

Media formulations containing Polypeptone Peptone are specified in standard methods for various applications.⁶⁻¹¹

Principles of the Procedure

Polypeptone Peptone is a mixture of peptones made up of equal parts of pancreatic digest of casein and peptic digest of animal tissue. Polypeptone Peptone includes the high content of amino acids and small polypeptides characteristic of pancreatic digest of casein and the larger polypeptides characteristic of peptic

digest of animal tissue. Polypeptone Peptone provides nitrogen, amino acids and vitamins in microbiological culture media.

Typical Analysis

Refer to Product Tables in the Reference Guide section of this manual.

Directions for Preparation from Dehydrated Product

Refer to the final concentration of Polypeptone Peptone in the formula of the medium being prepared. Add product as required.

Procedure

See appropriate references for specific procedures using Polypeptone Peptone.

Expected Results

Refer to appropriate references and procedures for results.

References

1. Tsuchiya and Kimura. 1978. Appl. Environ. Microbiol. 35:631.
2. Son, Heo, Kim and Lee. 2001. Biotechnol. Appl. Biochem. 33(Pt 1):1.
3. Lee, Lee, Kwon, Lee and Chang. 2000. Appl. Microbiol. Biotechnol. 54:23.
4. Yang, Takeyama, Tanaka and Matsunaga. 2001. Enzyme Microbiol. Technol. 29:13.
5. Taniyama, Yoshida and Furuta. 1988. J. Immunol. 141:4061.
6. Horowitz (ed.). 2000. Official methods of analysis of AOAC International, 17th ed. AOAC International, Gaithersburg, Md.
7. U.S. Food and Drug Administration. 1995. Bacteriological analytical manual, 8th ed. AOAC International, Gaithersburg, Md.
8. Downes and Ito (ed.). 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
9. United States Pharmacopeial Convention, Inc. 2001. The United States pharmacopeia 25/ The national formulary 20 – 2002. United States Pharmacopeial Convention, Inc., Rockville, Md.
10. Clesceri, Greenberg and Eaton (ed.). 1998. Standard methods for the examination of water and wastewater, 20th ed. American Public Health Association, Washington, D.C.
11. U.S. Department of Agriculture. 1998. Microbiology laboratory guidebook, 3rd ed. Food Safety and Inspection Service, USDA, Washington, D.C.

Availability

BBL™ Polypeptone™ Peptone

AOAC BAM COMPF SMWW USDA USP

Cat. No. 211910 Dehydrated – 454 g
297108 Dehydrated – 10 kg

User Quality Control

Identity Specifications

BBL™ Polypeptone™ Peptone

Dehydrated Appearance:	Light to dark, yellow to tan, fine, homogeneous, free of extraneous material.
Solution:	2.0% solution, soluble in purified water. Solution is light to medium, yellow to tan, clear to slightly hazy.
Reaction of 2.0% Solution at 25°C:	pH 6.8-7.5

Cultural Response

BBL™ Polypeptone™ Peptone

Prepare a sterile solution of peptone agar without (plain) and with 5% sheep blood (SB) using 10 g of Polypeptone Peptone, 2.5 g of sodium chloride and 6.5 g of agar in 500 mL of purified water. Adjust final pH to 7.2-7.4. Inoculate and incubate plates at 35 ± 2°C for 2-3 days (incubate streptococci with 3-5% CO₂).

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY PLAIN	RECOVERY WITH SB
<i>Salmonella choleraesuis</i> subsp. <i>choleraesuis</i> serotype Typhi	19430	10 ³ -10 ⁴	Good	N/A
<i>Staphylococcus aureus</i>	6538P	10 ³ -10 ⁴	Good	N/A
<i>Streptococcus pneumoniae</i>	6305	10 ³ -10 ⁴	N/A	Good, alpha hemolysis
<i>Streptococcus pyogenes</i>	49117	10 ⁴ -10 ⁵	Good	Good, beta hemolysis

Potato Dextrose Agar • Potato Dextrose Broth

Intended Use

Potato Dextrose Agar conforms with specifications of *The United States Pharmacopeia (USP)*.

Potato Dextrose Agar is used for the cultivation and enumeration of yeasts and molds.

Potato Dextrose Broth is used for cultivating yeasts and molds.

Summary and Explanation

Potato Dextrose Agar is recommended by the American Public Health Association for plate counts of yeasts and molds in the examination of foods and dairy products.^{1,2} It is recommended in the *USP* for use in the performance of Microbial Limit Tests.³ It is also used for the stimulation of sporulation (slide preparations), maintenance of stock cultures

of certain dermatophytes and for differentiation of atypical varieties of dermatophytes by pigment production.⁴

Potato Dextrose Broth is a general-purpose broth medium for yeasts and molds (Potato Dextrose Agar without the agar).

