

# TETRATHIONATE BROTH Dry Bag

## Presentation

Bag of 10 liters with 460g of dehydrated Tetrathionate broth. Includes a  $0.22\mu m$  autoclavable non-sterile microbiological filter. Does not include water pumping equipment for bag filling.

# Sterilization method

Gamma irradiation.

# Application

Medium recommended for the selective enrichment of *Salmonella* from food and other samples.

# Principle

Many *Salmonella* cells may suffer injury during food processing, making their recovery for subsequent identification important. Additionally, within the same sample, there may exist different microorganisms capable of inhibiting the growth of *Salmonella* species. ISO 6579:2017 recommends selective enrichment of food samples to ensure destruction of competing flora and recovery of *Salmonella* species.

The Müller Kauffmann Tetrathionate Novobiocin Broth is used as a selective enrichment medium for *Salmonella* that may be present in small quantities in food. The medium contains sodium thiosulfate, which, in the presence of iodine, produces tetrathionate that suppresses the growth of coliforms and other enteric bacteria present in the sample. *Salmonella*, *Proteus*, and some other bacterial species can reduce tetrathionate and are not inhibited by it. The addition of novobiocin ensures suppression of *Proteus* growth. Bile promotes *Salmonella* growth but inhibits other accompanying bacteria. Brilliant green suppresses Gram-positive bacteria. Calcium carbonate is the buffering agent for sulfuric acid formed during tetrathionate reduction.

#### How to use

Before hydrating the bag, sterilize the microbiological filter by moist heat at 121°C for 15 minutes. The filter can withstand autoclaving up to 10 times. The production of the medium requires the use of a water pumping equipment, such as a peristaltic pump. Follow the procedure below to hydrate the bag, using aseptic manipulation technique to prevent contamination of the culture medium:

- 1. Using a laminar flow hood, remove the dry bag from its packaging.
- 2. Shake the bag to distribute the powder inside. Place the bag on the hood's surface.
- 3. Carefully remove the cap from the bag's hose connector and place it in a sterile Petri dish to prevent contamination.
- 4. Connect the bag's hose to the sterile microbiological filter.
- 5. Connect the filter to a purified water pumping device.

- 6. Open the red valve on the bag and the valve on the filter to allow air to escape.
- 7. Start the water pumping device to fill the bag. Once water enters the bag, close the filter valve.
- 8. While filling, shake the bag to dissolve the powder. After filtering the total volume of water, turn off the device. Close the red valve, disconnect the filter from the bag's hose, and cap the hose connector.
- 9. Distribute the medium into suitable sterile containers. Proceed with the laboratory's analysis methodology.
- 10. Add 0.2ml of iodine solution for tetrathionate (iodine/potassium iodide) to the tube with medium. Inoculate the sample according to the technique adopted by the laboratory. Incubate for the time and temperature required by the technique. Proceed with the analysis according to techniques established by the laboratory.
- 11. After the incubation period, seed onto a selective *Salmonella* agar plate (e.g., XLD, Hektoen, Brilliant Green, SS, etc.), streaking on the surface of the medium using isolation seeding techniques.

# Quality Control

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Test	Result
Sterility	Absence of microbial growth
Salmonella enterica	Good growth in colonies pink
ATCC 14028	with black centers after
	subculturing on XLD agar
Escherichia coli	Partial or inhibited growth after
ATCC 25922	subculture on non-selective
	agar
Enterococcus faecalis	Partial or inhibited growth after
ATCC 29212	subculture on non-selective
	agar
Appearance	Dry medium: white straw-
	coloured granules.
	4.6% Solution: liquid medium,
	opaque, light green, with white
	precipitate.
pH at 25°C	8.0 ± 0.2

#### **Results interpretation**

Microbial growth is verified through colony formation in the culture medium. In the presence of growth, perform microscopic analysis, subculture in selective medium, or biochemical tests to identify isolated genera and species, if necessary.

# Precautions and special care

The water used to fill the bag must meet the grade of water used in preparing culture media. As soon as the water begins to enter the bag, check for any air pressure formation in the filter. If air pressure builds up, quickly open and close the valve of the filter to allow the air to escape.

Product intended for *in vitro* diagnostic use only.

Restricted for use by professionals. Do not inhale or ingest.

Do not use the product beyond the expiration date, with signs of contamination, or if it has changed color. In the presence of contamination, the product should be immediately discarded. Do not use the product if the packaging is damaged or tampered with.

#### Storage

Store between 10-35°C in a dry place and protect from light.

#### Shelf-life

30 days from the date of manufacture for the hydrated medium stored at 2-25  $^{\circ}\text{C}.$ 

#### Disposal of the product

After use, the product must be handled at the generating unit before environmentally appropriate final disposal, in accordance with official regulations.

#### **Quality Guarantee**

bioBoaVista guarantees the quality of its products as long as they are used according to their respective instructions and in accordance with national and international references. bioBoaVista does not take responsibility for the use of its products for purposes other than those described and approved by the company. All clinical diagnoses should be analyzed in conjunction with clinical evidence and not solely based on laboratory results.

#### References

1. ISO 6579-1:2017. Microbiology of food chain – Horizontal method for the detection, enumeration and sorotyping of *Salmonella*. Part 1: Detection of *Salmonella* spp.

2. ISO 11133:2014. Microbiology of food, animal feed and water - Preparation, production, storage and performance testing of culture media.

3. Manual de Métodos de Análise Microbiológica de Alimentos, Livraria Varela, 3ª ed., 2007.

4. Merck Microbiology Manual. 12th ed.