产品名称：**L-TRANS-环氧琥珀酸- ILE-PRO-OME 丙醛**
产品别名：**CA-074 methyl ester**

生物活性：

Description	CA-074 methyl ester is a specific inhibitor of Cathepsin B, which has potent bioactivities such as neuroprotective, anti-cancer, and anti-inflamatory effects.
In Vitro	CA-074Me (5 μM and 50 μM) inhibits RANKL-induced osteoclastogenesis in BMM cells derived from C57BL/6J and NOD/ShiLtJ mice. CA-074Me exerts its anti-osteoclastogenic effect within 24 hours post-RANKL stimulation in vitro. CA-074Me does not exert its anti-osteoclastogenic effect via the MAPK-ERK signaling cascade. CA-074Me inhibits c-FOS upregulation and subsequent NFATc1 autoamplification following RANKL stimulation.[2]. CA-074Me reduces apoptosis induced by CVB1[3].
In Vivo	Hippocampal CA1 neuronal programmed necrosis induced by global cerebral I/R injury is prevented by CA074-me (1 μg, 10 μg) both pre-treatment and post-treatment. The rupture of lysosomal membrane and the leakage of cathepsin-B, and this is strongly inhibited by CA074-me pre-treatment. The overexpression and nuclear translocation of RIP3 and the reduction of NAD+ level after I/R injury are also inhibited, while the upregulation of Hsp70 is strengthened by CA074-me pre-treatment[1]. CA-074Me (30 mg/kg) is capable of inhibiting osteoclastogenesis and bone degradation in vivo[2]. In the CVB+CA-074Me (4 mg/kg/day i.m.) guinea pigs group, the scores of inflammation significantly decrease in comparison with the CVB+None group. In CVB+CA-074Me group, the number of CD8+T cells decrease in comparison with the sham group[3].
Solvent&Solubility	***In Vitro:*** **DMSO : ≥ 215 mg/mL (540.92 mM)** **H2O : 26.66 mg/mL (67.07 mM; Need warming)** * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.5159 mL	12.5796 mL	25.1591 mL
	5 mM	0.5032 mL	2.5159 mL	5.0318 mL
	10 mM	0.2516 mL	1.2580 mL	2.5159 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]. Xu Y, et al. Protective mechanisms of CA074-me (other than cathepsin-B inhibition) against programmed necrosis induced by global cerebral ischemia/reperfusion injury in rats. Brain Res Bull. 2016 Jan;120:97-105. [2]. Patel N, et al. CA-074Me compound inhibits osteoclastogenesis via suppression of the NFATc1 and c-FOS signaling pathways. J Orthop Res. 2015 Oct;33(10):1474-86. [3]. Zhang L, et al. Treatment with CA-074Me, a Cathepsin B inhibitor, reduces lung interstitial inflammation and fibrosis in a rat model of polymyositis. Lab Invest. 2015 Jan;95(1):65-77.

实验参考：

	RANKL (0.08 mg/kg) with and without CA-074Me (10 mg/kg or 30 mg/kg) are mixed in sterile,

Animal Administration	nonimmunogenic 1% Extracel-HP gel. The gel is composed of thiol-modified sodium hyaluronate, thiol-modified heparin, thiol-modified gelatin, and degassed deionized sterile water. The hydrogel mixture is prepared in an aseptic hood using a sterile syringe. The control sham hydrogel contained sterile Phosphate Buffered Saline (PBS) without any cytokines. The osteolysis group is given 0.08 mg/kg RANKL in a hydrogel to induce pathologic bone loss. The hydrogel-only, hydrogel-RANKL, and hydrogel-RANKL-CA-074Me mixture is injected into 8-week old male mice calvarium in an aseptic hood (n = 5) following general anesthesia (80 mg/kg of ketamine and 7 mg/kg of xylazine). After four days, the calvaria are excised, fixed in 4% formaldehyde for 24 h, decalcified in 20% EDTA for one week, and sectioned into slides from paraffin blocks. The slides undergo Tartrate-Resistant Acid Phosphatase (TRAP) staining to identify osteoclasts. [2]
Kinase Assay	After seven days of cell culture and osteoclast generation, the media is removed and washed three times with PBS. BMMs are fixed with a fixing solution supplied by the manufacturer. The cells are incubated at 37°C with a solution containing deionized water, Fast Garnet GBC, Napthol phosphate, Acetate, and Tartrate for 1 h. The staining solution is removed, washed with PBS (3×), and air-dried. TRAP positive cells with three or more nuclei across whole culture area are counted as multinucleated osteoclasts using light microscopy. [2]
References	[1]. Xu Y, et al. Protective mechanisms of CA074-me (other than cathepsin-B inhibition) against programmed necrosis induced by global cerebral ischemia/reperfusion injury in rats. Brain Res Bull. 2016 Jan;120:97-105. [2]. Patel N, et al. CA-074Me compound inhibits osteoclastogenesis via suppression of the NFATc1 and c-FOS signaling pathways. J Orthop Res. 2015 Oct;33(10):1474-86. [3]. Zhang L, et al. Treatment with CA-074Me, a Cathepsin B inhibitor, reduces lung interstitial inflammation and fibrosis in a rat model of polymyositis. Lab Invest. 2015 Jan;95(1):65-77.