

## KLIGLER IRON AGAR ISO 10273

**CAT N°: 1364**

For the presumptive test of *Yersinia enterocolitica*

### FORMULA IN g/l

Pancreatic Digest of Casein	20.0	Glucose	1.0
Lactose	10.0	Ferrous Sulfate	0.2
Sodium Chloride	5.0	Sodium Tiosulfate Anhydrous	0.2
Meat Peptone	3.0	Phenol Red	0.025
Yeast Extract	3.0	Bacteriological Agar	15.0

**Final pH 7.4 ± 0.2 at 25°C**

### PREPARATION

Suspend 57.5 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. Dispense into tubes and sterilize in autoclave at 121°C for 15 minutes. Allow to cool in a slanted position in order to obtain butts of 1.5 - 2.0 cm depth. The prepared medium should be stored at 8-15°C. The color is amber, slightly opalescent.

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

### USES

Kligler Iron Agar is recommended by the ISO normative 10273 for presumptive tests of *Yersinia enterocolitica*.

Peptone Digest of Casein and Meat Peptone provide nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group. Glucose and Lactose are the fermentable carbohydrates producing acid, indicated by the Phenol red indicator. The color changes that result are yellow for acid production and red for alkalization. Sodium thiosulfate is reduced to hydrogen sulfide, which reacts with the iron salt to give the black iron sulfide. Sodium Thiosulfate is the H<sub>2</sub>S indicator. Ferrous Sulfate provides sources of sulfates and metallic ions. Bacteriological agar is the solidifying agent.

Once the colonies are purified in the Nutrient Agar (Cat.1060), spread them on Kligler Iron Agar and incubate at 30°C for 24-48 hours.

Interpretation of color changes:

#### **BUTTS;**

YELLOW – glucose-positive (fermentation of glucose)

RED or WITHOUT CHANGE – glucose-negative (Glucose doesn't ferment)

BLACK – Formation of H<sub>2</sub>S

BUBBLES – Formation of Gas

#### **SLANTED POSITION:**

YELLOW – Lactose-positive (use of Lactose).

RED or WITHOUT CHANGE – Lactose-negative (no use of Lactose).

### MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 30°C and observed after 24-48 hours.

Microorganisms	Growth	Slanted Position	Recovery	Base	H <sub>2</sub> S	Gas
<i>Yersinia enterocolitica</i> ATCC 27729	Good	Red	Good	Yellow	-	-
<i>Escherichia coli</i> ATCC 25922	Good	Yellow	Good	Yellow	-	+
<i>Proteus vulgaris</i> ATCC 6380	Good	Red	Good	Yellow	+	-
<i>Salmonella enteritidis</i> ATCC 13076	Good	Red	Good	Yellow	+	+
<i>Shigella flexneri</i> ATCC 12022	Good	Red	Good	Yellow	-	-
<i>Citrobacter freundii</i> ATCC 8090	Good	Yellow	Good	Yellow	+	+

## BIBLIOGRAPHY

ISO 10273 Microbiology of Food and animal feeding stuffs – Horizontal method for the detection of presumptive pathogenic *Yersinia enterocolitica*.



## STORAGE

Once opened keep powdered medium closed to avoid hydration.

