TRAM-34

Cat. No.: HY-13519

CAS No.: 289905-88-0

Molecular Formula: $C_{22}H_{17}ClN_2$

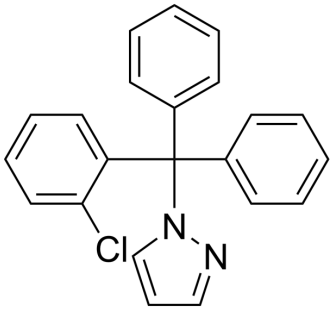
Molecular Weight: 345

Target: Potassium Channel

Pathway: Membrane Transporter/Ion Channel

Storage:

Powder	-20°C	3 years
	4°C	2 years
In solvent	-80°C	2 years
	-20°C	1 year



SOLVENT & SOLUBILITY

In Vitro

DMSO : 25 mg/mL (72.46 mM; Need ultrasonic)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.8986 mL	14.4928 mL	28.9855 mL
	5 mM	0.5797 mL	2.8986 mL	5.7971 mL
	10 mM	0.2899 mL	1.4493 mL	2.8986 mL

Please refer to the solubility information to select the appropriate solvent.

In Vivo

1. Add each solvent one by one: 20% SBE-β-CD in saline
   Solubility: 5 mg/mL (14.49 mM); Suspended solution; Need ultrasonic
2. Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline
   Solubility: ≥ 2.08 mg/mL (6.03 mM); Clear solution
3. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
   Solubility: 2.08 mg/mL (6.03 mM); Suspended solution; Need ultrasonic
4. Add each solvent one by one: 10% DMSO >> 90% corn oil
   Solubility: ≥ 2.08 mg/mL (6.03 mM); Clear solution

BIOLOGICAL ACTIVITY

Description

TRAM-34 is a highly selective blocker of intermediate-conductance calcium-activated K^+ channel (IKCa1) (K_d=20 nM).

IC_{50} & Target

Kd: 20 nM (IKCa1)[1]

In Vitro

TRAM-34 selectively blocks the IKCa1 current (K_d=25 nM), TRAM-34 also blocks IKCa1 currents in human T84 colonic

epithelial cells with equivalent potency (K_d=22 nM). TRAM-34 inhibits the cloned and the native IKCa1 channel in human T lymphocytes with a K_d of 20-25 nM and is 200- to 1,500-fold selective over other ion channels. The dose-response curve reveals a K_d of 20±3 nM and a Hill coefficient of 1.2 with 1 μM calcium in the pipette[1].
TRAM-34, a specific inhibitor of K_{Ca} 3.1 channels increased or decreased cell proliferation depending on the concentration. At intermediate concentrations (3-10 μM) TRAM-34 increased cell proliferation, whereas at higher concentrations (20-100 μM) TRAM-34 decreased cell proliferation. The enhancement of cell proliferation caused by TRAM-34 is blocked by the oestrogen receptor antagonists ICI182,780 and Tamoxifen. TRAM-34 also increases progesterone receptor mRNA expression, decreased oestrogen receptor-α mRNA expression and reduced the binding of radiolabelled oestrogen to MCF-7 oestrogen receptor, in each case mimicking the effects of 17β-oestradiol[2].
MCE has not independently confirmed the accuracy of these methods. They are for reference only.

In Vivo

Mice (n=5) injected intravenously with a single dose of TRAM-34 (0.5 mg/kg; 29 μM) appeared clinically normal during the 7-day study. The body-weight data of the TRAM-34-treated group (day 1:17.8 g; day 7: 27.0 g) are similar to control mice injected with the vehicle (day 1: 17.4 g; day 7: 23.4 g). Collectively, data from these limited toxicity studies suggest that TRAM-34 is not acutely toxic at ≈500-1,000 times the channel-blocking dose[1].
Treatment with TRAM-34 results in a significant reduction in hematoxylin & eosin (H&E) defined lesion area with the mean infarct size being reduced from 22.6±3.6% in the controls (n=8) to 11.3±2.8% in rats treated with 10 mg/kg TRAM-34 (n=6, mean±s.e.m., P=0.039) and to 8.1±1.9% in rats treated with 40 mg/kg TRAM-34 (n=8; P=0.004). The treatment also tended to reduce brain shrinkage. However, the results are only statistically significant with 40 mg/kg TRAM-34 (P=0.013), but not for the 10 mg/kg group (P=0.11)[3].
MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Kinase Assay [2]

