

**TECHNICAL SHEET**

|   |                                     |                                  |       |      |     |                  |
|---|-------------------------------------|----------------------------------|-------|------|-----|------------------|
| <b>B1089</b>  | <b>KLIGLER IRON AGAR</b>            |                                  |       |      |     |                  |
| <b>Formula</b>  |                                     |                                  |       |      |     |                  |
| <b>Ingredients :</b>  |                                     |                                  |       |      |     |                  |
|   |                                     | <b>gms/lit.</b>                  |       |      |     |                  |
| Meat extract B#   | 3.00                                |                                  |       |      |     |                  |
| Yeast extract   | 3.00                                |                                  |       |      |     |                  |
| Casein enzymic hydrolysate  | 20.00                               |                                  |       |      |     |                  |
| Sodium chloride   | 5.00                                |                                  |       |      |     |                  |
| Lactose   | 10.00                               |                                  |       |      |     |                  |
| Glucose anhydrous   | 1.00                                |                                  |       |      |     |                  |
| Ferrous ammonium sulphate,6H <sub>2</sub> O   | 0.50                                |                                  |       |      |     |                  |
| Sodium thiosulphate, pentahydrate   | 0.50                                |                                  |       |      |     |                  |
| Phenol red  | 0.025                               |                                  |       |      |     |                  |
| Agar  | 15.00                               |                                  |       |      |     |                  |
| #- Equivalent to Beef extract   |                                     |                                  |       |      |     |                  |
| Final pH (at 25°C): 7.4 ± 0.2   |                                     |                                  |       |      |     |                  |
| <b>Directions:</b>  |                                     |                                  |       |      |     |                  |
| Suspend 57.7 gms (the equivalent weight of dehydrated medium per Litre) in 1000 ml. distilled water. Boil to dissolve the medium completely. Dispense in tubes & sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the tubes to cool in slanted position to form slope of about 1inch butts. Best reactions are obtained on freshly prepared media.   |                                     |                                  |       |      |     |                  |
| <b>Principle :</b>  |                                     |                                  |       |      |     |                  |
| Kligler Iron Agar permits the differentiation of the gram-negative bacilli both by their ability to ferment lactose and to produce hydrogen sulfide. Meat Extract B, Yeast Extract, Casein enzymic hydrolysate provide nitrogen, vitamins and minerals. Ferrous ammonium sulphate, or 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. |                                     |                                  |       |      |     |                  |
| <b>QC Tests – (I)Dehydrated Medium</b>  |                                     |                                  |       |      |     |                  |
|   | Colour :                            | Light pink to light reddish pink |       |      |     |                  |
|   | Appearance:                         | Homogeneous Free Flowing powder  |       |      |     |                  |
| <b>(II)Rehydrated medium</b>  |                                     |                                  |       |      |     |                  |
|   | pH (post autoclaving/heating) :     | 7.4 ± 0.2                        |       |      |     |                  |
|   | Colour (post autoclaving/heating) : | Reddish orange to red            |       |      |     |                  |
|   | Clarity (post autoclaving/heating): | Clear to slightly opalescent     |       |      |     |                  |
| <b>(III)Q.C. Test Microbiological</b>   |                                     |                                  |       |      |     |                  |
| Cultural characteristics observed after 24 – 48 hrs at 35-37°C.   |                                     |                                  |       |      |     |                  |
|   | MICROORGANISM (ATCC )               | GROWTH                           | SLANT | BUTT | GAS | H <sub>2</sub> S |
|   | Escherichia coli (25922)            | Luxuriant                        | A     | A    | +   | -                |
|   | Enterobacter aerogenes (13047)      | Luxuriant                        | A     | A    | +   | -                |
|   | Proteus vulgaris (13315 )           | Luxuriant                        | K     | A    | -   | +                |
|   | Salmonella typhi ( 6539 )           | Luxuriant                        | K     | A    | -   | +                |
|   | Salmonella enteritidis (13076 )     | Luxuriant                        | K     | A    | +   | +                |
|   | Klebsiella pneumoniae (13883 )      | Luxuriant                        | A     | A    | +   | -                |
|   | Shigella flexneri (12022)           | Luxuriant                        | K     | A    | -   | -                |
|   | Pseudomonas aeruginosa (27853)      | Luxuriant                        | K     | K    | -   | -                |
|   | Yersinia enterocolitica (27729)     | Luxuriant                        | K     | A    | V   | -                |
|   | Key : A = acid production (yellow)  |                                  |       |      |     |                  |
|   | K = alkaline reaction (red)         |                                  |       |      |     |                  |
|   | + = positive or blacking            |                                  |       |      |     |                  |
|   | V = variable                        |                                  |       |      |     |                  |

Refer disclaimer overleaf

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|                         |   |                                |           |            |                    |
|-------------------------|---|--------------------------------|-----------|------------|--------------------|
| <b>Precautions :</b>    | 1. For Laboratory Use.<br>2. Follow proper, established laboratory procedures in handling and disposing of infectious materials.  |                                |           |            |                    |
| <b>Limitations :</b>    | 1. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.<br>2. H <sub>2</sub> S – producing organisms may produce a black precipitate to such a degree that the reaction in the butt is completely masked. If H <sub>2</sub> S is produced, dextrose is fermented even if it is not observed.<br>3. Further biochemical tests and serological typing must be performed for definite identification and confirmation of organisms.<br>4. Do not use as inoculating loop to inoculate a tube of Kligler Iron Agar. While stabbing the butt, mechanical splitting of the medium occurs, causing a false positive result for gas production.<br>5. Best reactions are obtained on freshly prepared medium.<br>6. A pure culture is essential when inoculating Kligler Iron Agar. If inoculated with a mixed culture, irregular observations may occur.<br>7. Hydrogen sulfide determinations using Kligler Iron Agar should be limited to the members of the Enterobacteriaceae. Other organisms may require more sensitive methods for detection of H <sub>2</sub> S production.<br>8. Tubes should be incubated with caps loosened to allow a free exchange of air, which is necessary to enhance the alkaline condition on the slant. |                                |           |            |                    |
| <b>Use :</b>            | For identification of Pseudomonas species. It can also be used for differential identification of gram-negative enteric bacilli on the basis of fermentation of dextrose, lactose and H <sub>2</sub> S production.  |                                |           |            |                    |
| <b>Storage :</b>        | Dehydrated medium- below 30°C Prepared medium– Between 2 to 8°C.  |                                |           |            |                    |
| <b>Packing :</b>        | 500 gm. bottle  |                                |           |            |                    |
| <b>Product profile:</b> | Reconstitution  | Quantity on Preparation (500g) | pH (25°C) | Supplement | Sterilization      |
| <b>B1089</b>            | 57.7g/l   | 8.66L                          | 7.4 ± 0.2 | NIL        | 121°C / 15 minutes |

**Disclaimer:**

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related BIOMARKLABORATORIES publications.

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