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产品名称：
4-HYDROXY-3,3-DIMETHYL-2H-BENZO[G]INDOLE-2,5(3H)-DIONE
产品别名：BVT948

生物活性：

Description	BVT948 is a protein tyrosine phosphatase (PTP) inhibitor which can also inhibit several cytochrome P450 (P450) isoforms and lysine methyltransferase SETD8 (KMT5A).
IC_{50} & Target	PTP[1], P450[1], SETD8[2]
In Vitro	Results show that the effect of BVT948 (BVT.948) is to strengthen the insulin signal and has no effects on the duration of the signal. BVT948 appears to be an effective inhibitor of both protein tyrosine phosphatases (PTP activity and P450 activity)[1]. BVT948 efficiently and selectively suppresses cellular H4 lysine 20 (H4K20me1) at doses lower than 5 μM within 24 h. The cells treated with BVT948 recapitulate cell-cycle-arrest phenotypes similar to what are reported for knocking down SETD8 by RNAi[2]. Treatment of MCF-7 cells with 0.5, 1 or 5 μM of BVT948 for 24 h does not cause any significant changes in cell viability. BVT948 inhibits TPA-induced MMP-9 up-regulation in a dose-dependent manner. Treatment with BVT948 inhibits TPA-stimulated NF-κB binding activity, but not AP-1 binding activity. BVT948 does not affect the MAPK phosphorylation by TPA. Treatment with BVT948 diminishes the TPA-induced cell invasion by 50%[3].
In Vivo	Results show that 3 μmol/kg BVT948 (BVT.948) significantly enhances glucose clearance from the blood stream in response to insulin compare with vehicle-treated controls[1].
Solvent&Solubility	***In Vitro:*** **DMSO : 100 mg/mL** (**414.52 mM;** Need ultrasonic)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	4.1452 mL	20.7262 mL	41.4525 mL
	5 mM	0.8290 mL	4.1452 mL	8.2905 mL
	10 mM	0.4145 mL	2.0726 mL	4.1452 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 (10.36 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (10.36 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。
References	[1]. Liljebris C, et al. Oxidation of protein tyrosine phosphatases as a pharmaceutical mechanism of action: a study using 4-hydroxy-3,3-dimethyl-2H-benzo[g]indole-2,5(3H)-dione. J Pharmacol Exp Ther. 2004 May;309(2):711-9. [2]. Blum G, et al. Small-molecule inhibitors of SETD8 with cellular activity. ACS Chem Biol. 2014 Nov 21;9(11):2471-8. [3]. Hwang BM, et al. Protein tyrosine phosphatase controls breast cancer invasion through the expression of matrix metalloproteinase-9. BMB Rep. 2013 Nov;46(11):533-8.

实验参考：

Cell Assay	L6 myocytes are maintained in minimum essential medium-alpha (α-MEM) supplemented with 10% fetal bovine serum and 100 IU/mL penicillin-streptomycin at 37°C in 5% CO_2. Cells are seeded into 24-well plates, and the medium is replaced with α-MEM containing 2% fetal calf serum to induce differentiation into myotubes. The medium is changed every other day, and cytidine (0.24 mg/mL medium) is added to the cultures at days 7 to 9 to suspend cycling cells. The cells are used in experiments after overnight serum starvation at days 11 to 16. They are treated with or without 25 μM BVT948 (BVT.948) for 30 min followed by 5 min of insulin (25 nM) stimulation. After freezing with liquid N_2, the cells are lysed with a Tris-HCl buffer, pH 7.4, containing 1% Nonidet-40, 0.25% sodium deoxycholate, 150 mM NaCl, 1 mM EDTA, 1 mM EGTA, 1 mM sodium orthovanadate, 10 mM β-glycerophosphate, 5 mM sodium pyrophosphate, and complete protease inhibitor cocktail The cell extracts are centrifuged at 14,000 g for 10 min, and the supernatants are used in the Delfia assay[1].
Animal Administration	Male mice 12 to 14 weeks old are used in this study. They are divided into equal groups (n=9) based on blood glucose levels. At time 0, the mice are injected with vehicle (NaCl with 10% DMSO) or BVT948 (BVT.948) (0.3 and 3 μmol/kg) and 1 U/kg insulin intraperitoneally. Blood glucose is determined from tail vein sampling at 0, 30, 60, and 120 min using a glucometer[1].
Kinase Assay	To determine the reversibility of the inhibition of protein tyrosine phosphatases (PTP) activity by BVT948 (BVT.948), 50 ng of PTP1B is incubated in 100 μL of assay buffer with 20 μM BVT948 for 10 min in a concentration device. The sample is then centrifuged at 14,000 rpm at 4°C for 12 min. The concentrate is subsequently washed three times with 100 μL of assay buffer followed by centrifugation. After washing, 190 μL of assay buffer is added to the sample, increasing the volume to 200 μL. Twenty microliters are used in assays measuring enzyme activity remaining using para-nitrophenyl phosphate (pNPP) as a substrate. Controls includes enzyme, which is treated with inhibitor but not washed, and enzyme, which is not treated with BVT948 but is put through the incubation and washing procedures[1].
References	[1]. Liljebris C, et al. Oxidation of protein tyrosine phosphatases as a pharmaceutical mechanism of action: a study using 4-hydroxy-3,3-dimethyl-2H-benzo[g]indole-2,5(3H)-dione. J Pharmacol Exp Ther. 2004 May;309(2):711-9. [2]. Blum G, et al. Small-molecule inhibitors of SETD8 with cellular activity. ACS Chem Biol. 2014 Nov 21;9(11):2471-8. [3]. Hwang BM, et al. Protein tyrosine phosphatase controls breast cancer invasion through the expression of matrix metalloproteinase-9. BMB Rep. 2013 Nov;46(11):533-8.