

BRUCELLA AGAR LKV

INTENDED USE

Remel Brucella Agar LKV is a solid medium recommended for use in qualitative procedures for selective primary isolation of gram-negative anaerobic microorganisms.

SUMMARY AND EXPLANATION

Brucella Agar, originally developed for the cultivation of *Brucella* spp., is prepared according to the formula of the American Public Health Association.¹ Finegold et al. reported Brucella Agar base supplemented with whole or laked blood supports the growth of fastidious anaerobic bacteria better than Wilkins-Chalgren Agar supplemented with blood.² Brucella Agar LKV also contains menadione (Vitamin K) and hemin for added enrichment.³ Ellner formulated reducible anaerobic media by adding cysteine, palladium chloride, and dithiothreitol to Brucella Agar.⁴ Reducible agar improves the recovery of anaerobes by lowering the oxidation-reduction potential of the medium.

PRINCIPLE

Brucella Agar LKV is a nutritious medium which contains peptone, hemin, vitamin K, and dextrose to support the growth of anaerobic organisms. The oxidation-reduction potential (Eh), or reducing tendency, is the measure of a medium's capacity to release electrons. Reducing agents such as cysteine, palladium chloride, and dithiothreitol reduce the Eh of a medium. Sodium chloride is a source of essential electrolytes and maintains osmotic equilibrium. Sodium bisulfite is a reducing substance which helps maintain reduced conditions and a low pH. Kanamycin and vancomycin allow for selective isolation of certain anaerobes by inhibiting gram-positive and facultative anaerobic bacteria. Sheep blood provides growth factors and other enriching agents. Agar is a solidifying agent.

REAGENTS (CLASSICAL FORMULA)*

Casein Peptone.....	15.0 g	Sodium Bisulfite.....	0.1 g
Meat Peptone.....	5.0 g	Kanamycin.....	100.0 mg
Sodium Chloride.....	5.0 g	Vitamin K.....	10.0 mg
Yeast Extract.....	2.0 g	Vancomycin.....	7.5 mg
Dextrose.....	1.0 g	Hemin.....	5.0 mg
Cysteine HCl.....	0.5 g	Laked Sheep Blood.....	5 %
Palladium Chloride.....	0.33 g	Agar.....	15.0 g
Dithiothreitol.....	0.1 g	Demineralized Water.....	1000.0 ml

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 non-selective media.
3. Streak the specimen as soon as possible after it is received in the laboratory.
4. If material is being cultured directly from a swab, roll the swab over a small area of the agar surface and streak for isolation.
5. Incubate anaerobically at 33-37°C for up to 72 hours.
6. Confirm anaerobic growth by Gram stain and subculture to a blood agar plate incubated in ambient air..

QUALITY CONTROL

All lot numbers of Brucella Agar LKV have been tested using the following quality control organisms and have been found to be acceptable. This quality control testing meets or exceeds CLSI standards.⁵ 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® 25285
**Prevotella melaninogenica* ATCC® 25845
Escherichia coli ATCC® 25922
Staphylococcus aureus ATCC® 25923

*CLSI recommended organism

INCUBATION

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

RESULTS

Growth
Growth
No Growth
No Growth

BIBLIOGRAPHY

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3. Summanen, P., E.J. Baron, D.M. Citron, C.A. Strong, H.M. Wexler, and S.M. Finegold. 1993. Wadsworth Anaerobic Bacteriology Manual. 5th ed. Star Publishing Co., Belmont, CA.
4. Ellner, P.D., P.A. Granato, and C.B. May. 1973. Appl. Microbiol. 26:904-913.
5. Clinical and Laboratory Standards Institute. 2004. Quality Assurance for Commercially Prepared Microbiological Culture Media; Approved Standard, 3rd ed. M22-A3. CLSI, Wayne, PA.

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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