

Kligler Iron Agar (i23224)

Kligler Iron Agar is used for the differentiation of members of the *Enterobacteriaceae* on the basis of their ability to ferment dextrose and lactose and to liberate sulfides.

Industry: Clinical

Principles & Uses

Kligler Iron Agar is a comprehensive medium used to differentiate between lactose fermenters and non-fermenters, particularly in identifying *Salmonella Typhi* from other *Salmonellae*. The medium, a combination of lead acetate and Russell's Double Sugar Agar, includes peptones, yeast extract, lactose, and dextrose. Phenol red serves as the pH indicator, showing a color change in response to acid produced during sugar fermentation. The presence of ammonium iron (III) citrate and sodium thiosulfate enables the detection of hydrogen sulfide production, evidenced by a black color in the butt or a ring formation.

Lactose non-fermenters, like *Salmonella* and *Shigella*, exhibit a yellow slant due to dextrose fermentation, but revert to alkaline (red slant) when the dextrose is exhausted in the aerobic environment. This reversion doesn't occur in the anaerobic butt, which remains acidic (yellow). Lactose fermenters produce yellow slants and butts due to sufficient acid production.

For identification, suspected organisms from plating media are inoculated onto Kligler Iron Agar. The medium provides essential nutrients and supports differentiation based on carbohydrate fermentation, pH changes, and hydrogen sulfide production. It should be used promptly for optimal results.

Composition (gr/L)

Peptone from Casein 15 g, Peptone from Meat 5 g, Meat Extract 3 g, Yeast Extract 3 g, Sodium Chloride 5 g, Lactose 10 g, Dextrose 1 g, Phenol Red 0.024 g, Sodium Thiosulfate 0.5 g, Ammonium Iron (III) Citrate 0.5 g, Agar 12 g.

Final pH at 25°C 7.4 ± 0.2

Preparation from dehydrated Powder

Suspend 55 g of the medium in one liter of purified water. Autoclave at 121°C for 15 minutes. Cool in a slanted position such that deep butts are formed. For best results, the medium should be used on the date of preparation or melted and resolidified before use.

Quality Control

Dehydrated Appearance: Fine, homogeneous, free of extraneous material

Prepared Appearance: Medium to dark, orange to red, with or without a tint of brown, clear to slightly hazy.

Reaction of 5.5% Solution at 25°C: pH 7.4 ± 0.2

Cultural Response

Medium was prepared according to label and after inoculation with microorganisms below, was incubated at 35 ± 2°C for 24 hours.

Organism (ATCC*)	Recovery	Slant	Butt	H ₂ S
<i>Salmonella enterica</i> (14028)	Good	Alkaline	Acid w/o gas	+
<i>Escherichia coli</i> (25922)	Good	Acid	Acid w/ gas	-
<i>Pseudomonas aeruginosa</i> (27853)	Good	Alkaline	Alkaline w/o gas	-

*ATCC is a registered trade mark of the American Type Culture Collection.



Salmonella enterica (left). *Escherichia coli* (middle). Prepared Culture Media (right).

Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.