

Instructions for Use

VOGES-PROSKAUER (VP) TEST REAGENTS

Cat. no. Z91	Voges-Proskauer Reagent A	15ml
Cat. no. Z92	Voges-Proskauer Reagent B	15ml

INTENDED USE

Hardy Diagnostics Voges-Proskauer Reagents are recommended for use in determining the Voges-Proskauer (VP) reaction of bacteria.

SUMMARY

Certain bacteria produce stable acid end products when cultivated in specific media. After fermentation of glucose, particular enteric bacteria metabolize pyruvic acid to acetylmethyl carbinol. This end product reacts with alpha-naphthol in the presence of 40% potassium hydroxide to produce a red color complex.

REAGENT FORMULA

Ingredients per liter:*

Voges-Proskauer Reagent A:	
Alpha-Naphthol, 5%	50.0gm
Absolute Ethanol	1000.0ml

Voges-Proskauer Reagent B:	
Potassium Hydroxide	400.0gm
Deionized Water	1000.0ml

* Adjusted and/or supplemented to meet performance criteria.

STORAGE AND SHELF LIFE

Upon receipt store at 2-30°C. Products should not be used if there are any signs of contamination, deterioration, or if the expiration date has passed. Do not expose to excessive heat or moisture.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "[Storage](#)" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual Universal Precautions for blood. Do not ingest, inhale, or allow to come into contact with skin.

This product is for *in vitro* diagnostic use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "[Guidelines for Isolation Precautions](#)" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections*.

Sterilize all biohazard waste before disposal.

Refer to the document "[Precautions When Using Media](#)" for more information.

Sterilize all biohazard waste before disposal.

Warning! Voges-Proskauer Reagent A can cause irritation.

Warning! Voges-Proskauer Reagent B is extremely caustic; it will cause burns.

Avoid contact with skin. If contact occurs, rinse thoroughly with copious amounts of water.

PROCEDURE

Specimen Collection: This product is not intended for primary isolation of patient specimens. It should be used in conjunction with other biochemical tests to identify cultures of isolated organisms.

Method of Use: Inoculate MR-VP (Cat. no. K37) medium with the test organism. Incubate 24 to 48 hours at 35°C. Aseptically transfer 1ml of the incubated broth to a clean test tube. Add 15 drops of Voges-Proskauer Reagent A followed by 5 drops of Voges-Proskauer Reagent B. Shake gently to aerate. Examine for the appearance of a red color within 20 minutes. If the 24 hour test is negative, repeat the test with a 48 hour culture of the test organism. If equivocal results are obtained, repeat the test with cultures incubated for 5 days at 25-30 degrees C.

If VP Reagents are to be used with commercial identification tests, such as API[®], Microgen[™] GnA+B-ID System, Enteropluri-Test, consult the manufacturer's literature.

INTERPRETATION OF RESULTS

Positive reactions are denoted by the development of a red color within 20 minutes. Negative reactions remain yellow to amber. Development of a copper color should be considered negative. Hold negatives for an additional 45 minutes since the maximum color development occurs 1 hour after the reagents are added. Do not read the test after more than 1 hour of incubation.

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification of bacteria and/or fungi.

The Voges-Proskauer test may be used in the identification of gram-negative bacteria. Additional biochemical testing

using pure culture is recommended for complete identification.

Prolonged incubation (greater than 72 hours at 35°C.) of certain VP-positive strains of *Enterobacteriaceae* may result in false-negative reactions due to the breakdown of acetylmethyl carbiol.

The order of the addition of reagents is extremely important. Reversal of the order may result in weak positive reactions or false-negative reactions.

Refer to the document "[Limitations of Procedures and Warranty](#)" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, needles, incubators, and incinerators, etc., as well as biochemical and serological reagents, are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Reaction
<i>Enterobacter cloacae</i> ATCC® 23355	Positive VP; red color change
<i>Escherichia coli</i> ATCC® 25922	Negative VP; no color change

USER QUALITY CONTROL

It is recommended that each new lot or shipment of reagent be tested with known positive and negative controls.^(3,4)

PHYSICAL APPEARANCE

- Voges-Proskauer Reagent A should appear clear, and reddish-brown in color.
- Voges-Proskauer Reagent B should appear clear and colorless.



Escherichia coli (ATCC® 25922) grown in MR-VP Broth (Cat. no. K37). Incubated aerobically for 24 hours at 35°C. A 1mL aliquot was removed and 0.6mL of alpha-naphthol (Cat. no. Z91) and 0.2mL of 40% KOH (Cat. no. Z92) was added to the tube. No



Enterobacter cloacae (ATCC® 23355) grown in MR-VP Broth (Cat. no. K37). Incubated aerobically for 24 hours at 35°C. A 1mL aliquot was removed and 0.6mL of alpha-naphthol (Cat. no. Z91) and 0.2mL of 40% KOH (Cat. no. Z92) was added to the tube.

development of a pink-red color within 10-60 minutes was indicative of a negative VP reaction.

Development of a pink-red color within 10-60 minutes was indicative of a positive VP reaction.

REFERENCES

1. Versalovic, J., et al. *Manual of Clinical Microbiology*. American Society for Microbiology, Washington, D.C.
2. Tille, P., et al. *Bailey and Scott's Diagnostic Microbiology*, C.V. Mosby Company, St. Louis, MO.
3. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.
4. Isenberg, H.D. *Clinical Microbiology Procedures Handbook*, Vol. I, II & III. American Society for Microbiology, Washington, D.C.
5. MacFaddin, J.F. *Biochemical Tests for Identification of Medical Bacteria*, Lipincott Williams & Wilkins, Philadelphia, PA.

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Microgen™ GnA+B-ID System is a trademark or Microgen® Bioproducts, U.K.

IFU-10844[A]



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[Ordering Information](#)

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