# QuantiFluo™ HRP Reagent (QFHRP-25mL)

## Liquid Substrate Reagent for Detecting Horseradish Peroxidase Activity

## DESCRIPTION

QuantiFluo™ Horseradish Peroxidase Reagent is a stable 10-Acetyl-3,7-dihydroxyphenoxazine (also called Ampliflu Red) liquid substrate reagent formulated for detecting trace amounts of horseradish peroxidase HRP activity, e.g. in ELISA. This reagent produces a highly fluorescent product ($\lambda_{ex/em}$ = 530/590nm) when reacted with $H_2O_2$ by a peroxidase. It detects as little as 0.26 pg HRP per well in standard 96-well plate assays, or 0.008 U/L (one unit will form 1.0 mg of purpurogallin from pyrogallol in 20 seconds at pH 6.0 at 20°C). The fluorescent product can also be visualized under a light source (530 ± 20 nm).

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**QuantiFluo™ HRP Reagent:** 25 mL Reagent
100 µL 3% Stabilized $H_2O_2$

The provided reagent is sufficient for 500 assays (96-well plate format).

## INSTRUCTIONS FOR USE

Prior to assay, bring reagent to room temperature. Add 1 µL 3% hydrogen peroxide per 1 mL HRP Reagent. For ELISA, use 50 µL Reagent/$H_2O_2$ per well in 96-well plate assays or 20 µL per 384-well plate. Tap plate briefly, and incubate 30 min at room temperature. If desired, stop the reaction with 10 mM KOH (56 mg KOH/100 mL $H_2O$). Read fluorescence at $\lambda_{ex/em}$ = 530/590nm.

**Handling:** this product is shipped on ice. For long-term storage, keep reagent at -20°C. Shelf life of 1 year after receipt.

**Bulk order** of any size (>125 mL) is available. Please contact info@bioassaysys.com.

## LITERATURE

1. Towne V et al (2004). Complexities in horseradish peroxidase-catalyzed oxidation of dihydroxyphenoxazine derivatives: appropriate ranges for pH values and hydrogen peroxide concentrations in quantitative analysis. Anal Biochem. 334(2):290-6.

2. Li W, Jin W. (2006). Measurement of peroxidase activity in single neutrophils by combining catalyzed-enzyme reaction and epi-fluorescence microscopy. Talanta. 70(2):251-6.

## PRECAUTIONS

Reagents are for research use only. Normal precautions for laboratory reagents should be exercised while using the reagents.

Avoid contact and inhalation. Standard laboratory safety procedures should be followed when handling this product. Safety procedures include wearing OSHA approved safety glasses, gloves and protective clothing.

If eye or skin contact occurs, immediately wash affected area with soap and copious amounts of water for at least 15 minutes. Assure adequate flushing by separating the eyelids with fingers. Call a physician to seek medical advice.

If swallowed, wash out mouth with water provided person is conscious and call a physician to seek medical advice.

If inhaled, move individual to fresh air and call a physician to seek medical advice.

## TRANSPORT INFORMATION:

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| DOT (US): | Not regulated |
| UN Number: | Not regulated |
| Class: | Not regulated |
| Packaging Group: | Not regulated |