上海源叶生物科技有限公司
Shanghai yuanye Bio-Technology Co., Ltd
电话：021-61312973　　传真：021-55068248
网址：www.shyuanye.com
邮箱：shyysw@sina.com

产品名称：USP7-IN-1
产品别名：USP7-IN-1

生物活性：

Description	USP7-IN-1 is a selective and reversible inhibitor of ubiquitin-specific protease 7 (USP7), with an IC_{50} of 77 μM, and can be used for the research of cancer.
IC_{50} & Target	IC50: 77 μM (USP7)[1]
In Vitro	USP7-IN-1 (Example 2) is a selective and reversible inhibitor of USP7, with an IC_{50} of 77 μM, and shows no inhibition of USP8, USP5, Uch-L1, Uch-L3 or caspase 3. USP7-IN-1 inhibits the proliferation of HCT116 cells, with a GI_{50} of 67 μM[1].
Solvent&Solubility	***In Vitro:*** **DMSO : ≥ 40 mg/mL (93.92 mM)** * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.3479 mL	11.7396 mL	23.4791 mL
	5 mM	0.4696 mL	2.3479 mL	4.6958 mL
	10 mM	0.2348 mL	1.1740 mL	2.3479 mL

*请根据产品在不同溶剂中的溶解度选择合适的溶剂配制储备液；一旦配成溶液，请分装保存，避免反复冻融造成的产品失效。

储备液的保存方式和期限：-80°C, 6 months; -20°C, 1 month。-80°C 储存时，请在 6 个月内使用，-20°C 储存时，请在 1 个月内使用。

References	[1]. SELECTIVE AND REVERSIBLE INHIBITORS OF UBIQUITIN SPECIFIC PROTEASE 7. WO 2013030218 A1

实验参考：

Cell Assay	HCT116 colon cancer cells are maintained in Mc Coy's 5A medium containing 10% FBS, 3 mM glutamine and 1 % penicillin/streptomycin. Cells are incubated at 37°C in a humidified atmosphere containing 5% CO_2. Cell viability is assayed using the MTS technique in 96-well culture plates. MTS (3-(4,5-dimethyl-thiazol-2-yl)-5-(3-carboxy- methoxyphenyl)-2-(4-sulfophenyl)-2H-tetra-zolium) is a MTT-derived tetrazolium that is reduced in metabolically active cells into a soluble, cell-permeant formazan. The amount of formazan, detected by its absorbance at 492 nm is proportional to the number of living, metabolically active cells. 10^3 HCT116 cells are seeded per well. 24 hours later, the medium is changed and the cells treated in triplicate with the concentrations of each compound from 100 μM to 50 nM. The compounds (including USP7-IN-1) are diluted in 100% DMSO, whose final concentration on cells is kept at 0.5%. Cells are incubated with the compounds for 72 hours, and their viability then assayed by the addition of MTS for 2 hours. Absorbance at 492 nm is measured directly from the 96-well culture plates. GI_{50} (Growth Inhibition 50) concentrations for each compound are calculated using a sigmoidal variable slope fit. Values represent mean of three independent experiments[1].
	USP7 is diluted in USP buffer (50 mM Tris HCl; 0.5 mM EDTA (Ethylenediaminetetraacetic acid); 5 mM DTT; 0.01 % Triton X-100; Bovine Serum Albumin 0.05 mg/mL pH7.6). Compounds stocks (10 mM) are

Kinase Assay	stored at -20°C in DMSO. Compounds (including USP7-IN-1) are tested at different concentrations: from 200 μM to 91 nM. Reactions are performed as duplicates in Black 384 well plates (10 μL final reaction volume). The substrate concentration for USP7 is 300 nM Ub-AMC. The concentrations of the enzyme (USP7) in specificity assays is 100 pM. The concentrations are determined in order to perform specificity assays under initial velocities at fixed substrate concentration. Compounds are pre-incubated with enzymes for 30 minutes at 25°C. Reactions are initiated by addition of substrate to the plates containing the enzymes (+/- compounds) diluted in assay buffer. Reactions are incubated for 60 minutes at 37°C. Reactions are stopped by adding acetic acid (100 mM final). Readings are performed on a Pherastar Fluorescent Reader. λ Emission 380 nm; λ Excitation = 460 nm. Data (mean values +/- standard deviation) are analyzed as % of control (no compound) and plotted as percentage versus the Log of the compound concentration using GraphPad. Data are fitted to a sigmoidal model (variable slope)[1].
References	[1]. SELECTIVE AND REVERSIBLE INHIBITORS OF UBIQUITIN SPECIFIC PROTEASE 7. WO 2013030218 A1