

pNPP

CHROMOGENIC SUBSTRATES

ELISA AP Substrate

Cat. no. 4400

Product Characteristics

pNPP (p-nitrophenylphosphate) is a chromogenic substrate for Alkaline Phosphatase (AP). In the presence of AP, pNPP is hydrolyzed rapidly to p-nitrophenol and inorganic phosphate. Liberated p-nitrophenol is seen as a clear yellow color that is measured at 405/620 nm.

pNPP is a ready-to-use and aqueous formulation. The buffer solution contains stabilized pNPP and appropriate stabilizing components. pNPP is highly active compared to pNPP ONE (cat. No. 4401). The substrate gives superior results due to its sensitivity, allowing very low detection limits.

Composition & Properties

Ready-to-use substrate: It is an active and stable substrate formulation. No other reagents should be added.

Working Procedure

1. The desired amount of substrate is poured into a sealed container and allowed to reach room temperature in the dark.
2. Recommended volumes are 100 µl substrate solution per microtiter well. Development time is typically 15-30 minutes. Optimal time is determined by the user and will vary according to the procedure and the incubation temperature.
3. 100 µl of 0.1 M NaOH is then added and mixed thoroughly.
4. In kinetic assays, the color is read directly at 405 nm. For endpoint assays the absorbance is read at 405/620 nm.

Tips & Tricks

- For use in hot climate countries, due to superior background characteristics, we recommend pNPP ONE (cat. no. 4401).
- Recommended stop solution is an equal volume of 0.1 M NaOH.
- In case a higher assay sensitivity is required, a 1.0 M NaOH may be used as stop solution offering an approx. increase of OD of 3%. In both cases a 1 hour stop stability is guaranteed.

Handling & Storage

- Store solution at 2-8 °C in the dark.
- Avoid exposure to light and heat.
- Re-dispense only into bottles made of High Density Polyethylene (HDPE), amber color.
- Dispensing guidelines are available upon request.

Our immunoassay solutions are environmentally conscious, creating a safer work environment while helping our customers fulfill significant regulatory requirements.

We call it ECO-TEK!



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