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Instructions for Use

SABOURAUD DEXTROSE (SABDEX) MEDIA

Cat. no. P36	SabDex Agar, Contact Plate	10 plates/bag
Cat. no. X40	SabDex Agar, 50ml HardyFlask™, 12ml Slant	20 flasks/box
Cat. no. L40	SabDex Agar, 20x125mm Tube, 10ml Slant	20 tubes/box
Cat. no. K54	SabDex Broth, 16x125mm Tube, 10ml	20 tubes/box
Cat. no. W20	SabDex Agar, Emmons, 15x100mm Plate, 26ml	10 plates/bag
Cat. no. X57	SabDex Agar, Emmons, 50ml HardyFlask™, 10ml	20 flasks/box
Cat. no. W72	SabDex Agar with Chloramphenicol, 15x100mm Plate, 26ml	10 plates/bag
Cat. no. X41	SabDex Agar with Chloramphenicol, 50ml HardyFlask™, 12ml Slant	20 flasks/box
Cat. no. Q34	SabDex Agar with Chloramphenicol, 20x125mm Tube, 18ml Deep	20 tubes/box
Cat. no. G159	SabDex Agar with Chloramphenicol and Gentamicin, 15x60mm Plate, 12ml	10 plates/bag
Cat. no. W73	SabDex Agar with Chloramphenicol and Gentamicin, 15x100mm Plate, 26ml	10 plates/bag
Cat. no. J107	SabDex Agar with Chloramphenicol and Gentamicin / Dermatophyte Test Medium (DTM), 15x100mm Biplate, 15ml/15ml	10 plates/bag
Cat. no. W74	SabDex Agar with Chloramphenicol and Tetracycline, 15x100mm Plate, 26ml	10 plates/bag
Cat. no. W250	SabDex Agar, 25x100mm Plate, 60ml, deep fill	5 plates/bag

INTENDED USE

Hardy Diagnostics Sabouraud Dextrose Agar, Sabouraud Dextrose Broth, and Sabouraud Dextrose Agar, Emmons are recommended for the isolation, cultivation, and maintenance of non-pathogenic and pathogenic species of fungi and yeasts. Sabouraud Dextrose Agar with Chloramphenicol, Sabouraud Dextrose Agar with Chloramphenicol and Gentamicin, and Sabouraud Dextrose Agar with Chloramphenicol and Tetracycline are recommended for the selective isolation of fungi and yeasts from clinical and nonclinical specimens.

Cat. no. P36 is not intended to be used for the diagnosis of human disease.

SUMMARY

Sabouraud Dextrose Agar was formulated by Sabouraud in 1892 for culturing dermatophytes.[13] The pH is adjusted to approximately 5.6 in order to enhance the growth of fungi, especially dermatophytes, and to slightly inhibit bacterial growth in clinical specimens.[2] This medium is recommended for mold and yeast counts by the Association of Official Analytical Chemists and the *Compendium of Methods for the Microbiological Examination of Foods*.[3,6,14] Sabouraud Dextrose Broth is a modification of the original formulation made without agar. Sabouraud Dextrose Agar, Emmons is

a modification of the original formulation. Emmons originally formulated this modification, which reduces the amount of dextrose, and neutralizes the medium to a pH of approximately 7.0.[9] Chloramphenicol, gentamicin, and tetracycline are selective agents added to inhibit bacterial overgrowth of competing microorganisms while permitting the successful isolation of fungi and yeasts.

Sabouraud Dextrose Medium contains digests of animal tissues (peptones) which provide a nutritious source of amino acids and nitrogenous compounds for the growth of fungi and yeasts. Dextrose is added as the energy and carbon source. Chloramphenicol and/or tetracycline may be added as broad spectrum antimicrobials to inhibit the growth of a wide range of gram-positive and gram-negative bacteria. Gentamicin is added to further inhibit the growth of gram-negative bacteria.

Sabouraud Dextrose Medium is not recommended for the cultivation of dermatophytes, dematiaceous fungi, and mucormycetes (formally zygomycetes). Also, it is a poor promoter of conidiation (see "Limitations" section below).

SabDex Agar Contact Plate (Cat. no. P36), is recommended for use in the cultivation of microorganisms from environmental surfaces. The contact plate has a specified grid molded into the bottom of the plate. They are used primarily to monitor microbial contamination, for enumeration of microbial colonies growing on a variety of surfaces, and to assist in determining surface sanitation.

FORMULA

Ingredients per liter of deionized water:*

Sabouraud Dextrose Agar:	
Dextrose	40.0gm
Pancreatic Digest of Casein	5.0gm
Peptic Digest of Animal Tissue	5.0gm
Agar	15.0gm

Final pH 5.6 +/- 0.2 at 25ºC.

In addition,

Sabouraud Dextrose Broth is the same formulation as above, without agar added.
Final pH 5.6 +/- 0.2 at 25ºC.

Sabouraud Dextrose Agar with Chloramphenicol contains 50.0mg of chloramphenicol.
Final pH 5.6 +/- 0.3 at 25ºC.

