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# TSC Agar (Tryptose Sulfite Cycloserine Agar), Base

Medium proposed by HARMON et al. (1971) for the isolation and enumeration of the vegetative and spore forms of Clostridium perfringens in foodstuffs and other materials.

The culture medium complies with the recommendations of the International Organization for Standardization (ISO) 7937 (2004). It also conforms with the APHA recommendations for the examination of foods (1992).

## Mode of Action

The superior nutrient base provides optimal conditions for the development of clostridia. Colonies producing hydrogen sulfide are characterized by blackening due to the reaction with sulfite and iron salt. In TSC Agar cycloserine inhibits the accompanying bacterial flora and causes the colonies, which develop, to remain smaller. It also reduces a diffuse and thus disturbing blackening around the Cl. perfringens colonies. SFP Agar contains polymyxin and kanamycin as selective inhibitors of accompanying flora. It is slightly less selective than TSC Agar.

## Typical Composition (g/litre)

Tryptose 15.0; peptone from soymeal 5.0; yeast extract 5.0; sodium disulfite 1.0; ammonium iron(III) citrate 1.0; agar-agar 12.0.

Also to be added:

cycloserine 0.4 or polymyxin 0.003; kanamycin 0.012.

## Preparation

Suspend 39.0 g/litre, dispense into suitable vessels, autoclave (15 min at 121 °C). Add the necessary substances, mix, pour plates.

SFP Agar: Prior to autoclaving, add 3 mg polymyxin sulfate/litre and 12 mg kanamycin disulfate/litre to the culture medium base. These antibiotics can also be added to the sterile, liquefied culture medium in the form of filter-sterilized solutions.

TSC Agar: Cool the liquefied culture medium base to approx.

50 °C, add 0.4 g cycloserine/litre (10 ml of filter-sterilized 5 % solution). Alternatively you can use Clostridium perfringens Supplement, Merck Cat. No. 1.00888.0001.

- Whereas the prepared culture medium base can be stored for several months, the ready-to-use selective culture media must be stored under anaerobic condition and used within 4 days after preparation.
  pH: 7.6 ± 0.2 at 25 °C.

## Experimental Procedure and Evaluation

Inoculate the medium by the pour-plate method or by spreading the sample material on the surface of the plates.

Incubation: 18-24 hours at 37 °C or 44 °C under anaerobic conditions (e.g. Anaerocult® A, Anaerocult® A mini, or Anaerocult® P).

Cl. perfringens produces black colonies. Further tests should be performed for purposes of identification.

## Literature


American Public Health Association: Compendium of methods for the microbiological examination of foods. - 3rd. (1992).

DIN Deutsches Institut für Normung e.V.: Mikrobiologische Untersuchung von Fleisch und Fleischerzeugnissen. Bestimmung von Clostridium perfringens. Plattenguß-Verfahren (Referenzverfahren). - DIN 10165.

EMSWILER, B.S., PIERSON, C.J., a. KOTULA, A.W.: Comparative study of two methods for detection of Clostridium perfringens in ground beef. - Appl. Envir. Microbiol., 33; 735-737 (1977).

HARMON, S.M.: Collaborative study of an improved method for the enumeration and confirmation of Clostridium perfringens in foods. - J. AOAC, 59; 606-612 (1976).

HARMON, S.M., KAUTER, D.A., a. PEELER, J.T.: Comparison of media enumeration of Clostridium perfringens. - Appl. Microbiol., 21; 922-927 (1971).

HAUSCHILD, A.H.W., a. HILSHEIMER, R.: Evaluation and modifications of media for enumeration of Clostridium perfringens. - Appl. Microbiol., 27; 78-82 (1974).

HAUSCHILD, A.H.W., HILSHEIMER, R., a. GRIFFITH, D.W.: Enumeration of faecal Clostridium perfringens spores in egg-yolk-free Tryptose-Sulfite-Cycloserine Agar. - Appl. Microbiol., 27; 527-530 (1974).

