

TMV 141 - KLIGLER IRON AGAR (VEG.)

INTENDED USE

For differential identification of gram negative enteric bacilli from clinical and non-clinical samples on the basis of fermentation of dextrose and lactose as well as H₂S production.

PRODUCT SUMMARY AND EXPLANATION

This medium is prepared by using vegetable peptones in place of animal based peptones which makes the medium free of BSE/TSE risks free. Kligler developed a Lead Acetate Medium for differentiation of typhoid-paratyphoid group. Kligler further evaluated this medium by combining the principles of Russell Double Sugar Medium. Bailey and Lacey substituted phenol red for the Andrade's indicator from the previous formulation permitting the differentiation of gram-negative bacilli on their ability to ferment dextrose, lactose and hydrogen sulphide production. Kligler Iron Veg Agar differentiates lactose fermenters from nonfermenters. Kligler Iron HiVeg Agar is the modification of Kligler Iron Agar with the use of vegetable peptones and serves the same purpose. It differentiates Salmonella serotype Typhi from other Salmonellae and also Salmonella serotype Paratyphi A form Salmonella serotype Scottmuelleri and Salmonella serotype Enteritidis. Sodium thiosulphate and ferrous sulphate accelerate hydrogen sulfide (H2S) production. Phenol red is the pH indicator. Fermentation of dextrose is indicated by yellow butt and that of lactose by yellow slant and hydrogen sulfide (H₂S) production is indicated by blackening in the butt. Pure cultures of suspected organisms from plating media such as MacConkey Veg Agar, Bismuth Sulphite Veg Agar, Deoxycholate Citrate Veg Agar, SS Veg Agar etc. are inoculated on Kligler Iron Veg Agar for identification.

COMPOSITION

| Ingredients | Gms / Ltr |
|---------------------|-----------|
| Veg special peptone | 15.0 |
| Veg extract | 3. 0 |
| Yeast extract | 3. 0 |
| Veg peptone No. 3 | 5. 0 |
| Lactose | 10.0 |
| Dextrose | 1. 0 |
| Ferrous sulphate | 0. 2 |
| Sodium chloride | 5. 0 |
| Sodium thiosulphate | 0. 3 |
| Phenol red | 0.024 |
| Agar | 15.0 |

PRINCIPLE

Kligler Iron Agar (Veg.), in addition to Veg special peptone Veg 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.

INSTRUCTION FOR USE

- Dissolve 57.5 grams in 1000 ml distilled water.
- Heat to boiling to dissolve the medium completely.













- Mix well and distribute into tubes.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool the tubes to set as slants with 1 inch butts.
- Best reactions are obtained on freshly prepared media.
- Use a straight wire to inoculate the butt, do not use screw-capped tubes or bottles.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Light yellow to pink homogeneous free flowing powder.

Appearance of prepared medium: Red coloured, clear to slightly opalescent gel forms in tubes as slants.

pH (at 25°C) : 7.4±0.2

INTERPRETATION

Cultural characteristics observed after an incubation.

| Microorgan ism | АТСС | Inoculu m (CFU/mI) | Growth | Gas | H₂S | Slant | Butt | Incubati on Tempera ture | Incubat ion Period |
|----------------------------------|-------|------------------------------|-----------|----------------------|---|---|--|-----------------------------------|--------------------------|
| Escherichia coli | 25922 | 50-100 | 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 | 50-100 | 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 | 50-100 | 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 | 50-100 | 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 | 50-100 | 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 | 50-100 | 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 Schottmuell eri | 10719 | 50-100 | 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 |









| <i>Salmonella</i> Typhi | 6539 | 50-100 | 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 | 50-100 | 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 | 50-100 | 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 |

PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

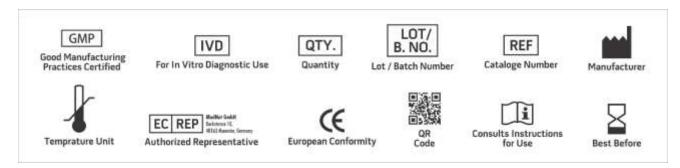
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

- 1. Kligler I.J., 1917, Am. J. Publ. Health, 7:1042.
- 2. Kligler J.J., 1918, J. Exp. Med., 28:319.
- 3. Russell F.F., 1911, J. Med. Res., 25:217.
- 4. Bailey S.F. and Lacey G.R., 1927, J. Bact., 13:182.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. *For Lab Use Only **Revision: 08 Nov., 2019**





