TMS 02- KLIGLER IRON AGAR SLANT

INTENDED USE

For differential identification of Gram-negative enteric bacilli on the basis of the fermentation of dextrose, lactose and H_2S Production.

PRODUCT SUMMARY AND EXPLANATION

Kligler Iron Agar is a combination of the lead acetate medium described by Kligler and Russels Double Sugar Agar and is used as a differentiation medium for typhoid, dysentery and allied bacilli. Bailey and Lacey substituted phenol red for andrade indicator previously used as pH indicator. Kligler Iron Agar differentiates lactose fermenters from the non-fermenters. It differentiates *Salmonella typhi* from other Salmonellae and also *Salmonella paratyphi* A from *Salmonella scottmuelleri* and *Salmonella enteritidis*. Fermentation of dextrose results in production of acid, which turns the indicator from red to yellow. Since there is little sugar i.e. dextrose, acid production is very limited and therefore a reoxidation of the indicator is produced on the surface of the medium, and the indicator remains red. However, when lactose is fermented, the large amount of acid produced, avoids reoxidation and therefore the entire medium turns yellow.

COMPOSITION

Ingredients	Gms / Ltr
Agar	15.000
Peptone	15.000
Lactose	10.000
Proteose peptone	5.000
Sodium chloride	5.000
Beef extract	3.000
Yeast extract	3.000
Dextrose	1.000
Sodium thiosulphate	0.300
Ferrous sulphate	0.200
Phenol red	0.024

PRINCIPLE

Kligler Iron Agar, in addition to peptone, Beef extract and yeast extract, contains lactose and glucose (dextrose), which enables the differentiation of species of enteric bacilli. Phenol red is the pH indicator, which exhibits a colour change in response to acid produced during the fermentation of sugars. The combination of ferrous sulphate and sodium thiosulphate enables the detection of hydrogen sulfide production, which is evidenced by a black color either throughout the butt, or in a ring formation near the top of the butt. Lactose non-fermenters (e.g., *Salmonella* and *Shigella*) initially produce a yellow slant due to acid produced by the fermentation of the small amount of glucose (dextrose). When glucose (dextrose) supply is exhausted in the aerobic environment of the slant, the reaction reverts to alkaline (red slant) due to oxidation of the acids produced. The reversion does not occur in the anaerobic environment of the butt, which therefore remains acidic (yellow butt). Lactose fermenters produce yellow slants and butts because of lactose fermentation. The high amount of acids thus produced helps to maintain an acidic pH under aerobic conditions. Tubes showing original colour of the medium indicates the fermentation of neither glucose (dextrose) nor lactose. Gas production (aerogenic reaction) is detected as individual bubbles or by splitting or displacement of the agar by the formation of cracks in the butt of the medium.

INSTRUCTION FOR USE

Inoculate the bacterial culture with an inoculating needle by streaking the slants.

QUALITY CONTROL SPECIFICATIONS

Appearance	:	Red coloured, clear to slightly opalescent gel forms in tubes as slants
Quantity of Medium	:	8 ml of medium in glass tube.
pH (at 25°C)	:	7.4±0.2

INTERPRETATION

Cultural characteristics observed after an incubation.

Microorganism	ATCC	Growth	Gas	H_2S	Slant	Butt	Incubation Temperature	Incubation Time
Escherichia coli	25922	Luxuriant	Positive reaction	Negative reaction, no blackening of medium	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Klebsiella aerogenes	13048	Luxuriant	Positive reaction	Negative reaction, no blackening of medium	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Citrobacter freundii	8090	Luxuriant	Positive reaction	Positive reaction, blackening of medium	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Proteus vulgaris	6380	Luxuriant	Negative reaction	Positive reaction, blackening of medium	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Klebsiella pneumoniae	13883	Luxuriant	Positive reaction	Negative reaction, no blackening of medium	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Salmonella paratyphi A	9150	Luxuriant	Positive reaction	Negative reaction, no blackening of medium	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Salmonella typhi	6539	Luxuriant	Negative reaction	Positive reaction, blackening of medium	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Salmonella enteritidis	13076	Luxuriant	Positive reaction	Positive reaction, blackening of medium	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Shigella flexneri	12022	Luxuriant	Negative reaction	Negative reaction, no blackening of medium	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Pseudomonas aeruginosa	27853	Luxuriant	Negative reaction	Negative reaction, no	Alkaline reaction, red colour of the medium	Alkaline reaction, red colour of the medium	35-37°C	18 - 48 hours.

				blackening of medium				
Yersinia enterocolitica	27729	Luxuriant	Variable reaction	Negative reaction, no blackening of medium	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.
Enterobacter cloacae	13047	Luxuriant	Positive reaction	Negative reaction, no blackening of medium	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	35-37°C	18 - 48 hours.

PACKAGING:

Kit of 10 Ready-To-Use Slants containing 8 ml medium in each glass tube.

STORAGE

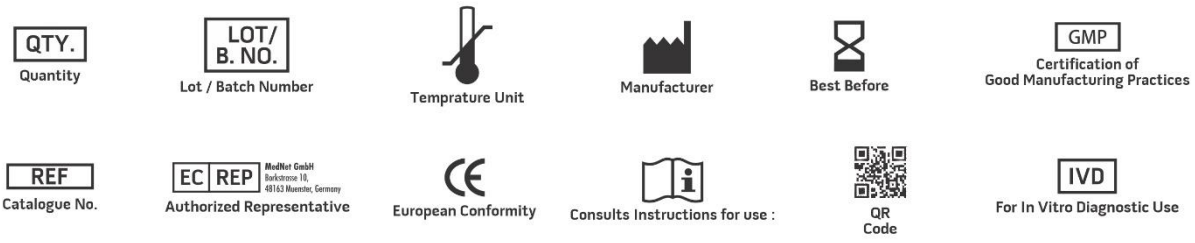
On receipt, store tubes in the dark at 2 – 8°C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Tubed media stored as labeled until just prior to use may be inoculated up to the expiration date and incubated for the recommended incubation times. Allow the medium to warm to room temperature before inoculation. **Product Deterioration:** Do not use tubes if they show evidence of microbial contamination, discoloration, drying or other signs of deterioration.

DISPOSAL

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques.

REFERENCES

1. Bailey S. F. and Lacey G. R., 1927, J. Bacteriol., 13:183.
2. Ewing, 1986, Edwards and Ewings Identification of the Enterobacteriaceae, 4th Ed., Elsevier Science Publishing Co., Inc., N.Y.
3. Kligler I. J., 1917, Am. J. Publ. Health, 7:1041.
4. Russell F. F., 1911, J. Med. Res., 25:217.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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