



# Product Manual

## **HIGHDEF<sup>®</sup> black IHC chromogen (HRP)**

Catalog #: ADI-950-171

**FOR RESEARCH USE ONLY.  
NOT FOR USE IN DIAGNOSTIC PROCEDURES.**

## INTENDED USE

Substrate/chromogen in conjunction with horseradish peroxidase (HRP)-based immunostaining or *in situ* hybridization (ISH) systems. With a 1-day working stability, this substrate/chromogen system can be used in any automation system as well as manual use.

## INTRODUCTION

HIGHDEF<sup>®</sup> black IHC chromogen (HRP) is a substrate-chromogen system designed to be used for either immunohistochemistry (IHC) or ISH when utilizing horseradish peroxidase. HIGHDEF<sup>®</sup> black IHC chromogen (HRP) produces a sharply contrasting ebony black color that can easily be distinguished from other stains.

## KIT CONTENTS

Description
HIGHDEF <sup>®</sup> black IHC chromogen (HRP) Substrate Buffer
HIGHDEF <sup>®</sup> black IHC chromogen (HRP) Chromogen
Empty Mixing Bottle

## STORAGE

Store at 2-8°C away from light. Do not use beyond the expiration date stated on the label.

## WORKING SOLUTION

Aliquot 1mL of HIGHDEF<sup>®</sup> black IHC chromogen (HRP) substrate Buffer in a mixing bottle. Add one drop (~20µL) of concentrated HIGHDEF<sup>®</sup> black IHC chromogen (HRP) chromogen solution. Replace tip, mix, and allow solution to reach room temperature before using.

**Note:** *The HIGHDEF<sup>®</sup> black IHC chromogen (HRP) chromogen-substrate working solution is light sensitive and should be kept away from light as much as possible. Working solution is stable for 1 day, for optimal results prepare fresh reagent.*

## PROCEDURE

Protocol	STAINING PROCEDURE:	INCUBATION TIMES
<p>Following peroxidase incubation, wash tissue sections with wash buffer, then follow protocol of choice:</p>		
<p><b>Pre-Mix Working Solution: (Automation)</b></p>	<p>HIGHDEF<sup>®</sup> black IHC chromogen (HRP) has a working solution stability of 1 day and can be loaded directly onto instrument as a single solution. Reduce exposure to light to achieve optimal staining. Working solution is applied directly to slide.</p>	<p><b>Working Solution: 10 - 20 min</b></p>
<p><b>On Board Mixing (Automation)</b></p>	<p>Instruments that have on-board mixing capability can load the chromogen and substrate-buffer components independently. Working solution is made mixing reagents 1:50 in on-board mixing station before application to slide.</p>	<p><b>Working Solution: 10 - 20 min</b></p>
<p><b>Manual Use</b></p>	<p>Mix substrate-chromogen and buffer in a 1:50 ratio and apply directly to slide.</p>	<p><b>Working Solution: 10 - 20 min</b></p>

## COUNTERSTAIN

Counterstain with Hematoxylin or other counterstain for good contrast. Wash with distilled or de-ionized H<sub>2</sub>O followed by immuno wash buffer.

## MOUNTING

Slides should be air dried or dehydrate in alcohol or xylene (or xylene substitute), then permanently mounted (e.g. using HIGHDEF<sup>®</sup> IHC mount, Prod. no. ADI-950-261).



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## TRADEMARKS AND PATENTS

Several Enzo Life Sciences products and product applications are covered by US and foreign patents and patents pending.

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