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Produktinformation

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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

USP7 Inhibitor Screening Assay Kit

Catalog # 79256
Size: 96 reactions

BACKGROUND: Deubiquitinases (DUBs) play important roles in pathways that are dysregulated in cancer, such as DNA repair, cell growth, and apoptosis. This makes DUBs attractive potential drug targets for many cancers and neurodegenerative diseases. Ubiquitin-specific-processing proteases (USPs) are the largest class of DUBs. USP7, also known as herpesvirus-associated ubiquitin-specific protease (HAUSP), acts as a tumor suppressor by stabilizing p53 tumor suppressor as well as a regulator of gene expression in herpes simplex virus (HSV) and Epstein-Barr virus (EBV) infections.

DESCRIPTION: The *USP7 Inhibitor Screening Assay Kit* is designed to measure USP7 activity for screening and profiling applications. The *USP7 Inhibitor Screening Assay Kit* comes in a convenient format, with a 96-well plate, fluorogenic, ubiquitinated substrate, assay buffer, and purified USP7 enzyme for 96 enzyme reactions. USP7 is incubated with a sample containing assay buffer and Ub-AMC Substrate, and then the plate is read, measuring fluorescence using a fluorescence reader.

COMPONENTS:

Catalog #	Component	Amount	Storage	
80395	USP7, HisFLAGtags enzyme	1 µg	-80°C	***(Avoid freeze/ thaw cycles!)***
81150	Ub-AMC Substrate	5 µl	-80°C	
79274	5x USP7 Assay Buffer*	3 x 1 ml	-80°C	
79685	black, low binding NUNC black microtiter plate	1	Room Temp.	

*****Add 13 µl of 0.5 M DTT before use.

MATERIALS REQUIRED BUT NOT SUPPLIED:
Luminometer or fluorescent microplate reader capable of reading fluorescence
Adjustable micropipettor and sterile tips
Rotating or rocker platform

APPLICATIONS: Great for studying enzyme kinetics and HTS applications.

CONTRAINDICATIONS: DMSO >1%, strong acids or bases, ionic detergents, high salt

STABILITY: One year from date of receipt when stored as directed.

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REFERENCE:

1. Pfoh, R., *et al.* Deubiquitinases and the new therapeutic opportunities offered to cancer. *Endocrine-Related Cancer*. 2015;**22(1):**T35-T54.

ASSAY PROTOCOL:

All samples and controls should be tested in duplicate.

1) Add 13 µl of 0.5 M DTT before use. Dilute **5x USP7 Assay Buffer** in distilled water to create **1x USP7 Assay Buffer**. Make only a sufficient quantity needed for the assay; store excess **5x USP7 Assay Buffer** in aliquots at -20°C.

2) Dilute **Ub-AMC Substrate** 400-fold in **1x USP7 Assay Buffer**. Add 20 µl of **Ub-AMC Substrate** to each well designated "Test Inhibitor," "Positive Control," and "Blank."

3) Add 5 µl of inhibitor solution to each well designated "Test Inhibitor." For the "Positive Control," and "Blank," add 5 µl of the same solution without inhibitor (**inhibitor buffer**). *Note: The USP7 Inhibitor Screening Assay Kit is compatible with up to 1% final DMSO concentration.*

4) Add 25 µl of **1x USP7 Assay Buffer** to the well designated "Blank."

5) Thaw **USP7 enzyme** on ice. Upon first thaw, briefly spin tube containing **USP7 enzyme** to recover full content of the tube. Aliquot **USP7 enzyme** into single use aliquots. Store remaining undiluted enzyme in aliquots at -80°C immediately. *Note: USP7 enzyme - is sensitive to freeze/thaw cycles. Avoid multiple freeze-thaw cycles.*

6) Dilute **USP7 enzyme** to 0.4 ng/µl (10 ng/25 µl) with **1x USP7 Assay Buffer.**

	Blank	**Positive Control**	**Test Inhibitor**
Ub-AMC Substrate	20 µl	20 µl	20 µl
Test Inhibitor	-	-	5 µl
Inhibitor buffer (no inhibitor)	5 µl	5 µl	-
1x USP7 Assay Buffer	25 µl	-	-
USP7 enzyme (0.4 ng/µl)	-	25 µl	25 µl
Total	**50 µl**	**50 µl**	**50 µl**

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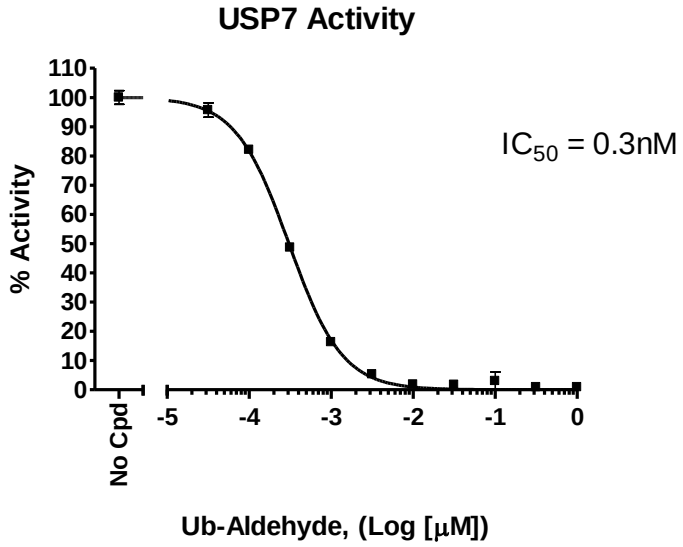
7) Initiate reaction by adding 25 µl of diluted **USP7 enzyme** (prepared as described above) to the wells labeled "Test Inhibitor" and "Positive Control". Incubate 30 minutes at room temperature on a rotating platform.

8) Immediately read sample in a luminometer or microtiter-plate reader capable of reading fluorescence intensity at 360 nm excitation and 460 nm emission. "Blank" value is subtracted from all readings.

Example of Assay Results:



Inhibition of USP7 enzyme activity by Ubiquitin Aldehyde, measured using the *USP7 Inhibitor Screening Assay Kit*, BPS Bioscience Catalog #79256. *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
USP7, His-FLAG-tags	80397	100 µg
Ubiquitin AMC	81150	50 µg
Ubiquitin-Rhodamine	81151	50 µg
USP2, FLAG-tag	80352	50 µg
USP5, FLAG-tag	80355	50 µg
USP8, FLAG-tag	88888	50 µg
USP10, FLAG-tag	80360	50 µg
USP14, FLAG-tag	80364	50 µg