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TECHNICAL PRODUCT INFORMATION

Anaerobic Media

Catalog No.	Media		Catalog No.	Media
P5010	Anaerobic BA (CDC Formulation)		P5050	K-V Laked Anaerobic Agar
P5011	K-V Anaerobic Blood Agar		P5120	K-V Laked Anaerobic/CNA Anaerobic Agar
P5060	Anaerobic Lecithin Lactose Agar		P5015	Bacteroides Bile Esculin
P5020	Columbia Anaerobic Agar		P1658	TSC Agar
P5030	CNA Anaerobic Agar			

INTENDED USE:

MEDIUM	GENERAL USE	COMMENTS
CDC Anaerobic Blood Agar	Cultivation of anaerobes	An enriched non-selective medium for isolation of a wide variety of obligatory anaerobic organisms, especially ones found in clinical specimens.
Columbia Anaerobic Agar	Cultivation of anaerobes	Addition of hemin and Vitamin K, plus 5% Sheep Blood, to a general purpose medium supplies growth factors needed by certain strains of *Bacteroides, Clostridium, Fusobacterium* and *Actinomyces.*
CNA Anaerobic Blood Agar's	Selective isolation of gram positive anaerobes	Addition of Colistin and Nalidixic Acid suppresses growth of gram negative anaerobes, *Proteus, Klebsiella* and *Pseudomonas* species. Certain non-spore forming anaerobic rods may grow, e.g. *Bacteroides fragilis* and Propionibacterium *acnes.*
Lecithin Lactose	Isolation of *Clostridium perfringens*	Recommended for isolation and differentiation of histotoxic clostridia from clinical specimens. Culture media containing egg yolk with the addition of milk and lactose, allows grouping of clostridia on the basis of lecithinase production, hydrolysis of casein and lactose fermentation. Neomycin sulfate is included to make the medium selective.
K-V & K-V Laked Blood Agars	Isolation of gram negative anaerobic bacilli, *Bacteroides*	Kanamycin and Vancomycin allow selective isolation of gram negative anaerobic bacilli by inhibiting facultative and obligate anaerobic gram positive organisms. The laked sheep blood improves pigmentation of the *Porphyromonas-Provotella* species. (Orange to brick red fluorescence under long wave UV light.)
Bacteroides Bile Esculin	Isolation and presumptive identification of *Bacteroides fragilis*	Gentamicin and oxgall allow selective Inhibition of facultative anaerobes and most gram negative organisms. *B. fragilis* group give positive bile esculin reactions. Include a non-selective culture medium; some strains of *B. fragilis* may be inhibited by the selective properties of the Bacteroides Bile Esculin Agar.
TSC Agar	Isolation of *Clostridium perfringens* especially in food	Perfringens Agar Base (TSC) is a nutrient medium to which egg yolk emulsion and Cycloserine has been added. After overnight incubation, both black, lecithinase positive and black, lecithinase negative colonies should be considered as presumptive *Clostridium perfringens*. Addition of Cycloserine allows for increased selectivity and specificity for *Clostridium perfringens*.

HISTORY/SUMMARY:

Anaerobic bacteria were first recognized by Pasteur when he noticed that bacteria which produced butyric acid were non-motile when exposed to air and regained their motility in the absence of air.[1]

Smith and Hungate[2] investigating the degree of sensitivity of anaerobic microorganisms to oxygen found that anaerobic microorganisms vary from those that are able to grow on the surface of agar plates exposed to air, to organisms unable to grow if as little as 0.3% oxygen is present. The presence of "organic peroxides" (or peroxide-like compounds) and the redox potential (Eh) of media are important factors in the determination of whether anaerobic organisms will grow in, or on, a particular medium. The addition of egg yolk, blood, or starch to the medium will reduce the inhibitory effects of the peroxides.

Ellner[3], using Columbia Agar Base, formulated reducible anaerobic media designed to improve recovery of anaerobes with minimal difficulty. The reducing agents used were cysteine, palladium chloride and dithiothreitol.

FORMULAS:

Ingredients	Per liter of lab grade water
ANAEROBIC BLOOD AGAR (CDC FORMULATION)	
Tryptic Soy Agar	40.0 g
Yeast Extract	5.0 g
L-Cystine	400.0 mg
Hemin	5.0 mg
Vitamin K	10.0 mg
Sheep Blood	50.0 mL
Final pH 7.3 ± 0.2 at 25°C	

Ingredients	Per liter of lab grade water
ANAEROBIC LECITHIN LACTOSE AGAR	
Columbia Agar Base	40.0 g
Bromcresol Purple	25.0 mg
Neomycin Sulfate	0.15 g
Sodium Azide	0.20 g
Lecithin Emulsion	0.66 g
Final pH 6.9 ± 0.2 at 25°C	

COLUMBIA ANAEROBIC AGAR	
Columbia Agar Base	44.0 g
Hemin	5.0 mg
Vitamin K	0.5 mg
Sheep Blood	50.0 mL
Final pH 7.3 ± 0.2 at 25°C	

ANAEROBIC CNA AGAR	
Columbia CNA Agar	44.0 g
Hemin	5.0 mg
Vitamin K	0.5 mg
Sheep Blood	50.0 mL
Final pH 7.0 ± 0.2 at 25°C	