Sabouraud Dextrose Agar with Chloramphenicol and Gentamicin contains 50.0mg of chloramphenicol and 5.0mg gentamicin.
Final pH of 5.6 +/- 0.3 at 25ºC.

Sabouraud Dextrose Agar with Chloramphenicol and Tetracycline contains 50.0mg of chloramphenicol and 10.0mg of tetracycline.
Final pH of 5.6 +/- 0.3 at 25ºC.

Sabouraud Dextrose Agar, Emmons has only 20.0gm of dextrose.
Final pH of 6.9 +/- 0.2 at 25ºC.

*Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Storage: Upon receipt store plated products (Cat. no. P36, W20, W72, G159, W73, J107, W74, and W250) at 2-8ºC away from direct light. SabDex Agar tubed and bottled products (Cat no. X40, L40, and K54), SabDex with Chloramphenicol tube and HardyFlask® (Cat. no. X41 and Q34), and SabDex Agar, Emmons (Cat. no. X57) should be stored at 2-30ºC away from direct light.

Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "Storage" for more information.

PRECAUTIONS

For Cat. nos. X40, L40 K54, W20, X57, W72, X41, Q34, W73, J107, W74, and W250.

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual Universal Precautions for blood. Do not ingest, inhale, or allow to come into contact with skin.

This product is for *in vitro* diagnostic use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

For Cat. no. G159, and P36.

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

PROCEDURE

Specimen Collection: Consult listed references for information on specimen collection.[1,2,4,5,7-10] Infectious material should be submitted directly to the laboratory without delay and protected from excessive heat and cold. If there is to be a delay in processing, the specimen should be inoculated onto an appropriate transport media and refrigerated until inoculation.

Consult the listed references for information regarding the processing and inoculation of specimens.[1,2,4,5,7-10]

Method of Use: Allow media to warm to room temperature, and the agar surface to dry before inoculating. Inoculate and streak the specimen as soon as possible after collection. If the specimen to be cultured is on a swab, roll the swab over a small area of the agar surface. Streak for isolation with a sterile loop. Incubate plates in an inverted position. MycoSeal™ (Cat. no. SS9225) may be used to seal plate lids to keep moisture from evaporating from plated media, while still allowing for atmospheric circulation. Examine plates for typical colonial and hyphal morphology and color.

Note: Inoculate two samples for isolation of fungi that cause systemic mycoses; incubate one at 25-30ºC and the other set at 35 +/- 2ºC. Examine cultures weekly for a period of up to four to six weeks; solid media should be incubated under conditions of increased humidity during prolonged incubation.

For melting bottled media and agar deeps: Liquefy the medium by autoclaving at 121ºC for 1-3 minutes Cool the medium to 45-50ºC and pour into sterile petri dishes. Allow the agar to solidify for at least 30 minutes prior to use. Alternatively, a covered, boiling waterbath (100ºC) can be used. There should be enough water in the waterbath to reach the top of the media line. Heat in a waterbath until melted through. A covered waterbath will help to reach and maintain the media temperature prior to dispensing.

Note: After autoclaving, do not heat media using a hot plate, heat block or waterbath for longer than 3 hours at 45-50ºC. Melt only enough media that can be poured within a 3 hour time period. For optimal performance, sterile solidified medium should be remelted only once prior to use.

For tubed media: Inoculate the slant or broth and replace the caps loosely to allow for air circulation. Media should be protected from light and incubated aerobically; solid media should be incubated under conditions of increased humidity during prolonged incubation. Examine SabDex Broth for growth by comparing turbidity to an uninoculated control. Subculture onto an appropriate agar medium when growth is observed.

INTERPRETATION OF RESULTS

Identification of fungi is performed by observing various aspects of colony morphology, characteristic microscopic structures, rate of growth, media which supports the organism's growth, and source of specimen. Yeasts are identified by various biochemical tests. Consult the listed references for information regarding the identification and further testing of fungi and yeast cultures.[1,2,4,5,7-10]

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification of bacteria and/or fungi.

Sabouraud Dextrose is not a good medium to promote conidiation of filamentous fungi. Potato Flake Agar (Cat. no. W59) may be used for any mold which fails to thrive or produce identifying structures on SabDex medium.

Dermatophytes, dematiaceous fungi, and mucormycetes (formally zygomycetes) may grow poorly, if at all, on Sabouraud Dextrose Media. Inhibitory Mold Agar (Cat. no. W25) or Potato Dextrose Agar (Cat. no. W60) may promote more luxuriant growth.

For the growth of mucormycetes (formally zygomycetes), medium containing cycloheximide should not used because these fungi are sensitive to cycloheximide. If all other media fail, sterile bread in a test tube may recover mucormycetes (formally zygomycetes) since these microbes are commonly associated with bread.

A non-selective and selective medium should be inoculated in parallel for isolation of fungi from potentially contaminated specimens.

