

## Product Data Sheet

### **AK AGAR NO. 2 (SPORULATING AGAR) (ARRET AND KIRSHBAUM MEDIUM)**

**Product No.** GB-DCM-00020-1A

#### **Product Description**

Detection of penicillin and other antibiotic residues in milk is of primary importance in the dairy industry. This is done by the Penicillin Milk Test procedure. AK Agar formulated by Arret and Kirshbaum is used for the production of spores of *Bacillus subtilis* ATCC 6633, which is used in the Penicillin Milk Test procedure. A fresh slant culture of *Bacillus subtilis* is washed with sterile physiological saline onto the surface of Roux bottles containing 300 ml sterile medium. The bottles are incubated at 35°C for 5 days and the resulting growth is suspended into 50 ml of sterile physiological saline. The growth is washed by centrifuging the suspension and discarding the supernatant. The sediment obtained is re-suspended in fresh sterile saline and heated at 70°C for 30 minutes to kill vegetative cells and obtain the spore suspension. This spore suspension can be stored for months for use in detection of penicillin/ antibiotic residues in milk and dairy products.

#### **Product Specifications**

<b>Ingredients</b>	<b>Gms / Ltr</b>
Tryptone	6.000
Gelatin peptone	4.000
Yeast extract	3.000
Beef extract	1.500
Dextrose (Glucose)	1.000
Manganous sulphate	0.300
Agar	15.000

#### **PRINCIPLE**

This medium is highly nutritious due to the presence of yeast extract, beef extract, gelatin peptone and tryptone in addition to being a source of vitamins and essential amino acids. Dextrose serves as the source of energy as well as the fermentable carbohydrate. Manganous sulphate stimulates sporulation.

## QUALITY CONTROL SPECIFICATIONS

Appearance of Powder: Cream to yellow homogeneous free flowing powder.  
 Appearance of Prepared medium: Light amber coloured clear to slightly opalescent gel forms in Petri plates.  
 pH (at 25°C): 6.6±0.2

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Sporulation	Incubation Temperature	Incubation Period
Bacillus megaterium	25848	50-100	Good-luxuriant	≥50%	Positive	35-37°C	5 Days
Bacillus subtilis subsp. spizizenii	6633	50-100	Good-luxuriant	≥50%	Positive	35-37°C	5 Days

## 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.

## INSTRUCTION FOR USE

- Dissolve 30.8 grams in 1000 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely
- Dispense in 300 ml amounts in Roux or other suitable bottles.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50°C. Note: Do not autoclave till the medium has been completely dissolved.

## DISPOSAL

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

**This product is for research use only.**