CypK

Cat. No.: HY-134669

CAS No.: 1610703-09-7

Molecular Formula: $C_{12}H_{20}N_2O_4$

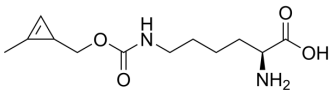
Molecular Weight: 256.3

Target: Others

Pathway: Others

Storage:

Powder	-20°C	3 years
In solvent	-80°C	6 months
	-20°C	1 month



SOLVENT & SOLUBILITY

In Vitro

DMSO : 30 mg/mL (117.05 mM; ultrasonic and adjust pH to 2 with HCl)

Preparing Stock Solutions	Mass / Solvent / Concentration	1 mg	5 mg	10 mg
	1 mM	3.9017 mL	19.5084 mL	39.0168 mL
	5 mM	0.7803 mL	3.9017 mL	7.8034 mL
	10 mM	0.3902 mL	1.9508 mL	3.9017 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: ≥ 3 mg/mL (11.71 mM); Clear solution
2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
   Solubility: ≥ 3 mg/mL (11.71 mM); Clear solution
3. Add each solvent one by one: 10% DMSO >> 90% corn oil
   Solubility: ≥ 3 mg/mL (11.71 mM); Clear solution

BIOLOGICAL ACTIVITY

Description

CypK (N-Cyclopropene-L-Lysine), a cyclopropene derivative of lysine, is efficiently incorporated into antibodies through genetic-code expansion. CypK is a minimal bioorthogonal handle for the creation of stable therapeutic protein conjugates[1][2].

In Vitro

Guidelines (Following is our recommended protocol. This protocol only provides a guideline, and should be modified according to your specific needs).
CypK assay[1] (express the antibody):
1. Thaw a vial of HEK suspension cells in a 250 mL flask containing 50 mL of expression medium supplemented with 100

units/mL penicillin, 100 µg/mL streptomycin, and 250 ng/mL amphotericin B. Keep the cells at 37 ☒ with 8% CO_2 in humidified incubators equipped with a shaker at 125 rpm. Split cells to 0.3-0.5 x 10^6 cells/mL (every 2-3 days) at least 2 times before transfecting.

2. When a density of 2.5 x 10^6 cells/mL is reached (2-3 days after splitting), prepare a fresh solution of 100 mM CypK. For this purpose, weigh 64 mg of CypK, add 2.5 mL of 0.1 sodium hydroxide, vortex, spin down to recover all undissolved particles and sonicate.

3. Add 2.5 mL of CypK (100 mM in 0.1 M NaOH) to 42.5 mL of expression medium supplemented with antibiotics. Mix well, add 250 µL of 0.1 M HCl, and sterilize using a 0.22 µm filter.

4.Dilute 50 µg of HC and LC pKym1 plasmids to 2.5 mL with reduced serum medium. In a separate tube, dilute 135 µL of transfection reagent to 2.5 mL with reduced serum medium.

5. Five minutes after preparing the solutions, mix the plasmids and the transfection reagent solution and incubate for 20 min to allow the formation of complexes between the DNA and the transfection reagent.

6. In the meantime, centrifuge 125 million cells at the target density for 5 min at 500 x g, resuspend with the expression medium containing CypK and add the DNA–transfection reagent mixture.

7. After incubating cells for 20 h, add 250 µL of transfection reagent enhancers included in the kit.

8. Harvest antibodies from the supernatant 6-7 days after addition of CypK (no change of medium is required during expression).

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

REFERENCES

[1]. Oller-Salvia B. Genetic Encoding of a Non-Canonical Amino Acid for the Generation of Antibody-Drug Conjugates Through a Fast Bioorthogonal Reaction. J Vis Exp. 2018 Sep 14;(139):58066.

[2]. Oller-Salvia B, et, al. Rapid and Efficient Generation of Stable Antibody-Drug Conjugates via an Encoded Cyclopropene and an Inverse-Electron-Demand Diels-Alder Reaction. Angew Chem Int Ed Engl. 2018 Mar 5;57(11):2831-2834.

Tel: 609-228-6898 Fax: 609-228-5909 E-mail: tech@MedChemExpress.com

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