# **KANAMYCIN BILE ESCULIN (KBE) AGAR**

## **INTENDED USE**

Remel Kanamycin Bile Esculin (KBE) Agar is a solid medium recommended for use in qualitative procedures for the selective isolation and presumptive identification of the *Bacteroides fragilis* group.

### SUMMARY AND EXPLANATION

Members of the *B. fragilis* group are the anaerobic bacteria most frequently isolated in human clinical infections.<sup>1</sup> This group includes *B. fragilis*, *B. thetaiotaomicron*, *B. ovatus*, *B. distasonis*, *B. vulgatus*, and others. Selective media are recommended for isolation of anaerobes because clinical specimens for anaerobic culture frequently contain mixed flora, including facultative gram-negative rods that readily overgrow the anaerobes.<sup>2</sup> KBE Agar was formulated by Chan and Porschen for the purpose of selective isolation and presumptive identification of the *B. fragilis* group on the basis of resistance to bile and kanamycin and hydrolysis of esculin.<sup>3,4</sup>

#### PRINCIPLE

Members of the *B. fragilis* group hydrolyze esculin to produce esculetin. Ferric ammonium citrate reacts with esculetin to produce a brown-black complex in the agar surrounding the colony. The selectivity of this medium is provided by addition of kanamycin and bile. Kanamycin in a concentration of 1000  $\mu$ g/ml inhibits facultative anaerobes and also anaerobic gram-negative bacteria other than *B. fragilis* group. Bile (20%) is stimulatory to *B. fragilis* group but inhibitory to other anaerobic bacteria. Vitamin K is a growth factor. Hemin also is a growth factor and allows testing for catalase production.<sup>5</sup>

## **REAGENTS (CLASSICAL FORMULA)\***

Bile 20% (Oxgall)	20.0	g
Casein Peptone	15.0	g
Sodium Chloride	5.0	ğ
Soy Peptone	5.0	ğ
Fsculin	1.0	ă

Kanamycin	1.0	g
Ferric Ammonium Citrate	0.5	ğ
Vitamin K	.10.0	mg
Hemin	5.0	mg
Agar	. 15.0	ğ
Demineralized Water1	0.000	mĬ

pH 7.0 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

## PROCEDURE

- 1. Prior to use, reduce the plates for a minimum of 24 hours by placing them in an anaerobic environment at room temperature.
- 2. Inoculate specimens for anaerobic culture on both selective and nonselective media as soon as possible after receipt in the laboratory; streak plates for isolation.
- 3. Incubate anaerobically at 33-37°C for 48-72 hours.
- 4. Confirm anaerobic growth by subculture to an aerobic blood agar plate.

#### QUALITY CONTROL

All lot numbers of Kanamycin Bile Esculin (KBE) Agar have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

#### CONTROL

Bacteroides fragilis ATCC<sup>®</sup> 25285 Bacteroides thetaiotaomicron ATCC<sup>®</sup> 29148 Clostridium perfringens ATCC<sup>®</sup> 13124 Escherichia coli ATCC<sup>®</sup> 25922 Staphylococcus aureus ATCC<sup>®</sup> 25923

## INCUBATION

Anaerobic, up to 48 h @ 33-37°C Anaerobic, up to 48 h @ 33-37°C Anaerobic, up to 48 h @ 33-37°C Aerobic, 18-24 h @ 33-37°C Aerobic, 18-24 h @ 33-37°C

#### RESULTS

Growth, blackening of media Growth, blackening of media Inhibition (partial to complete) Inhibition (partial to complete) Inhibition (partial to complete)

#### LIMITATIONS

- Organisms other than *B. fragilis* group may grow on Kanamycin Bile Esculin (KBE) Agar (e.g., enterococci, *Fusobacterium* spp., facultative gramnegative rods, yeast, etc.). Additional testing, including a Gram stain, is required to presumptively identify isolates as members of *B. fragilis* group.<sup>3,6</sup> Consult appropriate references for further instructions.<sup>1,2</sup>
- 2. Some penicillin-resistant strains of *Bacteroides* are inhibited by 20% bile and may not grow on this medium.<sup>3,4</sup>

#### BIBLIOGRAPHY

- 1. Murray, P.R., E.J. Baron, J.H. Jorgensen, M.L. Landry, and M.A. Pfaller. 2007. Manual of Clinical Microbiology. 9th ed. ASM Press, Washington, D.C.
- 2. Dowell, V.R., Jr. G.L. Lombard, F.S. Thompson, and A.Y. Armfield. 1977. CDC Laboratory Manual. U.S. Dept. of H.H.S., CDC, Atlanta GA.
- 3. Chan, P.C.K. and R.K. Porschen. 1977. J. Clin. Microbiol. 6:528.
- 4. Draper, D.L. and A.L. Barry. 1977. J. Clin. Microbiol. 5:439.
- 5. Wilkins, T.D., S.L. Chalgren, F. Jimenez-Ulate, C.R. Drake, Jr., and J.L. Johnson. 1976. J. Clin. Microbiol. 3:359-363.
- 6. Vargo, V., M. Korzeniowski, and E.H. Spaulding. 1974. J. Clin. Microbiol. 27:480-483.

Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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