## **INSTRUCTIONS**



# 1-Step PNPP

37621 0755.2

Number Description

**1-Step PNPP**, 100mL, contains a non-mercury, non-azide preservative

**Storage:** Upon receipt store product at 4°C. Product is shipped at ambient temperature.

### Introduction

Thermo Scientific 1-Step 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 405nm. 1-Step PNPP is supplied pre-mixed with substrate buffer and is ready to use.

## Procedure for using 1-Step PNPP

- 1. Equilibrate the 1-Step PNPP to room temperature.
- 2. Gently mix the 1-Step PNPP end over end.
- 3. Add 100µL of the 1-Step PNPP to each well. Mix thoroughly by gently agitating the plate.
- 4. Incubate plate at room temperature for 15-30 minutes or until sufficient color develops.
- 5. Add 50µL of 2N NaOH to stop the reaction. Mix thoroughly by gently agitating the plate.
- 6. Measure absorbance of each well at 405nm.

## **Related Thermo Scientific Products**

31493	$\mathbf{EZ} ext{-}\mathbf{Link}^{^{\mathrm{TM}}}$ Maleimide Activated Alkaline Phosphatase Kit
21323	Streptavidin, Alkaline Phosphatase Conjugated, 3mg
15075	Reagent Reservoirs, 200/pkg.
15082	Microtube Racked System, 960 tubes
15036	Sealing Tape for 96-Well Plates, 100/pkg.
37538	StartingBlock™ (PBS) Blocking Buffer, 1L
37542	StartingBlock (TBS) Blocking Buffer, 1L

#### **General References**

Feller, A.C., et al. (1983). Combined immunohistochemical staining for surface IgD and T-lymphocyte subsets with monoclonal antibodies in human tonsils. Histochem J 15:557-62.

Yolken, R.H. (1982). Enzyme immunoassays for the detection of infectious agents in body fluids: Current limitations and future prospects. *Rev Infect Dis* **4:**35-68.

Malik, N.J. and Daymon, M.E. (1982). Improved double immunoenzyme labeling using alkaline phosphatase and horseradish peroxidase. *J Clin Path* **35:**1092-4.



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