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Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Data Sheet
TPH1 Inhibitor Screening Assay Kit
Catalog # 72053
Size: 96 reactions

BACKGROUND: Tryptophan 5-hydroxylase 1 and 2 (TPH1 and TPH2) are enzymes that catalyze the monooxygenation of tryptophan to 5-hydroxytryptophan (5-HTP), which is subsequently converted to serotonin. By catalyzing the rate-limiting step in the biosynthesis of serotonin, these enzymes also play key roles in the signaling of other neurotransmitters downstream of serotonin synthesis, like melatonin. Additionally, TPH enzymes may play a role in immune regulation through tryptophan depletion.

DESCRIPTION: The TPH1 Inhibitor Screening Assay Kit is designed to measure TPH1 enzyme inhibition in a 96 reaction format. This fluorescence-based assay kit is especially suitable for high throughput screening applications. The procedure is quick, straightforward, and simple – just mix two solutions, incubate, add quenching solution, and measure the fluorescence.

COMPONENTS:

Catalog #	Component	Amount	Storage	
71192	TPH1, His-tag	50 µg	-80°C	***(Avoid freeze/ thaw cycles!)***
	TPH Enzyme Solution	5 ml	-80°C	
	TPH Reaction Solution	6 ml	-80°C	
	TPH Quench Solution	1 ml	-20°C	
	Black, 96 Well Plate		Room Temp	

MATERIALS REQUIRED BUT NOT SUPPLIED:
Microplate reader capable of measuring Fluorescence
Adjustable micropipettor and sterile tips

APPLICATIONS: Great for screening small molecular inhibitors for drug discovery and HTS applications.

STABILITY: At least 6 months from date of receipt when stored as directed.

REFERENCES:
1. Moran G. R., *et al. J. Biol. Chem.* 1998; **273:**12259-12266.
2. Nowak E. C., *et al., J. Exp. Med.* 2012; **209:**2127-2135.

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ASSAY PROTOCOL:

All samples and controls should be tested in duplicate.

1) Thaw **TPH Enzyme Solution** on ice. Remove only a sufficient quantity needed for the assay. Keep solution on ice. Store remaining solution in aliquots at -80°C.

2) Thaw **TPH Reaction Solution** on ice. Remove only a sufficient quantity needed for the assay. Keep solution on ice. Store remaining solution in aliquots at -80°C.

3) Keep plate cold on ice. Add 10 µl of inhibitor solution to each well labeled "Test Inhibitor." Add 10 µl of the same solution without inhibitor (Inhibitor Buffer) to each well labeled "Negative Control" and "Positive Control."

	Negative Control	Positive Control	Test Inhibitor
Test Inhibitor	-	-	10 µl
Inhibitor Buffer (no inhibitor)	10 µl	10 µl	-
Enzyme Solution	40 µl	-	-
TPH1 Solution (8 ng/µL)	-	40 µl	40 µl
Reaction Solution	50 µl	50 µl	50 µl
Total	**100 µl**	**100 µl**	**100 µl**

4) Add 40 µl of **TPH Enzyme Solution** to each well labeled "Negative Control."

5) Thaw **TPH1** enzyme on ice. Upon first thaw, briefly spin tube containing enzyme to recover full contents of the tube. Aliquot **TPH1** into single use aliquots. Store remaining undiluted enzyme in aliquots at -80°C. Note: **TPH1** is very sensitive to freeze/thaw cycles. Do not re-use thawed aliquots or diluted enzyme.

6) Dilute **TPH1** enzyme to 8 ng/µl (320 ng/reaction) in **TPH Enzyme Solution**. Keep on ice until ready to use.

7) Add 40 µl of diluted **TPH1** solution to each well labeled "Positive Control" and "Test inhibitor."

8) Initiate reaction by adding 50 µl of **TPH Reaction Solution** to each well. Remove plate from ice and incubate for 4 hours at 4°C.

9) After incubation, add 10 µl of **TPH Quench Solution** to each well. After quenching, plate can be handled at room temperature.

10) Read the fluorescent intensity in a microtiter-plate reader.

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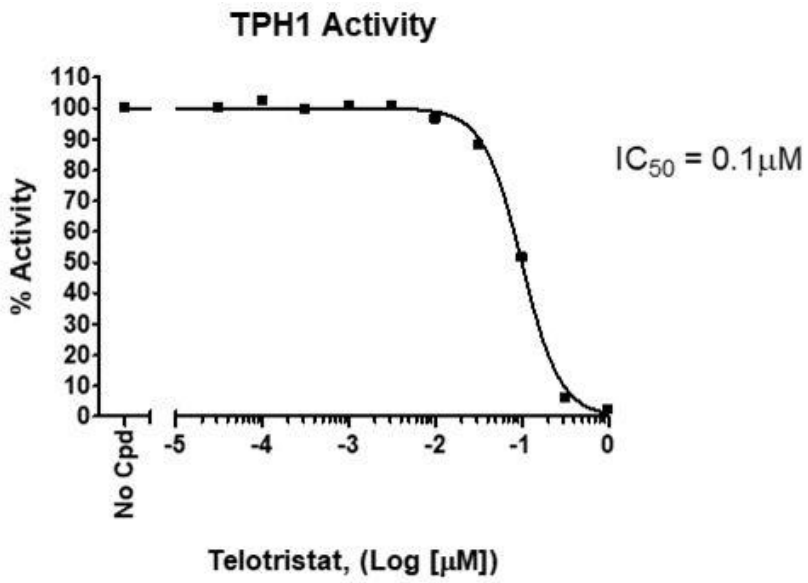
Instrument Settings

Reading Mode	λ (nm)
Excitation Wavelength	300
Emission Wavelength	360

CALCULATING RESULTS:

Subtract average "Negative Control" value from average "Positive Control" value to obtain total Δ Fluorescence. Subtract average "Negative Control" value from each "Test Inhibitor" value to obtain Δ Fluorescence of test compounds.

EXAMPLE OF ASSAY RESULTS:



Inhibition of TPH1 by Telotristat, measured using the *TPH1 Inhibitor Screening Assay Kit*, BPS Bioscience #72053. *Data shown is lot-specific. For lot-specific information, please contact BPS Bioscience, Inc. at info@bpsbioscience.com*

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RELATED PRODUCTS:

Product	Catalog #	Size
TPH1, His-tag	71192	50 µg
TPH2, His-tag	71193	50 µg
TPH2 Inhibitor Screening Assay Kit	72054	96 rxns.
Human TDO, His-tag	71195	50 µg
Mouse TDO, His-tag	71241	50 µg
Human IDO1, His-tag	71182	50 µg
Human IDO2, His-tag	71194-2	100 µg
Human IDO1 Inhibitor Screening Assay Kit	72021	96 rxns
Human IDO2 Inhibitor Screening Assay Kit	72022	96 rxns
Human TDO Inhibitor Screening Assay Kit	72023	96 rxns.