



# Copan

- Incubate the plates at 35°C ± 2°C for 18-24 hours.
- Read the plate and record the results to release the lot.

STRAINS	GROWTH AT ZERO TIME (CFU)	GROWTH AT 18-24 H (CFU)
<i>S. typhimurium</i> ATCC® 14028	172	CONFLUENT GROWTH
<i>E. coli</i> ATCC® 25922	CONFLUENT GROWTH	PARTIAL TO TOTAL INHIBITION
<i>S. sonnei</i> ATCC® 9290	202	CONFLUENT GROWTH

The QC procedure described below can be applied by End User, in compliance to CLSI M22-A3<sup>3</sup>.

- Prepare a 0.5 McFarland bacterial suspension in nonbacteriostatic saline, from a fresh culture of microorganism in test (growth or inhibition organisms).
- Inoculate the Selenite Broth with 10 µl of undiluted 0.5 McF.
- Recap the tube.
- Vortex for 10 seconds at 2500 rpm.
- Incubate the Selenite Broth tube at 35°C -37°C for 18-24 hours.
- After incubation, check the turbidity and streak for isolation of Selenite Broth onto appropriate culture medium.
- Incubate the plates at 35°C -37°C for 18-24 hours.

The Selenite Broth should be tested in aerobic condition at 35-37°C with ATCC® strains *S. typhimurium* ATCC® 14028, *S. sonnei* ATCC® 9290 and *E. coli* ATCC® 25922 as reported in CLSI M22-A3<sup>3</sup>. Selective media perform satisfactorily if the quality control organisms exhibits adequate growth, expected colony size, typical morphology, and inhibition of growth of certain organism<sup>3</sup>.

## RESULTS

Results obtained will largely depend on proper and adequate sample collection, as well as timely transport and processing in the laboratory.

## REFERENCES

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2. UK Standards for Microbiology Investigations. of fecal specimens for Enteric Pathogens. Bacteriology [B 30] Issue no: 8.1 | Issue date: 24.04.14.
3. Clinical and Laboratory Standards Institute (CLSI). 2004. Quality Control for commercially Prepared Microbiological Culture Media. Approved Standard – Third Edition M22-A3.
4. M. Miller and S.A. Miller 2017. A Guide to Specimen Management in Clinical Microbiology, 3rd ed. ASM Press, Washington, DC.
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8. E. W. Koneman, S.D. Allen, V.R. Dowell Jr., W.M. Janda, H.M. Sommers, W. C. Winn Jr. The Enterobacteriaceae in: Diagnostic Microbiology. Third Ed. J.B. Lippincott Company 1988.
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11. Isenberg, H. D., 2010. Clinical Microbiology Procedures Handbook, 3rd ed. ASM Press Washington, DC.
12. Isenberg, H.D., 1998. Essential Procedures for Clinical Microbiology. Chapter 14.12, Page 787. Packaging and Shipping Infectious Substances.
13. Clinical and Laboratory Standards Institute (CLSI). 2014. Quality Control Microbiological Transport System; Approved Standard – Second Edition M40-A2.

## INDEX OF SYMBOLS

Symbol	Meaning	Symbol	Meaning
	Manufacturer		Use-by date
	In vitro diagnostic medical device		Consult the operating instructions supplied with the device or available in electronic format, and which can be identified by the e-IFU indicator on the packaging label
<b>Rx Only</b>	For professional use		
	Do not re-use		Batch code (Lot)
	Catalogue number		Contains sufficient for <n> tests
	Temperature limit		Caution

# Copan



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## Instructions for Use

## Copan SELENITE Broth - Product Insert &amp; How to Use Guide

## INTENDED USE

Copan Selenite Broth is a selective enrichment broth for clinical samples used for the culturing and the subsequent isolation of enteric pathogens including *Salmonella* spp. and *Shigella* spp.

## SUMMARY AND PRINCIPLES

Diarrhoea can be defined as an increased frequency of bowel movements with at least three discharges a day accompanied by a decreased stool consistency. It is caused by a wide range of pathogens (bacteria, viruses and parasites) through different pathogenic mechanisms. *Salmonella* spp. and *Shigella* spp. infections come from the multiplication of intestinal bacteria. Most *Salmonella* spp. infections in humans are related to the consumption of animal based food, water contaminated by animals or interpersonal contact. Gastroenteritis is the most common infection that occurs. A small amount of *Salmonella* spp. may be present in the faeces of asymptomatic carriers. *Shigella* spp. infections have a wide variety of symptoms that range from mild diarrhoea to dysentery. Shigellas are transmitted directly from person to person, environmental multiplication is insignificant and asymptomatic infections are rare. The aetiological diagnosis of *Salmonella* spp and *Shigella* spp diarrhea is done by seeding the fecal sample directly onto a moderately selective solid culture medium such as Hektoen enteric agar/XLD (Xylose lysine deoxycholate), a more selective medium such as SS (Salmonella Shigella Agar) or by passing from an enrichment phase in a liquid medium such as Selenite Broth. Using an enrichment broth can increase the recovery of *Salmonella* spp and *Shigella* spp by 10% and is particularly recommended in populations such as service personnel and nutritionists<sup>1,2,5</sup>.

