



QUALITY CONTROL PROCEDURES (Optional)

I INTRODUCTION

Selenite-F Broth is an enrichment medium for the isolation of *Salmonella* and some species of *Shigella*.

II PERFORMANCE TEST PROCEDURE

1. Inoculate representative samples with the cultures listed below.
 - a. Using sterile disposable 1.0 mL pipettes, inoculate tubes with 1.0 mL of 18- to 24-h **Trypticase™** Soy Broth cultures of *Salmonella* Typhimurium and *Shigella sonnei* diluted to contain 10^2 – 10^3 CFU/mL.
 - b. To each of the tubes inoculated as above, add 1.0 mL of an 18- to 24-h TSB culture of *Escherichia coli* diluted to contain 10^2 – 10^3 CFU/mL. An uninoculated tube of the Selenite-F Broth is included in the subculturing and incubation as a growth control.
 - c. Vortex all tubes well.
2. Incubate all tubes with loosened caps at 35 ± 2 °C in an aerobic atmosphere for 18–24 h. After incubation, vortex tubes and streak MacConkey II Agar plates with one loopful from each of the tubes.
3. Incubate the MacConkey II Agar plates at 35 ± 2 °C for 18–24 h in an aerobic atmosphere. Examine the MacConkey II Agar plates for amount of growth of lactose-positive (pink colonies) and lactose-negative (colorless colonies) organisms.
4. Expected Results

CLSI Organisms	ATCC®	Recovery on MacConkey II Agar
* <i>Salmonella enterica</i> subsp. <i>enterica</i> serotype Typhimurium	14028	Fair to heavy growth of colorless colonies
* <i>Shigella sonnei</i>	9290	Fair to heavy growth of colorless colonies
* <i>Escherichia coli</i>	25922	Partial to complete inhibition (pink colonies)

NOTE: Growth control shows no growth on MacConkey II Agar.

*Recommended organism strain for User Quality Control.

III ADDITIONAL QUALITY CONTROL

1. Examine tubes as described under "Product Deterioration."
2. Visually examine representative tubes to assure that any existing physical defects will not interfere with use.
3. Determine the pH potentiometrically at room temperature for adherence to the specification of 7.0 ± 0.2 .
4. Incubate uninoculated representative tubes at 20–25 °C and 30–35 °C and examine after 7 days for microbial contamination.

PRODUCT INFORMATION

IV INTENDED USE

Selenite-F Broth is used as an enrichment medium for the isolation of *Salmonella* from feces, urine, water, foods and other materials of sanitary importance.

V SUMMARY AND EXPLANATION

Selenite-F Broth was devised by Leifson,¹ who demonstrated that selenite was inhibitory for coliforms and certain other microbial species, such as fecal streptococci, present in fecal specimens, and thus, was beneficial in the recovery of *Salmonella* species. He found that the inhibited strains would eventually breakthrough, but if subcultures were made from the enrichment broth after 8–12 h incubation, the isolation of *Salmonella* was possible without overwhelming growth of many members of the intestinal flora.

Enrichment media are routinely employed for detection of pathogens in fecal specimens since the pathogens usually represent only a small percentage of the intestinal flora. Selenite-F Broth and the related medium, Selenite Cystine Broth, are recommended for use in the recovery of *Salmonella* with subcultures being made after 12–18 h incubation. For detection of *Shigella*, GN Broth is a satisfactory enrichment medium.² *Campylobacter* Thioglycollate Medium with 5 Antimicrobics is recommended for specimens suspected to contain *Campylobacter jejuni* when low numbers of organisms are expected because of delayed transport to the laboratory or because the acute stage of disease has passed.³

VI PRINCIPLES OF THE PROCEDURE

The casein peptone provides essential nitrogenous and carbon compounds. The lactose in the medium serves to maintain a uniform pH. When selenite is reduced by the growth of bacteria, alkali is produced and such increase in pH would lessen the toxicity of the selenite and result in overgrowth of extraneous bacteria. The acid produced by lactose fermentation serves to maintain a neutral or slightly decreased pH. The function of the phosphate is two-fold; it serves to maintain a stable pH and also lessens the toxicity of the selenite, thus increasing the capacity of the medium. Sodium selenite inhibits many species of gram-positive and gram-negative bacteria including enterococci and coliforms.

VII REAGENTS

Selenite-F Broth

Approximate Formula* Per Liter Purified Water

Pancreatic Digest of Casein	5.0 g
Lactose	4.0 g
Sodium Selenite	4.0 g
Sodium Phosphate	10.0 g

*Adjusted and/or supplemented as required to meet performance criteria.

