

Product Insert

CHOCOLATE AGAR (CHOC)

Products

AS-214	Chocolate Agar (CHOC)	1 plate / pkg
AS-244	Chocolate Agar (CHOC)	4 plates / pkg

Intended Use

Chocolate Agar (CHOC) is recommended for use in the isolation and cultivation of fastidious aerobic and anaerobic organisms.

Summary

CHOC agar is an enriched non-selective media supplemented with cofactor V (nicotinamide adenine dinucleotide; NAD) and cofactor X (hemin), and facilitates the growth of fastidious anaerobes. CHOC agar is recommended for aerotolerance testing of obligate anaerobes. Heated horse blood added to the media gives it a “chocolate” appearance. This media is prepared, dispensed, and packaged under oxygen-free conditions to prevent the formation of oxidized products prior to use.

Formulation*

Pancreatic Digest of Casein	10.00	g
Soy Peptone	3.00	g
Meat Peptone	10.00	g
Dextrose	1.00	g
Yeast Extract	2.00	g
Sodium Chloride	5.00	g
Sodium Bisulfite	0.10	g
Agar	15.00	g
VX Supplementation	10.00	mL
Horse Blood	50.00	mL
DI Water	1.00	L

Final pH: 7.0 ± 0.2 at 25 °C

Final weight: 16.0 g ± 1.6 g

*Approximate formula. Adjusted and/or supplemented as required to meet performance criteria.

Precautions

For *IN VITRO DIAGNOSTIC USE* only. Utilize approved biohazard precautions and aseptic technique when using this product. This product is for use only by properly trained and qualified personnel. Sterilize all biohazard waste prior to disposal.

Storage and Shelf Life

Storage: Upon receipt, store at 2 – 8°C in original package until use. Avoid overheating or freezing. Do not use media if there are signs of deterioration (shrinking, cracking, or discoloration due to oxidation of media) or contamination. The expiration date applies to the product in its original packaging and stored as directed. Do not use product past the expiration date shown on the container.

Shelf Life: 6 months from date of manufacture.

Procedure

Specimen Collection: Protect specimens for anaerobic culture from oxygen during collection, transportation, and processing. Consult appropriate references for detailed instructions concerning collection and transportation of anaerobes.

Methods for Use: CHOC agar should be inoculated directly with clinical specimen or a broth that has been previously inoculated from a clinical specimen. CHOC agar should be inoculated and incubated in 5% - 10% CO₂ for 24 – 48 hours for aerotolerance testing. The CHOC can be split to permit multiple isolates on a single plate. CHOC should be inoculated first to ensure that a result of “no growth” is based on atmospheric requirement and not dependent on the amount of inoculum. CHOC is used in reference to blood agar plates because *Haemophilus influenzae* and other fastidious microorganisms grow anaerobically and would not be detected on an aerotolerance test conducted solely on blood agar plates. Additional periods of incubation may be necessary to recover some organisms. Detailed instructions for processing anaerobic and aerobic cultures can be found in the appropriate references

Materials Required, But Not Provided

Standard microbiological supplies and equipment such as loops, saline blanks, slides, staining supplies, microscope, incinerator / autoclave, incubators, anaerobic chamber / anaerobic jars, disinfectant, other culture media, and serological / biochemical reagents.

Interpretation of Results

If used properly, this media should support good growth of most pathogens found in clinical infections including *Haemophilus influenzae*, *Neisseria gonorrhoeae*, and other fastidious aerobes and anaerobes in the correct environment. Microorganisms that grow on CHOC and incubated in 5% - 10% CO₂ are not obligate anaerobes and findings should correlate with the microorganisms identified in the aerobic culture and direct Gram stain.

Limitations

CHOC agar will not provide complete information for identification of bacterial isolates. Additional test procedures and media are required for complete identification. It is important to note that some anaerobic microorganisms can be aerotolerant, such as certain species of *Clostridium*, *Actinomyces*, *Propionibacterium*, *Bifidobacterium*, and *Lactobacillus*. These aerotolerant microorganisms typically grow well in anaerobic conditions but poorly in carbon dioxide or air. Consult reference materials for additional information.

Quality Control

The following organisms are routinely used for quality assurance performance testing at Anaerobe Systems.

Organism Tested	ATCC #	Results in 5% CO ₂	Results in anaerobic conditions	Time
<i>Fusobacterium necrophorum</i>	25286	No Growth	Growth	24 – 48 hrs.
<i>Neisseria gonorrhoeae</i> *	43069	Growth	Inhibited Growth	48 hrs.
<i>Haemophilus influenzae</i> *	10211	Growth	Growth	24 – 48 hrs.
<i>Streptococcus pneumoniae</i>	49619	Growth	Growth	24 – 48 hrs.
<i>Staphylococcus aureus</i>	25923	Growth	Growth	24 – 48 hrs.
<i>Escherichia coli</i>	25922	Growth	Growth	24 hrs.

* Organisms specified by CLSI: M22-A3 for quality control performance testing of Chocolate Agar.

User Quality Control: The final determination to the extent and quantity of user laboratory quality control must be determined by the end user.

If sterility testing is to be performed on this product, a representative sample of the lot(s) should be incubated anaerobically and aerobically for 48 – 96 hours.

If the nutritive capacity of this media is to be tested for performance, it is recommended that the following ATCC organisms be evaluated for growth.

Organism	ATCC #	Expected Growth
F. necrophorum	25286	24 hours*
N. gonorrhoeae	43069	24 hours
H. influenzae	10211	24 hours
S. pneumoniae	49619	24 hours

*Growth in anaerobic conditions only.

Physical Appearance: CHOC agar should appear opaque and brown in color.

References

1. Murray, R. P., et al. 1999. *Manual of Clinical Microbiology*. American Society for Microbiology Publishing, Washington, D.C. 20005.
2. Finegold, S. M., and E. J. Baron. 1986. *Bailey and Scotts Diagnostic Microbiology*. C. V. Mosby Co., St.Louis, MO.
3. Jousimies-Somer, H. R., Summanen, P., Citron, D. M., Baron, E. J., Wexler, H. M. and S. M. Finegold. 2002. *Wadsworth – KYL Anaerobic Bacteriology Manual*. Star Publishing Co., Belmont, CA 94002.
4. CLSI. *Quality Control for Commercially Prepared Microbiological Culture Media; Approved Standard- Third Edition*. (2004). CLSI document M22-A3. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.
5. CLSI. *Principles and Procedures for Detection of Anaerobes in Clinical Specimens; Approved Guideline*. (2014). CLSI document M56-A. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.

Revision Date: 06/27/18