

KPL *p*NPP Microwell Substrate System

Catalog No.	Size
5120-0056 (50-80-00)	100 Tablets + 100 mL 5X Buffer

DESCRIPTION

KPL *p*NPP Microwell Substrate System develops a soluble yellow product when reacted with alkaline phosphatase-labeled conjugates in microwell plates. It is not recommended for membrane or immunohistochemical staining assays.

FORM

5120-0056 (50-80-00) consists of the following:

- 100 (5 mg) tablets of *p*-nitrophenylphosphate (*p*-NPP)
- 100 mL KPL DEA (Diethanolamine) Buffer (5X)

STORAGE/STABILITY

Store at 2-8°C. Stable for a minimum of 1 year from date of receipt when stored at 2-8°C.

CONTENT

KPL DEA Buffer is 5M Diethanolamine. Each tablet contains 5 mg of *p*-nitrophenylphosphate (*p*NPP).

USE

Preparation:

Mix 1 mL KPL DEA Buffer with 4 mL of reagent quality water. Add 1 *p*NPP tablet and mix thoroughly to dissolve tablet. The tablet should be completely dissolved within 10 minutes. Substrate solution should be clear to slightly yellow. Warm to room temperature before use.

Recommended Substrate Volume:

Following incubation with phosphatase labeled conjugate, wash plate thoroughly. Add 100 µL freshly mixed substrate to each well. As the color develops, tap plate gently to mix.

ABSORBANCE MEASUREMENTS

Kinetic Assays:

KPL *p*NPP phosphatase substrate produces a yellow color. Read at a wavelength between 405 - 410 nm.

Endpoint Assays:

The substrate remains yellow after the addition of a stopping solution. Read at a wavelength between 405 - 410 nm.

Stopping Solution:

To stop the KPL *p*NPP reaction, add an equal volume (100 µL) of 5% EDTA solution. Additionally, KPL APstop™ Solution may be used as well.

When to Stop Substrate Reaction:

The point at which the substrate reaction is stopped is often determined by the ELISA reader being used. Many ELISA readers record adsorbance up to 2.0 O.D. The O.D. values of the plate should be monitored and the substrate stopped before positive wells are no longer recordable.

Reducing Substrate Intensity:

To reduce the intensity of the substrate reaction, it is recommended that the conjugate and/or antibodies in the immunoassay be further diluted. **Dilution of the substrate is not recommended.**

PRODUCT SAFETY AND HANDLING

This product is considered non-hazardous as defined by the Hazard Communication Standard (29 CFR 1910.1200). Avoid contact with skin and eyes. In case of contact or spillage, clean with copious amounts of water. Product may be disposed via sanitary sewer.

RELATED PRODUCTS

Cat. No.

KPL DEA Buffer	5120-0058 (50-80-02)
KPL Coating Solution	5150-0014 (50-84-00)
KPL 10% BSA	5140-0006 (50-61-00)
Diluent/Blocking Solution	
KPL Wash Solution	5150-0008 (50-63-00)
KPL BluePhos® Microwell Phosphatase Substrate	5120-0059 (50-88-00)
KPL APstop Solution	5150-0026 (50-89-00)

The product listed herein is for research use only and is not intended for use in human or clinical diagnosis.