Rappaport Vassiliadis Salmonella Enrichment Broth

MU1491

Rappaport Vassiliadis Salmonella Enrichment Broth is recommended for selective enrichment of *Salmonella* species from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP.

Composition**

Ingredients	Gms / Litre
Soya peptone	4.500
Sodium chloride	8.000
Dipotassium phosphate	0.400
Potassium dihydrogen phosphate	0.600
Magnesium chloride, hexahydrate	29.000
Malachite green	0.036
pH after sterilization (at 25°C)	5.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 27.11 grams of dehydrated medium (the equivalent weight of dehydrated medium per litre) in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Dispense as desired into tubes and sterilize by autoclaving at #115°C for 15 minutes or as per validated cycle.

corresponds to 10 lbs pressure

Principle And Interpretation

Rappaport Vassiliadis Salmonella Enrichment Medium is designed according to the revised formulation by Van Schothorst et al (1) and is recommended for the selective enrichment of Salmonellae from pharmaceutical products. This medium can also be used in direct enrichment of samples containing low inoculum. Present medium is a modification of the Rappaport Vassiliadis Enrichment Broth described by Van Schothorst and Renauld (2). It is prepared as per the formulation in United States Pharmacopoeia (3) and is in accordance with the harmonized methodology of USP/EP/BP/JP (3,4,5,6) has been found to be superior to other *Salmonella* selective medias. Addition of magnesium chloride to the medium was reported by Peterz et al (7). *Salmonella* species can be isolated from human faeces without pre-enrichment by using this medium.

Salmonella generally survive at little high osmotic pressure, grow at slightly low pH and are resistant to malachite green compared to other bacteria. These characteristics are exploited in this medium for selective enrichment of *Salmonella* . Magnesium chloride present in the medium raises the osmotic pressure. Natural sugars of soya peptone provide essential growth nutrients and enhance the growth of *Salmonella* (8). Phosphate buffers the medium to maintain constant pH. Sodium chloride maintains the osmotic balance. Malachite green inhibits many gram-positive bacteria, while selectively enriches *Salmonella* . The relatively lower concentration of nutrition, also aids selective enrichment of *Salmonella* .This medium was reported to be superior to *Salmonella* selective medium like Tetrathionate Broth and Selenite enrichment broth and to Tetrathionate-Brilliant Green Broth for the detection of Salmonellae in milk samples. The enriched culture of Rappaport Vasiliadis Salmonella Enrichment Broth (MU1491) can be further subcultured and isolated on Brilliant Green Agar (MU016) or Xylose Lysine Deoxycholate Agar (MU031).

Quality Control

Appearance

Light yellow to light blue homogeneous free flowing powder

Colour and Clarity of prepared medium

Greenish blue coloured clear to slightly opalescent solution with a slight precipitate in tubes.

pH
5.00-5.40

Cultural Response
Growth Promotion is carried out in accordance with USP. Cultural response was observed after an incubation at 30-35°C for specified time. Recovery is carried out using Xylose Lysine Deoxycholate Agar (MU031) after enrichment in Rappaport Vassiliadis Salmonella Enrichment Broth.

Growth promoting properties
Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating <=100 cfu (at 30-35°C for <=18 hours).

Inhibitory properties
No growth of the test microorganism occurs for the specified temperature for not less than longest period of time specified inoculating >=100 cfu (at least 100 cfu) (at 30-35°C for >= 24 hours).

Cultural Response

Organism	Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Colour of colony	Incubation temperature
Growth promoting						
Salmonella Typhimurium ATCC 14028	50 -100	luxuriant	>=35	>=70 %	red with black centers	<=18 hrs
Salmonella Abony NCTC 6017	50 -100	luxuriant	>=35	>=70 %	red with black centers	<=18 hrs
Inhibitory						
Staphylococcus aureus ATCC 6538	>=10^3	inhibited	0	0 %		>=24 hrs
Additional Microbiological testing						
Escherichia coli ATCC 25922	50 -100	none-poor	0 -10	0 -10 %	yellow	18 -24 hrs
Escherichia coli ATCC 8739	50 -100	none-poor	0 -10	0 -10 %	yellow	18 -24 hrs
Salmonella Enteritidis ATCC 13076	50 -100	luxuriant	>=35	>=70 %	red with black centre	18 -24 hrs
Salmonella Paratyphi B ATCC 8759	50 -100	luxuriant	>=35	>=70 %	red with black centre	18 -24 hrs
Staphylococcus aureus ATCC 25923	>=10^3	inhibited	0	0 %		>=24 hrs
Pseudomonas aeruginosa ATCC 9027	>=10^3	inhibited	0	0%		>=24 hrs
Pseudomonas aeruginosa ATCC 27853	>=10^3	inhibited	0	0 %		>=24 hrs
Enterococcus faecalis ATCC 29212	>=10^3	inhibited	0	0 %		>=24 hrs
E.coli +S.Typhimurium (mixed culture)						
Salmonella Typhimurium ATCC 14028	50 -100	luxuriant	>=35	>=70 %	red with black centre	18 -72 hrs

Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference
1. Van Schothorst M., Renauld A. and VanBeek C., 1987, Food Microbiol., 4:11.
2. Van Schothorst M. and Renauld A., 1983, J. Appl. Bact., 54:209.
3. The United States Pharmacopoeia, 2011, The United States Pharmacopoeial Convention. Rockville, MD.
4. British Pharmacopoeia, 2011, The Stationery office British Pharmacopoeia
5. European Pharmacopoeia, 2011, European Dept. for the quality of Medicines.

6. Japanese Pharmacopoeia, 2008.
7. Peterz M., Wiberg C. and Norberg P., 1989, J. Appl. Bact., 66:523
8. McGibbon L., Quail E. and Fricker C.R. 1984, Inter. J. Food Microbiol . 1:171

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