

Biolife

Technical Sheet - Instructions for use

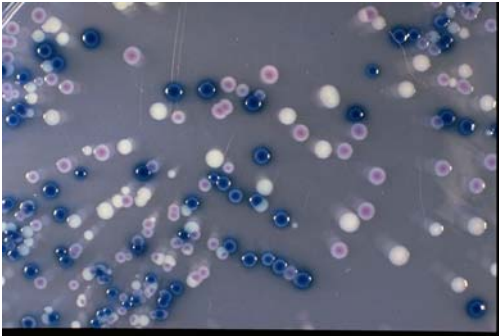
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ChromArt

CHROMOGENIC URINE AGAR IV CLEAR

Chromogenic dehydrated culture medium and ready to use plates without opaque background for isolation, enumeration and direct identification of the main microorganisms of the urinary tract

CULTURAL RESPONSE



Mixed culture of: *E.coli* (pink-magenta red colonies), *K.pneumoniae* (dark blue colonies), *Enterococcus* sp. (turquoise blue colonies), *S.aureus* (white colonies).

TYPICAL FORMULA

Dehydrated medium (g/L) CHROMOGENIC URINE AGAR IV CLEAR

Peptones and growth factors	25,500
Chromogenic mix	0,300
Agar	16,000

Ready to use plates (g/L) CHROMOGENIC URINE AGAR IV CLEAR

Peptones and growth factors	25,500
Chromogenic mix	0,300
Horse Serum (ml/L)	20,00
Agar	16,000

INTENDED USE

Chromogenic dehydrated culture medium and ready to use plates without opaque background for isolation, enumeration and direct identification of the main microorganisms of the urinary tract: *E.coli*; KES; *Proteus*; enterococci, staphylococci, yeasts.

PRINCIPLE OF THE METHOD AND EXPLANATION

Chromogenic Urine Agar IV Clear is a diagnostic medium useful for the isolation, counting and direct presumptive rapid identification of urinary tract pathogens: *E.coli*, *Klebsiella*, *Enterobacter*, *Serratia* (KES), *Proteus*, *Morganella*, *Providencia*, Enterococci, Staphylococci, yeasts.

Main characteristics and advantages:

- Very good productivity achieved by a new formula containing selected and standardized peptones and detoxifying agents.
- Optimized agar concentration to inhibit the swarming of the colonies.

The differentiation between the bacterial species or genus is achieved by:

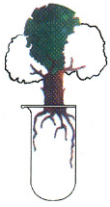
- A chromogenic substrate for β -galactosidase (GAL) which is split with the liberation of an insoluble pink-red dye.
- A chromogenic glucopyranoside derivative which is split by β -glucosidase (GLU) with the formation of an insoluble blue-green dye.
- Tryptophan for the detection of tryptophan deaminase (TDA) useful for the test of the indole for the confirmation of *E.coli*.

Strains that produce β -glucosidase, such as enterococci and the *Klebsiella/Enterobacter/Serratia* (KES) group, form colonies that generate a green/blue coloration as a result of hydrolysis of the indoxyl substrate. *Escherichia coli* strains appear as pink-magenta colonies because of β -galactosidase production. Tryptophan is also present in the medium to detect members of the *Proteae* group, which generate a diffuse

PREPARATION OF THE DEHYDRATED CULTURE MEDIUM

Suspend 41,8 g in 1000 ml of cold purified water. Heat to boiling with frequent agitation and sterilise by autoclaving at 121°C for 15 minutes. Keeping under agitation, cool to about 45 ° C and distribute in Petri dishes. The medium appears amber coloured. The ready plates, without horse serum, shall be used within ten days from the date of preparation.

For prolonged storage of the pre-poured plates add horse serum: cool the autoclaved medium to 45-50°C and add under aseptic conditions, 20 mL/L of Horse Serum. Mix well and distribute into sterile petri dishes. pH : 7.2 ± 0,2



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SPECIMEN

Specimens consist of urine samples collected according to conventional methods.

APPLICATION

Chromogenic Urine Agar IV Clear can be used according to the usual laboratory practices for urine bacterial count, by spreading the specimen on the agar surface and incubating at 37°C for 18-24 hours.

READING AND INTERPRETATION OF RESULTS

The cultivated colonies can be identified with the following scheme:

Escherichia coli : pink-magenta colonies (β -galactosidase positive, β -glucosidase negative)
Indole test positive: *E.coli*
Indole test negative: proceed to the identification with conventional methods.

Klebsiella – Enterobacter - Serratia (KES): blue/blue-violet colonies:
(β -galactosidase positive, β -glucosidase positive)
Microscopic examination: gram negative bacilli
For genus/species identification, proceed with conventional identification methods.

Enterococcus spp.: turquoise blue colonies (β -galactosidase neg. , β -glucosidase pos.)
Microscopic examination: gram positive cocci.

