

Antibiotic Assay Media

Antibiotic Medium 1 • Antibiotic Medium 2

Antibiotic Medium 3 • Antibiotic Medium 4

Antibiotic Medium 5 • Antibiotic Medium 8

Antibiotic Medium 9 • Antibiotic Medium 10

Antibiotic Medium 11 • Antibiotic Medium 12

Antibiotic Medium 13 • Antibiotic Medium 19

Intended Use

Antibiotic Assay Media are used for determining antibiotic potency by the microbiological assay technique.¹⁻³

These media, where noted, meet *United States Pharmacopeia (USP)* performance specifications.

Summary and Explanation

The activity (potency) of an antibiotic can be demonstrated under suitable conditions by its inhibitory effect on microorganisms.² Reduction in antimicrobial activity may reveal changes not demonstrated by chemical methods.² Antibiotic assays are performed by the cylinder plate method and the turbidimetric “tube” assay. The cylinder plate method, first described by Abraham et al.⁴ for the assay of penicillin, was later modified by Foster and Woodruff⁵ and by Schmidt and Moyer.⁶

Antibiotic assay media are prepared according to the specifications of the *USP*², *European Pharmacopeia*⁷ and *AOAC International*.³ The antibiotic media are identified numerically with names assigned by Grove and Randall in *Assay Methods of Antibiotics*.¹ Antibiotic Medium 19 corresponds to the use described in *Outline of Details for Official Microbiological Assays of Antibiotics*.⁸

Antibiotic Medium 12 is prepared from the Grove and Randall formula.¹ They recommended its use for preparing test plates for the cylinder plate assay of the antifungal agents, nystatin and anisomycin, using only a seed layer containing *Saccharomyces cerevisiae* as the test organism. It is used for the assay of amphotericin B.

Antibiotic Medium 1 and Antibiotic Medium 4 are used in a cylinder plate method for detecting penicillin in nonfat dry milk.⁹

The use of standardized culture media and careful control of all test conditions are fundamental requisites in the microbiological assay of antibiotics in order to achieve satisfactory test results.

Principles of the Procedure

Cylinder Plate Assay

This method is based on the diffusion of an antibiotic solution from a cylinder placed on the surface of an inoculated agar medium. The diameter of a zone of inhibition after incubation depends, in part, on the concentration or activity of the antibiotic. This method is used in the assay of commercial preparations of antibiotics, as well as in the quantitative determination of antibiotics in body fluids, animal feeds and other materials.

Turbidimetric Assay

The turbidimetric method is based on the inhibition of growth of a microbial culture in a fluid medium containing a uniform solution of an antibiotic.² Turbidimetric determinations have the advantage of requiring a short incubation period, providing test results after 3 or 4 hours. However, the presence of solvents or other inhibitory materials may influence turbidimetric assays more markedly than cylinder plate assays. Use of this method is appropriate only when test samples are clear.

Formulae

Difco™ Antibiotic Medium 1 (Penassay Seed Agar)

Approximate Formula* Per Liter	
Beef Extract.....	1.5 g
Yeast Extract.....	3.0 g
Pancreatic Digest of Casein.....	4.0 g
Peptone.....	6.0 g
Dextrose.....	1.0 g
Agar.....	15.0 g

Difco™ Antibiotic Medium 2 (Penassay Base Agar)

Approximate Formula* Per Liter	
Beef Extract.....	1.5 g
Yeast Extract.....	3.0 g
Peptone.....	6.0 g
Agar.....	15.0 g

Difco™ Antibiotic Medium 3 (Penassay Broth)

Approximate Formula* Per Liter	
Beef Extract.....	1.5 g
Yeast Extract.....	1.5 g
Peptone.....	5.0 g
Dextrose.....	1.0 g
Sodium Chloride.....	3.5 g
Dipotassium Phosphate.....	3.68 g
Monopotassium Phosphate.....	1.32 g

