

KAA CONFIRMATORY AGAR

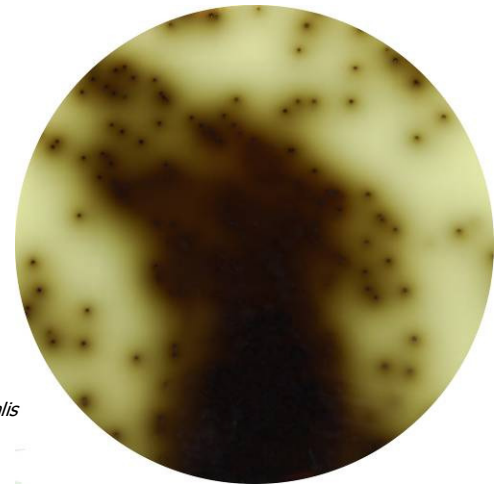
CAT N°: 1027

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

FORMULA IN g/l

Tryptone	20.00	Ammonium Ferric Citrate	0.50
Yeast Extract	5.00	Sodium Azide	0.15
Sodium Chloride	5.00	Kanamycin Sulfate	0.020
Sodium Citrate	1.00	Bacteriological Agar	15.00
Esculin	1.00		

Final pH 7.0 ± 0.2 at 25°C



Enterococcus faecalis
ATCC 11700

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. The prepared medium should be stored at 8-15°C. The color is tounasol-grey.

The dehydrated medium should be homogeneous, free-flowing and beige in color. If there are any physical changes, discard the medium.

Caution: This medium is toxic if swallowed, inhaled or comes into contact with skin. Wear gloves and eye/face protection.

USES

KAA CONFIRMATORY AGAR (Kanamycin, Aesculin, Azide) is a selective medium for the isolation and confirmation of intestinal enterococci 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 glucoside esculin to give glucose and esculin. These microorganisms present black zones around the colonies from the reaction of the resulting esculin 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.

Streak to obtain isolated colonies and incubate at 35 ± 2°C for 24 - 48 hours. Intestinal enterococci grow forming small, translucent colonies surrounded by a black halo. This medium is recommended by CeNAN for food and drinks analysis.

MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 35± 2°C and observed after 24-48 hours.

Microorganisms	Growth	Color Change	Esculin hydrolisis
<i>Enterococcus faecalis</i> ATCC 11700	Good	Olive green-black	+
<i>Enterococcus faecium</i> ATCC 8043	Good	Olive green-black	+
<i>Staphylococcus aureus</i> ATCC 6538	Moderate		
<i>Escherichia coli</i> ATCC 11775	Inhibited		
<i>Lactococcus lactis</i> ATCC 19435	Slightly inhibited	Olive green-black to colorless	

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., Pflieger, F. U-IBEN CH: Zum Vorkomment von D-streptokokken in Käse. 1985..

STORAGE

Once opened keep powdered medium closed to avoid hydration.

