

Product Information Sheet

Part Number: EG-5000

Introduction

EnzoGate is specifically formulated for stabilizing and maintaining the enzymatic activity of the horseradish peroxidase (HRP) enzyme and its conjugates. It is especially suitable for use in ELISA applications. In real time stability studies, EnzoGate has been shown to maintain HRP conjugate activity for up to three years when stored at 2°C - 8°C.

Preparing Working Solution of HRP Conjugate

1. Reconstitute the HRP conjugate in accordance to the manufacturer's instructions.
 - Visually check that all solids have completely dissolved.
 - The solution may be filtered through a 0.2 µm filter.
2. Dilute and adjust the reconstituted HRP conjugate in EnzoGate to the ready-to-use concentration.
 - Ready-to-use concentrations may vary slightly from buffer to buffer and from lot to lot. It is recommended to prepare and test a small batch before preparing batches in bulk.

Accelerated Study

It is recommended to perform an accelerated stability study before preparing batches in bulk or when adding other solutions to EnzoGate. The study involves monitoring the absorbance values of various control levels using the HRP conjugate prepared in EnzoGate for a specific period at 37°C.

- Using the Accelerated Study Table below, determine the desired length and testing intervals for the study. It is recommended to test the HRP conjugate in EnzoGate for at least 4 weeks.
- Prepare the HRP conjugate in EnzoGate. Prepare enough reagent to be able to test controls for the desired length of the study. It is recommended to use fresh HRP conjugate.
- Prepare enough of all of the other components that will be used in the study (controls, TMB solution, stop solution, sample diluent, microwells...). It is important to use the same reagents for the entire study.
- Compare the absorbance values of the controls from the ending and other periods of the study to the beginning of the study. Use the Accelerated Study Table to determine the stability of the HRP conjugate in EnzoGate.

Accelerated Study Table

Incubation Time at 37°C	Equivalent to 4°C
1 week	2.5 months
2 weeks	5 months
3 weeks	7.5 months
4 weeks	10 months
5 weeks	12.5 months

Accelerated Study Table. The table shows the equivalent time of being stored at 4°C, when incubated at 37°C. For example, when incubated at 37°C for 1 week, it is equivalent to being stored at 4°C for 2.5 months.

Typical Performance

Four (4) control levels ranging from low absorbance to high absorbance were assayed for a length of 37 days in an IgG detection ELISA. The HRP conjugate was prepared in EnzoGate and kept at 37°C throughout the 37 days of testing. The absorbance values for Control Level 1, Control Level 2, Control Level 3, and Control Level 4 on Day 0 were 0.088, 0.290, 0.660, and 1.786, respectively. The absorbance values for Control Level 1, Control Level 2, Control Level 3, and Control Level 4 on Day 37 were 0.105, 0.310, 0.688, and 1.759, respectively. The activity of the HRP conjugate was maintained throughout the study. The study shows excellent stability of the HRP conjugate. EnzoGate provided stability of the HRP conjugate for the equivalent of 13.2 months at 4°C with little change in enzyme activity.

Controls Tested	Day Incubated at 37°C							
	0	1	7	15	20	25	30	37
Control Level 1	0.088	0.086	0.099	0.107	0.120	0.110	0.106	0.105
Control Level 2	0.290	0.283	0.275	0.273	0.301	0.289	0.299	0.310
Control Level 3	0.660	0.640	0.650	0.658	0.735	0.704	0.692	0.688
Control Level 4	1.786	1.772	1.700	1.682	1.809	1.775	1.768	1.759

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