**Proteins** 

# CA-5f

Cat. No.: HY-112698 CAS No.: 1370032-19-1 Molecular Formula:  $C_{24}H_{24}N_{2}O_{3}$ Molecular Weight: 388.46

Target: Autophagy; Apoptosis Pathway: Autophagy; Apoptosis

Storage: Powder -20°C

3 years 4°C 2 years

In solvent -80°C 2 years

> -20°C 1 year

**Product** Data Sheet

## **SOLVENT & SOLUBILITY**

In Vitro

DMSO: 77.5 mg/mL (199.51 mM; Need ultrasonic)

	Solvent Mass Concentration	1 mg	5 mg	10 mg
Preparing Stock Solutions	1 mM	2.5743 mL	12.8713 mL	25.7427 mL
	5 mM	0.5149 mL	2.5743 mL	5.1485 mL
	10 mM	0.2574 mL	1.2871 mL	2.5743 mL

Please refer to the solubility information to select the appropriate solvent.

In Vivo

- 1. Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline Solubility: ≥ 2.08 mg/mL (5.35 mM); Clear solution
- 2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE- $\beta$ -CD in saline) Solubility: 2.08 mg/mL (5.35 mM); Suspended solution; Need ultrasonic
- 3. Add each solvent one by one: 10% DMSO >> 90% corn oil Solubility: ≥ 2.08 mg/mL (5.35 mM); Clear solution

# **BIOLOGICAL ACTIVITY**

Description	CA-5f is a potent late-stage macroautophagy/autophagy inhibitor via inhibiting autophagosome-lysosome fusion. CA-5f increases LC3B-II (a marker to monitor autophagy) and SQSTM1 protein, and also increases ROS production. Anti-tumor activity <sup>[1]</sup> .
IC <sub>50</sub> & Target	${\sf Macroautophagy/autophagy}^{[1]}$
In Vitro	CA-5f (0-40 μM, 6 hour) concentration- and time-dependently elevates the level of LC3B-II (a marker to monitor autophagy)

and SQSTM1 protein both in A549 cells and  ${\tt HUVECs}^{[1]}.$ 

CA-5f (20  $\mu$ M, 6 hours) inhibits the degradation of autophagosomes when treated alone or in combination Bafilomycin A1 (100 nM) or Chloroquine (30  $\mu$ M) in A549 cells and HUVECs<sup>[1]</sup>.

CA-5f (20  $\mu$ M) neither impairs the hydrolytic function nor the quantity of lysosomes [1].

CA-5f (20  $\mu$ M, 96 hours) inhibits the growth of A549 cells, and less cytotoxic to normal HUVECs<sup>[1]</sup>.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

## Cell Viability Assay<sup>[1]</sup>

Cell Line:	A549, HUVECs
Concentration:	20 μΜ
Incubation Time:	96 hours
Result:	Exhibited more cytotoxicity against A549 cells compared with normal HUVECs.

# Western Blot Analysis $^{[1]}$

Cell Line:	A549 cells and HUVECs	
Concentration:	0-40 μΜ	
Incubation Time:	6 hours	
Result:	Elevated LC3B-II (a marker to monitor autophagy) and SQSTM1 protein levels in a concentration- and time-dependent manner.	

### In Vivo

CA-5f (40 mg/kg, i.p., every 2 days for up to 30 days) is well tolerated, and potently inhibits the growth of tumor in nude mice bearing A549 lung cancer cells<sup>[1]</sup>.

CA-5f (40 mg/kg, i.p.) suppresses autophagic flux and induces apoptosis in nude mice bearing A549 lung cancer cells<sup>[1]</sup>. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

Animal Model:	Nude mice bearing A549 lung cancer ${\sf cells^{[1]}}$	
Dosage:	40 mg/kg	
Administration:	Injected via caudal vein, every 2 days for up to 30 days	
Result:	Significantly suppressed tumor volume and weight in mice, increased the number of apoptotic cells in mice.	

### **REFERENCES**

[1]. Zhang L, et al. Identification of compound CA-5f as a novel late-stage autophagy inhibitor with potent anti-tumor effect against non-small cell lung cancer. Autophagy. 2019 Mar;15(3):391-406.

Caution: Product has not been fully validated for medical applications. For research use only.

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