

# PRODUCT INFORMATION & MANUAL

# Peroxidase Activity Assay Kit (Colorimetric) NBP3-24554

For research use only. Not for diagnostic or therapeutic procedures.

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Novus kits are guaranteed for 6 months from date of receipt

#### (FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS !)

#### Peroxidase (POD) Activity Assay Kit (Serum Samples)

Catalog No: NBP3-24554 Method: Colorimetric method Specification: 50Assays (Can detect 48 samples without duplication) Measuring instrument: Spectrophotometer Sensitivity: 0.5 U/mL Detection range: 0.5-300U/mL

This manual must be read attentively and completely before using this product.

If you have any problems, please contact our Technical Service Center for help.

Phone: 240-252-7368(USA) Fax: 240-252-7376(USA)

Please kindly provide us the lot number (on the outside of the box) of the kit for more efficient service.

### Application

This kit can be used to measure the POD activity in animal serum samples.

# **Product introduction**

This kit is based on the reaction of hydrogen peroxide catalyzed by peroxidase, it detects the enzymatic activity by measuring the diversification of the absorbency value at 420 nm.

### Kit component

	Specification Storage					
Reagent 1	Liquid, 60 mL $\times$ 2 vials	4℃, 12 months				
Reagent 2	Powder, 2 vials	4°C, 12 months				
Preparation of Reagent 2 application solution: Add 10 mL of double distilled water before use.						
Store the prepared solution at $4^{\circ}$ C in the dark.						
Reagent 3	Liquid, 5 mL $\times$ 1 vial 4°C, 12 months					
Preparation of Reagent 3 application solution: Dilute for 15 times with double distilled water						
before use. Measure the OD value at 240 nm (1 cm diameter cuvette, set blank with double distilled						
water)., if the OD value is about 0.4, then the Reagent 3 application solution is prepared. If the OD						
value is too high, then dilute the reagent with double distilled water. If the OD value is too low, then						
add appropriate amount of Reagent 3. (Generally, the dilution ratio is 25.)						
Reagent 4	Reagent 4Liquid, 50 mL $\times$ 1 vial4°C, 12 months					

## **Experimental instrument**

Test tube, Micropipettor, Vortex mixer, Low-speed centrifuge,  $37^{\circ}$ C water bath/gas bath, Visible range spectrophotometer (420nm)

#### **Operation steps**

	Blank tube	Sample tube				
Reagent 1 (mL)	2.4	2.4				
Reagent 2 application solution (mL)	0.3	0.3				
Reagent 3 application solution (mL)	0.2	0.2				
Double-distilled water (mL)	0.1					
Sample (mL)		0.1				
Incubate for accurately 30 min at 37°C.						
Reagent 4 (mL)	1.0	1.0				

Mix fully and centrifuge for 10 min at 3500 rpm. Take the supernatant, then measure the OD values of each tube at 420 nm wavelength with 1cm diameter cuvette and set to zero with double-distilled water.

#### **Calculation of results**

POD activity (*U/mL*)

 $= \frac{\text{OD}_{\text{Sample}} - \text{OD}_{\text{Blank}}}{12 \times \text{Colorimetric diameter (1 cm)}} \times \frac{\text{Total reaction volume } (mL)}{\text{Sampling volume } (mL)} \times 1000$ 

 $\div$  Reaction time (30 min)  $\times\,$  Dilution factor of sample before tested

# **POD Standard Curve**

# **Reagent and preparation**

Dilute the POD Standard to different concentrations with double distilled water.

# **Operation table**

	Blank tube	Standard tube					
Reagent 1 (mL)	2.4	2.4					
Reagent 2 application solution (mL)	0.3	0.3					
Reagent 3 application solution (mL)	0.2	0.2					
Double-distilled water (mL)	0.1						
POD Standard (mL)		0.1					
Incubate for accurately 30 min at 37°C.							
Reagent 4 (mL)	1.0	1.0					

# **Detection results**

OD <sub>Blank</sub>	0.113											
Activity unit	0.025	0.05	0.1	0.111	0.125	0.143	0.167	0.2	0.25	0.333	0.5	1
OD	0.217	0.309	0.447	0.47	0.498	0.529	0.562	0.6	0.634	0.673	0.712	0.735
Absolute OD	0.104	0.196	0.334	0.357	0.385	0.416	0.449	0.487	0.521	0.56	0.599	0.622

# Standard curve

