SABOURAUD BASE MEDIA

PRODUCTS:

Plated Media:[a]

Sabouraud Dextrose Agar	303434, P2302 (double)
Sabouraud Dextrose Agar with Chloramphenicol	P2305, P2306
Sabouraud Dextrose Agar with Chloramphenicol and Gentamicin	P2312
Sabouraud Dextrose Agar with Chloramphenicol, Cycloheximide, and Gentamicin (CCG)	P2315
Sabouraud Agar, Emmons	P2304
Sabouraud Agar, Emmons, with Chloramphenicol	P2313 (double)
Sabouraud Agar, Emmons, with Gentamicin	P2307

Tubed and Bottled Media:[a]

Sabouraud Dextrose Agar	T3500, T7319, T7320, T7321
Sabouraud Dextrose Agar	B5610, B8680, B8683
Sabouraud Dextrose Agar with Chloramphenicol	T7323
Sabouraud Dextrose Agar with Chloramphenicol and Cycloheximide	T7324
Sabouraud Liquid (Broth)	T7325, T8625, B8655
Sabouraud Agar, Emmons	T3502, T7322, T8610 (deep)
Sabouraud Agar, Emmons, with Gentamicin	T7326
Sabouraud Agar with Lecithin and Polysorbate 80	T8615 (deep), B8685

[a]see catalog for ordering options

PURPOSE:

Sabouraud media are used for the isolation, cultivation, and maintenance of saprophytic and pathogenic fungi. The addition of antimicrobics inhibits the growth of bacterial flora.

PRINCIPLE:

Sabouraud media was described by Sabouraud[8] in 1892 and was used for the identification of fungi based on their morphological characteristics. Sabouraud Dextrose Agar is a standard media used to support the growth of yeasts and molds. It supplies peptone as the protein source and dextrose as the carbohydrate source for nourishment. Bacterial suppression occurs due to the low pH.[1,3] This media is especially suited for primary isolation of fungi from normally sterile sites such as cerebrospinal fluid.

Later, Emmons[4] modified the media by decreasing the dextrose content and adjusting the pH closer to the neutral range. This modification enhances sporulation and is particularly useful for the subculture of fungi that do not develop fruiting structures on other media, and so is useful in their identification. It also serves as a good holding media for stock cultures.[1,7]

Sabouraud Broth is the fluid counterpart of the agar. This media is used for the primary isolation of fungi from fluid specimens that are normally sterile and for the identification of *Candida* species based on their growth pattern and production of gas. It is particularly helpful in differentiating *Candida krusei* and *Candida tropicalis* from other *Candida* species.

The Sabouraud Base media can be made selective by the addition of one or more antimicrobics, (i.e. Chloramphenicol, Cycloheximide, Gentamicin). These antimicrobics inhibit bacterial growth and are useful for primary isolation of fungi from contaminated sites such as respiratory sources. Sabouraud with chloramphenicol is suited for the recovery of dermatophytes from cutaneous specimens. Cycloheximide prevents overgrowth of several rapidly growing fungi, allowing the slower-growing molds to grow. Gentamicin may be incorporated into the media to inhibit the growth of gram-negative and gram-positive bacteria.

FORMULAS*:

Approximate, per liter deionized filtered water.

(1) Sabouraud Dextrose Agar:

Pancreatic Digest of Casein	5.0 g
Peptic Digest of Animal Tissue	5.0
Dextrose	40.0
Agar	15.0

Final pH 5.6 ± 0.2 at 25°C

(2) Sabouraud Dextrose Agar with Lecithin and Polysorbate 80:
Same as **(1)** with 0.7g of Lecithin and 5.0g of Polysorbate 80.

(3) Sabouraud Dextrose Agar with Chloramphenicol:
Same as **(1)** with 50.0 mg of Chloramphenicol.

(4) Sabouraud Dextrose Agar with Chloramphenicol and Cycloheximide:
Same as **(1)** with 100.0 mg of Chloramphenicol and 500.0 mg of Cycloheximide.

(5) Sabouraud Dextrose Agar with Chloramphenicol and Gentamicin:
Same as **(1)** with 50.0 mg of Chloramphenicol, and 8.0 mg of Gentamicin.

(6) Sabouraud Dextrose Agar with Chloramphenicol, Cycloheximide, and Gentamicin:
Same as **(1)** with 100.0 mg of Chloramphenicol, 500.0 mg of Cycloheximide and 8.0 mg of Gentamicin.

(7) Sabouraud Broth:
Pancreatic Digest of Casein ..... 5.0 g
Peptic Digest of Animal Tissue 5.0
Dextrose 20.0
Final pH 5.6 ± 0.2 at 25°C

(8) Sabouraud Agar, Emmons:
Pancreatic Digest of Casein ..... 5.0 g
Pancreatic Digest of Animal Tissue 5.0
Dextrose 20.0
Agar 17.0
Final pH 6.9 ± 0.2 at 25°C

(9) Sabouraud Agar, Emmons, with Chloramphenicol:
Same as **(8)** with 50.0 mg of Chloramphenicol.

(10) Sabouraud Agar, Emmons, with Gentamicin:
Same as **(8)** with 5.0 mg of Gentamicin.

* Adjustments and/or supplements may be required to meet performance standards.

PRECAUTIONS:*

For *in vitro* diagnostic use. Observe approved biohazard precautions.

Storage: Upon receipt store at 2-8°C away from direct light. Media should not be used if there are signs of contamination, deterioration (shrinking, cracking, evaporation, or discoloration), or if the expiration date has passed.

