

## Specification

Solid medium for the enumeration and cultivation of fungi according to the Pharmacopeial Harmonised Method and ISO standard.

## Presentation

10 Prepared bottles  
Bottles 250 ml  
with: 200 ± 5 ml

**Packaging Details**  
1 box with 10 bottles 250 ml  
Non injectable cap

**Shelf Life** 16 months  
**Storage** 8-25°C

## Composition

Composition (g/l):

D(+)-Glucose..... 40.0  
Peptone from casein ..... 5.0  
Meat Peptone..... 5.0  
Agar..... 15.0

## Description /Technique

### Description

Sabouraud Dextrose Agar is a modification of the classical Sabouraud medium for the cultivation of fungi. This new formula helps to maintain the morphology of fungi, providing a reliable medium for both cultivation and differentiation.

Its selectivity is due to a low pH and a high glucose concentration, which together with incubation at a relatively lower temperature (25-30 °C) favours the growth of fungi while discouraging that of bacteria.

The mixture of peptones employed has been selected to provide the fungi with all their nitrogen requirements.

Since Sabouraud medium's low pH can partially hydrolyze the agar, only the required amount should be prepared and it should not be re-melted. Any overheating will also diminish its gelling capacity.

Should a higher selectivity be required, a variety of inhibitors or selective agents may be added after sterilization, while the medium is still in the molten form. It can also be made differential by adding suitable indicator agents. Some of the inhibitory and differential mixtures most commonly used are listed below:

- Penicillin: at 20.000 u/L, for bacterial inhibition.
- Penicillin and Streptomycin: at 20.000 u/L and 40.000 u/L used for the isolation of Histoplasma in dogs.
- Penicillin and Neomycin: at 20.000 u/L and 40 mg/L for bacterial inhibition.
- Streptomycin and Chloramphenicol: at 40 mg/L and 500 mg/L, for the isolation of Trichophyton verrucosum.
- Colistin, Novobiocin and Cycloheximide: at 8 mg/L, 0.1 mg/L and 30 mg/L, for the isolation of Candida albicans.
- Potassium Tellurite: at 150 mg/L, used for the primary isolation of fungi from scales and scabs.
- Cupric Sulfate, Crystal Violet and Brilliant Green: at 500 mg/L, 2 mg/L and 5 mg/L each, for bacterial inhibition.
- Triphenyltetrazolium chloride (TTC): at 100 mg/L, is the basis of a Pagano-Levin medium for the isolation of Candida albicans, which remains non-pigmented, among other pink coloured pathogenic yeasts.
- Chloramphenicol: for bacterial inhibition at 50,00 mg/L
- Oxytetracycline: for bacterial inhibition at 100 mg/L

### Technique:

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results. Dispense liquid medium in appropriate containers if the original container is of large volume..

This medium is also well suited for air environmental sampling (total compatibility with most commercially available air samplers) or for other types of environmental sampling (fingers or gloves of operators, swab streaking,...).

Melt the medium contained in the bottles in a water bath or in a microwave oven, avoiding overheating, before pouring into Petri dishes when cooled to room temperature.

Once solidified on a flat surface, Spread the plates by streaking methodology or by spiral method.

Incubate the plates right side up aerobically at 20-25°C for up to 5 days.

(Incubation times greater than those mentioned above or different incubation temperatures may be required depending on the sample, on the specifications,... This medium can be inoculated directly or after enrichment with broth).

After incubation, enumerate all the colonies that have appeared onto the surface of the agar.

Each laboratory must evaluate the results according to their specifications.

Calculate total microbial count per ml of sample by multiplying the average number of colonies per plate by the inverse dilution factor if streaked a diluted sample. Report results as Colony Forming Unit (CFU's) per ml or g along with incubation time and temperature.

## Quality control

### Physical/Chemical control

Color : Straw-coloured yellow      pH:  $5.6 \pm 0.2$  at 25°C

### Microbiological control

Melt Medium - Prepare Plates - According to harmonized pharmacopoeial monographs, ISO standards and test methods

Spiral Spreading: Practical range 50 - 100 CFU (Productivity).

Aerobiosis. Incubation at 20-25°C. Reading  $\leq 5$  days.

### Microorganism

*Aspergillus brasiliensis* ATCC® 16404, WDCM 00053

*Candida albicans* ATCC® 10231, WDCM 00054

*S. cerevisiae* ATCC® 9763, WDCM 00058

### Growth

Good ( $\geq 70$  %)

Good ( $\geq 70$  %)

Good ( $\geq 70$  %)

### Sterility Control

Incubation 48 hours at 30-35°C and 48 hours at 20-25°C: NO GROWTH

Check at 7 days after incubation in same conditions

## Bibliography

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