

LYSINE IRON AGAR (LIA)

- For in vitro use only -

Catalogue No. TL97

Our Lysine Iron Agar Slants are used for the differentiation of enteric organisms based on their ability to decarboxylate or deaminate lysine and to produce hydrogen sulfide.

LIA was developed by Edwards and Fife for the detection of *Salmonella arizonae* cultures. Johnson et al. utilized LIA along with Kligler Iron Agar to differentiate between certain groups within the *Enterobacteriaceae* family.

Pancreatic digest of gelatin and yeast extract provide nitrogen, vitamins, amino acids, and other essential factors required for bacterial growth. Bromcresol purple acts as the color indicator in the medium to detect carbohydrate fermentation, and decarboxylation-deamination lysine reactions. Bromcresol purple changes to yellow when the pH of the medium falls below 5.2 and appears purple when the medium is above 6.8. The presence of sodium thiosulfate and ferric ammonium citrate allows for hydrogen sulfide detection. Sodium thiosulfate acts as the substrate for enzymatic reduction and the resultant colorless hydrogen sulfide gas reacts with ferric ammonium citrate to produce ferrous sulfide, an insoluble black precipitate that blackens the medium.

The presence of lysine allows for the detection of the bacterial enzymes lysine decarboxylase and lysine deaminase. All Enterobacteriaceae ferment glucose resulting in the butt of the medium turning vellow; if the organism produces lvsine decarboxylase, cadarvine is formed and neutralizes the organic acid formed by glucose fermentation and the medium reverts back to the alkaline state. Therefore a positive result for lysine decarboxylase is the butt of the tube changing purple or remaining neutral. Lysine deamination is signalled by the development of a red color along the slant. This red coloration occurs by an unknown mechanism whereby one of the final end products from deamination reacts with bromcresol purple.

Formula per Litre of Medium

| Pancreatic Digest of Gelatin | 5.0 g |
|------------------------------|--------|
| Yeast Extract | 3.0 g |
| Dextrose | 1.0 g |
| Lysine | 10.0 g |
| Ferric Ammonium Citrate | 0.5 g |
| Sodium Thiosulfate | 0.04 g |
| Agar | |
| Bromcresol Purple | 0.02 g |
| | |

$pH 7.4 \pm 0.2$

Recommended Procedure

- 1. Allow medium to adjust to room temperature prior to inoculation.
- 2. Take a well-isolated colony from a pure culture plate and pick the centre using a straight inoculating needle.
- 3. Inoculate the LIA Slant by stabbing the middle of the tube twice to the bottom of the tube and streaking the slant with a fishtail motion.
- 4. Incubate the tubes at 35°C.
- 5. Examine tubes and interpret results after 18 to 24 hours of incubation.

Interpretation of Results

Lysine decarboxylation:

Positive (+): purple slant / purple butt with or without H2S blackening

Negative (-): purple slant / yellow butt

Lysine deamination: Positive (+): red slant / yellow butt

Negative (-): no red slant

Another system of differentiation is based on H_2S production. A positive H_2S reaction appears as blackening in the medium. H_2S production may not be detected for organisms that are negative for lysine decarboxylase, such as *Proteus* species, since acid production from dextrose fermentation may suppress its formation.

Gas production is a by-product of some metabolic cycles and can be observed as gas bubbles or cracks in the medium; gas production is often irregular or suppressed on this medium.

Quality Control

After checking the medium for correct pH, colour, depth, and sterility, the following organisms are used to determine the performance of the completed medium.

| Organism | Expected Results | | |
|--------------------------------------|------------------|------|------------------|
| | Slant | Butt | H ₂ S |
| Salmonella typhimurium ATCC 14028 | K | K | + |
| Proteus mirabilis ATCC 12453 | R | А | - |
| Shigella flexneri ATCC 12022 | K | А | - |

K = Alkaline (purple) A = Acid (yellow) R = Red

Storage and Shelf Life

Our LIA Agar Slants should be protected from light and stored in an upright position at 4°C to 8°C. Under these conditions the medium has a shelf life of 16 weeks from the date of manufacture.

References

- Edwards PR, Fife MA. Lysine-iron agar in the detection of *Arizona* cultures. Appl Microbiol 1961; 9:478-80.
- MacFaddin JF. Media for isolationcultivation-maintenance of medical bacteria, Vol I. Baltimore: Williams & Wilkins, 1985.

- 3. Ewing WH. Edwards and Ewing's identification of *Enterobacteriaceae*. 4th ed. New York: Elsevier, 1986.
- 4. Forbes BA, Sahm DF, Weissfeld AS. Bailey and Scott's diagnostic microbiology. 10th ed. St. Louis: Mosby, 1998.
- MacFaddin JF. Biochemical Tests for Identification of Medical Bacteria. 3rd ed. Lippincott Williams and Wilkins. Philadelphia, 2000.

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