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ProductInformation

2-Nitrophenyl b-D-galactopyranoside

Product Number **N1127** Storage Temperature -20 °C

Product Description

Molecular Formula: $C_{12}H_{15}NO_8$ Molecular Weight: 301.3 CAS Number: 369-07-3 Melting Point: 193-194 °C ¹ Specific Rotation: -67° (1 g/100 ml water at 25 °C) Synonym: ONPG

This product (ONPG) is a colorimetric substrate for β -galactosidase. This product is hydrolyzed to o-nitrophenol by the enzyme. o-Nitrophenol is yellow in alkaline solution and can be quantitated at 410-420 nm.² The molar extinction coefficient is 2.13 x 10⁴ at pH 10.2.³ We have used absorbance readings at 410 nm in the determination of the activity of β -galactosidase using this product as the substrate. The K_m for β -galactosidase (from *E. coli*) is 9.5 x 10⁻⁴ M in Tris buffer, pH 7.6, at 20 °C.⁴ The V_{max} for β -galactosidase for this substrate is 134 µmoles/min/mg protein.⁵

Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

Preparation Instructions

A stock solution of this product can be prepared in deionized water at a concentration of 3 mg/ml.

Alternatively, a stock solution may be prepared in phosphate buffer. When used as a substrate for β -galactosidase, a solution of approximately 20.5 mg/ml is prepared in 100 mM sodium phosphate buffer, pH 7.3. The solution is gently warmed to completely dissolve the product.

References

- 1. J. Am. Chem. Soc., 75, 1758 (1953).
- Molecular Cloning: A Laboratory Manual, 2nd ed., 3, Sambrook, J., et al., Cold Spring Harbor Laboratory (1989), p. 1666.
- 3. Adv. Carbohydr. Chem., 16, 239 (1961).
- 4. Enzyme Handbook, **II**, Barman, T. E., Springer-Verlag (Berlin-Heidelberg: 1969), p. 581.
- 5. Wallenfels, K., β-Galactosidase (Crystalline). Methods in Enzymology, **5**, 212-218 (1962).

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