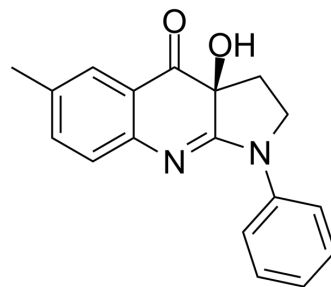


## (-)-Blebbistatin

Cat. No.:	HY-13441
CAS No.:	856925-71-8
Molecular Formula:	C <sub>18</sub> H <sub>16</sub> N <sub>2</sub> O <sub>2</sub>
Molecular Weight:	292.33
Target:	Myosin
Pathway:	Cytoskeleton
Storage:	4°C, stored under nitrogen

\* The compound is unstable in solutions, freshly prepared is recommended.



### SOLVENT & SOLUBILITY

In Vitro	DMSO : 10 mg/mL (34.21 mM; Need ultrasonic)						
	H <sub>2</sub> O : < 0.1 mg/mL (insoluble)						
	Preparing Stock Solutions	Solvent Concentration	Mass	1 mg	5 mg	10 mg	
				1 mM	3.4208 mL	17.1040 mL	34.2079 mL
				5 mM	0.6842 mL	3.4208 mL	6.8416 mL
10 mM				0.3421 mL	1.7104 mL	3.4208 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: ≥ 1 mg/mL (3.42 mM); Clear solution						
	2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline) Solubility: 1 mg/mL (3.42 mM); Suspended solution; Need ultrasonic						
	3. Add each solvent one by one: 10% DMSO >> 90% corn oil Solubility: ≥ 1 mg/mL (3.42 mM); Clear solution						

### BIOLOGICAL ACTIVITY

Description	(-)-Blebbistatin is a selective inhibitor of the ATPase activity of non-muscle myosin II <sup>[1][2]</sup> .
IC <sub>50</sub> & Target	IC50: 0.5 to 5 μM (myosin II) <sup>[1]</sup>
In Vitro	Blebbistatin potently inhibits several striated muscle myosins as well as vertebrate nonmuscle myosin IIA and IIB with IC <sub>50</sub> values ranging from 0.5 to 5 μM. Smooth muscle myosin is only poorly inhibited (IC <sub>50</sub> =80 μM) <sup>[1]</sup> . Blebbistatin does not compete with nucleotide binding to the skeletal muscle myosin subfragment-1. The inhibitor preferentially binds to the ATPase intermediate with ADP and phosphate bound at the active site, and it slows down phosphate release. It blocks the

myosin heads in a products complex with low actin affinity<sup>[2]</sup>. In culture-activated hepatic stellate cells, blebbistatin is found to change both cell morphology and function. Stellate cells become smaller, acquire a dendritic morphology and have less myosin IIA-containing stress fibres and vinculin-containing focal adhesions. Blebbistatin impairs silicone wrinkle formation, reduces collagen gel contraction and blocks endothelin-1-induced intracellular Ca<sup>2+</sup> release. It promotes wound-induced cell migration<sup>[3]</sup>.

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

#### In Vivo

Blebbistatin dose-dependently and completely relax both KCl- and carbachol-induced rat detrusor and endothelin-1-induced human bladder contraction. Pre-incubation with 10 µM blebbistatin attenuates carbachol responsiveness by 65% while blocking electrical field stimulation-induced bladder contraction reaching 50% inhibition at 32 Hz<sup>[4]</sup>.

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

## PROTOCOL

#### Cell Assay<sup>[3]</sup>

Freshly isolated HSCs are replated on 96-well plate. At day 3, medium is replaced by serum-free medium and cells are starved overnight, treated with or without blebbistatin (25 µM) for 2 h followed by stimulation with platelet-derived growth factor-BB (20 ng/mL). After an overnight incubation, the WST-1 cell proliferation assay are performed<sup>[3]</sup>.

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

## CUSTOMER VALIDATION

- Circulation. 2023 Nov 14.
- Adv Sci (Weinh). 2022 Mar 3;e2104682.
- Adv Sci (Weinh). 2020 Jun 17;7(15):1903583.
- Theranostics. 2019 Apr 13;9(9):2555-2571.
- Cell Death Dis. 2023 Oct 10;14(10):663.

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## REFERENCES

[1]. Cristina Lucas-Lopez, et al. Absolute Stereochemical Assignment and Fluorescence Tuning of the Small Molecule Tool, (-)-Blebbistatin.

[2]. Ponsaerts R, et al. The myosin II ATPase inhibitor blebbistatin prevents thrombin-induced inhibition of intercellularcalcium wave propagation in corneal endothelial cells. Invest Ophthalmol Vis Sci. 2008 Nov;49(11):4816-27.

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

Tel: 609-228-6898

Fax: 609-228-5909

E-mail: [tech@MedChemExpress.com](mailto:tech@MedChemExpress.com)

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