Also Known As

DIFCO™ BRAND PRODUCT NAME	ALTERNATIVE DIFCO™ NAME	BBL™ BRAND PRODUCT NAME	USP ²	AOAC ³
Antibiotic Medium 1	Penassay Seed Agar	–	Medium 1	Agar Medium A
Antibiotic Medium 2	Penassay Base Agar	–	Medium 2	Agar Medium C
Antibiotic Medium 3	Penassay Broth	Antibiotic Assay Broth	Medium 3	Broth Medium A
Antibiotic Medium 4	Yeast Beef Agar	–	Medium 4	Agar Medium B
Antibiotic Medium 5	Streptomycin Assay Agar	–	Medium 5	Agar Medium E
Antibiotic Medium 8	–	–	Medium 8	Agar Medium D
Antibiotic Medium 9	Polymyxin Base Agar	–	Medium 9	–
Antibiotic Medium 10	Polymyxin Seed Agar	–	Medium 10	–
Antibiotic Medium 11	Neomycin Assay Agar	–	Medium 11	Agar Medium J
Antibiotic Medium 12	–	–	–	–
–	–	Sabouraud Liquid Broth, Modified	Medium 13	Broth Medium B
Antibiotic Medium 19	–	–	Medium 19	–

User Quality Control

Identity Specifications

	DEHYDRATED APPEARANCE	SOLUTION	PREPARED APPEARANCE	PH AT 25° C
Difco™ Antibiotic Medium 1	Beige, homogeneous, free-flowing.	3.05% solution, soluble in purified water upon boiling. Solution is light to medium amber, slightly opalescent.	Light to medium amber, slightly opalescent.	6.55 ± 0.05
Difco™ Antibiotic Medium 2	Light tan, homogeneous, free-flowing.	2.55% solution, soluble in purified water upon boiling. Solution is light to medium amber, very slightly to slightly opalescent.	Light-medium amber, slightly opalescent.	6.55 ± 0.05
Difco™ Antibiotic Medium 3	Tan, free-flowing, homogeneous.	1.75% solution, soluble in purified water upon boiling. Solution is light to medium amber, clear.	Light to medium amber, clear.	7.0 ± 0.05
BBL™ Antibiotic Assay Broth (Antibiotic Medium 3)	Fine, homogeneous, free of extraneous material.	1.75% solution, soluble in purified water upon boiling. Solution is very pale to light, yellow to tan, clear to slightly hazy.	Pale to light, yellow to tan, clear to slightly hazy.	7.0 ± 0.2
Difco™ Antibiotic Medium 4	Light tan, free-flowing, homogeneous.	2.65% solution, soluble in purified water upon boiling. Solution is light amber, very slightly opalescent.	Light amber, very slightly to slightly opalescent.	6.55 ± 0.05
Difco™ Antibiotic Medium 5	Light tan, free-flowing, homogeneous.	2.55% solution, soluble in purified water upon boiling. Solution is light to medium amber, very slightly to slightly opalescent.	Light to medium amber, slightly opalescent.	7.9 ± 0.1
Difco™ Antibiotic Medium 8	Light tan, free-flowing, homogeneous.	2.55% solution, soluble in purified water upon boiling. Solution is light to medium amber, very slightly to slightly opalescent.	Light to medium amber, slightly opalescent.	5.85 ± 0.05
Difco™ Antibiotic Medium 9	Light beige, free-flowing, homogeneous.	5.0% solution, soluble in purified water upon boiling. Solution is light to medium amber, slightly opalescent, may have a slight flocculent precipitate.	Light to medium amber, slightly opalescent with slight flocculent precipitate.	7.25 ± 0.05
Difco™ Antibiotic Medium 10	Beige, homogeneous, moist with a tendency to clump.	5.2% solution, soluble in purified water upon boiling. Solution is light to medium amber, very slightly to slightly opalescent.	Light to medium amber, slightly opalescent.	7.25 ± 0.05
Difco™ Antibiotic Medium 11	Beige, homogeneous, free-flowing.	3.05% solution, soluble in purified water upon boiling. Solution is light to medium amber, very slightly to slightly opalescent.	Light to medium amber, slightly opalescent.	7.95 ± 0.05
Difco™ Antibiotic Medium 12	Tan, homogeneous, free-flowing.	6.25% solution, soluble in purified water upon boiling. Solution is light to medium amber, very slightly to slightly opalescent.	Light to medium amber, slightly opalescent.	6.1 ± 0.1
BBL™ Sabouraud Liquid Broth, Modified (Antibiotic Medium 13)	Fine, homogeneous, free of extraneous material.	3.0% solution, soluble in purified water upon boiling. Solution is light to medium, yellow to tan, clear to slightly hazy.	Light to medium, yellow to tan, clear to slightly hazy.	5.7 ± 0.1
Difco™ Antibiotic Medium 19	Light tan, homogeneous, free-flowing.	6.0% solution, soluble in purified water upon boiling. Solution is medium amber, very slightly to slightly opalescent.	Medium amber, slightly opalescent.	6.1 ± 0.1

