

Cat. 1027

KAA Confirmatory Agar

For the isolation and confirmation of intestinal enteroccoci in foods according to Mossel.

Practical information

Aplications Categories
Confirmation Enterococci
Selective isolation Enterococci

Industry: Food

Principles and uses

KAA Confirmatory Agar (Kanamycin, Aesculin, Azide) is a selective medium for the isolation and confirmation of intestinal enteroccoci in foods. KAA Confirmatory Agar is used to confirm positives from KAA Presumptive Broth (Cat. 1209) tubes.

Kanamycin, Sodium azide and Sodium citrate have a great inhibitory effect on the accompanying bacterial flora, they inhibit the growth of Gram-positive and Gram-negative bacteria, and the medium is highly selective for esculin-hydrolyzing enterococci. Esculin and Ferric Ammonium citrate are esculin indicators which detect the esculin-hydrolysing bacteria. They hydrolyze the esculin to give glucose and esculetin. These microorganisms present black zones around the colonies from the reaction of the resulting esculetin with the iron ions. Tryptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group essential for bacterial growth. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Bacteriological agar is the solidifying agent.

The presence of intestinal enterococci, is an indicator for faecal contamination, especially when the contamination occurred long ago and the less resistant coliform bacteria, including Escherichia coli, are already dead when the analysis is carried out.

Intestinal enterococci grow forming small, translucent colonies surrounded by a black halo. This medium is recommended by CeNAN for food and drinks analysis.

Formula in g/L

Bacteriological agar	15	Esculin	1
Ferric ammonium citrate	0,5	Kanamycin sulfate	0,02
Sodium azide	0,15	Sodium chloride	5
Sodium citrate	1	Tryptone	20
Yeast extract	5		

Preparation

Suspend 48 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 50 °C, mix well and dispense into plates.

Instructions for use

Streak to obtain isolated colonies and incubate at 35±2 °C for 24 - 48 hours.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Beige	Tournasol-grey	7,0±0,2

Microbiological test

Incubation conditions: (35±2 °C / 24-48 h).

Microrganisms	Specification	Characteristic reaction	
Enterococcus faecalis ATCC 11700	Good growth	Olive green-black colonies, positive esculin hydrolisis	
Escherichia coli ATCC 11775	Total inhibition		
Staphylococcus aureus ATCC 6538	Moderate growth		
Enterococcus faecium ATCC 8043	Good growth	Olive green-black colonies, positive esculin hydrolisis	

Storage

Temp. Min.:2 °C Temp. Max.:25 °C

Bibliography

M.R. Pascual Anderson. Técnicas para Examen Microbiológico de Alimentos y Bebidas (Centro Nacional de Alimentación y Nutrición CeNAN) Madrid, 1982.

Brandl, E. Aspergerger H., Pfleger, F. U-IBEN CH: Zum Vorkomment von D-streptokokken in Käse. 1985.

