Blood Agar + Nalidixic Acid (Base)

356-4534

DEFINITION
Selective medium used in the analysis of food products for the isolation of streptococci (including *S. Pneumoniae*), *Listeria monocytogenes* and *Erysipelothrix rhusiopathiae* while at the same time demonstrating hemolytic response.

PRINCIPLE
The nutrient substances provided by pastose, pastone, meat-liver peptic peptone and meat extract favor the growth of the bacteria mentioned above.
The presence of nalidixic acid leads to total inhibition of sensitive gram-negative bacteria and partial inhibition of that of staphylococci.

PRESENTATION
Dehydrated
500 g **code 356-4534**

STORAGE
- Dehydrated: + 15°C to 25°C, in carefully-sealed bottles in a cool, dry place.
- Expiration date and batch number are shown on the package.

THEORETICAL FORMULA

Pastose	5 g
Pastone	5 g
Meat-liver peptic peptone	5 g
Meat extract	7 g
Sodium chloride	5 g
Nalidixic acid	40 mg
Pastagar B	13 g
Distilled water	1,000 ml

Final pH (25°C) = 7.2 ± 0.2

OTHER PRODUCTS REQUIRED (NOT SUPPLIED)
- Distilled water
- Horse blood (**code 355-6641**)

See corresponding Technical Sheet(s)

EQUIPMENT REQUIRED (NOT SUPPLIED)
(non-exhaustive)
- Hotplate
- Mixer-homogenizer
- Vortex-type shaker
- 100 ml Pyrex bottles with autoclave-proof stoppers
- Sterile pipettes (0,1 ml, etc)
- Pasteur pipettes (**code 355-0751**) or inoculating loop
- Sterile spreaders
- Water-bath precise to ± 1°C
- Autoclave
- All usual laboratory equipment.

PREPARATION OF DEHYDRATED MEDIUM
Always shake before use
Dissolve 40 g of powder in 1 liter of distilled water. Wait for 5 minutes, then mix until a homogenous suspension is obtained. Heat gently, shaking frequently, then bring to boiling point until completely dissolved. Dispense 100 ml per bottle and sterilize in autoclave at 121°C ± 1°C for 15 minutes.

Reconstitution ratio: 40 g/l.
500 g of powder makes 12.5 liters of medium.

PROTOCOL
• Inoculation and incubation
This medium is generally used for the preparation of a blood agar:
- Add 5%-10% of sterile horse blood (**code 355-6641**) to the sterile base, melted and cooled to between 44°C and 47°C.
- After shaking carefully, avoiding the creation of air bubbles, pour the mixture into Petri dishes.
- After inoculation by spreading over the surface or by isolation, incubate at 37°C ± 1°C for 18 -24 hours.

READING AND INTERPRETATION
Streptococcus: 2 to 3 mm diameter, white or colorless, smooth, round, either haemolytic (alpha, beta) or not.

Listeria: 1 to 2 mm diameter, white or colorless, smooth, either slightly haemolytic or not.

PRECAUTIONS
- Take care not to shake the regenerated medium violently or it will be re-oxygenated.
- Do not add the blood to a base medium at a temperature exceeding 47°C.
- Comply with Good Laboratory Practice.

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PERFORMANCES / QUALITY CONTROL OF THE TEST

The growth performances of the media are verified with the following strains:

STRAINS	Results after 24h culture at 37°C
Streptococcus pyogenes ATCC 19615	Good growth β-hemolysis
Group C *Streptococcus* CIP A 7	Good growth β-hemolysis
Enterococcus faecalis var zymogenes ATCC 29212	Good growth β-hemolysis
Streptococcus bovis CIP 5623	Good growth Negative hemolysis
Streptococcus pneumoniae ATCC 6303	Good growth α-hemolysis turning green
Escherichia coli ATCC 25922	No culture
Staphylococcus aureus ATCC 25923	Partial inhibition

QUALITY CONTROL OF MANUFACTURER

Every product manufactured and marketed by Bio-Rad is subject to a quality-assurance procedure at all stages, from the reception of raw materials to the marketing of the end-product. Each batch of finished product undergoes quality control and is marketed only if it satisfies the acceptability criteria.

Documentation relative to the production and control of each batch is kept on file.

KEY WORDS

Blood agar + nalidixic acid / *Streptococcus* / *Listeria* / Food products / Nalidixic acid / Hemolytic response / Medium.

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