

## TECHNICAL DATA SHEET

### Kligler Iron Agar

#### Principle

Kligler Iron Agar composed of Meat extract (equivalent to Beef extract), yeast extract, peptone, proteose peptone, lactose, dextrose, ferrous sulfate, sodium chloride, sodium thiosulfate and phenol red. This combination permits the differentiation of the gram-negative bacilli both by their ability to ferment dextrose or lactose and to produce hydrogen sulfide. Meat extract, yeast extract, peptone, and proteose peptone provide nitrogen, vitamins and minerals. Ferrous sulfate and sodium thiosulfate are the indicators of hydrogen sulfide production. Phenol red is the pH indicator. Sodium chloride maintains the osmotic balance of the medium. Agar is the solidifying agent.

**Use:** For the differentiation and identification of gram-negative bacilli based on the fermentation of dextrose and lactose and production of H<sub>2</sub>S.

#### Contents\*

| Ingredients        | Gram/Litre |
|--------------------|------------|
| Meat extract#      | 3.000      |
| Yeast extract      | 3.000      |
| Peptone            | 15.000     |
| Proteose peptone   | 5.000      |
| Lactose            | 10.000     |
| Dextrose           | 1.000      |
| Ferrous sulfate    | 0.200      |
| Sodium chloride    | 5.000      |
| Sodium thiosulfate | 0.300      |
| Phenol red         | 0.024      |
| Agar               | 12.000     |
| pH at 25°C         | 7.4 ±0.2   |

\* Formula adjusted for optimum performance and parameters

#Equivalent to Beef extract

**Directions:** Dissolved 54.50 grams in 1000 ml distilled water. Boil to dissolve the medium completely and distribute aseptically in test tubes. Sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 min, cool it to 42-45 °C and kept in slanting position with large butt at bottom allow to solidify and inoculate test sample aseptically.

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## Specimens' types analyzed

Pharmaceutical samples, clinical and non-clinical samples, food and dairy samples etc.

## Precautions to be taken

These microbial media are intended for the in-vitro use only. All the handling, experiments, storage, and discarding should be performed with the help of skilled and knowledgeable technicians and as per the established guidelines. The material should be disposed only after proper sterilization by autoclaving. Please go through the MSDS of the media to avoid any accidents or in emergency.

## Performance and Evaluation

The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

## Quality Control

|                                   |   |
|-----------------------------------|---|
| Appearance                        | Pink beige colored free flowing, homogeneous powder         |
| Reaction of 5.45% solution        | 7.4 ±0.2 at 25 °C   |
| pH                                | 7.20- 7.60  |
| Gelling                           | Firm comparable with 1.2% agar gel                          |
| Color and clarity of ready medium | Slightly orange-red, opalescent gel with slight precipitate |
| Growth Promotion properties       | Best at ≤ 100 CFU at 32-37 °C for 18-72 h                   |
| Indicative properties             | Optimum at ≤ 100 CFU at 32-37 °C for 18-48 h                |
| Negative control                  | Performed using sterile distilled water                     |

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## Different Microbial Response: Cultural characteristics observed after an incubation at 35-37°C for 18 - 48 hours. (Inoculum CFU 50-100)

| Organism                                   | Growth    | Gas               | H <sub>2</sub> S  | Slant             | Butt              |
|--|-----------|-------------------|-------------------|-------------------|-------------------|
| <i>Salmonella typhimurium</i> (ATCC14028)  | Luxuriant | Negative reaction | Positive reaction | Alkaline reaction | Acidic reaction   |
| <i>Salmonella enteritidis</i> (ATCC 13076) | Luxuriant | Positive reaction | Positive reaction | Alkaline reaction | Acidic reaction   |
| <i>Escherichia coli</i> (ATCC 8739)        | Luxuriant | Positive reaction | Negative reaction | Acidic reaction   | Acidic reaction   |
| <i>Proteus vulgaris</i> (ATCC 6380)        | Luxuriant | Negative reaction | Positive reaction | Alkaline reaction | Acidic reaction   |
| <i>Klebsiella aerogenes</i> (ATCC 13048)   | Luxuriant | Positive reaction | Negative reaction | Acidic reaction   | Acidic reaction   |
| <i>Pseudomonas aeruginosa</i> (ATCC 27853) | Luxuriant | Negative reaction | Negative reaction | Alkaline reaction | Alkaline reaction |

**Storage and Shelf Life:** The product is highly hygroscopic; keep the container tightly closed at all times and store it properly as per the conditions mentioned on the label. The declared expiry is valid only when stored as per the conditions mentioned on the label.

**Note:** Sterilize media immediately after reconstitution.

**Disposal:** To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving after completion of task.

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## Reference

1. Atlas, R. M. (2005). *Handbook of media for environmental microbiology*. CRC press.
2. *Difco Manual* (1998). 11<sup>th</sup> Edition. Difco Laboratories., Division of Becton Dickinson and Company, Sparks, Maryland, USA.
3. Rand, M. C., Arnold E. Greenberg, and Michael J. Taras, (1976), *Standard methods for the examination of water and wastewater*. Prepared and published jointly by American Public Health Association, American Water Works Association, and Water Pollution Control Federation.

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