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# **ProductInformation**

Mutanolysin Assay Substrate, Streptococcus Cell Walls

Product Number **M 3440** Storage Temperature –20 to 0 °C

Synonym: Streptococcus cell wall suspension

### **Product Description**

The Mutanolysin Assay Substrate is a lyophilized suspension of washed *Streptococcus faecalis* STF-3 (ATCC 12784)<sup>1</sup> cell walls. The activity of the cell wall hydrolyzing enzyme, mutanolysin (Product Nos. M 9901 and M 4782), can be determined in a continuous spectrophotometric assay using the resuspended cell walls as a substrate.<sup>2</sup>

One vial provides enough substrate for ten assays following the Sigma enzymatic assay of mutanolysin.

The product is lyophilized from a suspension in 50 mM MES buffer, pH 6.0, with 1 mM MgCl<sub>2</sub> • 6H<sub>2</sub>O.

## **Precautions and Disclaimer**

This product is for laboratory research use only. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

# **Preparation Instructions**

Prepare the Substrate Suspension by diluting one vial with 10 ml of assay buffer to an absorbance at 600 nm ( $A_{600}$ ) of 0.45 to 0.65. The assay buffer is 50 mM MES, pH 6.0, with 1 mM MgCl<sub>2</sub> • 6H<sub>2</sub>O. Keep suspension on ice until use.

# Storage/Stability

The product ships on wet ice and it is recommended to store the product at –20 to 0 °C. The lyophilized product remains suitable for the determination of mutanolysin activity for at least 12 months when stored properly.

The resuspended product should be kept on ice prior to use. The resuspended material may divided into aliquots and frozen. This material should be used within 3 months. Repeated thawing and freezing of the resuspended product is **not** recommended.

#### **Procedure**

Transfer 1 ml of the Substrate Suspension into a cuvette and place in a spectrophotometer thermostatted at 37 °C. Monitor the  $A_{600}$  until constant. Dilute the mutanolysin sample to approximately 200 units per ml. Add the diluted sample (20 to 40  $\mu$ l) to the cuvette and mix by inversion. Determine the maximum linear rate ( $A_{600}$ /minute) and calculate the enzyme activity.

Note: The complete Sigma QC procedure for the assay of mutanolysin may be viewed on our website (<a href="www.sigmaaldrich.com">www.sigmaaldrich.com</a>). It may be accessed by clicking on the QC Methods in the product information section of the website listings for M 3440 or for mutanolysin (Product Nos. M 9901 and M 4782).

### References

- American Type Culture Collection, Manassas VA, U.S.A.
- 2. Calandra, G.B., and Cole, R.M., Infection and Immunity, **28**, 1033-1037 (1980).
- 3. Siegal, J.L. et al., Infect. Immun., **31**, 808-815 (1981).

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