International Organization for Standardization (ISO): Meat and meat products. - Enumeration of Clostridium perfringens (Reference method). - Working Draft ISO/TC 34/SC 6 (1978).

ORTH, D.S.: Comparison of sulfite-polymyxin-sulfadiazine medium and tryptose-sulfite-cycloserine medium without eggyolk for recovering Clostridium perfringens. - Appl. Envir. Microbiol., 33; 986-988 (1977).

SHAHIDI, S.A., a. FERGUSON, A.R.: New quantitative, qualitative and confirmatory media for rapid analysis food for Clostridium perfringens - Appl. Microbiol., 21; 500-506 (1971).


## Ordering Information

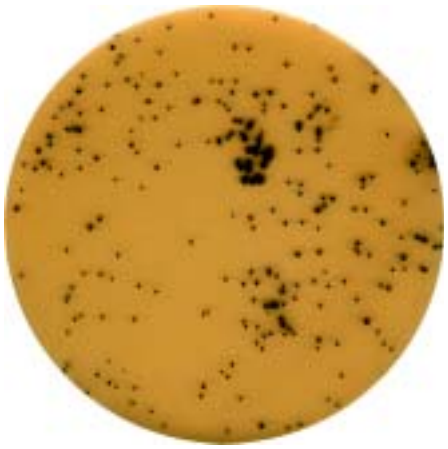
| Product | Merck Cat. No. | Pack size |
|---|---|---|
| TSC Agar (Tryptose Sulfite Cycloserine Agar), Base | 1.11972.0500 | 500 g |
| Anaerobic jar | 1.16387.0001 | 1 ea |
| Anaeroclip® | 1.14226.0001 | 1 x 25 |
| Anaerocult® A | 1.13829.0001 | 1 x 10 |
| Anaerocult® A mini | 1.01611.0001 | 1 x 25 |
| Anaerocult® P | 1.13807.0001 | 1 x 25 |
| Anaerotest® | 1.15112.0001 | 1 x 50 |
| Clostridium perfringens Supplement | 1.00888.0001 | 16 vials |
| Plate basket | 1.07040.0001 | 1 ea |
| UV Lamp (366 nm) | 1.13203.0001 | 1 ea |
| D-Cycloserine | EMD Biosciences | |
| Kanamycin disulfate | EMD Biosciences | |
| Polymyxin-B-sulfate | EMD Biosciences | |

# TSC Agar (Tryptose Sulfite Cycloserine Agar), Base

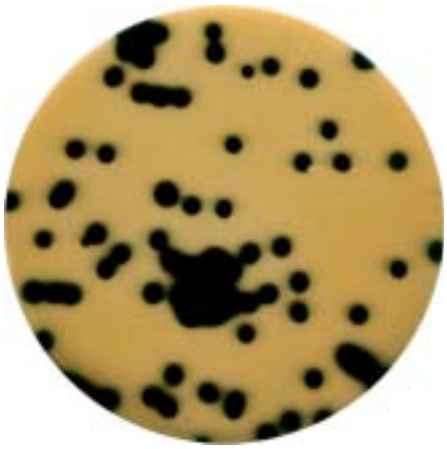
## Quality control

| Test strains | Growth | Black colonies | Fluorescence* |
|---|---|---|---|
| Clostridium perfringens ATCC 10543 | good / very good | + | + |
| Clostridium perfringens ATCC 13124 | good / very good | + | + |
| Clostridium tetani ATCC 19406 | none / fair | - | - |
| Clostridium novyi ATCC 17861 | none / fair | - | - |
| Pseudomonas aeruginosa ATCC 27853 | none / poor | - | - |
| Bacillus cereus ATCC 11778 | none / poor | - | - |
| Escherichia coli ATCC 25922 | none / fair | - | - |

*reading if Clostridium Supplement is used



Clostridium perfringens
ATCC 10543



Clostridium perfringens
ATCC 13124