

CM 21412 – PKU TEST AGAR W/ THIENYLALANINE

INTENDED USE

Foreestimationofphenylalanine in blood for detection of Phenylketonuria (PKU).

PRODUCT SUMMARY AND EXPLANATION

Phenylketonuria is a congenital defect caused due to absence of phenylalanine hydroxylase. As a result of this, phenylalanine accumulates in the blood, which is excreted via urine hence it is called as phenylketonuria. Subsequently this deficiency may cause brain damage resulting in mental retardation. Guthrie and Tiekemann devised a modified inhibition assay for early detection of PKU using blood / urine samples of newborn infants having low levels of phenylalanine by determining the serum phenylalanine levels or the level of phenylpyruvic acid in urine.

COMPOSITION

Ingredients	Gms / Ltr
L-Glutamic acid	0.500
DL-Alanine	0.500
Asparagine	0.500
Dextrose	10.000
Dipotassium phosphate	15.000
Monopotassium phosphate	5.000
Ammonium chloride	2.500
Ammonium nitrate	0.500
Sodium sulphate	0.500
Magnesium sulphate	0.050
Manganese chloride	0.005
Ferric chloride	0.005
Calcium chloride	0.0025
β-2-Thienylalanine	0.0033
Agar	15.000

PRINCIPLE

This medium consists of Dextrose which act as an energy source. Magnesium sulphate, manganese chloride, ferric chloride, calcium chloride serves as a source of ions. The amino acids present act a source of nitrogen in the medium. Phosphate buffers the medium. Agar act as a solidifying agent.

INSTRUCTION FOR USE

Dissolve 50.06 grams in 1000 ml purified/distilled water.



Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE OR OVERHEAT.
Cool to 50°C and add *Bacillus subtilis* spores.
Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder	: Cream to greenish yellow homogeneous free flowing powder.
Appearance of prepared medium	: Light yellow coloured clear to slightly opalescent gel forms in Petri plates.
pH (at 25°C)	: 7.0 ± 0.2

INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATC C	Inoculum (CFU/ml)	Growth w/ 2% Phenylalanine	Growth w/ 4% Phenylalanine	Growth w/ 6% Phenylalanine	Growth w/ 8% Phenylalanine	Growth w/ 10% Phenylalanine	Growth w/ 12% Phenylalanine	Recovery	Incubation Temperature	Incubation Period
<i>Bacillus subtilis</i>	6633	50-100	None-poor	Luxuriant	Luxuriant	Luxuriant	Luxuriant	Luxuriant	≥70%	35-37°C	12-16 Hours

PACKAGING:

In package of 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. Demain A. L., 1958, J. Bacteriol., 75:517.
2. Guthrie R., 1961, J. Am. Med. Assoc., 178:863.
3. Guthrie R. and Tiekemann H., 1960, London Conference on the Scientific study of Mental Deficiency, London.
4. Guthrie R. and Susi A., 1963, Pediatrics, 32:338.

