureus ATCC 25923 (00034*)	inhibited	>=10 <sup>4</sup>	

Key : (#) Formerly known as *Enterobacter aerogenes*, (\*) Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 2-8°C. 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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

## Reference

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11. The Indian Pharmacopoeia 2018, Govt. of India. The Controller of Publication, Delhi.
12. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention. Rockville, MD.
13. Van Schothorst M. et al, 1966, Vet Med., 13(3):273.

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