

Technical Data Sheet

CE Columbia Agar Base

acc. harm. EP/USP/JP and ISO

Ordering number: 1.10455.0500 / 1.10455.5000

Columbia Agar Base is a superior, complete medium proposed by Ellner et al. (1966) that can be used for the cultivation of even fastidious microorganisms and as a base for the preparation of various special culture media.

This medium complies with the specifications given by the harmonized methods of EP, USP, JP for Microbial Examination of Non-sterile Products: Tests for Specified Microorganisms.

IVD in vitro diagnosticum - For professional use only

Mode of Action

This culture medium can be utilized to prepare blood or boiled blood agar ("chocolate agar"), special inhibitors must be added for selective cultivation. Columbia agar base can be used to prepare lactose milk egg-yolk agar for the isolation of fastidious *Clostridia* (Ellner et al. 1966). Al-Jumaili and Bint (1981) recommended the addition of blood, cycloserine and cefoxitin to Columbia agar (base) for the isolation of *Clostridium difficile*. The inclusion of bacitracin makes the enriched Columbia Agar Medium selective for the isolation of *Haemophilus* species from clinical specimens, especially from upper respiratory tract. It can also be employed in the so-called *Corynebacterium diphtheriae* toxicity (virulence) test according to Hermann et al. (1958) when using the agar plate diffusion method described by Elek (1949). Greenwood et al. (1977) used it to prepare Vaginalis agar for the cultivation of *Gardnerella vaginalis*. Bannermann and Bille used it to make Acriflavin-Ceftacidim Agar (AC Agar) for the selective cultivation of *Listeria*.

Without enrichment, Columbia Blood Agar Base is used as a general purpose media.

Typical Composition

Peptone from Casein	10 g/l
Peptone from Meat	5 g/l
Heart Extract	3 g/l
Yeast Extract	5 g/l
Starch	1 g/l
NaCl	5 g/l
Agar-Agar	13 g/l

Preparation

Suspend 42 g/l. Autoclave (15 min at 121 °C). Cool to 45-50 °C before mixing in heat-sensitive additives.

The plates are clear and yellowish-brown. After blood is added, they are bright red and non-hemolytic.

The pH value at 25 °C is in the range of 7.1-7.5.

Preparation of blood agar: Mix 5 ml blood homogeneously with 95 ml sterile culture medium base. Pour plates.

Preparation of gentamicin blood agar: Mix 100 ml defibrinated sheep blood and 0.11 ml gentamicin solution homogeneously with 900 ml sterile culture medium base. Pour plates.

Preparation of boiled agar: Add 10 ml blood to 90 ml sterile culture medium base. Heat the mixture in a water bath for about 10 min to 80 °C swirling all the time until the medium becomes chocolate brown in color, pour plates.

Preparation of lactose milk egg-yolk agar: Dissolve 42 g dehydrated culture medium, 12 g lactose, 1 g agar-agar in 1 l demineralized water. Mix in 33 ml/l of a 0.1 % aqueous solution of neutral red, adjust the pH to 7.0 and autoclave (15 min at 121 °C). Cool to 45-50 °C, add approximately 35 ml egg-yolk emulsion/l and 10 g dried milk/l and mix homogeneously. Pour plates.

Specimen

e.g. Blood.

Clinical specimen collection, handling and processing, see general instructions of use.

Experimental Procedure and Evaluation

Depend on the purpose for which the medium is used.

Storage

The product can be used for sampling until the expiry date if stored upright, protected from light and properly sealed at +15 °C to +25 °C.

After first opening of the bottle the content can be used up to the expiry date when stored dry and tightly closed at +15 to +25° C.

Disposal

Please mind the respective regulations for the disposal of used culture medium (e.g. autoclave for 20 min at 121 °C, disinfect, incinerate etc.).



