

PNPP - phosphatase substrate

Catalog Numbers 34045, 34047, 34064, and 37620

Doc. Part No. 2160338 Pub. No. MAN0011219 Rev. B.0

 **WARNING!** Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from [thermofisher.com/support](https://www.thermofisher.com/support).

Product description

Thermo Scientific™ PNPP (*p*-nitrophenyl phosphate disodium salt) is a widely used substrate for detecting alkaline phosphatase in ELISA-based applications. The reaction produces a water-soluble yellow product that absorbs light at 405 nm.

Contents and storage

Product	Cat. No.	Amount	Storage
Phosphatase Substrate Kit: <ul style="list-style-type: none"> Diethanolamine Substrate Buffer (5X) (5.1 M diethanolamine, pH 9.8) PNPP Tablets: Each tablet contains 5 mg of <i>p</i>-nitrophenyl phosphate disodium salt with a balance of inert filler 	37620	Buffer: 225 mL Tablets: 105	Buffer: 4°C Tablets: -20°C
Diethanolamine Substrate Buffer (5X) (5.1 M diethanolamine, pH 9.8)	34064	225 mL	4°C
PNPP Tablets: Each tablet contains 5 mg of <i>p</i> -nitrophenyl phosphate disodium salt with a balance of inert filler	34047	105 tablets	-20°C
PNPP	34045	25 g	4°C

The tablets are shipped on wet ice, and the buffer is shipped at ambient temperature.

Use PNPP phosphatase substrate in an ELISA

 **CAUTION!** Avoid PNPP contact with eyes, skin, and clothing. For handling information, see the MSDS.

1. Equilibrate PNPP to room temperature before opening.
2. Dilute the Diethanolamine Substrate Buffer from 5X to 1X.
3. Dissolve 10 mg of PNPP or two PNPP Tablets in 10 mL of 1X Diethanolamine Substrate Buffer.
Note: Diethanolamine is effective at concentrations from 10 mM to 1 M.
4. Add 100 µL of the PNPP solution to each microplate well.
5. Incubate plate at room temperature for 30 minutes, or until sufficient color develops.
6. Add 50 µL of 2 N NaOH to each well to stop the reaction.
7. Measure the absorbance at 405 nm. Alternatively, monitor reaction in a kinetic ELISA reader.

Related products

Product	Cat. No.
EZ-Link™ Maleimide Activated Alkaline Phosphatase Kit	31493
Reagent Reservoirs, 200/pkg	15075
Microtube Racked System, 960 tubes	15082
Sealing Tape for 96-Well Plates, 100/pkg	15036
1-Step™ PNPP, 100 mL	37621
Streptavidin, Alkaline Phosphatase Conjugated, 3 mg	21323
NeutrAvidin™ Protein, Alkaline Phosphatase Conjugated, 2 mg	31002

Limited product warranty

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For descriptions of symbols on product labels or product documents, go to thermofisher.com/symbols-definition.

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