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, slides, colony counters, microscopes, MycoSeals™ (Cat. no. SS9225), incinerators, and incubators, etc., as well as serological and biochemical reagents, are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

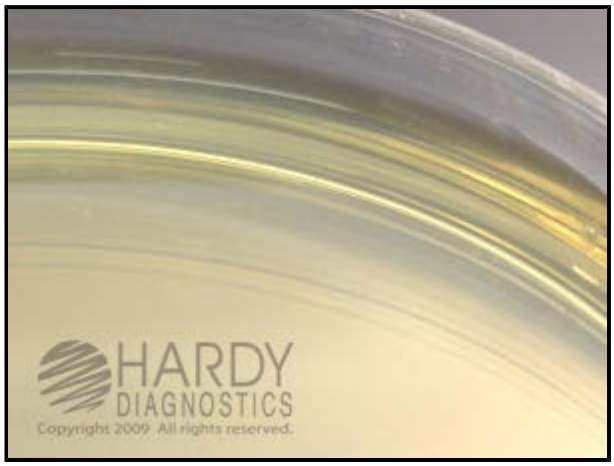
Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
SabDex Agar and SabDex Broth (Cat. no. P36, X40, L40, W250, and K54):					
Candida albicans ATCC® 10231	A	1-3 days	20-25°C	Aerobic	Growth
Aspergillus brasiliensis ATCC® 16404	G	1-5 days	20-25°C	Aerobic	Growth
Trichophyton mentagrophytes ATCC® 9533	G	1-5 days	20-25°C	Aerobic	Growth may take up to one week
SabDex Agar, Emmons (Cat. no. W20, and X57):					
Candida albicans ATCC® 60193	A	48hrs	20-25°C	Aerobic	Growth
Aspergillus brasiliensis ATCC® 16404	G	1-5 days	20-25°C	Aerobic	Growth
Trichophyton mentagrophytes ATCC® 9533	G	1-5 days	20-25°C	Aerobic	Growth
SabDex Agar with Chloramphenicol (Cat. no. W72, X41, and Q34) and SabDex Agar with Chloramphenicol and Tetracycline (Cat. no. W74):					
Aspergillus brasiliensis ATCC® 16404	G	1-5 days	20-25°C	Aerobic	Growth
Candida albicans ATCC® 10231	A	24-48hr	20-25°C	Aerobic	Growth
Trichophyton mentagrophytes ATCC® 9533	G	1-5 days	20-25°C	Aerobic	Growth
Escherichia coli ATCC® 25922	B	24hr	35°C	Aerobic	Partial to complete inhibition
SabDex Agar with Chloramphenicol and Gentamicin (Cat. no. G159, W73, and Side I of J107):					

Aspergillus brasiliensis ATCC® 16404	G	1-5 days	20-25°C	Aerobic	Growth
Candida albicans ATCC® 10231	A	24-48hr	20-25°C	Aerobic	Growth
Trichophyton mentagrophytes ATCC® 9533	G	1-5 days	20-25°C	Aerobic	Growth
Escherichia coli ATCC® 25922	B	24hr	35°C	Aerobic	Partial to complete inhibition
Staphylococcus aureus ATCC® 25923	B	24hr	35°C	Aerobic	Partial to complete inhibition

USER QUALITY CONTROL

PHYSICAL APPEARANCE

Sabouraud Dextrose Media should appear translucent, and light amber in color.



Uninoculated plate of Sabouraud Dextrose Agar.

REFERENCES

1. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.

2. Ajello, et al. 1963. *CDC Laboratory Manual for Medical Mycology*, PHS Publication No. 994. U.S. Gov't Printing Office, Washington, D.C.

3. U.S. Food and Drug Administration. *Bacteriological Analytical Manual.* AOAC, Arlington, VA. www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/ucm2006949.htm

4. Versalovic, J., et al. *Manual of Clinical Microbiology.* American Society for Microbiology, Washington, D.C.

5. Tille, P.M., et al. *Bailey and Scott's Diagnostic Microbiology*, C.V. Mosby Company, St. Louis, MO.

6. American Public Health Association. *Standard Methods for the Examination of Water and Wastewater*, APHA, Washington, D.C.

7. Haley, L.D., et al. 1980. *Cumitech 11: Practical Methods for Culture and Identification of Fungi in the Clinical*

Microbiology Laboratory, Coordinating ed., J.C. Sherris. American Society for Microbiology, Washington, D.C.

8. Isenberg, H.D. *Clinical Microbiology Procedures Handbook*, Vol. I, II & III. American Society for Microbiology, Washington, D.C.

9. Kwon-Chung, K.J. and J.E. Bennett. 1992. *Medical Mycology*. Lea and Febiger, Malvern, PA.

10. Larone, D.H. 2011. *Medically Important Fungi: A Guide to Identification*, 5th ed. American Society for Microbiology, Washington, D.C.

11. MacFaddin, J.F. 2000. *Biochemical Tests for Identification of Medical Bacteria*, 3rd ed. Lipincott Williams & Wilkins, Philadelphia, PA.

12. *Quality Assurance for Commercially Prepared Microbiological Culture Media*, M22. Clinical and Laboratory Standards Institute (CLSI - formerly NCCLS), Wayne, PA.

13. Sabouraud, R. 1892. *Ann. Dermatol. Syphil.*; 3:1061.

14. APHA Technical Committee on Microbiological Methods for Foods. *Compendium of Methods for the Microbiological Examination of Foods*, APHA, Washington, D.C.

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