

## Columbia C.N.A. agar base

Columbia C.N.A. Agar Base is used for selective isolation of pathogenic gram-positive cocci from clinical and nonclinical specimens.

REF: BS.1/CU01.100.0100  
REF: BS.1/CU01.500.0500

100 Gram  
500 Gram

REF: BS.1/CU01.250.0250

250 Gram

### CLINICAL SIGNIFICANCE

Columbia Agar Base is a nutritionally rich formula containing 5% defibrinated blood, which provides more nutrients and capability of displaying haemolytic reactions. Columbia Blood Agar Base is utilized as a base for preparation of media containing blood and in selective media preparations where various combinations of antimicrobial agents are used as additives. Ellner et al formulated the medium (1) and found that the combination of peptones used gave more rapid and abundant growth of *Streptococci*, *Staphylococci*, *Neisseria* and *Haemophilus* with better-defined haemolytic reactions. Columbia C.N.A. Agar Base is prepared with the same formula as Columbia Agar Base with the addition of 10 mg/litre of colistin and 15 mg/ litre of nalidixic acid to inhibit the growth of gram-negative bacteria and to support the growth of *Staphylococci*, haemolytic *Streptococci* and *Enterococci* when supplemented with 5% blood.

### METHOD PRINCIPLE

Biopeptone and tryptose B supports luxuriant growth of microorganisms and visualization of good haemolytic reactions. Sheep blood allows detection of haemolytic reactions and supplies X-factor necessary for the growth of many bacterial species. Horse blood supplies X-factor and V-factor, therefore is mostly preferred in most laboratories. Yeast extract and cornstarch serve as energy source and neutralizer respectively. It should be noted that this medium has relatively high carbohydrate content and, therefore, beta-hemolytic streptococci may produce a greenish hemolytic reaction that may be mistaken for alpha haemolysis. The addition of the antimicrobial agents, colistin (or polymyxin B) and nalidixic acid, renders the medium selective for gram-positive microorganisms (2). Colistin and nalidixic acid disrupt the cell membrane of gram-negative organisms, whereas nalidixic acid blocks DNA replication in susceptible gram-negative bacteria (3). Columbia C.N.A. Agar Base with addition of blood gives selective isolation of gram-positive cocci, *Staphylococci* and *Streptococci*, particularly when gram-negative bacilli are present and tend to overgrow on conventional blood agar plates. Also used for selective isolation of *Gardnerella vaginalis*. This medium supports growth of *Brucella abortus*, *Yersinia pestis*, *Clostridium perfringens* and all commonly occurring *Enterobacteriaceae* without addition of blood.

### MEDIA COMPOSITION

Item	Formula per liter of medium
- Biopeptone	20.00 gm
- Tryptose B	3.000 gm
- Corn starch	1.000 gm
- Sodium chloride	5.000 gm
- Colistin sulphate	0.010 gm
- Nalidixic acid	0.015 gm
- Agar	15.00 gm

### PRECAUTIONS AND WARNINGS

Media to be handled by entitled and professionally educated person. Do not ingest or inhale.

Good Laboratories practices using appropriate precautions should be followed in:

- Wearing personnel protective equipment (overall, gloves, glasses,).
- Do not pipette by mouth.
- In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.

- Respect country requirement for waste disposal.

**S56:** dispose of this material and its container at hazardous or special waste collection point.

**S57:** use appropriate container to avoid environmental Contamination.

**S61:** avoid release in environment.

For further information, refer to the Columbia C. N. A. agar base material safety data sheet.

### STORAGE AND STABILITY

**BioScien** Columbia C. N. A. agar base dehydrated media are stable until expiration date stated on label when properly stored 10-30°C. The prepared medium should be stored at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to avoid lump development due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in a dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

**Final pH 7.3 ± 0.2 at 25°C**

### MEDIA PREPARATION

- Suspend 44.02 grams in 1000 ml distilled water.
- Adjust pH to 7.3 ± 0.2 at 25°C
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.
- Cool to 45-50°C and aseptically add 5% v/v sterile, defibrinated blood.
- Mix well and pour into sterile Petri plates.

### Deterioration

The color of **BioScien** Columbia C. N. A. agar base is Cream to yellow homogeneous free flowing powder. If there are any physical changes, discard the medium.

The hydrated basal medium is Yellow coloured clear to slightly opalescent gel. After addition of 5% v/v sterile defibrinated blood: Cherry red coloured opaque gel forms in Petri plates, media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), and contaminations.

### SPECIMEN COLLECTION AND PRESERVATION

Clinical samples

### EQUIPMENT REQUIRED NOT PROVIDED








- Sterile cups
- Petri plates
- Sterile loops
- Incubator

### PERFORMANCE CHARACTERISTICS

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Cultural characteristics observed with added 5% v/v sterile, defibrinated blood after an incubation at 35-37°C for 40-48 hours.

Test Organisms	Growth
<i>Escherichia coli</i> ATCC 25922	Good - Luxuriant
<i>Neisseria meningitidis</i> ATCC 13090	Good - Luxuriant
<i>Staphylococcus aureus</i> ATCC 25923	Inhibited
<i>Staphylococcus epidermidis</i> ATCC 12228	Inhibited
<i>Streptococcus pneumoniae</i> ATCC 6303	Good - Luxuriant
<i>Streptococcus pyogenes</i> ATCC 19615	Good - Luxuriant

SYMBOLS IN PRODUCT LABELLING	
<b>IVD</b> For in-vitro diagnostic use	 Number of <n> test in the pack
<b>LOT</b> Batch Code/Lot number	 Caution
<b>REF</b> Catalogue Number	 Do not use if package is damaged
 Temperature Limitation	 Consult Instruction for use
 Expiration Date	
 Manufactured by	

## QUALITY CONTROL

To ensure adequate quality control, it is recommended that positive and negative control included in each run. If control values are found outside the defined range, check the system performance. If control still out of range please contact **BioScien** technical support.

## REFERENCES

1. Ellner et al, 1966, Am. J. Clin. Path., 45:502.
2. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Tenover F. C., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
3. Estevez, 1984, Lab. Med., 15:258