Simmons' Citrate/Agar

356-1834
356-4834

DEFINITION
Medium used for the differentiation of gram negative bacilli. This medium contributes to demonstration of the identification characteristics of *Enterobacteriaceae*.

STANDARDS
FOOD MICROBIOLOGY
- **NF ISO 10273 (December 2003):** Microbiology - General guidelines for the detection of suspected pathogenic *Yersinia enterocolitica.*

PRINCIPLE
The principle of the medium relies on the ability of certain micro-organisms to develop with citrate as the only carbon and energy source.
Fermentation of citrate is shown by the indicator turning blue.

PRESENTATION
- **Ready-to-use**
  7 ml x 25 tubes **code 356-1834**

- **Dehydrated**
  500 g **code 356-4834**

STORAGE
- Ready-to-use: + 2℃ to 8℃.
- Dehydrated: + 15℃ to 25℃, in carefully-sealed bottles in a cool, dry place.
- Expiration date and batch number are shown on the package.

THEORETICAL FORMULA

Sodium citrate	1 g
Sodium chloride	5 g
Magnesium sulfate (anhydrous)	0.14 g
Monammonia phosphate	1 g
Dipotassium phosphate	1 g
Bromothymol blue	80 mg
Agar	13 g
Distilled water	1,000 ml

Final pH (25℃) = 6.8 ± 0.2

OTHER PRODUCTS REQUIRED (NOT SUPPLIED)
- Distilled water

EQUIPMENT REQUIRED (NOT SUPPLIED)
(non-exhaustive)
- Distilled water
- Hotplate
- Mixer-homogenizer
- Test tubes (16 x 160 mm) with autoclave-proof stoppers
- Sterile pipettes **(code 355-0751)**
- Thermostatically-controlled incubator or incubation room, precise to ± 1℃
- Autoclave
- All usual laboratory equipment.

PREPARATION OF DEHYDRATED MEDIUM
Always shake before use.
Dissolve 21 g of powder in 1 liter of distilled water. Wait for 5 minutes, then mix until a homogenous suspension is obtained. Heat gently swirling frequently, then bring to boiling point until completely dissolved. Dispense 3 to 5 ml per tube and sterilize in autoclave at 121℃ (± 1℃) for 20 minutes. Leave to cool in an inclined position.

Reconstitution ratio: 21 g/l.
500 g of powder makes 23.8 liters of medium.

PROTOCOL
Inoculation and incubation
- Using a previously flame-sterilized pipette, inoculate the slope of the citrate medium with longitudinal, parallel streaks, using an isolated colony taken from nutrient agar. citrate (-)
- Incubate at 30℃ (*Yersinia enterocolitica*) or 37℃ ± 1℃ for 24 hours.
- In the event of negative reaction, re-incubate for a further 24 hours.

READING AND INTERPRETATION
When citrate is used, the medium turns blue.

Escherichia coli
Yersinia enterocolitica } citrate (-)

Citrobacter
Enterobacter } citrate (+)

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PRECAUTIONS
Comply with Good Laboratory Practice.

PERFORMANCES / QUALITY CONTROL OF THE TEST
The growth performances of the media are verified with the following strains:

STRAINS	Utilisation of citrate after 1 to 7 days culture at 37°C
Proteus vulgaris ATCC 13315	Negative
Salmonella Enteritidis ATCC 13076	Positive
Klebsiella pneumoniae ATCC 13883	Positive
Escherichia coli ATCC 25922	Negative
Shigella sonnei ATCC 25931	Negative

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
Faecal coliforms / *Escherichia coli* / Food products / Detection / Citrate IMVIC test / Bromothymol blue.

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