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Quality Control

Control Strains	ATCC #	Inoculum CFU	Incubation	Expected Results
<i>Escherichia coli</i>	8739	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %
<i>Staphylococcus aureus</i>	6538	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %, β -Hemolysis delayed
<i>Streptococcus pyogenes</i>	12344	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %, β -Hemolysis
<i>Streptococcus pyogenes</i>	21059	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %, β -Hemolysis
<i>Streptococcus pneumoniae</i>	6301	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %, α -Hemolysis
<i>Enterococcus faecalis</i>	19433	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %, no Hemolysis
<i>Bacillus cereus</i>	11778	10-100	24 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Recovery on blood agar ≥ 70 %, β -Hemolysis
<i>Clostridium sporogenes</i>	11437	10 - 100	48 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Bacitracin test positive
<i>Clostridium sporogenes</i>	19404	10 - 100	48 h at 30-35 °C	Recovery on medium without blood ≥ 70 %, Bacitracin test positive

Please refer to the actual batch related Certificate of Analysis

Literature

Al-Jumali, I.J. and Bint, A.J. (1981): Simple method of isolation and presumptive identification of *Clostridium difficile*. Zbl. Bakt. I. Abt. Orig. A. **250**: 152-146.

Bannermann, E.S. and Bille, J. (1988): A new selective medium for isolating *Listeria spp.* from heavily contaminated material. Appl. Environ. Microbiol. **43**: 165-167.

Black, W.A. and Van Buskirk, F. (1973): Gentamicin blood agar used as a general-purpose selective medium. Appl. Microbiol. **25**: 905-907.

Elek, S.D. (1949): The plate virulence test for diphtheria. J. Clin. Pathol. **3**: 250-258.

Ellner, P.D., Stoessel, C.I., Drakeford, E. and Vasi (1958): A new culture medium for medical bacteriology. Amer. J. Clin. Path. **29**: 181-183.

European Directorate for the Quality of Medicines and Healthcare. (2014): The European Pharmacopoeia. 8th Ed. Chapter 2.6.13 Microbiological examination of non-sterile products: Test for specified products. Strasbourg, France.

Greenwood, J.R., Pickett, M.J., Martin, W.J., and Mack, E.G. (1977): *Haemophilus vaginalis* (*Corynebacterium vaginale*): method for isolation and rapid biochemical identification. Health Lab. Sci.



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14: 102-106.

Hermann, G.J., Moore, M.S., and Parsons, E.J. (1958): A substitute for serum in the diphtheria *in vitro* toxigenicity test. Amer. J. Clin. Path. **29**: 181-183.

Hunt, D.E., Jones, J.V., a. Dowell, V.R. (1986): Selective medium for the isolation of *Bacteroides gingivalis*. J. Clin. Microbiol. **23**: 441-445.

Japanese Ministry of Health, Labour and Welfare. (2011): The Japanese Pharmacopoeia. 16th Ed. Chapter 4.05 Microbial Limit Test II. Microbiological examination of non-sterile products: Test for specified products. Japanese Ministry of Health, Labour and Welfare. Tokyo, Japan.

Karmali, M.A., Simor, A.E., Roscoe, M., Fleming, P.C., Smith, S.S., and Lane, J. (1986): Evaluation of a blood-free, charcoal-based selective medium for the isolation of *Campylobacter* organisms from feces. J. Clin. Microbiol. **23**: 456-459.

Kunze, M. (1971): COLUMBIA-Agar-Grundsubstrat als Nährmedium für Mykoplasmen. Zbl. Bakt. I. Abt. Orig. **216**: 271-272.

Petts, D. (1984): Colistin-oxolinic acid-blood agar: a new selective medium for streptococci. J. Clin. Microbiol. **19**: 4-7.

Thompson, J.S. (1985): Colistin-oxolinic acid blood agar: a selective medium for the isolation of *Gardnerella vaginalis*. J. Clin. Microbiol. **21**: 843.

United States Pharmacopoeia 38 NF 33 (2015): <62> Microbiological examination of non-sterile products: Tests for specified microorganisms.

Ordering Information

Product	Cat. No.	Pack size	Other pack sizes available
Columbia Agar Base	1.10455.5000	500 g	5 kg
Neutral Red Indicator	1.01369.0025	25 g	
Agar-Agar	1.01614.1000	1 kg	
Egg Yolk Emulsion	1.03784.0001	10 x 100 ml	
Lactose Monohydrate	1.07657.1000	1 kg	
Skim Milk Powder	1.15363.0500	500 g	

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