Rappaport-Vassiliadis Soya (RVS) Broth | AS-1338 |

Selective Enrichment for Salmonella spp. from Food, Environmental, and Clinical Specimens

Catalogue Number: AS-1338 | acc. harm. EP / USP / JP

Target Organism	Pharmacopoeial	Final pH	HS Code
Salmonella spp.	EP / USP / JP harmonised	5.2 ± 0.2 at 25 °C	3821.00.00

Overview

Rappaport Vassiliadis Broth (RVS) — also designated Rappaport-Vassiliadis Soya Peptone Broth — is a selective liquid enrichment medium formulated for the isolation of *Salmonella* spp. from food, animal feed, environmental samples, and clinical specimens. The medium was originally described by Vassiliadis et al. (1981) as a modification of the earlier Rappaport formula (1954), substituting soy peptone for casein digest to improve recovery of stressed and low-inoculum Salmonella.

The medium is harmonised across major international pharmacopoeias — EP 2.6.13, USP <62>, and JP Chapter 4.05 — for Microbial Examination of Non-Sterile Products: Tests for Specified Microorganisms (Salmonella). It is also the standard secondary enrichment medium prescribed by ISO 6579-1:2017 (food and animal feed), ISO 19250 (water), and APHA Standard Methods for detection of Salmonella.

Mode of Action

RVS Broth achieves Salmonella selectivity through a combination of four mechanisms: (1) **Malachite green** — a triphenylmethane dye that inhibits Gram-positive organisms and most Gram-negative non-Salmonella bacteria at the 0.036 g/L concentration used; (2) **Magnesium chloride** — raises osmotic pressure to ~1400 mOsm/kg, exploiting Salmonella's superior tolerance to elevated Mg^{2+} and osmotic stress compared to competing flora; (3) **Acidic pH (5.2)** — further inhibits most competing Gram-negative bacteria while Salmonella remains viable; (4) **Soy peptone** — provides a balanced amino acid and growth factor profile that preferentially supports Salmonella enrichment.

Typical Composition (per litre)

Ingredient	CAS Number	Function	Amount (g/L)
Soy Peptone (Tryptone Soya Peptone)	73049-73-7	Sole nitrogen and carbon source — balanced amino acids for Salmonella	5.0

Sodium Chloride (NaCl)	7647-14-5	Base osmotic balance	8.0
Potassium Dihydrogen Phosphate (KH_2PO_4)	7778-77-0	pH buffer (acidic component)	1.6
Dipotassium Hydrogen Phosphate (K_2HPO_4)	7758-11-4	pH buffer (alkaline component)	0.8
Magnesium Chloride Hexahydrate ($MgCl_2 \cdot 6H_2O$)	7791-18-6	Elevated osmotic pressure — selective for Salmonella	13.4
Malachite Green Oxalate	2437-29-8	Selective inhibitor — Gram-positive and non-Salmonella Gram-negatives	0.036
Purified Water	7732-18-5	Solvent	q.s. 1 L

Total (without water): 28.836 g/L | Final pH: 5.2 ± 0.2 at 25 °C

Key Features

- Pharmacopoeial harmonised — acc. EP 2.6.13, USP <62>, JP Chapter 4.05
- ISO 6579-1:2017 compliant — international standard for Salmonella in food and animal feed
- Dual selectivity mechanism — malachite green + elevated osmotic pressure ($MgCl_2$) + acidic pH
- Soy peptone base — superior Salmonella recovery compared to casein peptone variants
- Supports recovery of stressed, freeze-damaged, and heat-injured Salmonella cells
- Used as secondary selective enrichment following non-selective pre-enrichment (BPW)
- Applicable to food, animal feed, water, environmental swabs, and pharmaceutical non-sterile products

