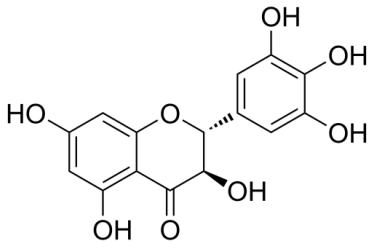
Data Sheet

Product Name:	Dihydromyricetin
Cat. No.:	CS-3809
CAS No.:	27200-12-0
Molecular Formula:	$C_{15}H_{12}O_8$
Molecular Weight:	320.25
Target:	Autophagy; DNA/RNA Synthesis; Influenza Virus; mTOR
Pathway:	Anti-infection; Autophagy; Cell Cycle/DNA Damage; PI3K/Akt/mTOR
Solubility:	DMSO : ≥ 100 mg/mL (312.26 mM)



BIOLOGICAL ACTIVITY:

Dihydromyricetin is a potent inhibitor with an **IC_{50}** of 48 μM on **dihydropyrimidinase**. Dihydromyricetin can activate autophagy through inhibiting **mTOR** signaling. Dihydromyricetin suppresses the formation of mTOR complexes (**mTORC1/2**). Dihydromyricetin is also a potent **influenza RNA-dependent RNA polymerase** inhibitor with an **IC_{50}** of 22 μM. IC50 & Target: IC50: 48 μM (dihydropyrimidinase)[1]

mTORC1/2[3] ***In Vitro:*** Dihydromyricetin, a flavonol, significantly inhibits the catalytic activities of dihydropyrimidinase toward both the natural substrate dihydrouracil and xenobiotic substrate 5-propyl-hydantoin. Dihydromyricetin exhibits a significant inhibitory effect on the activities of dihydropyrimidinase for both substrates, even more than Myricetin does. The IC_{50} values of Dihydromyricetin for dihydropyrimidinase determined from the titration curves using Dihydrouracil and 5-propyl-hydantoin are 48±2 and 40±2 μM, respectively[1]. Dihydromyricetin (DHM) supplementation significantly reverses the increased phosphorylation of mTOR at Ser^{2448} (p-mTOR) during D-gal administration, which suggests that Dihydromyricetin can activate autophagy through inhibiting mTOR signaling [2]. ***In Vivo:*** Changes in learning and memory capacity in rats administrated normal control group, D-gal group, D-gal+Dihydromyricetin (100 mg/kg) group, D-gal+Dihydromyricetin (200 mg/kg) group assessed by morris water maze (MWM) (n=10 per group). Dihydromyricetin (DHM) treatment significantly shortens the escape latency when compared with D-gal-induced model group[2].

PROTOCOL (Extracted from published papers and Only for reference)

Kinase Assay: [1]A rapid spectrophotometric assay is used to determine the enzymatic activity for hydantoinase, allantoinase, dihydroorotase, and imidase. Dihydrouracil, 5-propyl-hydantoin, and phthalimide are used as substrates. Unless explicitly stated otherwise, Dihydrouracil (2 mM) is used as the substrate in the standard assay of dihydropyrimidinase. Briefly, the decrease in absorbancy at 230, 248, and 298 nm is measured upon hydrolysis of Dihydrouracil, 5-propyl-hydantoin, and Phthalimide as the substrate at 25°C, respectively. To start the reaction, the purified dihydropyrimidinase (10-70 μg) is added to a 2 mL solution containing the substrate and 100 mM Tris-HCl (pH 8.0). Substrate hydrolysis is monitored with a UV/vis spectrophotometer. The extinction coefficient of each substrate is determined experimentally by direct measurement with a spectrophotometer. The extinction coefficients of Dihydrouracil, 5-propyl-hydantoin, and Phthalimide are 0.683 $mM^{-1}cm^{-1}$ at 230 nm, 0.0538 $mM^{-1}cm^{-1}$ at 248 nm, and 3.12 $mM^{-1}cm^{-1}$ at 298 nm, respectively. The initial rates of change are a function of enzyme concentration within the absorbance range of 0.01-0.18 min^{-1}. A unit of activity is defined as the amount of enzyme catalyzing the hydrolysis of 1 μmol substrate/min, and the specific activity is expressed in terms of units of activity per milligram of enzyme. The kinetic parameters K_m and V_{max} are determined from a non-linear plot by fitting the hydrolyzing rate from individual experiments to the Michaelis-Menten equation[1].