Principles of the Procedure

Potato starch and dextrose support luxuriant growth of fungi. Lowering of the pH of the medium to approximately 3.5 with sterile tartaric acid achieves the inhibition of bacterial growth. It is important, however, to avoid heating the medium after it has been acidified because this action results in the hydrolysis of the agar and impairs its ability to solidify.

User Quality Control

Identity Specifications

Difco™ Potato Dextrose Agar

Dehydrated Appearance: Light beige, free-flowing, homogeneous.
 Solution: 3.9% solution, soluble in purified water upon boiling. Solution is light amber, slightly opalescent.
 Prepared Appearance: Light amber, slightly opalescent.
 Reaction of 3.9% Solution at 25°C: pH 5.6 ± 0.2

Difco™ Potato Dextrose Broth

Dehydrated Appearance: Light beige, free-flowing, homogeneous.
 Solution: 2.4% solution, soluble in purified water upon boiling. Solution is very, very light amber, clear to very slightly opalescent.
 Prepared Appearance: Very, very light amber, clear to very slightly opalescent.
 Reaction of 2.4% Solution at 25°C: pH 5.1 ± 0.2

Cultural Response

Difco™ Potato Dextrose Agar

Prepare the medium per label directions. Inoculate and incubate at 25-30°C for 18-48 hours (up to 7 days for *T. mentagrophytes*).

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Aspergillus niger</i>	16404	10 ³ -10 ⁴	Good
<i>Candida albicans</i>	10231	10 ³ -10 ⁴	Good
<i>Saccharomyces cerevisiae</i>	9763	10 ³ -10 ⁴	Good
<i>Trichophyton mentagrophytes</i>	9533	Undiluted	Good

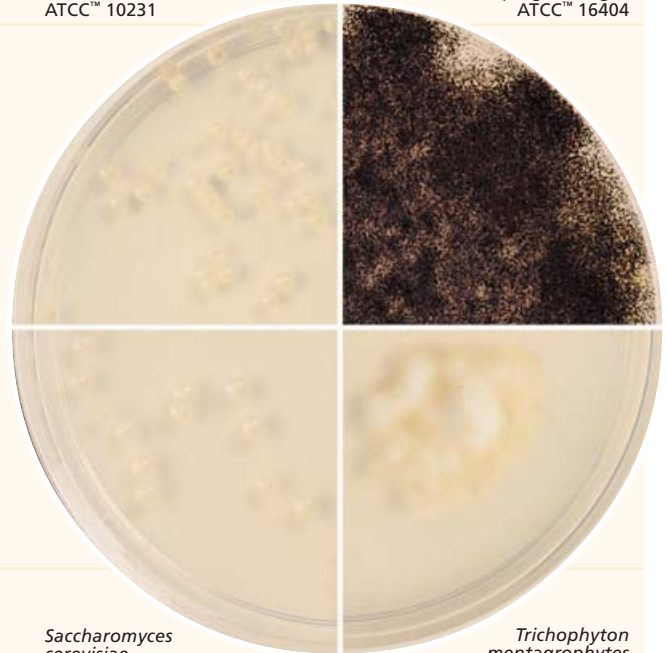
Difco™ Potato Dextrose Broth

Prepare the medium per label directions. Inoculate and incubate at 25 ± 2°C for 40-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Aspergillus niger</i>	16404	10 ² -10 ³	Good
<i>Candida albicans</i>	10231	10 ² -10 ³	Good
<i>Lactobacillus rhamnosus</i>	7469	10 ² -10 ³	Fair to good
<i>Saccharomyces cerevisiae</i>	9763	10 ² -10 ³	Good

Candida albicans
ATCC™ 10231

Aspergillus niger
ATCC™ 16404



Saccharomyces cerevisiae
ATCC™ 9763

Trichophyton mentagrophytes
ATCC™ 9533

Formulae

Difco™ Potato Dextrose Agar

Approximate Formula* Per Liter	
Potato Starch	4.0 g
Dextrose	20.0 g
Agar	15.0 g

Difco™ Potato Dextrose Broth

Consists of the same ingredients without the agar.

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

Directions for Preparation from Dehydrated Product

- Suspend the powder in 1 L of purified water:
 Difco™ Potato Dextrose Agar – 39 g;
 Difco™ Potato Dextrose Broth – 24 g.
 Mix thoroughly.
- Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
- Autoclave at 121°C for 15 minutes.