Cultural Response

Difco™ Antibiotic Medium 1 Difco™ Antibiotic Medium 2

Prepare the medium per label directions. Inoculate by the pour plate method and incubate at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Staphylococcus aureus</i>	6538P	30-300	Good

Difco™ Antibiotic Medium 3

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for up to 24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Enterococcus faecium</i>	10541	~10 ⁷	Good
<i>Escherichia coli</i>	10536	~10 ⁷	Good
<i>Klebsiella pneumoniae</i>	10031	~10 ⁷	Good
<i>Staphylococcus aureus</i>	6538P	~10 ⁷	Good

BBL™ Antibiotic Assay Broth (Antibiotic Medium 3)

Prepare the medium per label directions. Inoculate and incubate at 25 ± 2°C for the *Saccharomyces cerevisiae* and 35 ± 2°C for the remaining organisms for 7 days.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Bacillus subtilis</i>	6633	≤ 10 ³	Good
<i>Escherichia coli</i>	10536	≤ 10 ³	Good
<i>Kocuria rhizophila</i>	9341	≤ 10 ³	Good
<i>Saccharomyces cerevisiae</i>	9763	≤ 10 ³	Good
<i>Staphylococcus aureus</i>	6538P	≤ 10 ³	Good

Difco™ Antibiotic Medium 4

Prepare the medium per label directions. Inoculate by the pour plate method and incubate at 35 ± 2°C for 40-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Kocuria rhizophila</i>	9341	30-300	Good

Antibiotic Medium 5 Antibiotic Medium 8

Prepare the medium per label directions. Inoculate by the pour plate method and incubate at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Bacillus subtilis</i>	6633	30-300	Good

Antibiotic Medium 9 Antibiotic Medium 10

Prepare the medium per label directions. Inoculate by the pour plate method and incubate at 35 ± 2°C for 40-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Bordetella bronchiseptica</i>	4617	30-300	Good

Antibiotic Medium 11

Prepare the medium per label directions. Inoculate by the pour plate method and incubate at 35 ± 2°C for 18-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Kocuria rhizophila</i>	9341	30-300	Good
<i>Staphylococcus epidermidis</i>	12228	30-300	Good

Antibiotic Medium 12 Antibiotic Medium 19

Prepare the medium per label directions. Inoculate by the pour plate method and incubate at 30 ± 2°C for 40-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Saccharomyces cerevisiae</i>	2601	30-300	Good

BBL™ Sabouraud Liquid Broth, Modified (Antibiotic Medium 13)

Prepare the medium per label directions. Inoculate and incubate at 25 ± 2°C for 7 days (use one loopful of a fresh 3-7 day culture for *A. brasiliensis* and *T. mentagrophytes*).

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Aspergillus brasiliensis (niger)</i>	16404	Undiluted	Good
<i>Candida albicans</i>	60193	≤ 10 ³	Good
<i>Saccharomyces cerevisiae</i>	9763	≤ 10 ³	Good
<i>Trichophyton mentagrophytes</i>	9533	Undiluted	Good

BBL™ Antibiotic Assay Broth (Antibiotic Medium 3)

Approximate Formula* Per Liter	
Beef Extract.....	1.5 g
Yeast Extract.....	1.5 g
Pancreatic Digest of Gelatin.....	5.0 g
Dextrose.....	1.0 g
Sodium Chloride.....	3.5 g
Dipotassium Phosphate.....	3.68 g
Monopotassium Phosphate.....	1.32 g

Difco™ Antibiotic Medium 4 (Yeast Beef Agar)

Approximate Formula* Per Liter	
Beef Extract.....	1.5 g
Yeast Extract.....	3.0 g
Peptone.....	6.0 g
Dextrose.....	1.0 g
Agar.....	15.0 g

Difco™ Antibiotic Medium 5 (Streptomycin Assay Agar)

Same as Medium 2, except for the final pH after autoclaving.

Difco™ Antibiotic Medium 8

Same as Medium 2, except for the final pH after autoclaving.

Difco™ Antibiotic Medium 9 (Polymyxin Base Agar)

Approximate Formula* Per Liter	
Pancreatic Digest of Casein.....	17.0 g
Soy Peptone.....	3.0 g
Dextrose.....	2.5 g
Sodium Chloride.....	5.0 g
Dipotassium Phosphate.....	2.5 g
Agar.....	20.0 g

Difco™ Antibiotic Medium 10 (Polymyxin Seed Agar)

Approximate Formula* Per Liter	
Pancreatic Digest of Casein.....	17.0 g
Soybean Peptone.....	3.0 g
Dextrose.....	2.5 g
Sodium Chloride.....	5.0 g
Dipotassium Phosphate.....	2.5 g
Agar.....	12.0 g
Polysorbate 80.....	10.0 g

Difco™ Antibiotic Medium 11 (Neomycin Assay Agar)

Same as Medium 1, except for the final pH after autoclaving.

