ANDRADE LACTOSE PEPTONE WATER

Dehydrated culture medium

1 - INTENDED USE

Medium to aid in differentiation between genera and species of bacteria by their ability to ferment (degrade) lactose.

2 - COMPOSITION - TYPICAL FORMULA*
(AFTER RECONSTITUTION WITH 1 L OF WATER)

Tryptone	10.00 g
Lactose	10.00 g
Sodium chloride	5.00 g
Acid fuchsin	0.01 g

*The formula may be adjusted and/or supplemented to meet the required performances criteria.

3 - PRINCIPLE OF THE METHOD AND EXPLANATION OF THE PROCEDURE

When microorganisms ferment lactose an acid or acid with gas are produced. Depending upon the organisms involved, the end products may vary. The production of the acid lowers the pH of the test medium, which is detected by the colour change of the pH indicator. Colour change only occurs when a sufficient amount of acid is produced, as bacteria may utilize the peptone producing alkaline by-products.[1]
Andrade Lactose Peptone Water contains a peptone with a low carbohydrates content which is source of nitrogen and minerals for bacterial growth; sodium chloride maintains the osmotic balance. Lactose is the fermentable carbohydrate. Andrade's indicator is a solution of acid fuchsin that when added to peptone water is colourless or slightly pink at pH 7.4. It becomes pink-red at acidic pH and yellow at alkaline pH levels.
If the test is positive with a colour change of the medium to pink-red, it means that lactose has been fermented (degraded) by the tested organism. If the test is negative, a catabolic attack of peptones will occur with the formation of ammonia, the alkalinisation of the medium and a colour change of Andrade's indicator to yellow.

4- DIRECTIONS FOR MEDIUM PREPARATION

Suspend 25 g in 1000 mL of cold purified water. Heat to dissolve with frequent agitation. Distribute into tubes or bottles containing Durham tubes and sterilize by autoclaving at 121°C for 15 minutes.

5 - PHYSICAL CHARACTERISTICS

Dehydrated medium appearance	whitish, fine, homogeneous, free-flowing powder
Solution and prepared tubes appearance	pale pink, limpid
Final pH at 20-25 °C	7.4 ± 0.1

6 - MATERIALS PROVIDED - PACKAGING

Product	Type	REF	Pack
Andrade Lactose Peptone Water	Dehydrated medium	4010482	500 g (20 L)

7 - MATERIALS REQUIRED BUT NOT PROVIDED

Autoclave, water-bath, sterile loops and swabs, incubator and laboratory equipment as required, test tubes, Durham tubes, Erlenmeyer flasks, ancillary culture media and reagents for the identification of the colonies.

8 - SPECIMENS

Andrade Lactose Peptone Water is not intended for primary isolation from samples; it is inoculated with 18-24 h pure culture from solid media such as Tryptic Soy Agar or blood agar.

9 - TEST PROCEDURE

With a heavy inoculum, inoculate tubes of Andrade Lactose Peptone Water with pure culture using an inoculating loop or swab.
Incubate tubes with loosened caps, aerobically at 35-37°C for 18-48 hours.

10 - READING AND INTERPRETATION

After incubation observe the presence of growth (turbidity), bubbles and the colour change of the medium.
Positive reaction (lactose degradation): the medium turns pink-red and the formation of gas bubbles can be observed.
Negative reaction: the medium is turbid, yellow in colour.
After a positive reaction has been observed, discard the tube; by prolonging the incubation, an inversion of the reaction may be observed.

11 - USER QUALITY CONTROL

All manufactured lots of the product are released for sale after the Quality Control has been performed to check the compliance with the specifications. However, the end user can perform its own Quality Control in accordance with the local applicable regulations, in compliance with accreditation requirements and the experience of the Laboratory. Here below are listed some test strains useful for the quality control.

