MU1742: A Chemical probe for CK1δ and CK1ε

Version 1.0 (11th October 2021)

Web link for more details: https://www.thesgc.org/chemical-probes/mu1742

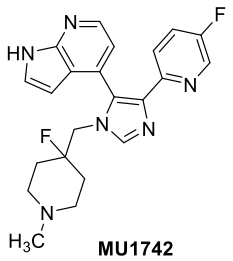
Overview

Casein kinases 1 (CK1) belong to the family of serine/threonine kinases. There are six CK1 isoforms (CK1α, CK1γ1, CK1γ2, CK1γ3, CK1*δ*, CK1ε) and several splice variants in humans. CK1 regulates Wnt, Hh and Hippo pathways, which are important for growth, development and homeostasis. CK1 kinases are involved in the regulation of various cellular states, processes and functions such as chromosome segregation, gene expression, cellular morphology, immune response and inflammation, membrane trafficking, cytokinesis, autophagy, cell stemness and differentiation, cell survival, proliferation and apoptosis. Although CK1 isoforms are similar in their structure and function (especially CK1*δ*/ε), they have also distinct and specific functions.

Summary

Chemical Probe Name	MU1742
Negative control compound	MU2027
Target(s) (synonyms)	CK1*δ*, CK1ε, CSNK1*δ*, CSNK1ε, Casein kinase 1 delta, Casein kinase 1 epsilon
Recommended cell assay concentration	For cellular assays, we recommend to keep MU1742 concentrations below 5 µM.
Suitability for *in* vivo use and recommended dose	Suitable for in vivo utilization. F (mice)= 57 %. Favourable PK profile was observed upon PO administration of 20 mg/kg, while also 100mg/kg dose was well received (mice).
Publications	
Orthogonal chemical probes	Jansen CK1 *δ* probe 6204
***In vitro* assay(s) used to characterise**	DSF, in vitro inhibition (Reaction Biology)
Cellular assay(s) for target-engagement	NanoBRET™, WB, Cell migration (chemotaxis inhibition), TopFlash (luciferase reporter assay)

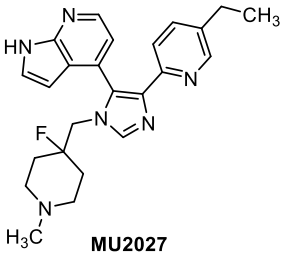
Chemical Probe & Negative Control Structures and Use



MU1742

CK1δ/ε chemical probe

SMILES: FC(C=C1)=CN=C1C2=C(C3=CC=NC4=C3C=CN4)N(CC5(F)CCN(C)CC5)C=N2
InChiKey: SWOIFXHMBKFCRM-UHFFFAOYSA-N
Molecular weight: 408.46
Storage. Stability not tested. Recommendation: Long term storage at -20 °C. Short term storage at room temperature.
Dissolution: It is possible to prepare at least 10 mM DMSO solution.
For aqueous solutions (in vivo experiments) we recommend to formulate MU1742 as a dihydrochloride salt (.2HCl).



MU2027

Negative control

SMILES: FC1(CCN(C)CC1)CN2C(C3=CC=NC4=C3C=CN4)=C(C5=NC=C(CC)C=C5)N=C2
InChiKey: SRSOVGVHVNSXTC-UHFFFAOYSA-N
Molecular weight: 418.52
Storage. Stability not tested. Recommendation: Long term storage at -20 °C. Short term storage at room temperature.
Dissolution: It is possible to prepare at least 10 mM DMSO solution.

Chemical Probe Profile

***In vitro* Potency & Selectivity:**

MU1742 was profiled against 415 protein kinases at 1 µM concentration (Reaction biology), showing that only CK1 kinases were strongly inhibited and no off target were observed below the threshold of 40 % residual activity. CK1 kinases were inhibited with following potency (IC_{50}): CK1α1 = 7.2 nM, CK1α1L = 520 Nm, CK1*δ* = 6.1 nM and CK1ε = 27.7 nM. MU1742 doesn't inhibit CK1γ isoforms.

Potency in Cells and Cellular Target Engagement:

Cellular target engagement for confirmed by NanoBRET assay with intact HEK 293 cells, indicating only weak potency towards CK1α1 (EC_{50} = 3 500 nM) but strong potency towards CK1δ and CK1ε (EC_{50} = 47 nM and 220 nM respectively). In cellulo activity was further confirmed using WB via effect on CK1δ/ε autophosphorylation, DVL3 phosphorylation or B-catenin stabilization (CK1α). In cellulo activity was further confirmed by Cell migration assay (chemotaxis inhibition) and TopFlash (luciferase reporter assay). MU1742 didn't exhibit any significant crude cytotoxic effect in JURKAT and HEK 293 cell lines up to 10 µM concentration over 24 hours (Alamar blue as an indicator).