Selection of Media for the Microbiological Assay of Antibiotics²

ANTIBIOTIC	ASSAY METHOD	ORGANISM	ATCC™	INOCULUM MEDIUM	CYLINDER PLATE BASE LAYER MEDIUM	CYLINDER PLATE SEED LAYER MEDIUM	TURBIDIMETRIC ASSAY MEDIUM
Amikacin	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Amphotericin B	Cylinder Plate	<i>Saccharomyces cerevisiae</i>	9763	19		19	
Bacitracin	Cylinder Plate	<i>Micrococcus luteus</i>	10240	1	2	1	
Candidicin	Turbidimetric	<i>Saccharomyces cerevisiae</i>	9763	19	2	13	
Capreomycin	Turbidimetric	<i>Klebsiella pneumoniae</i>	10031	1			3
Carbenicillin	Cylinder Plate	<i>Pseudomonas aeruginosa</i>	25619	1	9	10	
Cephalothin	Cylinder Plate	<i>Staphylococcus aureus</i>	29737	1	2	1	
Cephapirin	Cylinder Plate	<i>Staphylococcus aureus</i>	29737	1	2	1	
Chloramphenicol	Turbidimetric	<i>Escherichia coli</i>	10536	1			3
Chlortetracycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Cloxacillin	Cylinder Plate	<i>Staphylococcus aureus</i>	29737	1	2	1	
Colistimethate, sodium	Cylinder Plate	<i>Bordetella bronchiseptica</i>	4617	1	9	10	
Colistin	Cylinder Plate	<i>Bordetella bronchiseptica</i>	4617	1	9	10	
Cycloserine	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Demeclocycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Dihydrostreptomycin	Cylinder Plate	<i>Bacillus subtilis</i>	6633	32*	5	5	
Dihydrostreptomycin	Turbidimetric	<i>Klebsiella pneumoniae</i>	10031	1			3
Doxycycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Erythromycin	Cylinder Plate	<i>Kocuria rhizophila</i>	9341	1	11	11	
Gentamicin	Cylinder Plate	<i>Staphylococcus epidermidis</i>	12228	1	11	11	
Gramicidin	Turbidimetric	<i>Enterococcus hirae</i>	10541	3			3
Kanamycin	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Methacycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Nafcillin	Cylinder Plate	<i>Staphylococcus aureus</i>	29737	1	2	1	
Neomycin	Cylinder Plate	<i>Staphylococcus epidermidis</i>	12228	1	11	11	
Neomycin	Turbidimetric	<i>Klebsiella pneumoniae</i>	10031	1			39**
Netilmicin	Cylinder Plate	<i>Staphylococcus epidermidis</i>	12228	1	11	11	
Novobiocin	Cylinder Plate	<i>Staphylococcus epidermidis</i>	12228	1	2	1	
Nystatin	Cylinder Plate	<i>Saccharomyces cerevisiae</i>	2601	19		19	
Oxytetracycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Paromomycin	Cylinder Plate	<i>Staphylococcus epidermidis</i>	12228	1	11	11	
Penicillin G	Cylinder Plate	<i>Staphylococcus aureus</i>	29737	1	2	1	
Polymyxin B	Cylinder Plate	<i>Bordetella bronchiseptica</i>	4617	1	9	10	
Rolitetracycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Sisomicin	Cylinder Plate	<i>Staphylococcus epidermidis</i>	12228	1	11	11	
Streptomycin	Turbidimetric	<i>Klebsiella pneumoniae</i>	10031	1			3
Tetracycline	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Tobramycin	Turbidimetric	<i>Staphylococcus aureus</i>	29737	1			3
Troleandomycin	Turbidimetric	<i>Klebsiella pneumoniae</i>	10031	1			3
Tylosin	Turbidimetric	<i>Staphylococcus aureus</i>	9144	3			39**
Vancomycin	Cylinder Plate	<i>Bacillus subtilis</i>	6633	32*	8	8	

* Same as Medium 1, except for the additional ingredient of 300 mg of manganese sulfate.