CONTROL STRAINS	INCUBATION T°/ T / ATM	GLUCOSE
E. coli ATCC 25922	35-37° / 18-24H /AE	AG
S. Typhimurium ATCC 14028	35-37° / 18-24H /AE	K

AE: aerobic incubation; ATCC is a trademark of American Type Culture Collection; A: acid production, red-pink colour; G: gas production; K: alkalinity, yellow

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12 - PERFORMANCES CHARACTERISTICS

Prior to release for sale, a representative sample of all lots of dehydrated Andrade Lactose Peptone Water is tested for productivity and specificity of chromatic reactions by comparing the results with a previously approved Reference Batch.
Productivity is tested by dilution to extinction method, by inoculating 1 mL of appropriate decimal dilutions of target organisms in test tubes, incubating at 37°C for 24 hours and recording the highest dilution showing growth in Reference Batch (GrRB) and in Test Batch (GrTB). Productivity is tested with the following lactose fermenting strains *E.coli* ATCC 25922, *C.freundii* ATCC 40738, *S.aureus* ATCC 25923, *E.faecalis* ATCC 19433, and lactose non-fermenting strain S.Typhimurium ATCC 14028. The growth of tested strains is very good and the specific chromatic reactions (pink-red and the formation of gas bubbles for lactose positive strains-yellow medium for lactose negative strains) are in compliance with the specifications.

13 - LIMITATIONS OF THE METHOD

- The medium after autoclaving, when hot, is pink but the colour disappears upon cooling.[1]

14 - PRECAUTIONS AND WARNINGS

- This product is for microbiological control and for professional use only; it is to be used by adequately trained and qualified laboratory personnel, observing approved biohazard precautions and aseptic techniques.
- Dehydrated media must be handled with suitable protection. Before use, consult the Safety Data Sheet.
- This culture medium contains raw materials of animal origin. The *ante* and *post mortem* controls of the animals and those during the production and distribution cycle of the raw materials, cannot completely guarantee that this product doesn't contain any transmissible pathogen. Therefore, it is recommended that the culture medium be treated as potentially infectious, and handled observing the usual specific precautions: do not ingest, inhale, or allow to come into contact with skin, eyes, mucous membranes. Download the TSE Statement from the website www.biolifeitaliana.it, describing the measures implemented by Biolife Italiana for the risk reduction linked to infectious animal diseases.
- Apply Good Manufacturing Practice in the production process of prepared media.
- All laboratory specimens should be considered infectious.
- The laboratory area must be controlled to avoid contaminants such as culture medium or microbial agents.
- Sterilize all biohazard waste before disposal. Dispose the unused medium and the sterilized tubes inoculated with samples or microbial strains in accordance with current local legislation.
- Do not use the culture medium as active ingredient for pharmaceutical preparations or as production material intended for human and animal consumption
- The Certificates of Analysis and the Safety Data Sheet of the product are available on the website www.biolifeitaliana.it.
- The information provided in this document has been defined to the best of our knowledge and ability and represents a guideline for the proper use of the product but without obligation or liability. In all cases existing local laws, regulations and standard procedures must be observed for the examination of samples collected from human and animal organic districts, for environmental samples and for products intended for human or animal consumption. Our information does not relieve our customers from their responsibility for checking the suitability of our product for the intended purpose.

15 - STORAGE CONDITIONS AND SHELF LIFE

Upon receipt, store at +10°C /+30°C away from direct light in a dry place. If properly stored, it may be used up to the expiration date. Do not use beyond this date. Avoid opening the bottle in humid places. After use, the container must be tightly closed. Discard the product if the container and/or the cap are damaged, or if the container is not well closed, or in case of evident deterioration of the powder (colour changes, hardening, large lumps).
The user is responsible for the manufacturing and quality control processes of prepared media and for the validation of the shelf life of the finished products, according to the type (tubes/bottles) and the storage method (temperature and packaging).

16 - REFERENCES

1. MacFaddin JF. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria. Baltimore: Williams & Wilkins; 1985.

TABLE OF APPLICABLE SYMBOLS

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

REVISION HISTORY

Version	Description of changes	Date
Revision 1	Updated layout and content	2022/05

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

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