## PRODUCT DESCRIPTION

Selenite Broth is available in the following configuration:

Catalogue Number	Product Description	Pack Size
4C056N.A	SELENITE BROTH IN BULK – US MARKET: 2 ml of Selenite Broth in a 12X80 mm polypropylene screw-cap tube with internal conical shape.	50 devices per inner box 6X50 devices per box

## REAGENTS

Selenite Broth components (per liter)<sup>6</sup>:

Components	Grams
Pancreatic digest of Casein	5
Lactose	4
Sodium Selenite	4
Sodium Phosphate	10
Distilled water	1000

pH 7.0 ± 0.2 at 25°C (lot release time)

## REQUIRED MATERIALS BUT NOT INCLUDED

Appropriate materials for the cultivation and isolation of bacteria. Refer to laboratory reference manuals for recommended protocols for culture and identification techniques.

## STORAGE

This product is ready-to-use and no further preparation is necessary. The packaged and unopened device can be stored at 5 – 25°C until used or until the expiration date<sup>3</sup>. Do not overheat. Do not incubate or freeze prior to use. Improper storage will result in loss of efficacy. Do not use after expiration date, which is clearly printed on the outer and inner label.

## LIMITATIONS

- Performance testing with Copan SELENITE Broth was conducted using laboratory ATCC® strains spiked into the SELENITE Broth tube and not using human specimens.
- Proper specimen (condition, timing, and volume) collection from the patient is extremely critical for successful isolation and identification of infectious organisms. For the swirling procedure and specific guidance regarding specimen collection procedures, consult published reference manuals. Specimens should be collected as soon as possible after the clinical onset of disease<sup>4</sup>.
- Selenite Broth is a selective and enrichment broth for the isolation of samples of *Salmonella* spp. and *Shigella* spp. The product was not designed for the enrichment of other genera and species of bacteria.
- There is no dulcitol (melampirite or polyalcohol) in the Copan Selenite Broth; therefore, the growth of coliforms could be suppressed during the first 8-12 hours of incubation; longer incubation times may result in the excessive growth of mixed flora<sup>5</sup>.

## WARNING AND PRECAUTIONS

1. For In Vitro Diagnostic Use.
2. For professional use only. Use the device in accordance with the Package Insert.
3. Do not use if the tube is open, there is evidence of damage or leakage, deterioration or contamination (i.e. broth is turbid).
4. Do not use if the expiration date has passed.
5. Do not re-pack.
6. Do not re-use the device.
7. Use of this device in association with diagnostic kits and/or instrumentation should be validated prior to use.
8. All specimens must be considered potentially infectious. Wear protective gloves and other protection commensurate with universal precautions when handling clinical specimens. Observe CDC Biosafety recommendations, or other appropriate alternative.
9. Dangerous content: do not swallow or inhale and avoid contact with the human body.
10. Check the version of the operating instructions. The correct version is the one supplied with the device or available in electronic format, and can be identified by the e-IFU indicator on the packaging label.



11. Warning: Contains: sodium selenite in solution  
H317 May cause an allergic skin reaction.

- P261 Avoid breathing dust/fume/gas/mist/vapours/spray.  
P272 Contaminated work clothing should not be allowed out of the workplace.  
P302+P352 IF ON SKIN: Wash with plenty of water  
P333+P313 If skin irritation or rash occurs: Get medical advice/attention.  
P363 Wash contaminated clothing before reuse.  
P501 Dispose of contents/container in accordance with applicable regulations.

## PRODUCT DETERIORATION

Do not use Copan Selenite Broth if: (1). the product has visible signs of damage or contamination; (2). there is evidence of liquid leaking from the test tube; (3) it has passed the expiration date; (4). there are other visible signs of deterioration. (i.e. medium is turbid).

## PROCEDURE

An adequate collection of samples from the patient is an extremely critical factor for the successful isolation and identification of infectious organisms. Consult the published reference manuals for specific instructions on the sample collection procedures<sup>1-13</sup>. The Copan FecalSwab™ system with a regular size flocked swab can be used as the sampling system. Contact Copan or refer to the specific product instructions.