Applications

Food & Animal Feed Safety

- Secondary selective enrichment of Salmonella from pre-enriched BPW cultures — ISO 6579-1:2017
- Broad food matrices: raw meat, poultry, eggs, dairy, dried foods, spices, herbs, pet food
- Animal feed and raw material Salmonella screening per ISO 6579-1
- Environmental swab enrichment from food production facilities

Pharmaceutical Quality Control

- Salmonella testing of non-sterile pharmaceutical products — EP 2.6.13, USP <62>, JP Chapter 4.05
- Testing of herbal medicines, excipients, and raw botanical materials
- Finished product release testing for Salmonella absence per pharmacopoeial harmonised method

Water & Environmental Testing

- Salmonella enrichment from drinking water, wastewater, surface water — ISO 19250
- Environmental monitoring swabs from processing and manufacturing facilities
- Sewage sludge and biosolids Salmonella screening

Workflow — Salmonella Detection (ISO 6579-1)

Step	Medium	Incubation	Purpose
1. Non-selective pre-enrichment	Buffered Peptone Water (BPW)	37 °C, 18 ± 2 h	Resuscitate stressed/injured Salmonella cells
2. Selective enrichment	RVS Broth (AS-1338) ← this product	41.5 °C, 24 ± 3 h	Select and amplify Salmonella; suppress competing flora
3. Selective plating	XLD Agar + Chromogenic Salmonella Agar	37 °C, 24–48 h	Presumptive colony identification
4. Confirmation	Biochemical + serological	—	Confirmed positive result

Note: Transfer 0.1 mL of BPW pre-enrichment to 10 mL RVS Broth (1:100 inoculation ratio as specified in ISO 6579-1:2017).

Preparation Instructions

1. Dissolve 26.75 g (or as stated on label) of dehydrated RVS Broth in 1 litre of purified water.
2. Mix with gentle agitation at room temperature — do NOT heat. Malachite green and $MgCl_2$ are heat-sensitive.
3. Dispense into appropriate volumes: 10 mL per tube (for 0.1 mL inoculum) or 100 mL per flask.
4. Sterilise by autoclaving at 115 °C for 15 minutes. Do NOT autoclave at 121 °C — excessive heat degrades malachite green and alters osmolarity.
5. Cool to room temperature. Medium should appear bluish-green and clear.
6. Store at 2–8 °C, protected from light. Use within 3 months.

Critical: Autoclave at 115 °C (not 121 °C). Higher temperature irreversibly degrades malachite green, reducing selectivity and potentially causing false-negative results.

Storage & Stability

Dehydrated powder	15–30 °C, tightly sealed, dry, protected from light
Prepared medium	2–8 °C, protected from light; use within 3 months
Shelf life (powder)	As per labelled expiry date
Do not use if	Colour changed (should be bluish-green), turbid, or precipitate visible before inoculation

Customs & Trade Information

HS / AHECC Code	3821.00.00
Description	Prepared culture media for the development or maintenance of microorganisms
Country of Origin	Australia

Literature & References

Vassiliadis, P., Kalapothaki, V., Trichopoulos, D., Papadakis, J.A. & Serie, C. (1981). Salmonella isolation with Rappaport's Vassiliadis medium. J. Appl. Bacteriol., 51(2), 233–239.

Rappaport, F., Konforti, N. & Navon, B. (1954). A new enrichment medium for certain Salmonellae. J. Clin. Pathol., 9(3), 261–266.

ISO 6579-1:2017. Microbiology of the food chain — Horizontal method for the detection, enumeration and serotyping of Salmonella — Part 1: Detection. ISO, Geneva.

ISO 19250:2010. Water quality — Detection of Salmonella spp. ISO, Geneva.

European Pharmacopoeia, Chapter 2.6.13 — Microbial examination of non-sterile products: Tests for specified microorganisms.

USP <62> — Microbiological examination of non-sterile products: Tests for specified microorganisms.

JP Chapter 4.05 — Microbial Limit Test II.

APHA Standard Methods for the Examination of Water and Wastewater (2017), Method 9260 B.

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