** Same as Medium 3, except that the final pH after autoclaving is 7.9 ± 0.1.

Difco™ Antibiotic Medium 12

Approximate Formula* Per Liter

Beef Extract.....	2.5	g
Yeast Extract.....	5.0	g
Peptone.....	10.0	g
Dextrose.....	10.0	g
Sodium Chloride.....	10.0	g
Agar.....	25.0	g

BBL™ Sabouraud Liquid Broth, Modified (Antibiotic Medium 13)

Approximate Formula* Per Liter

Pancreatic Digest of Casein.....	5.0	g
Peptic Digest of Animal Tissue.....	5.0	g
Dextrose.....	20.0	g

Difco™ Antibiotic Medium 19

Approximate Formula* Per Liter	
Beef Extract.....	2.4 g
Yeast Extract.....	4.7 g
Peptone.....	9.4 g
Dextrose.....	10.0 g
Sodium Chloride.....	10.0 g
Agar.....	23.5 g

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

Directions for Preparation from Dehydrated Product

1. Suspend the powder in 1 L of purified water:
 - Difco™ Antibiotic Medium 1 – 30.5 g;
 - Difco™ Antibiotic Medium 2 – 25.5 g;
 - Difco™ Antibiotic Medium 3 – 17.5 g;
 - BBL™ Antibiotic Assay Broth (Antibiotic Medium 3) – 17.5 g;
 - Difco™ Antibiotic Medium 4 – 26.5 g;
 - Difco™ Antibiotic Medium 5 – 25.5 g;
 - Difco™ Antibiotic Medium 8 – 25.5 g;
 - Difco™ Antibiotic Medium 9 – 50 g;
 - Difco™ Antibiotic Medium 10 – 52 g;
 - Difco™ Antibiotic Medium 11 – 30.5 g;
 - Difco™ Antibiotic Medium 12 – 62.5 g;
 - BBL™ Sabourand Liquid Broth, Modified (Antibiotic Medium 13) – 30 g;
 - Difco™ Antibiotic Medium 19 – 60 g.Mix thoroughly.
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Autoclave at 121°C for 15 minutes.
4. To raise the pH of Antibiotic Medium 11 to 8.3 ± 0.1 , cool the base to 45-50°C and add NaOH.
5. Test samples of the finished product for performance using stable, typical control cultures.

Procedure

Test Organism Preparation

Maintain stock cultures on agar slants and make transfers at 1- or 2-week intervals. Prepare the inoculum for assay by washing growth from a fresh 24-48 hour agar slant using sterile purified water, saline or Antibiotic Medium 3 and further dilute the culture to obtain the desired organism concentration. In some turbidimetric assays, an 18- to 24-hour culture of the test organism in Antibiotic Medium 3, diluted to obtain the optimal number of organisms, is used.

When *Bacillus subtilis* is used as the test organism, inoculate it on Antibiotic Medium 1 and incubate at 37°C for 1 week, wash spores from the agar surface, and heat the spores at 56°C for 30 minutes. Wash the spores three times in purified water, heat again at 65°C for 30 minutes, and then dilute to the optimal concentration. This inoculum preparation should produce a sharp zone in the assay.

Antibiotic Medium 1 modified by the addition of 300 mg manganese sulfate ($MnSO_4 \cdot H_2O$) per liter often aids the

sporulation of *B. subtilis* and may be used in preparing the spore suspension.

When *B. cereus* var. *mycoides* is required, inoculate the organism on Antibiotic Medium 1 and incubate at 30°C for 1 week. Wash and prepare the spores as for *B. subtilis*, above.

Cylinder Plate Assay

Use 20 × 100 mm glass or plastic Petri dishes with sufficient depth so that cylinders used in the assay will not be pushed into the medium by the cover.

Use stainless steel or porcelain assay cylinders having the following dimensions (± 0.1 mm): 8 mm outside diameter, 6 mm inside diameter and 10 mm long.² Carefully clean the cylinders to remove all residues, using an occasional acid bath (i.e., with approximately 2N nitric acid or with chromic acid).² Four or six cylinders are generally used per plate, evenly spaced on a 2.8 cm radius.

To assure accurate assays, work on a level surface to obtain uniformly thick base and seed layers in the Petri dish. Allow the base layer to solidify and then overlay the seed layer containing a proper concentration of the test organism. The amount of medium in the layers varies for different antibiotics, with most assays specifying a 21 mL base layer and a 4 mL seed layer. In any case, dishes with flat bottoms are required to assure complete coverage of the bottom of the dish when small amounts of base medium are used. Tilt the plate to obtain even coverage of the base layer by the seed layer and allow it to solidify in a level position. Plates should be used the same day as prepared.

