Standard Methods Agar with Lecithin and Polysorbate 80

Intended Use

Standard Methods Agar with Lecithin and Polysorbate 80 is recommended for the detection and enumeration of microorganisms present on surfaces of sanitary importance.

Summary and Explanation

Standard Methods Agar with the neutralizers, lecithin and polysorbate 80, is formulated according to recommendations of the American Public Health Association.[1,2] It is primarily used in **RODAC**™ (Replicate Organism Detection and Counting) and contact plates for the enumeration of microorganisms on flat impervious surfaces. For this purpose the plates must be prepared carefully to ensure the presence of a meniscus of agar extending above the top of the poured plate. This requires approximately 17.0 mL of sterile medium per **RODAC** or contact plate.

The presence and number of microorganisms on a surface is determined by the appearance of colonies on the surface of the medium following application to the test surface.[3] Collection of "samples" from identical areas before and after treatment with disinfectant yields data useful in evaluating cleaning procedures in environmental sanitation.

User Quality Control

Identity Specifications

BBL™ Standard Methods Agar with Lecithin and Polysorbate 80

Dehydrated Appearance:	Medium fine, softly lumped powder "brown sugar appearance," free of extraneous material.
Solution:	2.92% solution, soluble in purified water upon boiling. Solution is light, yellow to tan, slightly to moderately hazy.
Prepared Appearance:	Light, yellow to tan, slightly to moderately hazy.
Reaction of 2.92% Solution at 25°C:	pH 7.0 ± 0.2

Cultural Response

BBL™ Standard Methods Agar with Lecithin and Polysorbate 80

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 42-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	APPEARANCE
Pseudomonas aeruginosa	10145	10^3-10^4	Good	Yellow to green pigment
Staphylococcus aureus	25923	10^3-10^4	Good	Cream to gold colonies

Principles of the Procedure

Casein peptone, yeast extract and dextrose are sources of nutrients required for the replication of microorganisms. The peptone provides nitrogenous compounds, including essential amino acids. Yeast extract is a rich source of B-complex vitamins. Dextrose is an energy source.

Lecithin and polysorbate 80, two commonly used neutralizers, are reported to inactivate residual disinfectants where the samples are being collected. Lecithin is incorporated to neutralize quaternary ammonium compounds, and polysorbate 80 is used to neutralize substituted phenolic disinfectants.[3-5]

Formula

BBL™ Standard Methods Agar with Lecithin and Polysorbate 80

Approximate Formula* Per Liter		
Pancreatic Digest of Casein	5.0	g
Yeast Extract	2.5	g
Dextrose	1.0	g
Agar	15.0	g
Lecithin	0.7	g
Polysorbate 80	5.0	g

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

Directions for Preparation from Dehydrated Product

1. Suspend 29.2 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. Autoclave at 121°C for 15 minutes. Cool to approximately 45°C.
4. In **RODAC** or Contact plates, use 16.5-17.5 mL per plate.
5. Test samples of the finished product for performance using stable, typical control cultures.

NOTE: The dehydrated medium has a characteristic "brown sugar" appearance and may seem moist.

Procedure

Liquefy the tubed medium in boiling water. Cool to 45-50°C and carefully pour in sterile **RODAC** plates. The agar in these plates after hardening should form a meniscus above the sides of the plates.

For use in the sampling of surfaces, remove the top of the plate. Apply the agar surface to a flat surface, pressing down gently but firmly and making certain that the entire agar meniscus touches the surface. Use a rolling uniform pressure on the back of the plate to effect contact. Lift the plate straight up from the surface, being careful not to allow it to slide along the surface. Replace the top of the plate. Incubate plates with the agar side up at 32°C for 24-48 hours depending upon whether contamination is heavy or light.[1,2]

Expected Results

After incubation, count the colonies and record as either number of colonies per **RODAC** plate or number of colonies per cm^2.[1,2] Subculture those colonies which are of interest so that positive identification can be made by means of biochemical testing and/or microscopic examination of organism smears.

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. Wehr and Frank (ed.). 2004. Standard methods for the examination of dairy products, 17th ed. American Public Health Association, Washington, D.C.
3. McGowan. 1985. *In* Lennette, Balows, Hausler and Shadomy (ed.), Manual of clinical microbiology, 4th ed. American Society for Microbiology, Washington, D.C.
4. Quisno, Gibby and Foter. 1946. Am. J. Pharm. *118*: 320.
5. Erlandson and Lawrence. 1953. Science *118*: 274.

Availability

BBL™ Standard Methods Agar with Lecithin and Polysorbate 80

COMPF SMD

Cat. No.	211643	Dehydrated – 500 g*
	221939	Prepared Contact Plates – Pkg. of 20*
	221032	Prepared Pour Tubes, 18 mL – Pkg. of 10*

**Store at 2-8°C.*