Cell Assay: [2]Hippocampus and cortex tissue samples are homogenized in lysis buffer containing 20 mM Tris (pH 7.5), 135 mM NaCl, 2 mM EDTA, 2 mM DTT, 25 mM β-glycerophosphate, 2 mM sodium pyrophosphate, 10% glycerol, 1% Triton X-100, 1 mM

sodium orthovanadate, 10 mM NaF, 10 μg/mL aprotinin, 10 μg/mL leupeptin, and 1 mM PMSF for 30 min on ice and centrifuged at 12000×g at 4°C for 30 min. The supernatant is collected and protein quantification is carried out using a BCA kit. The protein samples are boiled in the presence of sample buffer at 95°C for 5 min. The target protein is separated by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE), transferred to nitrocellulose membrane, and then probed by corresponding primary and secondary antibodies. Finally, the target protein is visualized by enhanced chemiluminescence (ECL) reagent exposure to X-ray film[2].

Animal Administration: Dihydromyricetin (DHM) is dissolved in distilled water (Rats)[2].[2]Rats[2]

Totally 40 male Sprague-Dawley (SD) rats (age: 8 weeks old; body weight: 160±20 g) are used. The rats are randomly divided into four groups including normal control group, D-gal model group, and D-gal combined with DHM at the doses of 100 and 200 mg/kg-d groups with 10 rats in each group. All rats are housed at the environment with room temperature of 22±2°C and a dark-light cycle (12 h: 12h), and provided the accessibility to food and water ad libitum. After adapting to new environment for 1 week, the rats from DHM groups are administered with DHM dissolved in distilled water at the designated dosages by gavage once a day at 8:00am for 6 consecutive weeks. The rats from the normal control group are administrated with distilled water. Except from the normal control group, the rats from other groups are subjected to subcutaneous injection of D-gal at the dose of 150 mg/kg.d for 6 consecutive weeks. Each administration of DHM should be 2 h ahead of D-gal injection.

References:

[1]. Huang CY. Inhibition of a Putative Dihydropyrimidinase from Pseudomonas aeruginosa PAO1 by Flavonoids and Substrates of Cyclic Amidohydrolases. PLoS One. 2015 May 19;10(5):e0127634.

[2]. Kou X, et al. Ampelopsin attenuates brain aging of D-gal-induced rats through miR-34a-mediated SIRT1/mTORsignal pathway. Oncotarget. 2016 Nov 15;7(46):74484-74495.

[3]. Chang H, et al. Ampelopsin suppresses breast carcinogenesis by inhibiting the mTOR signalling pathway. Carcinogenesis. 2014 Aug;35(8):1847-54.

[4]. Václav Zima, et al. Unraveling the Anti-Influenza Effect of Flavonoids: Experimental Validation of Luteolin and its Congeners as Potent Influenza Endonuclease Inhibitors. Eur J Med Chem. 22 August 2020, 112754.

CAIndexNames:

4H-1-Benzopyran-4-one, 2,3-dihydro-3,5,7-trihydroxy-2-(3,4,5-trihydroxyphenyl)-,(2R,3R)-

SMILES:

O=C1[C@H](O)[C@@H](C2=CC(O)=C(O)C(O)=C2)OC3=CC(O)=CC(O)=C13

Tel: 610-426-3128 Fax: 888-484-5008 E-mail: sales@ChemScene.com

Address: 1 Deer Park Dr, Suite Q, Monmouth Junction, NJ 08852, USA