SPS Agar (Sulfite Polymyxin Sulfadiazine)

Cat. 1082

For the isolation and enumeration of Clostridium perfringens from food and all types of foodstuffs

Practical information

Aplications	Categories
Selective enumeration	Clostridium perfringens

Industry: Food

Principles and uses

SPS Agar (Sulfite Polymyxin Sulfadiazine) is a moderately selective medium to recover Clostridium perfringens from fresh or preserved foods and food ingredients.

The medium was modified by Angelotti, incorporating sulfadiazine and polymyxin B sulphate to the more recent Mossel formula for the recovery of Clostridium perfringens.

Casein peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group. Ferric citrate and sodium sulfite are H2S indicators. C. perfringens reduces the sulfite to sulfide which in turn reacts with the iron and forms a black iron sulfide precipitate, seen as black colonies. Polymyxin B sulfate and sulfadiazine are inhibitors to organisms other than Clostridium spp. Bacteriological agar is the solidifying agent.

A few microorganisms other than C. perfringens also grow on SPS Agar so it is best to perform a Gram stain and look for spores. Many common microorganisms are totally or partially inhibited, but if they develop, they generally do not form black colonies nor spores, nor do they reduce nitrate and are non-motile Gram-positive vegetative bacilli.

Formula in g/L

Bacteriological agar	13	Casein peptone	15,5
Polymyxin B Sulfate	0,01	Sodium sulfite	0,5
Sulfadiazine	0,12	Yeast extract	10
Ferric citrate	0,5		

Preparation

Suspend 39,7 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 118 ºC for 15 minutes.

Instructions for use

- Prepare in a homogenizer, or other equipment, the material samples and make serial dilutions.
- Dispense the inoculum into sterile Petri dish.
- Pour the medium cooled to 50-55 °C over the inoculum and mix gently the inoculum and medium.
- It can be possible to dispense the medium into tubes and inoculate them by the stab method.
- Incubate anaerobically (The authors used a mixture of 90% nitrogen and 10% CO2) at 35±2 °C for 24-48 hours.
- The lack of motility and the capacity to reduce nitrate can be determined on Indole Nitrate Medium (Cat. 1504) with 2 g/L of added agar.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige	Amber, slightly opalescent	7,0 ± 0,2

Microbiological test

Incubation conditions: (35±2 ºC / 24-48 h)

Microrganisms	Specification	Characteristic reaction
Clostridium perfringens ATCC 13124	Good growth	Black colony
Clostridium sporogenes ATCC 19404	Moderate growth	Black colony
Escherichia coli ATCC 25922	Inhibited growth	
Staphylococcus aureus ATCC 6538	Moderate/inhibited growth	White colony

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

Angelotti, Nall, Foter y Lewis. Applied Microbiol. 10: 193. 1962. Mossel. J.SCI. Agr. 10: 662. 1959. Mossel de Bruin Van Diepen, Vendrig y Zoutwelle J. Applied Bact, 19: 142. 1956.