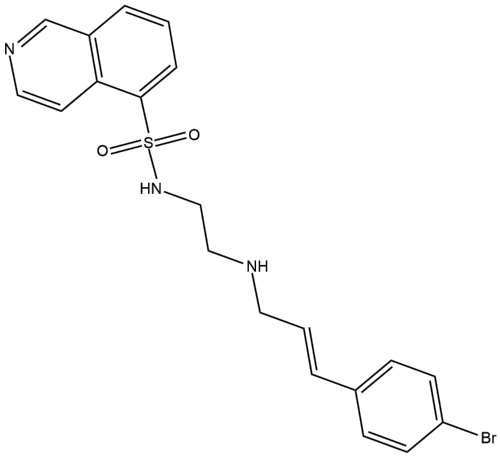
Product Data Sheet

Product Name: H 89
Cat. No.: GC19396

Chemical Properties

Cas. No.	127243-85-0		
SMILES	O=S(C1=CC=CC2=C1C=CN=C2)(NCCNC/C=C/C3=CC=C(Br)C=C3)=O		
Formula	$C_{20}H_{20}BrN_3O_2S$	M.Wt	446.36
Solubility	DMSO : 100 mg/mL (224.03 mM; Need ultrasonic)	Storage	Store at -20°C
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

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Kinase experiment: Kinase activities are assayed at 30°C for 2-5 min by measuring the transfer of 32P from [γ-32P]ATP to substrates. The reaction is terminated by adding 1 mL of 20% trichloroacetic acid, following the addition of 100 μg of bovine serum albumin as a carrier protein. The sample is centrifuged at 3000 rpm for 10 min, the pellet is resuspended in 5% trichloroacetic acid solution, the final pellet is dissolved in 1 mL of 1 N NaOH and the radioactivity is measured in a liquid scintillation counter.

Cell experiment: After 48 h in culture, PCl2D cells are cultured in test medium containing 30 μM H-89 for 1 h and then exposed to fresh medium that contained both 10 μM forskolin and 30 μM H-89. Cells are scraped off with a rubber policeman and sonicated in the presence of 0.5 mL of 6% trichloroacetic acid. To extract trichloroacetic acid, 2 mL of petroleum ether is added, the preparation mixed and centrifuged at 3000 rpm for 10 min. After aspiration of the upper layer, the residue sample solution is used for determination.

Animal experiment: Male albino mice weighing 20-25 g are obtained. Pentoxifylline (25, 50, 100 mg/kg), bucladesine (50, 100, 300 nM/mouse) and H-89 (0.05, 0.1, 0.2 mg/100 g) are administered intraperitoneally (i.p.) 30 min before intravenous (i.v.) infusion of PTZ. In combination groups, the first and second components are injected 45 and 30 min before PTZ infusion. In all groups, the respective control animalsreceive an appropriate volume of vehicle. For the i.v. infusion, the needle is inserted into the lateral tail vein, fixed to the tail vein by a narrow piece of adhesive tape, and the animal is allowed to move freely. PTZ solution is infused at a concentration rate of 1 mL/min.

Caution: Product has not been fully validated for medical applications. For research use only.
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Address: 10292 Central Ave. #205, Montclair, CA, USA

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References:

[1]. Chijiwa T, et al. Inhibition of forskolin-induced neurite outgrowth and protein phosphorylation by a newly synthesized selective inhibitor of cyclic AMP-dependent protein kinase, N-[2-(p-bromocinnamylamino)ethyl]-5-isoquinolinesulfonamide (H-89), of PC12D pheochromocytoma cells. J Biol Chem. 1990 Mar 25;265(9):5267-72.
[2]. Blazev R, et al. Effects of the PKA inhibitor H-89 on excitation-contraction coupling in skinned and intact skeletal muscle fibres. J Muscle Res Cell Motil. 2001;22(3):277-86.
[3]. Hosseini-Zare MS, et al. Effects of pentoxifylline and H-89 on epileptogenic activity of bucladesine in pentylenetetrazol-treated mice. Eur J Pharmacol. 2011 Nov 30;670(2-3):464-70.

Background

H-89 is a potent inhibitor of cyclic AMP-dependent protein kinase (protein kinase A) with IC50 of 48 nM and has weak inhibition on PKG, PKC, Casein Kinase, and others kinases.

H-89 inhibits protein kinase A, in competitive fashion against ATP. H-89 causes a dose-dependent inhibition of the forskolin-induced protein phosphorylation, with no decrease in intracellular cyclic AMP levels in PC12D cells. H-89 significantly inhibits the forskolin-induced neurite outgrowth from PC12D cells. H-89 (30 uM) inhibits significantly cAMP-dependent histone IIb phosphorylation activity in PC12D cell lysates[1]. H-89 (1-2 uM) significantly slows the repriming rate in rat skinned fibres, most likely due to it deleteriously affecting the T-system potential. H-89 (10-100 uM) inhibits net Ca2+ uptake by the SR and affectes the Ca32-sensitivity of the contractile apparatus in rat skinned fibres[2].

H-89 (0.2 mg/100g, i.p.) significantly increases seizure latency and threshold in PTZ-treated animals. H-89 (0.05, 0.2 mg/100 g, i.p.) prevents the epileptogenic activity of bucladesine (300 nM) with significant increase of seizure latency and seizure threshold[3].

References:
[1]. Chijiwa T, et al. Inhibition of forskolin-induced neurite outgrowth and protein phosphorylation by a newly synthesized selective inhibitor of cyclic AMP-dependent protein kinase, N-[2-(p-bromocinnamylamino)ethyl]-5-isoquinolinesulfonamide (H-89), of PC12D
[2]. Blazev R, et al. Effects of the PKA inhibitor H-89 on excitation-contraction coupling in skinned and intact skeletal muscle fibres. J Muscle Res Cell Motil. 2001;22(3):277-86.

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[3]. Hosseini-Zare MS, et al. Effects of pentoxifylline and H-89 on epileptogenic activity of bucladesine in pentylenetetrazol-treated mice. Eur J Pharmacol. 2011 Nov 30;670(2-3):464-70.

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