Warnings and Precautions: For *in vitro* Diagnostic Use.

Tubes with tight caps should be opened carefully to avoid injury due to breakage of glass.

Pathogenic microorganisms, including hepatitis viruses and Human Immunodeficiency Virus, may be present in clinical specimens. "Standard Precautions"⁴⁻⁷ and institutional guidelines should be followed in handling all items contaminated with blood and other body fluids. After use, prepared tubes, specimen containers and other contaminated materials must be sterilized by autoclaving before discarding.

Storage Instructions: On receipt, store tubes in the dark at 2–8 °C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Tubed media stored as labeled until just prior to use may be inoculated up to the expiration date and incubated for the recommended incubation times. Allow the medium to warm to room temperature before inoculation.

Product Deterioration: Do not use tubes if they show evidence of microbial contamination, discoloration, drying or other signs of deterioration.

VIII SPECIMEN COLLECTION AND HANDLING

Specimens suitable for culture may be handled using various techniques. For detailed information, consult appropriate texts.^{8,9}

Specimens should be obtained before antimicrobial agents have been administered. Provisions must be made for prompt delivery to the laboratory.

IX PROCEDURE

Material Provided: Selenite-F Broth

Materials Required But Not Provided: Ancillary culture media, reagents, quality control organisms and laboratory equipment as required.

Test Procedure: Observe aseptic techniques.

For feces and other solid materials, suspend 1–2 g of the specimen in the broth (approximately 10–15% by volume) and emulsify with an inoculating needle, if necessary.

Incubate tubes with loosened caps at 35 ± 2 °C for up to 24 h. Subcultures should be made after 12–18 h incubation, if possible. Coliforms will tend to overgrow the pathogens if incubated longer than 24 h.

User Quality Control: See "Quality Control Procedures."

Each lot of media has been tested using appropriate quality control organisms and this testing meets product specifications and CLSI standards, where relevant. As always, QC testing should be performed in accordance with applicable local, state, federal or country regulations, accreditation requirements, and/or your laboratory's standard quality control procedures.

A single electrode of sufficiently small size to fit into the tubes should be used to determine the pH potentiometrically of tubed media. The tip of the electrode should be placed below the surface of broth media.

X RESULTS

After incubation, there should be an increase in the number of pathogens that the medium is designed to select for and enrich.

Subculture onto appropriate selective and differential media (e.g., MacConkey Agar, Hektoen Enteric Agar, XLD Agar, XLT4 Agar and **CHROMagar™** Salmonella) to isolate pathogens for identification.

XI LIMITATIONS OF THE PROCEDURE

Enrichment broths should not be used as the sole isolation medium. They are to be used in conjunction with selective and nonselective plating media to increase the probability of isolating pathogens, especially when they may be present in small numbers. For identification, organisms must be in pure culture. Morphological, biochemical, and/or serological tests should be performed for final identification. Consult appropriate texts for detailed information and recommended procedures.⁸⁻¹⁰

XII PERFORMANCE CHARACTERISTICS

In a study by Kelly et al.,¹¹ 8,717 stool specimens were submitted to the laboratory for culture. The specimens were inoculated directly onto XLD Agar and into Selenite enrichment broth. After 12–18 h the Selenite broth was subcultured to XLD Agar. *Salmonella enterica* was identified in 312 (3.6%) of the stool specimens; 197 (63%) were from previously diagnosed cases and 115 (37%) were from newly identified cases. Of the 115 new *S. enterica* isolates, 68 were recovered from both the primary XLD plate and the Selenite broth. However, 47 (41%) only grew after Selenite enrichment.

XIII AVAILABILITY


Cat. No.	Description
221020	BD BBL™ Selenite-F Broth, 8 mL, Pkg. of 10 size K tubes
221021	BD BBL™ Selenite-F Broth, 8 mL, Ctn. of 100 size K tubes

XIV REFERENCES

1. Leifson, E. 1936. New selenite selective enrichment media for the isolation of typhoid and paratyphoid *Salmonella* bacilli. Am. J. Hyg. 24:423-432.
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11. Kelly, S., M. Cormican, L. Parke, G. Corbett-Feeney, and J. Flynn. 1999. Cost-effective methods for isolation of *Salmonella enterica* in the clinical laboratory. J. Clin. Microbiol. 37: 3369.

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