

# CLED / Mac Conkey

Bi-plate for the isolation, enumeration and identification of pathogenic bacteria from urine specimens.

# DESCRIPTION

CLED / Mac Conkey is a ready-to-use plate containing two distinct media used for microbiological urine analysis.

CLED Agar supports the growth of all urinary potential pathogens providing good colony differentiation.

Mac Conkey Agar is a selective medium giving excellent differentiation between lactose-fermenting and lactosenonfermenting gram-negative enteric bacilli. This medium meets United States Pharmacopeia (USP), European Pharmacopoeia (EP) and Japanese Pharmacopoeia (JP) performance specifications, where applicable.

(g/l)	<u>Mac Conkey Agar</u>	(g/l)
4.0	Pancreatic Digest of Gelatin	17.0
4.0	Enzymatic Digest of Casein	1.5
3.0	Enzymatic Digest of Animal Tissue	1.5
10.0	Lactose	10.0
0.128	Bile Salts Mixture	1.5
0.02	Sodium Chloride	5.0
15.0	Neutral Red	0.03
	Crystal Violet	0.001
	Agar	15.0
	Final pH 7.1 $\pm$ 0.2 at 25°C	
	(g/l) 4.0 3.0 10.0 0.128 0.02 15.0	(g/l)Mac Conkey Agar4.0Pancreatic Digest of Gelatin4.0Enzymatic Digest of Casein3.0Enzymatic Digest of Animal Tissue10.0Lactose0.128Bile Salts Mixture0.02Sodium Chloride15.0Neutral RedCrystal VioletAgarFinal pH 7.1 ± 0.2 at 25°C

# METHOD PRINCIPLE

Enzymatic digest of gelatin, enzymatic digest of casein, beef extract and enzymatic digest of animal tissue provide amino acids, nitrogen, carbon, vitamins and minerals required for organisms growth. Lactose is the fermentable carbohydrate. Agar is the solidifying agent. Lack of electrolytes suppresses the swarming of *Proteus* and *Shigella* species.

In CLED Agar, bromothymol blue is the pH indicator changing color from green to yellow when lactose fermentation lowers the pH. L-cystine is a growth supplement for cystine-dependent organisms.

In Mac Conkey Agar, bile salts and crystal violet are the selective agents, inhibiting Gram-positive organisms and allowing Gram-negative bacteria to grow. Sodium Chloride maintains the osmotic balance of the medium. Neutral red is the pH indicator.

### **TEST PROCEDURE**

Urine must be directly streaked over the agar surface no later than 2 h after collection or must be kept at 2-8°C (not longer than 24 h) to avoid microbial overgrowth. Use a calibrated loop (0.01 or 0.001 ml) to inoculate an undiluted, well-mixed urine sample in each of the two media on the plate. Incubate aerobically at  $35 \pm 2$ °C for 18-48 h.

### **RESULTS INTERPRETATION**

After incubation count the number of colonies on the plate. Each colony correspond to 100 or 1000 CFU/ml of urine, using a 0.01 ml or 0.001 ml loop, respectively. Observe the color and the morphology of the colonies for presumptive identification according to the ID table. Further tests should be performed for confirmation.

### ID Table.

Microorganism	Typical colony color and morphology on CLED Agar	Typical colony color and morphology on Mac Conkey Agar
Escherichia coli	Yellow, opaque	Pink with bile precipitate
Klebsiella spp, Enterobacter spp	Yellow to whitish-blue, mucoid	Pink, mucoid
Proteus spp	Translucent blue	Colorless to beige
<i>Salmonella</i> spp	Flat blue	Colorless
Pseudomonas aeruginosa	Green, matt surface and rough periphery	Colorless to pink, irregular
Enterococci	Yellow, about 0.5 mm in diameter	Partial to complete inhibition
Staphylococcus aureus	Deep yellow	Partial to complete inhibition
Coagulase negative staphylococci	Pale yellow	Partial to complete inhibition

### APPEARANCE

CLED Agar: slightly opalescent, green to blue-green. Mac Conkey Agar: slightly opalescent, pinkish-red.

### **STORAGE**

Store at 10-25°C away from light. Do not use the product beyond its expiry date on the label or if product shows any evidence of contamination or any sign of deterioration.

# SHELF LIFE

6 months.

# QUALITY CONTROL

Slides are inoculated with the microbial strains indicated in the QC table. Inoculum for productivity: 50-100 CFU Inoculum for selectivity:  $10^4$ - $10^6$  CFU Incubation conditions:  $35 \pm 2^{\circ}$ C for 18-24 hours.

# QC Table.

Microorganism		Growth on CLED Agar	Growth on Mac Conkey Agar
Escherichia coli	ATCC® 25922	Good, yellow colonies	Good, pink colonies with bile ppt
Klebsiella pneumoniae	ATCC® 13883	Good , yellow to whitish-blue colonies	Good, pink colonies
Proteus mirabilis	ATCC® 12453	Good, blue colonies, no swarming	Good, colorless colonies, no swarming
Salmonella Typhimurium	ATCC® 14028	Good, blue colonies	Good, colorless colonies
Pseudomonas aeruginosa	ATCC® 27853	Good, green colonies	Good, colorless to pink
Entercoccus faecalis	ATCC® 29212	Good, yellow colonies	Partially to completely inhibited
Staphylococcus aureus	ATCC® 25923	Good, small yellow colonies	Partially to completely inhibited

# WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. It is nevertheless recommended to consult the safety data sheet for its correct use. The product must be used by properly trained operators only.

# **DISPOSAL OF WAIST**

Disposal of waist mast be carried out according to national and local regulation in force.

### BIBLIOGRAPHY

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- Mackey J.P. and G.H. Sandys (1966) Diagnosis of urinary tract infections. Br. Med. J. 1:1173.
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- European Directorate for the Quality of Medicines and Healthcare. 2008. The European pharmacopoeia, 6th ed., Supp. 1, 4-1-08, online. European Directorate for the Quality of Medicines and Healthcare, Council of Europe, Strasbourg, France.
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PRESENTATION		Contents	Ref.
CLED / Mac Conkey	90 mm ready-to-use plates	20 plates	18502

### TABLE OF SYMBOLS

LOT	Batch code	IVD	<i>In vitro</i> Diagnostic Medical Device		Manufacturer	Use by	Fragile, handle with care
REF	Catalogue number		Temperature limitation	$\sum$	Contains sufficient for <n> tests</n>	Caution, consult Instruction For Use	Do not reuse





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