ORNITHINE DECARBOXYLASE BROTH TM 245

For detection of the ability of microorganisms to decarboxylate ornithine

Composition

Ingredients	Gms / Ltr.
L – Ornithine Mono HCl	5.00
Yeast extract	3.00
Dextrose	1.00
Bromo cresol purple	0.015

* Dehydrated powder, hygroscopic in nature, store in a dry place in tightly- sealed containers 25°C and protected from direct Sunlight.

Instructions for Use

Dissolve 9gms in 1000ml distilled water. Gently heat to boiling with gentle swirling and dissolve the medium completely. Dispense the medium in 5ml screw capped test tubes. Sterilize by autoclaving at 15 psi (121°C) for 15 minutes with loosely capped tubes. Cool the tubed medium in an upright position at room temperature before inoculation.

Appearance: Dark purple in colour, 0.9% of clear solution
pH (at 25°C): 6.8 ± 0.2

Principle

ORNITHINE DECARBOXYLASE BROTH for detection of the ability of microorganisms to decarboxylate ornithine. Identifying members of the Enterobacteriaceae, a group that contains many medically important bacteria, is often difficult. It is especially difficult to separate members of the genera *Klebsiella*, Enterobacter and Citrobacter. Measuring the ability of these bacteria to decarboxylate certain amino acids has proven a useful taxonomic tool. The conventional methods for detecting ornithine decarboxylase activity require an extended period of incubation. However, with a few simple modifications, accurate results were obtained within a few hours rather than several days. The broth medium was modified, primarily by omitting Dextrose and by decreasing the pH to 5.5. Later small amount of Dextrose has added as a source of carbohydrate to the medium. Yeast extract provides the Vitamin B source. The rapid ornithine decarboxylase test represents a simple, accurate technique which is well suited for the clinical microbiology laboratory. The amino acids that have proven to be most useful are lysine, arginine and ornithine. Under anaerobic conditions, specific decarboxylase enzymes are able to remove the carboxyl group from these amino acids and thus change the pH of the media to make it more alkaline. To create suitable anaerobic environments the bacteria are grown in broth tubes of the appropriate media, which have been capped after inoculation with sterile mineral oil. A 1-ml amount of this broth was inoculated with one colony and then overlaid with sterile mineral oil. Inoculate each culture into each of the three types of decarboxylase media. Add a layer (0.5 – 1 cm) of sterile mineral oil and incubate for 4 days at 37^0C. Within 2 to 4 hr, the pH increased if ornithine was decarboxylated, for a change in color of the medium, yellow is a negative result while various shades of purple indicate a positive result. Thus changing the color of the internal pH indicator (Bromo cresol purple), can detect this shift and provide an easy way to measure these reactions. to a dark purple. If the amino acid was not decarboxylated, the pH decreased to pH 5.0 to 5.2, enough to give a definite yellow color.

Interpretation

Cultural characteristics observed after inoculating (10^3CFU/ml), on incubation at 35 ± 2°C for 24 - 48 hours.

Microorganisms	ATCC	Decarboxylation	Medium colour
Salmonella typhi	6539	-	Yellow
Escherichia coli	25922	±	Variable
Enterobacter aerogenes	13048	+	Purple

References

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