



Fluid Selenite Cystine- Medium (Twin Pack)

MAP025

(MU025/ MM025)

Intended Use:

Used as enrichment medium for isolation of *Salmonellae* in food, dairy products and materials of sanitary importance in accordance with USP/IP.

Composition**

Ingredients	g / L
Part A	-
Tryptone	5.000
Lactose	4.000
Sodium phosphate	10.000
L-Cystine	0.010
Part B	-
Sodium acid selenite	4.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 4.0 grams of Part B in 1000 ml purified/distilled water. Add 19.01 grams of Part A. Mix well. Warm to dissolve the medium completely. Distribute in sterile test tubes. Sterilize in a boiling water bath or free flowing steam for 15 minutes. **DO NOT AUTOCLAVE**. Excessive heating is detrimental. Discard the prepared medium if large amount of selenite is reduced (indicated by red precipitate at the bottom of tube/bottle).

Principle And Interpretation

Selective inhibitory effects of selenite were first demonstrated by Klett (1). Guth (2) used it to isolate *Salmonella* Typhi. Leifson studied selenite and formulated a medium. Fluid Selenite Cystine Medium is a modification of Leifsons (3) formula with added cystine by North and Bartram (4). The formulation corresponds to that of recommended by the AOAC (5) for the detection of *Salmonellae* in foodstuff particularly egg products. It is included by APHA (6,7), USP (8) and IP (9). Recently ISO Committee also recommends this medium for the detection of *Salmonellae* (10). Selenite Cystine Broth is useful for detecting *Salmonella* in the non-acute stages of illness when organisms occur in low numbers in test samples and for epidemiological studies to enhance the detection of low numbers of organisms from asymptomatic or convalescent patients (11).

Tryptone provide nitrogenous substances. Lactose is the fermentable source of carbohydrate and also maintains the pH in medium as selenite is reduced by bacterial growth and alkali is produced. An increase in pH lessens the toxicity of the selenite and results in overgrowth of other bacteria. The acid produced by bacteria due to lactose fermentation that maintain a neutral pH counters this. Phosphate too maintains a stable pH and is a good buffering agent. L-cystine imparts ambient redox potential, which enhances and improves recovery of *Salmonellae* and few *Shigella* sp. which may be in small numbers in products to be tested. This medium to some extent prevents the growth of coliforms. Enriched broth is sub cultured on solid medium. Do not incubate the broth longer than 24 hours as inhibitory effect of selenite reduces after 6 - 12 hours of incubation (12).

Type of specimen

Food and dairy samples; Pharmaceutical sample

Specimen Collection and Handling:

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5,6,7). For pharmaceutical products, follow appropriate techniques for sample processing in case of viscous materials as mentioned under sterility (8,9).

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. Do not incubate the broth longer than 24 hours as inhibitory effect of selenite reduces after 6 - 12 hours of incubation (12).

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

Part A : Cream to light yellow homogeneous free flowing powder

Part B : Offwhite - white homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow clear to slightly opalescent solution

pH

7.0±0.2

Growth Promotion Test

As per USP/ IP

Cultural Response

As per IP : Cultural characteristics observed after enrichment in MAP025 for 36-38°C for 48 hours, and then subcultured on Xylose Lysine Deoxycholate Agar (Agar Medium K) and Brilliant Green, Phenol red, lactose monohydrate Sucrose Agar (Agar Medium L) and incubated at 36-38°C for 18-24 hours.

As per USP : Cultural characteristics observed after enrichment in MAP025 for 30-35°C for 18-24 hours, and then subcultured on Xylose Lysine Deoxycholate Agar (Agar Medium K) and Brilliant Green, Phenol red, lactose monohydrate Sucrose Agar (Agar Medium L) and incubated at 30-35°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Colour of Colony	Incubation period
Growth on Brilliant Green, Phenol red, lactose monohydrate Sucrose Agar (Agar Medium L)						
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50 -100	luxuriant	25 -100	≥50 %	pinkish white	24 -48 hrs
<i>Salmonella</i> Abony NCTC 6017 (00029*)	50 -100	luxuriant	25 -100	≥50 %	pinkish white	24 -48 hrs
Growth on Agar Xylose Lysine Deoxycholate Agar (Agar Medium K)						
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50 -100	luxuriant	25 -100	≥50 %	red with black centres	18 -24 hrs
<i>Salmonella</i> Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	25 -100	≥50 %	red with black centres	18 -24 hrs
<i>Escherichia coli</i> ATCC 8739 (00012*)	50 -100	fair	10 -30	20 -30 %	yellow	18 -24 hrs
<i>Escherichia coli</i> ATCC 25922 (00013*)	50 -100	fair	10 -30	20 -30 %	yellowish green	18 -24 hrs

Key: (*) Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (13,14).

Reference

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7. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products, 17th ed., APHA Inc., Washington, D.C.
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9. Indian Pharmacopoeia, 2022, Indian Pharmacopoeia Commission, Ministry of Health and Family Welfare Government of India.
10. International Organization for Standardization (ISO), 1993, Draft ISO/DIS 6579.
11. Murray PR, Baren EJ, Jorgensen JH, Pfaller MA, Tenover FC, Tenover RH (editors) 2003, Manual of clinical Microbiology, 8th ed., ASM, Washington, D.C.
12. Chattopadhyay W. and Pilford J. N., 1976, Med. Lab. Sci., 33:191.
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