

Instructions for Use

REF - KM0113

LB Broth (Lennox)

Dehydrated Culture Media

1. Intended Use

KM0113 LB Broth (Lennox) is used for the cultivation and maintenance of *Escherichia coli*.

2. Composition*

Ingredient	g/L
Enzymatic digest of casein	10.0
Yeast extract	5.0
Sodium chloride	5.0

*Adjusted/supplemented as needed to meet performance requirements

3. Summary and Explanation

KM0113 is based on the formulation stated by Lennox ⁽¹⁾. Suitable for non-selective cultivation of *E. coli* strains for cloning, DNA plasmid production and production of recombinant proteins. KM0113 provides all the nutrients necessary for the growth of these organisms and half the sodium chloride in LB Broth (Miller), allowing the researcher to select a medium with optimal salt concentration for a specific strain.

4. Principle

LB Broth (Lennox) contains half of the amount of sodium chloride found in LB Broth (Miller). Enzymatic digest of casein supplies the required nitrogen and carbon. Vitamin B complex required by recombinant strains of *Escherichia coli* are supplied by yeast extract. Sodium chloride maintains the osmotic balance and supplies sodium ions for membrane transport. The low salt formulation makes this medium ideal for cultures requiring salt-sensitive antibiotics.

5. Preparation Instructions

Suspend 20.0g/L of dried culture media in deionised/purified water and allow to soak whilst mixing for 10 minutes.

Autoclave to sterilise at 121°C for 15 minutes before cooling.

Aseptically dispense the specified volume into appropriate sterile containers.

6. Physical Characteristics

	Dehydrated Medium	Prepared Medium
Appearance and Colour	Beige fine powder	Straw liquid
pH	N/A	7.0 ± 0.2

7. Materials Provided

KM0113 can be provided in the formats detailed below. Each tub is labelled with product name, product code, lot number, expiry date, instructions, and appropriate warnings.

Product Code	Product Format
KM0113-500G-500	1 x 500g Dehydrated Culture Media Tub
KM0113-5KG-5000	1 x 5kg Dehydrated Culture Media Tub
KM0113-10KG-10000	1 x 10kg Dehydrated Culture Media Tub

8. Materials Needed but not Provided

Standard microbiological laboratory materials e.g., autoclave, sterile loops or swabs, collection containers, incubators, and quality control organisms.

9. Specimens

KM0113 LB Broth (Lennox) is suitable for the testing of the following specimens:

- Specimens should be isolated colonies or stock cultures.

Sampling and transport equipment must be used in accordance with the end user's suppliers' recommendations. Refer to appropriate standard method or local guidance on sample collection and subsequent processing.

10. Test Procedures and Interpretation of results

Allow the media to come to room temperature.

Based on sample type and information provided, check to see if the specimen needs to be pre-enriched prior to inoculation to the prepared medium.

Isolated Colonies or Stock Cultures:

Prepare a homogenised suspension using a ratio of 1:9 of stock solution to prepared KM0113 medium or inoculate 10ml with an isolated colony using a sterile loop. Incubate at $37 \pm 1^\circ\text{C}$ aerobically for 18-24 hours.

After incubation, examine broths for turbidity; examine any subcultures on agar for colonies (typical colony appearance outlined in Quality Control table below). Perform further biochemical, serological, or mass spectroscopy testing to confirm identity of presumptive positive isolates from subcultures. Refer to relevant local guidelines.

11. Quality Control

Organism	Incubation	Result (Specificity)
<i>E. coli</i> (NCTC 12241) <i>S. aureus</i> (NCTC 12981)	$37 \pm 1^\circ\text{C}$ aerobically for 18-24 hours $37 \pm 1^\circ\text{C}$ aerobically for 18-24 hours Subcultured onto E&O PP0280 Tryptone Soya Agar and incubated at $37 \pm 1^\circ\text{C}$ aerobically for 18-24 hours	Broth: Turbid. Subplate: Cream colonies Broth: Turbid. Subplate: White colonies

It is the responsibility of the user to perform Quality Control testing taking into consideration the intended use of the medium, and in agreement with any local relevant guidelines (e.g., frequency, strains used, atmosphere, incubation temperature).

12. Performance

To fully verify KM0113 LB Broth (Lennox) performance, dehydrated culture media samples were used to prepare LB Broth (Lennox) (10ml) and tested to assess colour change and turbidity/colony morphology and recovery level when incubated at $37 \pm 1^\circ\text{C}$ aerobically for 18-24 hours, before being subcultured on E&O PP0280 Tryptone Soya Agar and incubated at $37 \pm 1^\circ\text{C}$ aerobically for 18-24 hours. All samples of prepared media produced grew and showed good recovery and the correct morphology of the required test organisms: *Escherichia coli* (NCTC 12241) and *Staphylococcus aureus* (NCTC 12981). Therefore, it can be concluded that KM0113 LB Broth (Lennox), meets performance criteria when used according to the instructions outlined above. Trend analysis data available upon request.

13. Limitations of the Media

- Due to natural variation, some strains may grow poorly on this medium.

14. Precautions and Warnings

This product is considered non-hazardous under CLP regulations. Wear such PPE as recommended by laboratory COSHH assessment. During and after use, always handle all materials in a manner conforming to Good Laboratory Practices and consider that material under test should be regarded as a potential biohazard if mishandled.

Refer to KM0113 Material Safety Data Sheet.

15. Storage conditions and Shelf life

Store KM0113 dried media in the original container with the lid tightly closed at between 10 and 30°C in low humidity conditions away from direct sunlight. Kept under these conditions, dried culture media may be used up to date of expiry shown on the product label.

Dispose of in accordance with local and national authority requirements. Do not use if dried culture medium is not free-flowing, homogenous or the specified colour. Additionally, do not use medium if it has been stored inappropriately or the packaging has been damaged.

Do not use prepared media if it is contaminated, discoloured, out of date, cracked or a precipitate is present. Additionally, do not use prepared media if it has been stored inappropriately.

Dehydrated culture media does not need to be used all at once; it may be resealed and stored according to label instructions.

16. References

1. Lennox, E. (1955) Transduction of linked genetic characters of the host by bacteriophage P1. *Virology*. 1(190).

Version History*

001 09/10/23 - New Document Created

*Note: minor typographical, grammatical, and formatting changes are not included in the revision history.



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TABLE OF APPLICABLE SYMBOLS

IFU/KM0113 REV. 001

REF Catalogue number	LOT Batch code		Manufacturer	Use by
Temperature limitation	Contents sufficient for <n> tests	Consult Instructions for Use	Keep away from direct light	Store in a dry place