

V AGAR and V AGAR SELECTIVE

INTENDED USE

Remel V Agar and V Agar Selective are solid media recommended for use in qualitative procedures for the isolation and presumptive identification of *Gardnerella vaginalis*.

SUMMARY AND EXPLANATION

Gardner and Duker first isolated *G. vaginalis* from women with nonspecific vaginitis in 1955.¹ Dunkelberg et al. developed buffered peptone starch dextrose (PSD) agar which, unlike previously described media, did not contain blood or carbohydrates.² PSD agar is transparent, allowing observation of colony morphology by transmitted light which facilitates differentiation of *G. vaginalis* colonies from those of commensal microbial flora (e.g., streptococci, yeasts, lactobacilli, etc.).³ In 1976, Golberg et al. compared PSD Agar and Columbia Agar (first described by Ellner et al.) supplemented with colistin and nalidixic for isolation of *G. vaginalis*.^{4,5} Of 447 cervical or vaginal specimens inoculated in parallel, 100 *G. vaginalis* isolates were recovered. Colonies of *G. vaginalis* on Columbia colistin-nalidixic agar were found to be at least twice the diameter of those isolated on PSD. In 1978, Greenwood et al. introduced "V" (Vaginalis) Agar, a Columbia agar base which contained 5% whole human blood.⁶ They evaluated 78 strains of *G. vaginalis* for various characteristics including ability to produce beta-hemolysis on human blood agar and sheep blood agar.⁷ More than 90% of the strains tested were beta-hemolytic on human blood agar; none were beta-hemolytic on sheep blood agar.

PRINCIPLE

Casein and meat peptones supply nitrogenous compounds, carbon, sulfur, and trace elements necessary for the growth of *G. vaginalis*. Cornstarch is an energy source. Human blood is incorporated into the agar to allow for presumptive identification of *G. vaginalis*, which produces diffuse beta-hemolysis on V Agar and V Agar Selective. Colistin, nalidixic acid, and nystatin are selective agents which inhibit most gram-negative bacilli, yeast, and diphtheroids.

REAGENTS (CLASSICAL FORMULAE)*

Casein Peptone.....	12.0 g	Yeast Extract	3.0 g
Meat Peptone.....	5.0 g	Cornstarch	1.0 g
Sodium Chloride.....	5.0 g	Human Blood.....	5 %
Beef Extract.....	3.0 g	Agar	13.5 g
		Deminerlized Water.....	1000.0 ml

pH 7.3 ± 0.2 @ 25°C

The following ingredients are available per liter of medium:

Colistin	10.0 mg	Nystatin.....	12,500 U
Nalidixic Acid.....	10.0 mg		

pH 7.2 ± 0.2 @ 25°C

*Adjusted as required to meet performance standards.

PRECAUTIONS

WARNING! These media contains human blood. Each unit of blood used in the preparation of this product was tested by FDA-licensed procedures and found to be nonreactive for the presence of antibodies to human immunodeficiency virus (anti-HIV), hepatitis B surface antigen (HBsAg), antibody to hepatitis B core antigen (anti-HBc), antibody to hepatitis C virus (anti-HCV), and antibody to human T-cell lymphotropic virus, type 1 (anti-HTLV-1). Because no test can guarantee the absence of every infectious agent, all human specimens should be considered potentially infectious and handled accordingly. Information on handling human blood is provided in the CDC/NIH manual, *Biosafety in Microbiology and Biomedical Laboratories*.⁸

PROCEDURE

1. Inoculate and streak the specimen as soon as possible after it is received in the laboratory.
2. If specimen is being cultured directly from a swab, roll the swab over a small area of the agar surface and streak for isolation.
3. Incubate in 5-10% CO₂ at 33-37°C for 24-48 hours.
4. Examine plate for typical colony morphology. On V Agar and V Agar Selective, *G. vaginalis* produces small, opaque colonies surrounded by a diffuse zone of beta-hemolysis.

QUALITY CONTROL

All lot numbers of V Agar and V Agar Selective 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

V Agar:

Gardnerella vaginalis ATCC®14018

V Agar Selective:

Gardnerella vaginalis ATCC®14018

Candida albicans ATCC®10231

Escherichia coli ATCC®25922

Proteus mirabilis ATCC®12453

INCUBATION

CO₂, 24-48 h @ 33-37°C

CO₂, 24-48 h @ 33-37°C

Aerobic, 18-24 h @33-37°C

Aerobic, 18-24 h @33-37°C

Aerobic, 18-24 h @33-37°C

RESULTS

Growth, small colonies with beta hemolysis

Growth, small colonies with beta hemolysis

Inhibition (partial to complete)

Inhibition (partial to complete)

Inhibition (partial to complete)

LIMITATIONS

1. Obligately anaerobic strains of *G. vaginalis* have been reported; however, studies have demonstrated that anaerobic isolates occur with such infrequency that routine anaerobic incubation of genital cultures is unwarranted.⁹
2. Organisms other than *G. vaginalis* may grow on V Agar and V Agar Selective. Additional tests required for presumptive identification include: a Gram-stain demonstrating characteristic morphology (i.e., gram-positive, gram-negative, or gram-variable coccobacilli) and negative catalase and oxidase.¹⁰

BIBLIOGRAPHY

1. Gardner, H.L. and C.D. Dukes. 1955. Am. J. Obstet. Gynecol. 69:962-976.
2. Dunkelberg, W.E., Jr. and I. McVeigh. 1969. Antonie van Leeuwenhoek. J. Microbiol. Serol. 35:129-145.
3. Dunkelberg, W.E., Jr., R. Skaggs, and D.S. Kellogg, Jr. 1970. Appl. Microbiol. 19:47-52.
4. Ellner, P.D., C.J. Stoessel, E. Dukeford, and F. Vasi. 1966. Am. J. Clin. Pathol. 45:502-504.
5. Golberg, R.L. and J.A. Washington. 1976. J. Clin. Microbiol. 4:245-247.
6. Greenwood, J.R., M.J. Pickett, W.J. Martin, and E.G. Mack. 1977. Health Lab. Sci. 14:102-106.
7. Greenwood, J.R. and M.J. Pickett. 1979. J. Clin. Microbiol. 9:200-204.
8. Richmond, J.Y. and R.W. McKinney. 1999. Biosafety in Microbiological and Biomedical Laboratories. 4th ed. U.S. Dept. of H.H.S., CDC and National Institutes of Health, Washington, D.C.
9. Bailey, R.K., J.L. Voss, and R.F. Smith. 1979. J. Clin. Microbiol. 9:65-71.
10. Winn, W.C., Jr., S.D. Allen, W.M. Janda, E.W. Koneman, G.W. Procop, P.C. Schreckenberger, and G.L. Woods. 2006. Koneman's Color Atlas and Textbook of Diagnostic Microbiology. 6th ed. Lippincott Williams and Wilkins, Philadelphia, 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.

ATCC® is a registered trademark of American Type Culture Collection.

IFU 1950, Revised February 11, 2008

Printed in U.S.A.

remel

12076 Santa Fe Drive, Lenexa, KS 66215, USA

General Information: (800) 255-6730 Technical Service: (800) 447-3641 Order Entry: (800) 447-3635

Local/International Phone: (913) 888-0939 International Fax: (913) 895-4128

Website: www.remel.com Email: remel@remel.com