MCF-7 cell protein (250 μg) is incubated at room temperature for 2 h in TEDG buffer in the presence of 0.1 nM [2,4,6,7,16,17-^{3}H(N)]-oestradiol ([^{3}H]-E2) (110 Ci/mmol) in a total final volume of 500 μL. Non-specific binding is assessed in the presence of a 100-fold excess of non-radioactive E2. TRAM-34 and E2 standards are diluted in phenol red-free 5% DCC-FBS MEM containing supplements before being added to the cytosolic protein. A vehicle control comprised of 5% DCC-FBS MEM containing supplements with 0.7% DMSO. To separate ER-bound [^{3}H]-E2 from unbound [^{3}H]-E2, 250 μL of hydroxylapatite (HAP, 60% in TEDG buffer) is added, the mixture is vortexed every 5 min over 15 min and centrifuged at 1000×g for 10 min. The HAP-[^{3}H]-E2-ER complex is washed with TEDG buffer, centrifuged and the wash step repeated. To elute [^{3}H]-E2 from the HAP-[^{3}H]-E2-ER complex, 500 μL of 100% ethanol is added and the mixture then incubated for 15 min and centrifuged at 1034×g for 10 min. The separated [^{3}H]-E2 is removed and added to 2 mL of scintillation fluid. Radioactivity is quantified using a Beckman LS 5000TA scintillation counter. Competition of [^{3}H]-E2 with TRAM-34 is assayed in quadruplicate on four independent protein extractions. An apparent dissociation constant of 0.135±0.034 nM (n=3) and a maximum binding capacity of 48.3±5.4 fmol/mg (n=3) are determined by Scatchard analysis[2].
MCE has not independently confirmed the accuracy of these methods. They are for reference only.

Animal Administration [1][3]

Mice[1]
Five CF-1BR mice (17-19 g) are injected intravenously with a single 1.0-ml dose of 0.5 mg/kg TRAM-34 (in mammalian Ringer solution with 1% ethanol and 2.5% BSA). Five control mice are injected with an equal volume of the vehicle. Mice are observed for adverse effects immediately after dosing, at 4 h after injection and daily for 7 days.
Rats[3]
Adult male Wistar rats weighing 160 to 180 g are used. Rats receive TRAM-34 at 10 mg/kg, 40 mg/kg or vehicle (Miglyol 812 neutral oil at 1 μL/g) twice daily intraperitoneally for 7 days starting 12 hours after reperfusion. Neurological deficits are scored according to a 4-score test and a tactile and proprioceptive limb-placing test as follows: (1) 4-score test (higher score for more severe neurological deficits): 0=no apparent deficit; 1=contralateral forelimb is consistently flexed during suspension by holding the tail; 2=decreasing grip ability on the contralateral forelimb while tail pulled; 3=spontaneous movement in all directions but circling to contralateral side when pulled by the tail; 4=spontaneous contralateral circling or depressed level of consciousness. (2) 14-score limb-placing test (lower score for more severe neurological deficits): proprioception, forward extension, lateral abduction, and adduction are tested with vision or tactile stimuli. For visual limb placing, rats are held and slowly moved forward or lateral toward the top of a table. Normal rats placed both forepaws on

	the tabletop. Tactile forward and lateral limb placing are tested by lightly contacting the table edge with the dorsal or lateral surface of a rat's paw while avoiding whisker contact and covering the eyes to avoid vision. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

CUSTOMER VALIDATION

- Nat Commun. 2022 Jun 21;13(1):3544.
- Haematologica. 2017 Oct;102(10):e415-e418.
- Life Sci. 2023 Dec 4, 122326.
- J Inflamm Res. 2021 Mar 5;14:719-735.
- Cell Calcium. 2022 Jun;104:102571.

See more customer validations on www.MedChemExpress.com

REFERENCES

[1]. Wulff H, et al. Design of a potent and selective inhibitor of the intermediate-conductance Ca2+-activated K+ channel, IKCa1: a potential immunosuppressant. Proc Natl Acad Sci U S A. 2000 Jul 5;97(14):8151-6.

[2]. Roy JW, et al. The intermediate conductance Ca2+-activated K+ channel inhibitor TRAM-34 stimulates proliferation of breast cancer cells via activation of oestrogen receptors. Br J Pharmacol. 2010 Feb 1;159(3):650-8.

[3]. Chen YJ, et al. The KCa3.1 blocker TRAM-34 reduces infarction and neurological deficit in a rat model of ischemia/reperfusion stroke. J Cereb Blood Flow Metab. 2011 Dec;31(12):2363-74.

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