K-V LAKED ANAEROBIC AGAR	
Columbia Agar Base	44.0 g
Vitamin K	0.5 mg
Vancomycin	7.5 mg
Kanamycin	100.0 mg
Hemin	5.0 mg
Laked Sheep Blood	50.0 mL
Final pH 7.1 ± 0.2 at 25°C	

ANAEROBIC BLOOD AGAR W/KANAMYCIN & VANCOMYCIN	
Tryptic Soy Agar	40.0 g
Vitamin K[1]	0.5 mg
Hemin	5.0 mg
Vancomycin	7.5 mg
Kanamycin	100.0 mg
Final pH 7.1 ± 0.2 at 25°C	

BACTEROIDES BILE ESCULIN AGAR	
Pancreatic Digest of Casein	15.0 g
Papaic Digest of Soybean Meal	15.0 g
Sodium Chloride	5.0 g
Esculin	1.0 g
Ferric Ammonium Citrate	0.5 g
Oxgall	20.0 g
Hemin	0.01 g
Gentamicin	0.1 g
Vitamin K	0.01 g
Agar	15.0 g
Final pH 7.6 ± 0.2 at 25°C	

TSC AGAR	
Yeast Extract	5.0 g
Tryptose	15.0 g
Papiac Digest of Soybean Meal	5.0 g
Ferric Ammonium Citrate	1.0 g
Sodium Bisulfite	1.0 g
Agar	20.0 g
Cycloserine (1% Solution)	40.0 mL
Egg Yolk Emulsion	80.0 mL
Final pH 7.6 ± 0.2 @ 25°C	

PRECAUTIONS:
These media are for IN VITRO DIAGNOSTIC USE. They support the growth of pathogens and should be handled with caution by adequately trained personnel under the supervision of a microbiologist.

STORAGE:
Media should be stored at 2-8^{o}C. Adequate storage prolongs the life and quality of the product. Do not use the media beyond its expiration date.

SPECIMEN COLLECTION, STORAGE AND TRANSPORTATION:
Specimens should be transported to the laboratory without delay and protected from excessive heat and cold. If there is to be any delay in processing, a swab inoculated with the specimen should be placed in a suitable transport medium such as Amies or Stuarts. Specimens being transported for the isolation of anaerobic bacteria should be transported as soon as possible, preferably in a pre-reduced medium or gassed-out tube. The precautions will vary with the organisms being sought.

PROCEDURE:
The usual clinical microbiological equipment is required for procedures involving these products. Other media and equipment required will depend upon the identification scheme employed by the microbiologist.

Specimens for anaerobic culture should be quality controlled using those organisms known to be susceptible to the agents in the medium as well as well as those organisms each medium is designed to isolate.

PERFORMANCE CHARACTERISTICS:

Organisms	**CDC ANAEROBIC BA**	**COLUMBIA ANAEROBIC BA**	**CNA ANAEROBIC BA**	**LECITHIN LACTOSE**	**K-V LAKED**	**BACTEROIDES BILE ESCULIN**	**TSC**
Bacteriodes melaninogenicus	G	G	G	-	G	-	
Bacteroides fragilis	G	G	G	S	G	G,B	
Escherichia coli	G	G	S	S	S	-	
Peptostreptoccus anaerobius	G	G	G	-	S	-	
Clostridium perfringens	G	G	-	G,L,+A	S	NG	G,B,L+
Clostridium novyi	G	G	-	-	S	-	G
Clostridium sordellii	-	G	-	-	-	-	NG
Staphylococcus aureus	G	G	G	S	S	-	

G = Growth
NG = No Growth
L+ = Lecithinase Positive
A = Lactose Fermented
B = Blackening
S = Suppressed

QUALITY CONTROL:
It is recommended that the user confirm the performance characteristics of this medium. Careful selection of test organisms must be made so maximum information on product suitability is obtained. Proper environmental conditions must be chosen to further warrant effective results.

LIMITATIONS:
For best recovery of anaerobic organisms from clinical specimens use selective and non-selective media in parallel cultures. Ellner et al.[3] have reported that recovery rates of anaerobic organisms were lower with selective media than with non-selective media. When culturing a clinical specimen for anaerobes, it is recommended to also culture the specimen aerobically, for many times aerobic or facultative organisms may be associated with anaerobes.[1.3.4.9.]

Certain strains of organisms usually inhibited by the selective agents used might be resistant to the agents and thus grow on the medium which normally inhibits the organism. Likewise, certain strains or organisms usually resistant to the selective agents used might be sensitive to the agents and thus not grow on the medium which normally permits growth of the organism.

Since oxygen is toxic in varying degrees to anaerobes, it is essential to properly collect, transport, reduce their exposure to oxygen.

Identification of anaerobes requires additional study such as gram staining, biochemical testing, gas chromatographic procedures or immunological tests.

REFERENCES:

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2) Hungate and Smith, J. Bact., 75:713, 1958.
3) Ellner et al., App. Micro., 26:904-913, 1973.
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7) Aranki et al., App. Micro., 17:568, 1969.
8) Gibbons and MacDonald, J. Bact., 80:2164-170, 1960.
9) Dowell and Hawkins, Laboratory Methods in Anaerobic Bacteriology CDC Laboratory Manual, USD of HEW, Public Health Service Center for Disease Control, Atlanta, GA. 1974.
10) The Oxoid Manual 8th Edition, P. 2-168.