

# Sabouraud Chloramphenicol Dextrose Agar EP/USP/ISO

For the selective cultivation and isolation of yeasts and molds

# Practical information

Aplications Selective enumeration Categories Yeasts and molds

Industry: Pharmaceutical/Veterinary / Food / General cultivation / Antimicrobial susceptibility testing

Regulations: USP / ISO 11133 / ISO 16212 / European Pharmacopoeia



Cat. 1134

#### Principles and uses

Sabouraud Chloramphenicol Dextrose Agar is a selective medium that can be used for the cultivation of yeasts, molds (As pathogenic fungi, particularly those associated with skin infections) and aciduric microorganisms. This medium is also used for determining the microbial and fungal content of cosmetics and for the mycological evaluation of food. It is recommended by ISO 16212 for the enumeration of yeast and molds in cosmetic products.

The formula is based on the European Pharmacopoeia. Dextrose is the fermentable carbohydrate providing carbon and energy. Peptone mixture provides nitrogen, vitamins, minerals and amino acids essential for growth. Bacteriological agar is the solidifying agent. The high dextrose concentration and acidic pH makes this medium selective for fungi.

This medium is a modification of the Dextrose Agar described by Sabouraud, with the addition of Chloramphenicol that inhibits a great majority of bacterial contaminants. Chloramphenicol is an antibiotic which is used for isolating pathogenic fungi from heavily contaminated material, as it inhibits most contaminating bacteria. It is a recommended antibiotic for use with media due to its heat stability and wide bacterial spectrum.

This medium has a lower concentration of Chloramphenicol than the medium of the same name (Cat. 1090), making it less inhibitory to contaminant bacteria.

Sabouraud Dextrose Agar + Chloramphenicol is recommended by European Pharmacopoeia when the total yeast and molds count (TYMC) is expected to exceed the acceptance criterion due to the bacterial growth

## Formula in g/L

| Bacteriological agar | 15 | Chloramphenicol   | 0,05 |
|----------------------|----|---|------|
| Dextrose             | 40 | Mixture of peptic digest of animal tissue and pancreatic digest of casein (1:1) | 10   |

Typical formula g/L \* Adjusted and/or supplemented as required to meet performance criteria.

## Preparation

Suspend 65 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. Distribute and sterilize in autoclave at 118-121 °C for 15 minutes. AVOID OVEARHEATING.

#### Instructions for use

According to European Pharmacopoeia for the examination of TYMC in products:

Membrane filtration:

- Prepare the sample.

- Transfer the appropriate amount of the sample to a membrane filter.

- Place the membrane to the surface of Sabouraud Dextrose Agar.
- Incubate the plate of Sabouraud Dextrose Agar at 20-25 °C for 5-7 days.
- Plate-count methods:
- Prepare the sample.
- Inoculate the plates of Sabouraud Dextrose Agar conforming to the pour-plate method or the surface-spread method.
- Incubate the plates of Sabouraud Dextrose Agar at 20-25 °C for 5-7 days.
- Select the plates corresponding to a given dilution and showing the highest number of colonies less than 50.

According to European Pharmacopoeia for the test of Candida albicans in products:

- Prepare the product to be examined and use 10 mL or the quantity corresponding to not less than 1 g or 1ML to inoculate 100 mL of Sabouraud Dextrose Broth.

- Incubate at 30-35 °C for 3-5 days.

- Subculture on a plate of Sabouraud Dextrose Agar.
- Incubate at 30-35 °C for 24-48 hours.
- Growth of white colonies may indicate the presence of C. albicans. Confirm by identification tests.
- The product complies with the test if such colonies are not present or if the confirmatory tests are negative.

For the enumeration of yeast and molds according to ISO 16212:

Prepare the initial suspension from a sample of at least 1 g or 1 ml of the product. If needed, additional serial dilutions (e.g. 1:10 dilution) may be performed from the initial suspension using the same diluent (according to the expected level of contamination of the product).
The enumeration of the yeast and molds may be made by plate count methods and membrane filtration method.

Inoculate the initial suspension and/or sample dilution in Sabouraud Dextrose Agar + Chloramphenicol plates.

- Incubate at 25±2,5 °C for 3-5 days.

- After incubation, count the colonies.

#### Quality control

| Solubility | Appareance  | Color of the dehydrated medium | Color of the prepared medium | Final pH (25°C) |
|------------|-------------|--------------------------------|------------------------------|-----------------|
| w/o rests  | Fine powder | Beige                          | Amber, slightly opalescent   | 5,6 ± 0,2       |

# Microbiological test

| According to European Pharmacopoeia:<br>Aspergillus brasiliensis and Candida albicans.<br>Incubation conditions: (20-25 °C /<=5 days).<br>Inoculation conditions: (<=100 CFU).                                   |                        |                         |  |
|--|------------------------|-------------------------|--|
| According to ISO 11133; Sacharomyces cerevisiae:<br>Incubation conditions: (25±1 °C / 5 days).<br>Inoculation conditions: Productivity quantitative: (100±<br>Reference media: Media batch SDA already validated | -20.Min.50 CFU).<br>I. |                         |  |
| Rest of strains:<br>Incubation conditions: (30 °C / 3-7 days).   |                        |                         |  |
| Microorganisms   | Specification          | Characteristic reaction |  |
| Candida albicans ATCC 10231  | Good growth            | White colonies          |  |
| Aspergillus brasiliensis ATCC 16404  | Good growth            |                         |  |
| Escherichia coli ATCC 25922  | Inhibition             |                         |  |
| Staphylococcus aureus ATCC 25923   | Inhibition             |                         |  |
| Saccharomyces cerevisiae ATCC 9763   | Good gorwth, >70%      | Cream domed colonies    |  |

## Storage

Temp. Min.:2 °C Temp. Max.:25 °C

## Bibliography

Sabouraud R. 1892. Ann. Dermatol. Syphilol. 3:1061. Jarett, L., and A.C. Sonnenwirth (ed) 1980. Gradwohl's clinical laboratory methods and diagnosis, 8th ed. CV Mosby. Curry, A. S., J. G. Graf, and G. N. McEwen, Jr. (ed) 1993. CTFA Microbiology Guidelines. The Cosmetic, Toiletry, and Fragrance Association, Washington, D.C. European Pharmacopoeia. 9.3 ISO 16212: 2017 Cosmetics. Microbiology. Enumeration of yeast and molds.