Proteus-Morganella-Providencia: (brown colonies with brown halo: tryptophan deaminase positive, β -galactosidase negative, β -glucosidase negative)
Indole test negative: *Proteus mirabilis*.
Indole test positive: *Providencia* or *Morganella* or *Proteus* spp. indole + (proceed to the identification with conventional methods,

Staphylococci and yeasts: white colonies (β -galactosidase negative, β -glucosidase negative)
Microscopic examination: gram positive cocci or yeasts
Proceed to the identification with conventional methods.

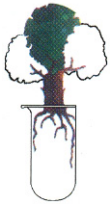
MATERIAL NOT SUPPLIED

Reagents, other culture media and necessary laboratory equipment.

QUALITY CONTROL

It is user's responsibility to carry out the quality control in accordance with the regulations in force and according to his own Laboratory experience. The following table shows some useful strains for quality control.

Test Strains	Incubation T° / t / Atm.	Growth characteristics
<i>E. coli</i> ATCC 25922	37°C - 24H-A	Good growth, pink colonies, indole positive
<i>E. coli</i> ATCC 8739	37°C - 24H-A	Good growth, pink colonies indole positive
<i>K. pneumoniae</i> ATCC 27736	37°C - 24H-A	Good growth, pale violet colonies
<i>E. cloacae</i> ATCC 13047	37°C - 24H-A	Good growth, grey blue colonies
<i>E. aerogenes</i> ATCC 13048	37°C - 24H-A	Good growth, pale blue colonies
<i>C. freundii</i> ATCC 8090	37°C - 24H-A	Good growth, pale blue-pink colonies
<i>C. diversus</i> ATCC 40738	37°C - 24H-A	Good growth, grey-pale blue
<i>P. mirabilis</i> ATCC 10005	37°C - 24H-A	Good growth, brown-orange colonies



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<i>S. aureus</i>	ATCC	25923	37°C - 24H-A	Good growth, white colonies
<i>S. saprophyticus</i>	ATCC	15305	37°C - 24H-A	Good growth, small pink colonies
<i>E. faecalis</i>	ATCC	19433	37°C - 24H-A	Good growth, turquoise blue/green colonies
<i>S. epidermidis</i>	ATCC	12228	37°C - 24H-A	Good growth, white colonies

Notes: A: aerobic incubation

LIMITATION OF THE METHOD

- The identification obtained with the medium should be considered as a presumptive identification. It must be confirmed with biochemical, immunological or other appropriate identification test. Gram staining and microscopic observation is recommended to confirm any doubtful colour reactions.
- It is reported that some strains of the bacterial genus reported above have abnormal biochemical patterns.
- *Citrobacter* spp. may be presumptively identified as *E. coli* (pink-magenta colonies) because some strains are β -galactosidase positive and β -glucosidase negative. The use of a spot indole test successfully eliminates some of these *E. coli* false positives (1). The use of susceptibility test or the detection of pyrrolidonyl aminopeptidase (PYR test) may facilitate the differentiation of pink colonies of *Citrobacter* spp. from *E. coli* (2).
- Between the *Proteus-Morganella-Providencia* group, *P. mirabilis* is indole negative and can be easily recognised
- Biochemical identification is needed for genus/species identification within *Klebsiella*, *Enterobacter*, *Serratia*, *Citrobacter* (KESC group).
- A pyrrolidonyl aminopeptidase (PYR test) might be necessary to differentiate enterococci from *S. agalactiae*
- *S. saprophyticus* and *S. xyloso* produces small pink colonies.
- Interpretation of the test results should be done considering the patient's history, the source of the specimen, colonial and microscopic reading and the results of other tests performed.
- Do not use plates that appear contaminated before sewing or have an excess of condensation water.
- Bring the plates to room temperature before use.

PRECAUTIONS

- The products described here are in vitro diagnostic for professional use and must be used in the laboratory by appropriately trained operators using approved asepsis and safety methods against pathogens.
- Sterilize the plates after use and before disposal. Dispose waste in accordance with current legislation.
- Download the Certificate of Analysis of the products from the site www.biolifeitaliana.it

STORAGE

Dehydrated medium: keep tightly closed, away from bright light, at 2 °C to 8 °C.

Store the ready to use plates in their original box at 2 °C to 8 °C.

When stored as directed the products remain stable until the expiry date shown on the label. Do not use beyond stated expiry date. Discard if there are signs of deterioration.

REFERENCES

1. J. D. Perry, L. A. Butterworth, A. Nicholson, M. R. Appleby, and K. E. Orr. J. Clin. Pathol. 2003 Jul; 56(7): 528–531.
2. D. Fallon, N. Andrews, D. Frodsham, B. Gee, S. Howe, A. Iliffe, K. J. Nye, and R. E. Warren J. Clin. Pathol. 2002 Jul; 55(7): 524–529.

PRODUCTS

Description	Type	Cat. N°	Pack size
CHROMOGENIC URINE AGAR IV CLEAR	Dehydrated medium	409810C2	500 g (12,7 L)
CHROMOGENIC URINE AGAR IV CLEAR	Ready to use plates (Ø90mm)	549810C	20 plates



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