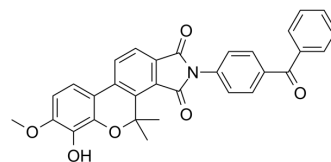


Ampkinone

Cat. No.:	HY-12831
CAS No.:	1233082-79-5
Molecular Formula:	C ₃₁ H ₂₃ NO ₆
Molecular Weight:	506
Target:	AMPK
Pathway:	Epigenetics; PI3K/Akt/mTOR
Storage:	Powder -20°C 3 years 4°C 2 years In solvent -80°C 2 years -20°C 1 year



SOLVENT & SOLUBILITY

In Vitro	DMSO : 50 mg/mL (98.81 mM; Need ultrasonic)					
	Preparing Stock Solutions	<div><div>Solvent</div><div>Concentration</div></div>	Mass	1 mg	5 mg	10 mg
		1 mM		1.9763 mL	9.8814 mL	19.7628 mL
		5 mM		0.3953 mL	1.9763 mL	3.9526 mL
		10 mM		0.1976 mL	0.9881 mL	1.9763 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: 2.5 mg/mL (4.94 mM); Suspended solution; Need ultrasonic					

BIOLOGICAL ACTIVITY

Description	Ampkinone is an indirect AMP-activated protein kinase (AMPK) activator.
IC ₅₀ & Target	AMPK
In Vitro	Ampkinone stimulates the phosphorylation of AMPK via the indirect activation of AMPK in various cell lines. Ampkinone-mediated activation of AMPK requires the activity of LKB1 and results in increased glucose uptake in muscle cells ^[1] . MCE has not independently confirmed the accuracy of these methods. They are for reference only.
In Vivo	Ampkinone-treated DIO mice significantly reduce total body weight and overall fat mass. Histological examination and measurement of lipid parameters show that Ampkinone effectively improves metabolic abnormalities in the DIO mice model ^[1] . MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Kinase Assay ^[1]

Total AMPK activity is measured using a synthetic SAMS peptide substrate and [γ -³²P]ATP. Briefly, 500 μ g of protein extract is incubated with anti-AMPK- α 1 and α 2 antibodies for 2 h at 4°C. Protein A/G sepharose and agarose are added, and the mixtures are incubated for 3 h at 4°C. After the samples are washed three times with RIPA buffer, the activity is assessed in AMPK reaction buffer containing 20 mM HEPES-NaOH (pH 7.0), 0.4 mM Dithiothreitol, and 0.01% Brij-35, with or without 300 μ M AMP. The immune complexes are added to 23 μ L of reaction buffer per assay, and then 7 μ L of SAMS substrate peptide (HMRSAMSGHLVKRR, 100 μ M final concentration) and 10 μ L of ATP mixture (an aliquot of 1 μ Ci/ μ L: 90 μ L of 75 mM magnesium chloride, 500 μ M unlabeled ATP in 20 mM MOPS, pH7.2, 25 mM β -glycerophosphate, 5 mM EGTA, 1 mM sodium orthovanadate, and 1 mM Dithiothreitol) are added, and the mixture is incubated at 30°C for 15 min. After 35 μ L is spotted onto the center of P81 paper, the paper is washed three times with 0.75% phosphoric acid and once with acetone for 5 min. The samples are read using a scintillation counter^[1].

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

Animal Administration ^[1]

Mice^[1]

C57BL/6J mice are used and housed individually in a room maintained at 25°C on a 12/12 h light/dark schedule. For diet-induced obesity (DIO) mice, 4-week-old male C57BL/6J mice are fed a high-fat diet (HFD, 60% calories from fat) ad libitum for 8 weeks. Ampkinone in polyethylene glycol (PEG400) or vehicle is administered subcutaneously at 10 mg/kg body weight per day for 1 month. Body weight and food intake are measured every 3 days. All animals are given an insulin tolerance test (ITT) and sacrificed at 30 days. Liver weight and fat masses are measured, sectioned, and stained with Oil-red O and H&E.

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

CUSTOMER VALIDATION

- Biomed Pharmacother. 2023 Dec 25;170:116060.
- FASEB J. 2019 Jun;33(6):7202-7212.

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REFERENCES

[1]. Oh S, et al. Antidiabetic and antiobesity effects of Ampkinone (6f), a novel small molecule activator of AMP-activated protein kinase. J Med Chem. 2010 Oct 28;53(20):7405-13.

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

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