PHENOL RED BROTH WITH CARBOHYDRATES

INTENDED USE

Phenol Red Broth Base and Phenol Red Broth with carbohydrates are used for the determination of fermentation reactions in the differentiation of microorganisms.

SUMMARY AND EXPLANATION

The ability of an organism to ferment a specific carbohydrate incorporated in a basal medium, resulting in the production of acid or acid and gas, has been used to characterize a specific species or group of bacteria, aid in the differentiation between genera and aid in species differentiation.[1,2]

In 1950, Vera recommended using pancreatic digest of casein in fermentation test media.[3] She found that casein peptone could be used with the pH indicator phenol red in fermentation tests with a high degree of accuracy.

PRINCIPLE

Phenol Red Broth Base is a complete medium without added carbohydrate. It is used as a negative control for fermentation studies or as a base for the addition of carbohydrates. Pancreatic digest of casein provides nutrients and is low in fermentable carbohydrate.[3] The pH indicator, phenol red, is used to detect acid production.

Phenol Red Broths, prepared with a final concentration of one-half percent carbohydrate, are convenient for the determination of fermentation reactions. Most of the end products of carbohydrate fermentation are organic acids which, in the presence of phenol red, produce a color change in the medium from red to yellow.[1] If gas is produced during the fermentation reaction, it is collected in the inverted Durham tube.

No yellow color should occur in the control tube. If it does, the results cannot be correctly interpreted since acid has been produced without fermentation.

REAGENTS (FORMULA)

Pancreatic Digest of Casein	10.0	g
Carbohydrate	5.0	g
Sodium Chloride	5.0	g
Phenol Red	18.0	mg
Deionized Water	1000.0	ml

PROCEDURE

Using a heavy inoculum, inoculate tubes of media with growth from an 18- to 24-hour pure culture using an inoculating loop. Incubate tubes with loosened caps at 35 ± 2°C for 18-48 hours either in an aerobic or anaerobic atmosphere depending on the organism being evaluated. Incubation up to 30 days may be necessary for a negative result.

EXPECTED RESULTS

Examine the unsupplemented tubes at intervals during the incubation process for growth. If supplemented with carbohydrate, observe for the presence of acid (yellow color) and gas (as evidenced by displacement of the liquid in the Durham tubes).

Consult appropriate references for typical reactions produced by various microbial species.[1,2,]

QUALITY CONTROL

All lot numbers have been tested and have been found to be acceptable. Customers can test products using the following quality control organisms. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, sample results should not be reported.

Organisms	Incubation	Results
Proteus vulgaris ATCC 8427	35 ± 2°C for 42-48 hours	Base (K), Dextrose (A), Mannitol (K)
Escherichia coli ATCC 25922	35 ± 2°C for 42-48 hours	Base (K), Dextrose (AG), Mannitol (AG)

ATCC® is a registered trademark of American Type Culture Collection.
K=grow with alkaline reaction, red color
A=grow with acid, yellow color
G=gas production

BIBLIOGRAPHY

1. MacFaddin. 2000. Biochemical tests for identification of medical bacteria, 3rd ed., Lippincott Williams & Wilkins, Baltimore, Md.

2. Forbes, Sahm and Weissfeld. 2007. Diagnostic microbiology, 12th ed. Mosby, Inc., St. Louis, Mo.

3. Vera. 1950. Am. J. Public Health, 40:1267

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