产品名称：UNC3866
产品别名：UNC3866

生物活性：

Description	UNC3866 is a potent antagonist of the CBX7-H3 interaction as determined by AlphaScreen (IC_{50}=66±1.2 nM) and is more than 100-fold selective for CBX7 over the other nine members of this methyl-lysine (Kme) reader panel.
IC_{50} & Target	IC50: 66±1.2 nM (CBX7)[1]
In Vitro	UNC3866, a potent antagonist of the methyl-lysine (Kme) reading function of the Polycomb CBX and CDY families of chromodomains. UNC3866 binds the chromodomains of CBX4 and CBX7 most potently with a K_d of 100 nM for each, and is 6- to 18-fold selective versus seven other CBX and CDY chromodomains while being highly selective versus >250 other protein targets. UNC3866 inhibits PC3 cell proliferation, a known CBX7 phenotype, while UNC4219, a methylated negative control compound, has negligible effects. UNC3866 is a potent and cellularly active antagonist of PRC1 chromodomains. UNC3866 is the most potent ligand reported for CBX7 with a K_d of 97±2.4 nM. UNC3866 is equipotent for CBX4, which is most similar to CBX7, while it is 18-, 6- and 12-fold selective for CBX4/7 over CBX2, -6 and -8, respectively. Additionally, UNC3866 is 65-fold selective for CBX4/7 over CDY1 and 9-fold selective for CBX4/7 over CDYL1b and CDYL2[1].
Solvent&Solubility	***In Vitro:*** **DMSO : ≥ 27 mg/mL (33.96 mM)** * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	1.2578 mL	6.2891 mL	12.5783 mL
	5 mM	0.2516 mL	1.2578 mL	2.5157 mL
	10 mM	0.1258 mL	0.6289 mL	1.2578 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。

In Vivo:

请根据您的实验动物和给药方式选择适当的溶解方案。以下溶解方案都请先按照 **In Vitro** 方式配制澄清的储备液，再依次添加助溶剂：

——为保证实验结果的可靠性，澄清的储备液可以根据储存条件，适当保存；体内实验的工作液，建议您现用现配，当天使用； 以下溶剂前显示的百分比是指该溶剂在您配制终溶液中的体积占比；如在配制过程中出现沉淀、析出现象，可以通过加热和/或超声的方式助溶

1.请依序添加每种溶剂： 10% DMSO→40% PEG300 →5% Tween-80 → 45% saline

Solubility: ≥ 2.5 mg/mL (3.14 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (3.14 mM，饱和度未知) 的澄清溶液。

以 1 mL 工作液为例，取 100 μL 25.0 mg/mL 的澄清 DMSO 储备液加到 400 μL PEG300 中，混合均匀；向上述体系中加入 50 μL Tween-80，混合均匀；然后继续加入 450 μL 生理盐水定容至 1 mL。

2.请依序添加每种溶剂： 10% DMSO→ 90% (20% SBE-β-CD in saline)

Solubility: ≥ 2.5 mg/mL (3.14 mM); Clear solution

	此方案可获得 ≥ 2.5 mg/mL (3.14 mM，饱和度未知) 的澄清溶液。 以 1 mL 工作液为例，取 100 μL 25.0 mg/mL 的澄清 DMSO 储备液加到 900 μL 20% 的 SBE-β-CD 生理盐水水溶液中，混合均匀。 3.请依序添加每种溶剂： 10% DMSO →90% corn oil Solubility: ≥ 2.5 mg/mL (3.14 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (3.14 mM，饱和度未知) 的澄清溶液，此方案不适用于实验周期在半个月以上的实验。 以 1 mL 工作液为例，取 100 μL 25.0 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。
References	[1]. Stuckey JI, et al. A cellular chemical probe targeting the chromodomains of Polycomb repressive complex 1. Nat Chem Biol. 2016 Mar;12(3):180-7.

实验参考：

Cell Assay	PC3 cells are seeded at 200 cells/well into 24-well plates. Cells are allowed to adhere overnight. The media (DMEM supplemented with 10 % FBS) is then exchanged with fresh media containing DMSO, UNC3866 or UNC4219. On day three, the media are exchanged with fresh media containing DMSO, UNC3866 or UNC4219. For dose-response studies, the EC_{50} is derived from a six-point titration ranging from 100 μM to 0.4 μM of UNC3866 or UNC4219. At day 0, 3 or 6, cells are fixed with ice-cold methanol for 30 sec. and rehydrated with PBS. Nuclei of the cells are stained with DAPI (0.05 μg/mL) and numerated using High Content Microscopy. For dose-response studies, the cell count of UNC3866- or UNC4219-treated cells is normalized to the average cell count of DMSO-treated cells. The EC_{50} is calculated using the “log[inhibitor] vs. the normalized response-Variable slope” equation in GraphPad Prism 5[1].
Kinase Assay	The effect of UNC3866 on the methyltransferase activity of G9a, EHMT1, SUV39H1, SUV39H2, SETDB1, SETD8, SUV420H1, SUV420H2, SETD7, MLL1 trimeric complex, MLL3 tetrameric complex, EZH2 trimeric complex, PRMT1, PRMT3, PRMT5-MEP50 complex, PRMT6, PRMT7, PRMT8, PRDM9, SETD2, SMYD2, SMYD3, BCDIN3D and DNMT1 is assessed by monitoring the incorporation of tritium-labeled methyl group to lysine or arginine residues of peptide substrates using Scintillation Proximity Assay (SPA). Assays are performed in a 20 μL reaction mixture containing ^{3}H-SAM at substrate concentrations close to the K_m values for each enzyme. Three concentrations (1 μM, 10 μM, and 50 μM) of UNC3866 are used in all selectivity assays. To stop the enzymatic reactions, 7.5 M Guanidine hydrochloride is added, followed by 180 μL of buffer (20 mM Tris, pH 8.0). The reactions are mixed and then transferred to a 96-well FlashPlate. The reaction mixtures in Flash plates are incubated for 1 hour and the CPM are measured using a TopCount plate reader. The CPM counts in the absence of compound for each data set are defined as 100% activity. In the absence of the enzyme, the CPM counts in each data set are defined as background (0%)[1].
References	[1]. Stuckey JI, et al. A cellular chemical probe targeting the chromodomains of Polycomb repressive complex 1. Nat Chem Biol. 2016 Mar;12(3):180-7.