Salmonella Selective Supplement

PRODUCT INFORMATION

C058-100mg - Cefsulodin Sodium, Powder, 100 mg

C058-250mg - Cefsulodin Sodium, Powder, 250 mg

C058-1g - Cefsulodin Sodium, Powder, 1g

DESCRIPTION

Salmonella Chromogenic Agar Base with Salmonella Selective Supplement is a selective and differential medium for the identification of *Salmonella* species from other organisms in the family *Enterobacteriaceae*.

BACKGROUND

Cefsulodin is a third generation cephalosporin antibiotic that has very specific activity against Pseudomonas aeruginosa. It has no significant activity against other Gram-negative bacteria and very limited activity against Gram-positive bacteria and anaerobic bacteria.

Novobiocin is an aminocoumarin antibiotic that is produced by the actinomycete *Streptomyces niveus*, which has recently been identified as a subjective synonym for *S. spheroides* a member of the order Actinobacteria

Mechanism of action

The molecular basis of action of novobiocin, and other related drugs clorobiocin and coumermycin A1 has been examined. Aminocoumarins are very potent inhibitors of bacterial DNA gyrase and work by targeting the GyrB subunit of the enzyme involved in energy transduction. Novobiocin as well as the other aminocoumarin antibiotics act as competitive inhibitors of the ATPase reaction catalysed by GyrB. The potency of novobiocin is considerably higher than that of the fluoroquinolones that also target DNA gyrase, but at a different site on the enzyme. The GyrA subunit is involved in the DNA nicking and ligation activity.

APPLICATION IN SALMONELLA CHROMOGENIC AGAR BASE

Salmonella Chromogenic Medium is designed to identify *Salmonella* species based on their utilisation of one chromogenic substrate. Their inability to utilise another chromogenic substrate, that most other members of the family *Enterobacteriaceae* can utilise, enables rapid and reliable identification of *Salmonella* species.

There are in excess of 2000 known species of *Salmonella*, some of which differ from the typical rod-shaped, Gram-negative motile bacterium. In the U.S. alone there are between 800,000 and 4 million reported cases of salmonellosis per year resulting in 500 deaths. Infections due to *Salmonella* are of particular concern in the very young, the elderly and in the severely immunosuppressed where salmonellosis is recognised as an AIDS defining condition. Incidence continues to rise and infections due to *Salmonella* remain a principal health issue. The widespread occurrence means there is a need for the rapid detection and identification of Salmonella in food and water to aid in the prevention and control of outbreaks.

Traditionally, media used to differentiate *Salmonella* species from other members of the family *Enterobacteriaceae* depend upon the ability of *Salmonella* species to produce hydrogen sulphide coupled with their inability to ferment lactose. These are, however, essentially inadequate methods, with a significant number of the 2000 plus species not exhibiting these characteristics. In recent times chromogenic media have been developed for the rapid and more reliable identification of Salmonella.

Salmonella Chromogenic Agar Base combines two chromogens for the detection of *Salmonella* sp., 5-Bromo-6-Chloro-3-Indolyl caprylate (Magenta-caprylate) and 5-Bromo-4-Chloro-3-Indolyl b-D galactopyranoside (X-gal). X-gal is a substrate for the enzyme b-D-galactosidase. Hydrolysis of the chromogen, Mag-caprylate, by lactose negative Salmonella species results in magenta colonies.

The medium contains bile salts to inhibit the growth of Gram-positive organisms and the addition of the Salmonella Selective Supplement is recommended to increase the selectivity of the medium. This uses novobiocin to inhibit *Proteus* growth and cefsulodin to inhibit growth of *Pseudomonads*.

Content concentrations

Typical Formula*	**mg/litre**
Salmonella Chromogenic Agar Base	
Special Peptone	10
Chromogenic mix	28

Agar	12
Final pH 7.2 ± 0.2 @ 25°C	
Salmonella Selective Supplement	
Cefsulodin	12
Novobiocin	5
* Adjusted as required to meet performance standards	

Table 1 - Typical Formula for Salmonella Chromogenic Agar Base and Salmonella Selective Supplement

METHOD

Preparation

Suspend appreciate amount of Salmonella Chromogenic Agar Base in distilled water and add the contents Salmonella Selective Supplement reconstituted as directed. Mix well and bring to the boil with frequent agitation. DO NOT AUTOCLAVE. DO NOT HOLD AT BOILING TEMPERATURE. Cool to 50°C, mix well and pour into sterile Petri dishes.

Protocol

1. Inoculate the plates with a food or clinical sample to produce single colonies. A *Salmonella* enrichment broth may be used prior to streaking out.

2. Incubate for 18-24 hours at 37°C.

3. Examine the plates for coloured colonies.

On Salmonella Chromogenic Medium, typical colonies will be coloured as follows:

Species	Colony Colour	Colony Diameter	Colony Morphology
Salmonella spp.	Magenta	1.0mm	Raised, smooth
Salmonella typhi	Magenta	1.0mm	Raised, smooth
Salmonella paratyphi	Magenta	1.0mm	Raised, smooth
Salmonella arizonae	Magenta / blue †	1.5mm	Raised, smooth
Salmonella gallinarum	Magenta	0.75mm	Raised, smooth
Salmonella indiana	Blue †	1.0mm	Raised, smooth
Escherichia coli	Blue	1.0mm	Raised, smooth
Enterobacter spp.	Blue	1.5mm	Raised, smooth
Klebsiella spp.	Blue	3.0mm	Raised, mucoid
Citrobacter spp.	Blue	1.5mm	Raised, mucoid
Proteus spp.	No growth / straw	0.25mm	-
Pseudomonas spp.	No growth	-	-
Shigella sonnei	Blue	4.0mm	Undulate
Shigella dysenteriae	Magenta	1.0mm	Raised

Quality control

Positive control:

Salmonella enteritidis ATCC® 13076: Good growth, purple colonies

Salmonella poona NCTC 4840: Good growth, purple colonies

Negative control:

Escherichia coli ATCC® 25922: Growth, blue colonies

Pseudomonas aeruginosa ATCC® 27853: Inhibited

REFERENCES

1. Gaillot, O. et al. (1999) J. Clin. Microbiol. 37: 762-765.

2. Rambach, A. (1990) Appl. Environ. Microbiol. 56: 301-303.

3. Gruenewald, R. (1991) J. Clin. Microbiol. 29: 2354-2356.