## Direct specimen collection:

## Fecal sample:

1. Unscrew the cap of the Selenite Broth tube.
2. Add the sample as described below:
  - Add a portion of the fecal sample using a swab or a special scoop. **NOTE:** The stool specimens in dry containers must be processed within two hours after collection. **NOTE:** make sure that the swab used to transfer the feces is completely covered with fecal material.
  - Transfer a portion of the feces into the Selenite Broth. Follow the guidelines used in your laboratory for the volumetric ratio.
  - Vigorously shake the swab or scoop in the Selenite Broth for at least 10 seconds, in order to release the collected material.
  - Close the Selenite Broth tube.
3. Homogenize the tube by vortexing it for 10 seconds at 2000-2500 rpm;
4. Incubate the Selenite Broth tubes at 35°C ± 2°C for 18-24 hours;
5. After 18-24 hours look for any turbidity in the broth. If negative, incubate for another 18-24 hours;
6. Inoculate a plate of selective and/or differential growth media with 1 to 10 µl of enriched broth.

The enriched sample in Selenite Broth can be seeded with automated sample management systems such as Wasp®/WaspLab®. For seeding, follow the procedure for seeding broths given in the Wasp® automation manual.

## Using Collection system (i.e. FecalSwab™) for specimen:

1. Take the tubes of SELENITE Broth and unscrew the cap.
2. Vortex the FecalSwab™ specimen tube for 10 seconds.
3. Unscrew the cap and transfer an amount of approximately 30 µl from the FecalSwab™ to the SELENITE tube or transfer directly the Flocked Swab of the FecalSwab™. **NOTE:** the FecalSwab™ cap can be transfer with its swab directly to the SELENITE Broth tube. The FecalSwab™ cap can be used to close the SELENITE tube: Re-cap the FecalSwab™ tube with the SELENITE's cap.
4. Using a loop or a micropipette, plate minimum 1 µl of SELENITE Broth onto appropriate bacteriology selective plate medium.
5. Incubate inoculated SELENITE Broth tubes at 35 ± 2°C.
6. After 6-8 hours or 18-24 hours, inoculate 1 to 10 µl of SELENITE Broth onto appropriate bacteriology culture plate medium.

Incubate SELENITE Broth according to laboratory Standard Operating Procedures and taking under consideration that SELENITE Broth formulation inhibit growth of Gram positive bacteria and some coliforms better up to 8-12 hours incubation, after this time coliforms are no longer suppressed and may overgrow the target pathogens.

## WASTE DISPOSAL

Waste disposal must be done according with national laws. Use the precautions for potential infectious material, when necessary.

## QUALITY CONTR EDURE

The enrichment procedure described below is applied by manufacturer to release the lots:

- Prepare a 0.5 McFarland bacterial suspension in PBS from a fresh culture of microorganism in test.
- Prepare a 10<sup>-5</sup> serial dilution from the 0.5 McFarland bacterial suspension.
- Inoculate the SELENITE broth with 200 µl of the dilution prepared.
- Recap all tubes.
- Vortex for 10 seconds at 2500 rpm.
- Plate immediately 100 µl of Selenite Broth inoculated (time zero count).
- Incubate the SELENITE broth tube at 35°C ± 2°C for 18-24 hours.
- After incubation, check the turbidity and plate 100 µl of Selenite Broth onto appropriate culture medium.
- Incubate the plates at 35°C ± 2°C for 18-2 hours.
- Read the plate and record the results to release the lot.

The inhibition procedure described below is applied by manufacturer to release the lots:

- Prepare a 0.5 McFarland bacterial suspension in PBS from a fresh culture of microorganism in test.
- Prepare a 1,5x 10<sup>1</sup> dilution from the 0.5 McFarland bacterial suspension.
- Inoculate the SELENITE Broth with 200 µl of the dilution prepared
- Recap all tubes.
- Vortex for 10 seconds at 2500 rpm.
- Plate immediately 100 µl of Selenite Broth inoculated (time zero count).
- Incubate the SELENITE Broth tube at 35°C ± 2°C for 6-8 hours.
- After incubation, check the turbidity and plate 100 µl of Selenite Broth onto appropriate culture medium.
- Incubate the plates at 35°C ± 2°C for 18-24 hours.
- Incubate the Selenite Broth tube at 35°C ± 2°C till the end of incubation (18-24 hours).
- After incubation, check the turbidity and plate again 100 µl of Selenite Broth onto appropriate culture medium.