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# **ProductInformation**

## **Phosphatase Substrates**

Capsules

Storage Temperature: -0 °C

Preweighed 40 mg Capsules, Prod. No. P 5744 Preweighed 100 mg Capsules, Prod. No. P 5869

Tablets

Storage Temperature: -0 °C 5 mg Tablets, Prod. No. S 0942 40 mg Tablets, Prod. No. S 1067

Powder

Storage Temperature: -0 °C

Powdered Substrate, Prod. No. P 4744

C<sub>6</sub>H<sub>4</sub>NO<sub>6</sub>PNa<sub>2</sub>•6H<sub>2</sub>O

FW 371.1

# **Product Description**

p-Nitrophenyl Phosphate (pNPP) is a soluble substrate for use with alkaline phosphatase in ELISA procedures. It may also be used for the determination of alkaline and acid phosphatase activity in physiological fluids and other aqueous solutions. This substrate produces a soluble end product that is yellow in color and can be read spectrophotometrically at 405 nm. The pNPP reaction may be stopped with 3M NaOH and read at 405 nm.

#### **Preparation Instructions**

Dissolve tablets or contents of capsules in either 0.1 M glycine buffer containing 1 mM  $MgCl_2$  and 1 mM  $ZnCl_2$ , pH 10.4 **or** 1M diethanolamine buffer containing 0.5 mM  $MgCl_2$ , pH 9.8, to the desired concentration (typically a pNPP concentration of 1 mg/ml is used).

To prepare 0.1 M glycine buffer, 1 mM MgCl<sub>2</sub>, 1 mM ZnCl<sub>2</sub>, pH 10.4: Add 7.51 g of glycine (Product No.

## p-Nitrophenol (Hydrolysis Product) Standards

p-Nitrophenol, Prod. No. N 0286 Storage Temperature: RT

p-Nitrophenol 10 mM Solution, Prod. No. N 7660 Storage Temperature: 2 to 8 °C

C<sub>6</sub>H<sub>5</sub>NO<sub>3</sub> FW 139.1

G 7126), 203 mg MgCl $_2$  (Product No. M 0250) and 136 mg ZnCl $_2$  (Product No. Z4875) to approximately 980 ml dH $_2$ O and mix. Adjust the pH to 10.4 with 19 N NaOH and adjust the volume to 1 L with dH $_2$ O.

To prepare 1 M diethanolamine buffer 0.5 mM MgCl $_2$ , pH 9.8: Add 97 ml diethanolamine (Product No. D 8885), 100 mg MgCl $_2$  (Product No. M 0250) and 0.2 g sodium azide (Product No. S 2002) to 800 ml dH $_2$ O, adjust the pH to 9.8 with 10 M HCl and adjust the volume to 1 L with dH $_2$ O.

The reaction may be stopped by the addition of 50  $\mu$ l of 3 N NaOH per 200  $\mu$ l of reaction mixture.

p-Nitrophenol standard solutions can be prepared in 0.02 to 1 N sodium hydroxide.

RG 1/03