

## References

1. Pine and Watson. 1959. J. Lab. Clin. Med. 54:107.
2. Ajello, Georg, Kaplan and Kaufman. 1963. CDC laboratory manual for medical mycology. PHS Publication No. 994. U.S. Government Printing Office, Washington, D.C.

## Availability

### BBL™ Actinomycetes Broth

Cat. No. 210920 Dehydrated – 500 g

# Actinomycete Isolation Agar Glycerol

## Intended Use

Actinomycete Isolation Agar is used with added glycerol for isolating and cultivating actinomycetes from soil and water.

Glycerol is used in preparing microbiological culture media.

## Summary and Explanation

Although some genera are important to human medicine, most of the actinomycetes are part of the indigenous flora of soil, water and vegetation. Actinomycetes may impart a musty odor to water or a muddy flavor to fish.<sup>1</sup> Actinomycetes can cause massive growths which will form a thick foam in the activated sludge process, causing a disruption in wastewater treatment.<sup>2,3</sup> Actinomycetes are gram-positive, acid-fast cells, growing as filaments that may branch and may form irregularly shaped rods and cocci.

Olsen<sup>4</sup> formulated Actinomycete Isolation Agar for isolating and cultivating actinomycetes from soil and water. The formula is supplemented with glycerol, a highly purified fermentable alcohol used occasionally for differentiating certain bacteria and in media for isolating and culturing fastidious bacteria.

## Principles of the Procedure

Actinomycete Isolation Agar contains sodium caseinate which is a source of nitrogen. Asparagine is an amino acid and a

source of organic nitrogen. Sodium propionate is a substrate used in anaerobic fermentation. Dipotassium phosphate provides buffering capability to maintain pH balance. Magnesium sulfate and ferrous sulfate provide sources of sulfates and metallic ions. Agar is the solidifying agent. The added glycerol is a source of carbon.

## Formulae

### Difco™ Actinomycete Isolation Agar

Approximate Formula\* Per Liter

Sodium Caseinate .....	2.0	g
Asparagine .....	0.1	g
Sodium Propionate .....	4.0	g
Dipotassium Phosphate .....	0.5	g
Magnesium Sulfate .....	0.1	g
Ferrous Sulfate .....	1.0	mg
Agar .....	15.0	g

### Difco™ Glycerol

Glycerin

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

## Directions for Preparation from Dehydrated Product

1. Suspend 22 g of the powder in 1 L of purified water. Mix thoroughly.
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Add 5 g of Glycerol.
4. Autoclave at 121°C for 15 minutes.
5. Test samples of the finished product for performance using stable, typical control cultures.

## Procedure

Inoculate medium and incubate at 30°C for up to 72 hours.

## Expected Results

Refer to appropriate references and procedures for results.

## References

1. Clesceri, Greenberg and Eaton (ed.). 1998. Standard methods for the examination of water and wastewater, 20th ed. American Public Health Association, Washington, D.C.
2. Lechevalier. 1975. Actinomycetes of sewage-treatment plants. Environ. Protection Technol. Ser., EPA-600/2-75-031, U. S. Environmental Protection Agency, Cincinnati, Ohio.
3. Lechevalier and Lechevalier. 1974. Int. J. Syst. Bacteriol. 24:278.
4. Olsen. 1960. Personal communication.

## Availability

### Difco™ Actinomycete Isolation Agar

Cat. No. 212168 Dehydrated – 500 g

### Difco™ Glycerol

Cat. No. 228210 Bottle – 100 g  
228220 Bottle – 500 g

## User Quality Control

### Identity Specifications

#### Difco™ Actinomycete Isolation Agar

Dehydrated Appearance: Light beige, free-flowing, homogeneous.

Solution: 2.2% solution, soluble in purified water upon boiling with 0.5% Glycerol. Solution is light to medium amber, opalescent to opaque with precipitation.

Prepared Appearance: Medium amber, opalescent.

Reaction of 2.2% Solution with 0.5% Glycerol at 25°C: pH 8.1 ± 0.2

### Cultural Response

#### Difco™ Actinomycete Isolation Agar

Prepare the medium per label directions. Inoculate and incubate at 30 ± 2°C for up to 72 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Streptomyces achromogenes</i>	12767	10 <sup>2</sup> -10 <sup>3</sup>	Good
<i>Streptomyces albus</i>	3004	10 <sup>2</sup> -10 <sup>3</sup>	Good
<i>Streptomyces lavendulae</i>	8664	10 <sup>2</sup> -10 <sup>3</sup>	Good