Limitations: Growth on these media is not sufficient for the identification of fungi. Further biochemical, physiological, or serological tests are required for definitive identification.

Antimicrobial agents contained in the media may inhibit aerobic actinomycetes and certain fungi. Aerobic actinomycetes such as *Nocardia* sp. are inhibited by chloramphenicol and gentamicin. Cycloheximide may inhibit some pathogenic fungi; therefore, media containing cycloheximide should never be used alone, but always used in conjunction with media that does not contain cycloheximide. Fungi inhibited by cycloheximide include: *Cryptococcus neoformans, Aspergillus* sp., *Pseudallescheria boydii, Trichosporon* sp., some *Candida* sp., and the zygomycete group. The yeast phase of some of the dimorphic fungi may be inhibited by cycloheximide, but the mycelial phase of these organisms is not inhibited if incubated at 25-30°C.

If using a swab for inoculation, it is best to inoculate the nonselective media first so that the selective agents are not transferred from one media to another.

The taping of plates may be necessary to avoid excessive dehydration and aerial dissemination of spores.

PROCEDURE:*

Specimen Collection: Infectious material should be submitted directly to the laboratory in a sterile container, or other appropriate means of transport. Consult appropriate references for specimen collection and transport.[5,6,7] Samples should be processed as

soon as possible upon arrival in the laboratory. Observe appropriate biohazard precautions when handling the specimen.

Method of Use: Prior to inoculation, the media should be brought to room temperature. Cutaneous specimens, biopsy, or autopsy specimens should be lightly embedded in the agar. Larger specimens should be macerated with sterile normal saline in a tissue grinder, then inoculated onto the media. Specimens from normally sterile sites should be streaked across the agar surface. If the site is possibly contaminated, the specimen should also be inoculated onto selective media.

To prepare plated media from bottled or tubed agar, heat the bottle or tube in a boiling water bath until the agar is melted, cool to 50°C, pour the melted media into a sterile petri dish, and cool at room temperature to solidify. Inoculate lightly.

Inoculated media should be incubated aerobically at 25°C. Media containing cycloheximide should be incubated at 25-30°C. Caps on slants or broth should be loose to ensure an aerobic environment. Most dermatophytes, with the exception of *Trichophyton verrucosum*, will grow well at 25°C. Media should be examined at regular intervals for up to six weeks before considering the specimen negative for fungi. Specimens for which fastidious organisms such as *Histoplasma capsulatum* and *Blastomyces dermatitidis* are suspected should also be inoculated onto an enrichment media such as BHI Agar.

Additionally, fungi may be transferred on Sabouraud media and characterized by their gross morphology (topography, texture, and pigmentation), as well as rate of growth and microscopic appearance.

Interpretation: Once growth occurs, note each specific type of colony morphology by gross appearance (topography, texture, and pigmentation). Subculture to appropriate media to perform specific biochemical and microscopic examination to secure a definitive identification of the organism.

Materials Required but Not Provided: Standard microbiological supplies and equipment such as those products commonly used in a microbiological laboratory are not provided.

QUALITY CONTROL:*

Microorganisms Used (ATCC #):	**Expected Results:**
Sabouraud Agar without antimicrobics:	
Trichophyton mentagrophytes (9533)	Growth
Candida albicans (10231)†	Growth
Aspergillus niger (16404)†	Growth
Sabouraud Agar with antimicrobics:	
same as above, plus:	
Candida albicans (60193)	Growth (Chlor/Gent, CCG)
Escherichia coli (25922)	Inhibition, partial (Chlor/Gent, only)
Escherichia coli (8739)	Inhibition (Emmons, with Chloramphenicol, only)
Staphylococcus aureus (25922)	Inhibition (CCG, only)
Candida krusei (14243)	Inhibition (Cycloheximide, CCG); Growth (Chloramphenicol, Gentamicin)
Aspergillus niger (16404)	Inhibition (Cycloheximide); Growth (Chloramphenicol, Gentamicin)
Sabouraud Broth:	
Candida albicans (10231)†	Growth
Candida krusei (14243)	Growth

†USP microorganisms

User Quality Control: Check for signs of contamination and deterioration. Sabouraud Agar media should appear translucent to slightly opalescent, and light amber to straw in color. Broth media should appear clear, and light amber to tan in color with no precipitate.

BIBLIOGRAPHY:

1. Campbell, M. C., and J. L. Stewart, *The Medical Mycology Handbook*, John Wiley & Sons, New York, 1980.
2. Centers for Disease Control Laboratory Manual for Medical Mycology, PHS, Publication No. 994, U.S. Govt. Printing Office, Washington, D. C., 1963.
3. Cooke, W. B., *Antibiot. Chemother.*, 4:657, 1954.
4. Emmons, C. W., et al., *Medical Mycology*, 2nd ed., Lea and Febiger, Philadelphia, 1970.

5. Koneman, E. W., et al., *Color Atlas and Textbook of Diagnostic Microbiology*, 5th ed., J. B. Lippincott, Philadelphia, 1997.
6. Murray, P.R., et al., *Manual of Clinical Microbiology*, 8th ed., American Society for Microbiology, Washington, D. C., 2003.
7. Rippon, J., *Medical Mycology*, W. B. Saunders, Philadelphia, 1974.
8. Sabouraud, R., *Ann. Dermatol. and Syphil.*, 3:1061, 1892.
9. Sabouraud, R., *Les Tegnes*, Masson et Cie, Paris, 1910, p. 553.

*For more detailed information, consult appropriate references.

bioMérieux, Inc.
Data #670
Revision Date: April 2009