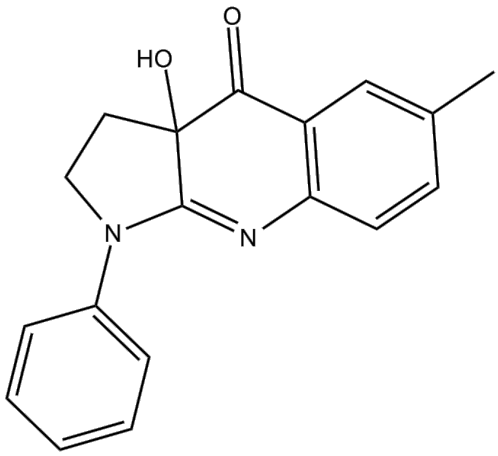
Product Data Sheet

Product Name: (±)-Blebbistatin
Cat. No.: GC12341

Chemical Properties

Cas. No.	674289-55-5		
Chemical Name	3a-hydroxy-6-methyl-1-phenyl-3,3a-dihydro-1H-pyrrolo[2,3-b]quinolin-4(2H)-one		
SMILES	OC12CCN(C1=NC3=C(C2=O)C=C(C=C3)C)C4=CC=CC=C4		
Formula	$C_{18}H_{16}N_2O_2$	M.Wt	292.34
Solubility	10mg/mL in DMSO, 20mg/mL in DMF	Storage	Store at -20°C,unstable in solution, ready to use.
General tips	For obtaining a higher solubility , please warm the tube at 37 °C and shake it in the ultrasonic bath for a while.Stock solution can be stored below -20°C for several months.		
Shipping Condition	Evaluation sample solution : ship with blue ice All other available size: ship with RT , or blue ice upon request.		

Structure



Protocol

Cell experiment [1]:

Cell lines: Rat ventricular myocytes

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Preparation Method	Fluo-5F–loaded myocytes were placed in an open chamber on an inverted stage microscope with epifluorescence objectives and perfused with physiologic salt-containing solution (PSS) at 0.5mL/min. Cells continuously received either PSS alone or PSS with 0.1, 0.5, 2, or 10μM (±)-Blebbistatin. Each cell was exposed to only one blebbistatin concentration. Contraction and calcium transients ($[Ca^{2+}]_i$) were evaluated at baseline and after 10min of blebbistatin application in steady-state conditions. Cell shortening was recorded using IonOptix Digital Sarcomere Length software (IonOptix).
Reaction Conditions	0.1, 0.5, 2, or 10μM; 10min
Applications	In single rat ventricular cells, (±)-Blebbistatin blocked cell shortening in a dose-dependent manner with an IC_{50} of 0.43μM. However, even at the highest concentration (10μM), it did not affect the $[Ca^{2+}]_i$.

Animal experiment [2]:

Animal models	Wistar rats
Preparation Method	Rats were randomly divided into groups that received intravesical instillation of Retinyl acetate (RA) or polysorbate 80 in saline. Three days after, each group was subdivided into groups that received (±)-Blebbistatin intravesically (125nM/kg) or DMSO, and immediately after cystometric assessment was performed.
Dosage form	125nM/kg; intravesical instillation

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Applications	(±)-Blebbistatin improves urine storage with no impairment of the voiding function. The significant reduction of the detrusor overactivity index, amplitude of nonvoiding contractions, and frequency of nonvoiding contractions in animals with detrusor overactivity (DO), following (±)-Blebbistatin administration.

References:
[1] Fedorov V V, Lozinsky I T, Sosunov E A, et al. Application of blebbistatin as an excitation–contraction uncoupler for electrophysiologic study of rat and rabbit hearts[J]. Heart rhythm, 2007, 4(5): 619-626.
[2]Wróbel A, Nowakowski Ł, Doboszewska U, et al. Blebbistatin reveals beneficial effects on the cystometric parameters in an animal model of detrusor overactivity[J]. Naunyn-schmiedeberg's Archives of Pharmacology, 2019, 392: 843-850.

Background

(±)-Blebbistatin is a racemic mixture containing two configurations, (+)-Blebbistatinb and (-)-Blebbistatin (Cat. No. GC13430). (±)-Blebbistatin can effectively inhibit the activity of non-muscle myosin II ATPase and the expression of various striated muscle myosins and non-muscle myosins in vertebrates[1, 2]. (±)-Blebbistatin is a commonly used excitation-contraction uncoupling agent in cardiovascular physiology[3]. (±)-Blebbistatin is a potential photodynamic drug that can kill cancer cells. Exposure to UV light with a wavelength below 488nm can also lead to rapid inhibition of (±)-Blebbistatin[4].

In vitro, (±)-Blebbistatin (0.1, 0.5, 2, 10μM) treated rat ventricular myocytes for 10min prevented cell shortening in a dose-dependent manner with an IC_{50} of 0.43μM, but did not affect intracellular calcium transients ($[Ca^{2+}]_i$)[5].

In vivo, (±)-Blebbistatin (125nM/kg) treated rats with detrusor overactivity (DO) by intravesical instillation significantly reduced the detrusor overactivity index, non-micturition contraction amplitude, and non-micturition contraction frequency, and improved urine storage without impairing urination function[6]. (±)-Blebbistatin (125nM/kg) treated DO rats by intra-arterial injection attenuated 13-cis-RA-induced changes in cystometric parameters and central and peripheral neurotrophic factor levels,

and also had antidepressant-like effects[7].

References:

[1]Lucas-Lopez C, Patterson S, Blum T, et al. Absolute Stereochemical Assignment and Fluorescence Tuning of the Small Molecule Tool,(–)-Blebbistatin[J]. 2005.
[2]Limouze J, Straight A F, Mitchison T, et al. Specificity of blebbistatin, an inhibitor of myosin II[J]. Journal of Muscle Research & Cell Motility, 2004, 25: 337-341.
[3]George S A , Lin Z , Efimov I R .Simultaneous triple-parametric optical mapping of transmembrane potential, intracellular calcium and NADH for cardiac physiology assessment[J].Communications Biology, 2024.
[4]Sakamoto T, Limouze J, Combs C A, et al. Blebbistatin, a myosin II inhibitor, is photoinactivated by blue light[J]. Biochemistry, 2005, 44(2): 584-588.
[5]Fedorov V V, Lozinsky I T, Sosunov E A, et al. Application of blebbistatin as an excitation–contraction uncoupler for electrophysiologic study of rat and rabbit hearts[J]. Heart rhythm, 2007, 4(5): 619-626.
[6]Wróbel A, Nowakowski Ł, Doboszewska U, et al. Blebbistatin reveals beneficial effects on the cystometric parameters in an animal model of detrusor overactivity[J]. Naunyn-schmiedeberg's Archives of Pharmacology, 2019, 392: 843-850.
[7]Wróbel A, Doboszewska U, Rechberger E, et al. Blebbistatin, a myosin II inhibitor, exerts antidepressant-like activity and suppresses detrusor overactivity in an animal model of depression coexisting with overactive bladder[J]. Neurotoxicity Research, 2019, 35: 196-207.