- To alter the reaction of the agar medium to pH 3.5, cool the base to 45-50°C and aseptically add an appropriate amount of sterile 10% tartaric acid to each liter of medium. Mix well. Do not reheat the medium.
- Test samples of the finished product for performance using stable, typical control cultures.

Procedure

Consult appropriate references for information concerning the processing and inoculation of specimens.^{1-3,5,6} Liquefy the medium in pour tubes by heating in boiling water. Cool to 45-50°C and pour into sterile Petri dishes. Allow to solidify for a minimum of 30 minutes.

Streak the specimen onto prepared media with a sterile inoculating loop to obtain isolated colonies. When used for determining yeast and mold counts, the medium should be adjusted to a pH of approximately 3.5 with sterile tartaric acid and used in the standard pour plate technique. Incubate the plates at 25-30°C in an inverted position (agar side up) with increased humidity.

Tubed slants are used primarily for the cultivation and maintenance of pure cultures. They should be inoculated with an inoculating loop and incubated under the same conditions as the plated medium.

For isolation of fungi from potentially contaminated specimens, a selective medium should be inoculated along with the nonselective medium. For isolation of fungi causing systemic mycoses, two sets of media should be inoculated, with one set incubated at 25-30°C and a duplicate set at 35 ± 2°C. All cultures should be examined at least weekly for fungal growth and should be held for 4-6 weeks before being reported as negative.

Inoculation of Potato Dextrose Broth with pure cultures of yeasts can assist in their identification.

Expected Results

After sufficient incubation, the plates, which were streak-inoculated, should show isolated colonies in streaked areas and confluent growth in areas of heavy inoculation. The colonies in pour plates should be counted and the results expressed as yeast and molds counts per gram or milliliter of material, taking into account the applicable dilution factor.

Growth from tubes inoculated with pure cultures may be used for biochemical and/or serological testing.

For broth, observe cultures for surface growth and pellicle formation.

Limitations of the Procedure

1. Heating Potato Dextrose Agar after acidifying hydrolyzes the agar and may destroy the solidifying properties.
2. Potato Dextrose Agar is not a differential medium. Perform microscopic examination and biochemical tests to identify isolates to genus and species if necessary.

References

1. Downes and Ito (ed.). 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
2. Marshall, (ed.). 1993. Standard methods for the examination of dairy products, 16th ed. American Public Health Association, Washington, D.C.
3. United States Pharmacopeial Convention, Inc. 2001. The United States pharmacopeia 25/The national formulary 20 – 2002. United States Pharmacopeial Convention, Inc., Rockville, Md.
4. MacFaddin. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, Md.
5. Murray, Baron, Pfaller, Tenover and Tenover (ed.). 1999. Manual of clinical microbiology, 7th ed. American Society for Microbiology, Washington, D.C.
6. Isenberg (ed.). 1992. Clinical microbiology procedures handbook, vol. 1. American Society for Microbiology, Washington, D.C.

Availability

Difco™ Potato Dextrose Agar

	AOAC	BAM	BS10	CCAM	CMPH	COMPF	MCM7	SMD	USP
Cat. No. 213300									
213400									
213200									

BBL™ Potato Dextrose Agar

	AOAC	BAM	BS10	CCAM	CMPH	COMPF	MCM7	SMD	USP
<i>United States and Canada</i>									
Cat. No. 296272									
297945									
221002									
297241									
299906									

Japan

Cat. No. 251545									
251821									
251544									

Difco™ Potato Dextrose Broth

Cat. No. 254920									
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*Store at 2-8°C.

Potato Flakes Agar • Potato Flakes CC Agar Potato Flakes Agar with Chloramphenicol and Gentamicin

Intended Use

These media are used in qualitative procedures for the cultivation of pathogenic and opportunistic fungi encountered in clinical mycology.

Summary and Explanation

Potato Flakes Agar induces sporulation, enhancing the production of morphological structures required for the identification of many pathogenic and opportunistic fungi.¹ The addition of chloramphenicol and cycloheximide (CC) or gentamicin provides selectivity for more effective isolation and identification of medically significant fungi.

Principles of the Procedure

The medium stimulates the production of morphological features, such as conidia configurations, improving the ability to identify fungi by their particular morphological structures. Correct identification of fungi causing human disease depends upon visualization of characteristic morphological features.

The antimicrobial agents chloramphenicol, cycloheximide and gentamicin are incorporated in various combinations to improve the recovery of pathogenic fungi from specimens heavily contaminated with bacteria and saprophytic fungi.² Chloramphenicol is a broad-spectrum antibiotic that inhibits a wide range of gram-positive and gram-negative bacteria. Cycloheximide is an anti-fungal agent that inhibits saprophytic