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产品名称：Inolitazone (dihydrochloride)
产品别名：Efatutazone dihydrochloride; CS-7017 dihydrochloride; RS5444 dihydrochloride

生物活性：

Description	Inolitazone dihydrochloride (Efatutazone dihydrochloride) is a novel high-affinity PPARγ agonist that is dependent upon PPARγ for its biological activity with IC50 of 0.8 nM for growth inhibition.
IC_{50} & Target	PPARγ
In Vitro	Inolitazone dihydrochloride (Efatutazone dihydrochloride) upregulates the cell cycle kinase inhibitor, $p21^{WAF1/CIP1}$. Silencing $p21^{WAF1/CIP1}$ rendered cells insensitive to Inolitazone. A 10 nM dose of Inolitazone activates PPARγ:RXRα-dependent transcription as demonstrated in a transient transfection assay utilizing a PPRE response element fused to a luciferase reporter gene (PPRE3-tk-luc). DRO cells are treated in culture with Inolitazone, Rosiglitazone, or Troglitazone at the indicated concentrations. DRO cells are transiently transfected with PPRE3-tk-luc to examine effective concentrations at which EC_{50} occurs. The EC_{50}s are 1 nM (Inolitazone), 65 nM (Rosiglitazone) and 631 nM (Troglitazone). Similarly, the calculated inhibitory concentration at IC_{50} is 0.8 nM for Inolitazone, 75 nM for Rosiglitazone, and 1412 nM for Troglitazone. Inolitazone specifically activates PPARγ, but not PPARα or PPARδ. Exposure of 10 nM Inolitazone following transient transfection with the appropriate PPAR isoform (γ, α, or δ) and PPAR response element linked to a luciferase reporter in RIE rat small intestinal cell line, which does not express PPARs, yields increased luciferase activity only in the presence of PPARγ and PPRE3-tk-luc[1]. DRO cells are growth inhibited by 10 nM Inolitazone (RS5444) through a PPARγ-dependent mechanism[2].
In Vivo	Inolitazone dihydrochloride (Efatutazone dihydrochloride) plus Paclitaxel demonstrate additive antiproliferative activity in cell culture and minimal ATC tumor growth. When Inolitazone is administered in the diet to athymic nude mice prior to DRO tumor cell implantation, tumor growth is inhibited in a dose responsive fashion. At the highest dose, 0.025% Inolitazone inhibits growth on day 32 by 94.4% as compared to that of control. In this treatment group, five of 10 animals do not develop demonstrable tumors. In the 0.0025% treatment group, tumor growth is inhibited by 62.3% compared to that of control on day 32 while the 0.00025% dose demonstrated no growth inhibitory activity as compared to control. Tumors is nest allowed to establish in the mouse and began 0.025% Inolitazone treatment of mice 1 week after DRO or ARO tumor cell implantation. Inolitazone treated animals demonstrate tumor growth inhibition of 68.9% in DRO tumors and 48.3% in ARO tumors as compared to that of their respective controls on day 35[1].

In Vitro:

DMSO : 25 mg/mL (**43.44 mM;** Need ultrasonic)

H_2O : < 0.1 mg/mL (insoluble)

Preparing Stock Solutions

Solvent / Mass / Concentration	1 mg	5 mg	10 mg
1 mM	1.7376 mL	8.6879 mL	17.3759 mL
5 mM	0.3475 mL	1.7376 mL	3.4752 mL
10 mM	0.1738 mL	0.8688 mL	1.7376 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

Solvent&Solubility	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 (4.34 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.34 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 (4.34 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.34 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 (4.34 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.34 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]. Copland JA, et al. Novel high-affinity PPARgamma agonist alone and in combination with paclitaxel inhibits human anaplastic thyroid carcinoma tumor growth via p21WAF1/CIP1. Oncogene. 2006 Apr 13;25(16):2304-17. [2]. Marlow LA, et al. Reactivation of suppressed RhoB is a critical step for the inhibition of anaplastic thyroid cancer growth. Cancer Res. 2009 Feb 15;69(4):1536-44.

实验参考：

Cell Assay	DRO90-1 (DRO) and ARO81 (ARO) cells are plated in 12-well culture plates in triplicate for each condition at an initial concentration of 2×10^4 cells/well. After overnight incubation, cells are treated with either Inolitazone, Rosiglitazone, Troglitazone, GW9662, or Paclitaxel diluted in DMSO at concentrations indicated in figure legends. All cells receive identical volumes of DMSO and are exposed to each drug for 6 days with medium and drug changed every 48 h. After 6 days, cells are washed with PBS, trypsinized and counted by Beckman Coulter Counter[1].
	Mice[1] Suspensions of 1×10^6/0.1 mL DRO or ARO cells in RPMI medium are injected subcutaneously in

Animal Administration	one flank of 3-4 week athymic female nude mice. Mice are changed to specialized diets either 1 week prior or 1 week after tumor implantation and randomly assigned to experimental or control groups with 10 mice per group. Diets consisted either placebo, 0.00025%, 0.0025%, or 0.025% Inolitazone formulated into the diet. Mice weighed between 20-25 g and consume on average 4 g of food per day. For combinatorial studies either placebo, 10 mg/kg or 15 mg/kg paclitaxel is injected i.p. twice weekly. Tumors are measured every 3-4 days for 35 days with calipers and tumor volumes are calculated by the formula: 0.5236(a×b×c), where a is the shortest diameter, b is the diameter perpendicular to a and c is the diameter height.
References	[1]. Copland JA, et al. Novel high-affinity PPARgamma agonist alone and in combination with paclitaxel inhibits human anaplastic thyroid carcinoma tumor growth via p21WAF1/CIP1. Oncogene. 2006 Apr 13;25(16):2304-17. [2]. Marlow LA, et al. Reactivation of suppressed RhoB is a critical step for the inhibition of anaplastic thyroid cancer growth. Cancer Res. 2009 Feb 15;69(4):1536-44.