Turbidimetric Assay

Use glass or plastic test tubes (i.e., 16 × 125 mm or 18 × 150 mm) that are relatively uniform in length, diameter and thickness and substantially free from surface blemishes.² Tubes that will be placed in the spectrophotometer should be matched and free of scratches or blemishes.² Clean the tubes thoroughly to remove all antibiotic residues and traces of cleaning solution and, prior to subsequent use, sterilize tubes that have been previously used.²

Prepare working dilutions of the antibiotic reference standards in specific concentrations. To a 1 mL quantity of each solution in a suitable tube, add 9 mL of inoculated broth, as required. Prepare similar solutions of the assay materials containing approximately the same amounts of antibiotic activity and place in tubes. Incubate the tubes for 3-4 hours at the required temperature, generally in a water bath. At the end of the incubation period, stop growth by adding 0.5 mL of 1:3 formalin. Determine the amount of growth by measuring light transmittance with a suitable spectrophotometer. Determine the concentration of the antibiotic by comparing the growth obtained with that given by reference standard solutions.

For a complete discussion of antibiotic assay methods, refer to appropriate procedures outlined in the references.^{2,3,7}

Expected Results

Refer to appropriate procedures for results.^{2,3,7}

References

1. Grove and Randall. 1955. Assay methods of antibiotics. Medical Encyclopedia, Inc. New York, N.Y.
2. United States Pharmacopeial Convention, Inc. 2008. The United States pharmacopeia 31/The national formulary 26, Supp. 1, 8-1-08, online. United States Pharmacopeial Convention, Inc., Rockville, Md.
3. Horwitz (ed.). 2007. Official methods of analysis of AOAC International, 18th ed., online. AOAC International, Gaithersburg, Md.
4. Abraham, Chain, Fletcher, Florey, Gardner, Heatley and Jennings. 1941. *Lancet* *iii*:177.
5. Foster and Woodruff. 1943. *J. Bacteriol.* 46:187.
6. Schmidt and Moyer. 1944. *J. Bacteriol.* 47:199.
7. Council of Europe. 2002. European pharmacopeia, 4th ed. Council of Europe, Strasbourg, France.
8. Kirshbaum and Arret. 1967. *J. Pharm. Sci.* 56:512.
9. Wehr and Frank (ed.). 2004. Standard methods for the examination of dairy products, 17th ed. American Public Health Association, Washington, D.C.

Availability

Difco™ Antibiotic Medium 1

AOAC EP USP

Cat. No. 226340 Dehydrated – 500 g

Difco™ Antibiotic Medium 2

AOAC USP

Cat. No. 227020 Dehydrated – 500 g

Difco™ Antibiotic Medium 3

AOAC EP USP

Cat. No. 224320 Dehydrated – 500 g
224310 Dehydrated – 2 kg

BBL™ Antibiotic Assay Broth (Antibiotic Medium 3)

AOAC EP USP

Cat. No. 210932 Dehydrated – 500 g

Difco™ Antibiotic Medium 4

AOAC USDA USP

Cat. No. 224410 Dehydrated – 500 g

Difco™ Antibiotic Medium 5

AOAC USDA USP

Cat. No. 227710 Dehydrated – 500 g

Difco™ Antibiotic Medium 8

AOAC USDA USP

Cat. No. 266710 Dehydrated – 500 g

Difco™ Antibiotic Medium 9

EP USP

Cat. No. 246210 Dehydrated – 500 g

Difco™ Antibiotic Medium 10

EP USP

Cat. No. 246310 Dehydrated – 500 g*

Difco™ Antibiotic Medium 11

AOAC USDA USP

Cat. No. 259310 Dehydrated – 500 g

Difco™ Antibiotic Medium 12

Cat. No. 266910 Dehydrated – 500 g

BBL™ Sabouraud Liquid Broth Modified (Antibiotic Medium 13)

AOAC USP

Cat. No. 210986 Dehydrated – 500 g
221014 Prepared Tubes (K Tubes) – Pkg. of 10

Difco™ Antibiotic Medium 19

EP USP

Cat. No. 243100 Dehydrated – 500 g

Europe

Cat. No. 254655 Prepared Bottles, 250 mL – Pkg. of 10

*Store at 2-8°C.