

# Modified Semisolid Rappaport Vassiliadis Medium (MSRV) ISO

Cat. 1376

For the detection of motile *Salmonella* species from food and environment samples.

## Practical information

Applications	Categories
Selective enrichment	Salmonella

Industry: Food

Regulations: ISO 11133 / ISO 6579

## Principles and uses

Modified Semisolid Rappaport Vassiliadis Medium (MSRV) is a selective medium used for the rapid detection of motile *Salmonella* spp.

Is a modification of Rappaport Vassiliadis enrichment broth for detecting motile *Salmonella* spp in feces, food products and environmental samples. In this medium the main detection is based on the motility and ability of *Salmonella* to migrate through selective medium ahead of competing motile microorganism, therefore producing opaque halos of growth.

The mobility of other microorganisms is inhibited by selective mediums (such as magnesium chloride, malachite green oxalate and novobiocin) as well as by the temperature of incubation at 42 °C.

Tryptose and acid casein peptone provide nitrogen, vitamins, minerals and amino acids essential for growth. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Magnesium chloride and malachite green oxalate are inhibitory to organisms other than *Salmonella* spp. Novobiocin is a selective agent that inhibits gram positive bacteria and avoids the development of *Proteus*.

This medium is not suitable for the detection of non motile strains of *Salmonella* whose of which their presence is very low (= 1 %).

It is recommended to conduct serological and biochemical tests for *Salmonella* species confirmation.

## Formula in g/L

Bacteriological agar	2,7	Magnesium chloride anhydrous	10,9
Novobiocin	0,01	Potassium dihydrogen phosphate	1,5
Sodium chloride	7,3	Enzymatic Digest of Plants & Animal Tissue	4,6
Malachite green oxalate	0,04	Acid hydrolysate of casein	4,6

## Preparation

Suspend 31,6 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. AVOID OVERHEATING. DO NOT AUTOCLAVE. Dispense into Petri plates.

## Instructions for use

\* For detection of *Salmonella* spp. in food, animal feed, animal faeces, and environmental samples according to ISO 6579:

- Preenrichment in non-selective liquid medium:

Inoculate the Buffered Peptone Water (Cat. 1402) with the sample or dilutions, and incubate at 34-38 °C for 18±2 h.

- Enrichment in/on selective media:

Inoculate, with the culture obtained in the pre-enrichment stage, the Rappaport Soy Broth (Vassiliadis)(Cat. 1174) or the Modified Semisolid Rappaport Vassiliadis medium (MSRV) (Cat. 1376), and the MKKTN Broth(Cat. 1173).

The Rappaport Soy Broth and the Modified Semisolid Rappaport medium are incubated at 41,5 °C for 24±3 h, and the MKKTN Broth at 34-38 °C for 24±3 h.

- Plating out on selective solid media:

From the selective enriched cultures, inoculate two selective isolation agar; XLD agar (Cat. 1274) and any other selective medium complementary to XLD agar (*Salmonella* Chromogenic Agar (Cat. 1122), Brilliant Green Agar (Cat. 1143), Bismuth Sulfite Agar (Cat. 1011), DCLS Agar(Cat. 1045), Desoxycholate Citrate Agar (Cat. 1067), Hektoen Enteric Agar (Cat. 1030), *Salmonella* Shigella Agar(Cat. 1064) and XLT4 Agar (Cat. 1159)).

Incubate the XLD plates inverted at 34-38 °C for 24±3 h.

Incubate the second selective medium in accordance with the manufacturer's instructions.

- Confirmation:

Subculture colonies of presumptive *Salmonella* and confirm their identity by biochemicals and serological tests.

Note: According to Annex D of ISO 6579-1: 2017, for the detection of enterica subspecies enterica serovars Typhi and Paratyphi, Selenite Cystine Broth (Cat. 1220) should be added as a selective enrichment medium and Bismuth Sulfite Agar (Wilson Blair) should be selected as a second selective medium (Cat. 1011).

\* For detection of *Salmonella* spp. in water samples according to ISO 19250:

- Preenrichment in non-selective medium:

Inoculate the Buffered Peptone Water (Cat. 1402) with the sample or dilutions, and incubate at 34-38 °C for 18±2 h.

- Enrichment in selective media:

Inoculate, with the culture obtained in the pre-enrichment stage, the Rappaport Soy Broth (Vassiliadis)(Cat. 1174) and the MKKTN Broth (Cat. 1173).

The Rappaport Soy Broth is incubated at 41,5±1 °C and the MKKTN Broth at 34-38 °C, both of them for 24±3 hours.

- Plating out on selective solid media:

From the selective enriched cultures, inoculate two selective isolation agar; XLD agar (Cat. 1274) and any other selective medium complementary to XLD agar ( For instance, Brilliant Green Agar (Cat. 1143) or Bismuth Sulfite Agar (Cat. 1011))

Incubate the XLD plates inverted at 34-38 °C for 24±3 hours.

Incubate the second selective medium in accordance with the manufacturer's instructions.

- Confirmation:

Subculture colonies of presumptive *Salmonella* and confirm their identity by biochemicals and serological tests.

## Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
Sin restos	Fine powder	Beige	Blue	5,1-5,4

## Microbiological test

According to ISO 11133:

Incubation conditions: (41,5±1 °C / 24±3 h).

Inoculation conditions: Productivity qualitative (10<sup>3</sup>-10<sup>4</sup> CFU) / Selectivity (10<sup>4</sup>-10<sup>6</sup> CFU) / Specificity (10<sup>3</sup>-10<sup>4</sup> CFU).

Microorganisms	Specification	Characteristic reaction
<i>Salmonella enteritidis</i> ATCC 13076	Grey-white turbid zone extending out from inoculated. After 24-48 h the turbid zone will be fully migrated over the plate.	Characteristic colonies after subculture in XLD
<i>Salmonella typhimurium</i> ATCC 14028	Grey-white turbid zone extending out from inoculated. After 24-48 h the turbid zone will be fully migrated over the plate.	Characteristic colonies after subculture in XLD
<i>Escherichia coli</i> ATCC 25922	Possible growth without a turbid zone	
<i>Enterococcus faecalis</i> ATCC 29212	No growth	

## Storage

Temp. Min.:2 °C

Temp. Max.:25 °C

## Bibliography

De SMEDT et al.; Rapid *Salmonella* Detection in Foods by Motility Enrichment on a Modified Semi-Solid Rappaport-Vassiliadis Medium. J. Food Protect. Vol. 49, 7; 510-514 (1986).

De SMEDT, a. BOLDERDIJK, R.F.; Dynamics of *Salmonella* Isolation with Modified Semi-Solid Rappaport-Vassiliadis Medium. J. Food Protect. Vol. 50, 8; 658-661 (1987).

ISO 6579 Microbiology of food and animal feeding stuffs – Horizontal method for the detection of *Salmonella* spp. Amendment 1: Broader range of incubation temperatures, amendment to the status of Annex D, and correction of the composition of MSR and SC.