

# **RAPPAPORT SOY BROTH (VASSILIADIS) ISO 6579, ISO 19250**

# CAT Nº: 1174

For the selective enrichment of Salmonella

### FORMULA IN g/L

*Magnesium Chloride (anhydrous)	13.40	Monopotassium Phosphate	1.26			
Sodium Chloride	7.20	Dipotassium Phosphate	0.18			
Soy Peptone	4.50	Malachite Green	0.036			
* Equivalent to 28.6 g/L Magnesium Chloride Hexahydrate						
Final pH 5.2 $\pm$ 0.2 at 25°C						

#### PREPARATION

Suspend 26.6 grams of the medium in 1 L of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 115°C for 15 minutes. DO NOT OVERHEAT. The prepared medium should be stored at 2-8°C. The color is blue.

The dehydrated medium should be homogeneous, free-flowing and beige in color. If there are any physical changes, discard the medium.

#### USES

RAPPAPORT SOY BROTH (VASSILIADIS) is recommended by ISO 6579, after the preenrichment step, for the selective isolation of *Salmonella spp*.

Rappaport medium was modified by Vassiliadis by reducing Malachite green concentration and increasing incubation temperature, thus offering a better stability of the pH of the prepared medium and optimizing the concentration of Magnesium chloride, resulting in an improved recovery of Salmonellae.

Soy peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Potassium phosphates balance the low pH of the medium, combined with the presence of Magnesium chloride to raise the osmotic pressure, and Malachite green to inhibit other organisms. Sodium chloride supplies essential electrolytes for transport and osmotic balance.

Procedure for the sampling of foods:

- Transfer 0.1 ml of Preenrichment Broth (25 g sample in 225 ml of Buffered Peptone Water (Cat. 1402) incubated at  $37\pm1^{\circ}$ C for  $18\pm2$  hours) to 10 ml of Rappaport Soy Broth Vassiliadis.

- Incubate for 24  $\pm$  3 hours at 41.5  $\pm$  1°C.

- Subculture to selective agar media, for example: XLD Agar (Cat. 1274) or Salmonella Chromogenic Agar (cat. 1122) and any other medium of your choice, and incubate at  $35 \pm 2^{\circ}$ C for 18 - 24 hours.

- Confirm in suitable plates and verify the biochemical and serological characteristics of the suspect colonies.

Procedure for the sampling of water:

- Transfer 0.1 ml of Preenrichment Broth (Buffered Peptone Water (Cat. 1402) incubated at  $36\pm2^{\circ}$ C for  $18\pm2$  hours) to 10 ml of Rappaport Soy Broth Vassiliadis.

- Incubate for  $24 \pm 3$  hours at  $41.5 \pm 1^{\circ}$ C.

- Subculture to selective agar media, for example: XLD Agar (Cat. 1274) or Salmonella Chromogenic Agar (cat. 1122) and any other medium of your choice, and incubate at  $35 \pm 2^{\circ}$ C for 18 - 24 hours.

- Confirm in suitable plates and verify the biochemical and serological characteristics of the suspect colonies.



# **MICROBIOLOGICAL TEST**

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of  $41.5 \pm 1^{\circ}$ C and observed after  $24 \pm 3$  hours.

Microorganisms	Medium Concentration	Growth
Escherichia coli ATCC 25922	99%	< 5%
Salmonella typhimurium ATCC 14028	1%	> 95%

#### According ISO 11133: 24±3 h/41, 5±1°C

Microorganisms	Inoculum (cfu)	Productivity Qualitative	Selectivity Qualitative	Characteristic Reaction
Salmonella typhimurium ATCC 14028 + Escherichia coli ATCC 8739 + Pseudomonas aeruginosa ATCC 27853	$\leq 100$ $\geq 10^{3}$ $\geq 10^{3}$	> 10 colonies on XLD or other medium of choice		Colonies with black centre and a lightly transparent zone of reddish colour due to the colour change of the medium
Salmonella enteritidis ATCC 13076 + Escherichia coli ATCC 8739 + Pseudomonas aeruginosa ATCC 27853	≤100 ≥ 10 <sup>3</sup> ≥ 10 <sup>3</sup>	> 10 colonies on XLD or other medium of choice		Colonies with black centre and a lightly transparent zone of reddish colour due to the colour change of the medium
Escherichia coli ATCC 8739	10 <sup>4</sup> / 10 <sup>6</sup>		Partial inhibition ≤ 100 colonies on TSA	
Enterococcus faecalis ATCC 29212	10 <sup>4</sup> / 10 <sup>6</sup>		< 10 colonies on TSA	

# **BIBLIOGRAPHY**

Rappaport F., Konforti N. and Navon B. (1 956) J. Clin Pathol., 9,261.

Peterz M. Wiberg C. and Norberg P. (1989) J. Appl. Bact. 66: 523-528.

UNE-EN-ISO 6579. Food Microbiology for human consumption and Animal Feed. Horizontal Method for the detection of Salmonella spp. ISO19250 Water quality-Detection of *Salmonella spp* 



# STORAGE

Once opened keep powdered medium closed to avoid hydration.

