ECO-TEK®

SUBSTRATES

pNPP and pNPP ONE, Ready-to-use

ELISA AP Substrates
Cat. No. 4400/4401

INTRODUCTION

The pNPP* **liquid mono-component substrates** are a highly active and stable formulations utilized for measuring AP- (Alkaline Phosphatase) probe activity. The substrates give superior results due to its sensitivity, allowing very low detection limits.
The 2 products, the **pNPP** and the **pNPP ONE**, offer an activity level with a difference in OD-values of up to 10% with the same incubation time. The **pNPP** offers the highest signal, however, the **pNPP ONE** is our recommendation for use in hot climate countries due to superior background characteristics.
Making pNPP-solutions might involve dissolving the powder in buffer or diluting pNPP concentrates. The stability of these solutions is normally one day. Our ready-to-use mono-component substrates are stable up to 36 months from manufacture date at 2-8°C without loss of activity.
In the presence of Alkaline Phosphatase, pNPP is hydrolyzed rapidly to p-nitrophenol and inorganic phosphate. Liberated p-nitrophenol is measured at 405⁄620 nm.

REAGENTS

pNPP liquid substrate, ready-to-use. The solution is a solvent free buffer solution containing stabilized pNPP and appropiate stabilizing components.
Recommended stopping solution is an equal volume of 0.1 M** sodium hydroxide solution.

THE 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. Optimum 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. Kinetic absorbances are read at 405 nm,- stopped reactions at 405⁄620 nm.

RECOMMENDED HANDLING AND STORAGE

The pNPP substrates are extremely sensitive to certain handling and storage conditions. Avoid exposure to light and heat. Redispense only into bottles made of High Density Polyethylene (HDPE), amber colour, i.e. Nalgene 2004, and keep the substrate at 2-8^{0}C in the dark. (Dispensing guidelines are available upon request).

Store solutions at 2-8^{0}C. The products support room temperature for several days.
pNPP Substrate: Stable for 24 months at 2-8°C from date of manufacturing.
pNPP ONE Substrate: Stable for 36 months at 2-8°C from date of manufacturing.

ECO-TEK® BRAND NAME

The **ECO-TEK** brand name stands for environmentally-friendly products.
These products are characterized by offering the highest level of sustainability from replacing toxic components in buffer solutions to wasting less water.

** p-NitroPhenyl Phosphate*
*** a stop solution of 0.1 M* NaOH requires no harzardous labelling. 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 10%. In both cases a 1 hour stop stability is guarranteed.*

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Instruction Manual

ECO-TEK®

The ECO-TEK brand name stands for environmentally-friendly IVD components that focus on human health and offer the highest level of sustainability, e.g. removing toxic ingredients and reducing water waste.