

# Muller Kauffmann Tetrathionate-Novobiocin Broth – MKTTN (i23390)

For the selective enrichment of salmonellae from food and animal feed stuffs.

Industry: Water / Food

### **Principles & Uses**

Tetrathionate Broth, a vital medium for enriching enteric pathogens and isolating *Salmonella* from heavily contaminated samples, involves the addition of iodine to generate tetrathionate from sulfate. This, combined with bile salts, strongly inhibits normal intestinal bacteria, creating an environment where *Salmonella*, capable of reducing tetrathionate, thrives. Reduction reactions release sulfuric acid, neutralized by carbonate to prevent harmful pH decrease.

Bile promotes *Salmonella* growth while hindering accompanying bacteria. Tetrathionate, produced by iodine, is crucial for suppressing normal intestinal flora. However, some *Proteus* species resist bile salts and may reduce tetrathionate. To counteract this, simultaneous addition of inhibitors like Brilliant Green Solution and novobiocin is recommended. These selective agents effectively suppress primarily Grampositive bacteria.

For specific Salmonella serovars such as Salmonella Typhi and Salmonella Paratyphi, additional steps or selective enrichment media, as described in EN ISO/FDIS 6579-1, may be necessary. The intricacies of this medium make it a valuable tool for precise and reliable microbiological analysis in challenging samples. After enrichment, crucial confirmatory tests, including subculturing on selective media like XLD, are imperative for accurate Salmonella detection, and laboratories must evaluate results according to their specifications.

#### Composition (gr/L)

Peptic digest of Animal Tissue 4.3 g, Peptone from Casein 8.6 g, Sodium Chloride 2.6 g, Calcium Carbonate 38.7 g, Sodium Thiosulphate 30.5 g, Ox Bile 4.78 g, Brillant Green 0.0096 g, Novobiocin 0.04 g. Final pH at  $25^{\circ}$ C 8.0  $\pm$  0.2

#### **Preparation from dehydrated Powder**

Suspend 89.5g of MKTTn in 1 litre of distilled water, mix well and bring to the boil. Cool to below 45°C. Immediately before use add 20ml of iodine-iodide solution prepared by dissolving 25 g of potassium iodide in 10ml of water, adding 20 g of iodine and then diluting to 100ml with sterile water. **DO NOT AUTOCLAVE.** 

#### **Quality Control**

Dehydrated Appearance: Cream to greenish yellow, homogeneous, free-flowing.

Prepared Appearance: Light green colored opalescent solution forms with heavy white precipitate.

Reaction of 8.95% Solution at 25°C: pH 8.0 ± 0.2

#### **Cultural Response**

The medium was inoculated using the organisms listed below, then incubated for 20 - 24 hours at 37  $^{\circ}$ C.

| Organism (ATCC*)               | Recovery   |
|--------------------------------|------------|
| Escherichia coli (25922)       | None-poor  |
| Shigella flexneri (12022)      | Inhibition |
| Salmonella Typhimurium (14028) | Excellent  |

\*ATCC is a registered trade mark of the American Type Culture Collection.



Salmonella typhimurium (left). Prepared Culture Media (right).



## Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.