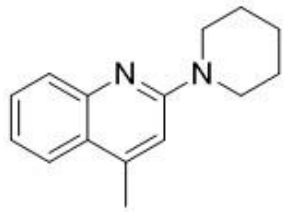
Product data sheet

MedKoo Cat#: 522358 Name: ML204 CAS#: 5465-86-1 (free base) Chemical Formula: $C_{15}H_{18}N_2$ Exact Mass: 226.147 Molecular Weight: 226.32	
Product supplied as:	Powder
Purity (by HPLC):	≥ 98%
Shipping conditions	Ambient temperature
Storage conditions:	Powder: -20°C 3 years; 4°C 2 years.
	In solvent: -80°C 3 months; -20°C 2 weeks.

1. Product description:
ML204 is a novel and potential TRPC4 Channel inhibitor. ML204 inhibited TRPC4β-mediated intracellular Ca(2+) rise with an IC(50) value of 0.96 μm and exhibited 19-fold selectivity against muscarinic receptor-coupled TRPC6 channel activation. ML204 represents an excellent novel tool for investigation of TRPC4 channel function and may facilitate the development of therapeutics targeted to TRPC4.

2. CoA, QC data, SDS, and handling instruction
SDS and handling instruction, CoA with copies of QC data (NMR, HPLC and MS analytical spectra) can be downloaded from the product web page under "QC And Documents" section. Note: copies of analytical spectra may not be available if the product is being supplied by MedKoo partners. Whether the product was made by MedKoo or provided by its partners, the quality is 100% guaranteed.

3. Solubility data

Solvent	Max Conc. mg/mL	Max Conc. mM
DMSO	30.0	132.66
Ethanol	37.0	163.49

4. Stock solution preparation table:

Concentration / Solvent Volume / Mass	**1 mg**	**5 mg**	**10 mg**
1 mM	4.42	22.09	44.19
5 mM	0.88	4.42	8.84
10 mM	0.44	2.21	4.42
50 mM	0.09	0.44	0.88

5. Molarity Calculator, Reconstitution Calculator, Dilution Calculator
Please refer the product web page under section of "Calculator"

6. Recommended literature which reported protocols for in vitro and in vivo study
In vitro study

1. Lee JH, Wu WH, Huang XY, Jun JY, Choi S. Transient Receptor Potential Canonical 4 and 5 Channel Antagonist ML204 Depolarized Pacemaker Potentials of Interstitial Cells of Cajal. J Neurogastroenterol Motil. 2020 Sep 30;26(4):521-528. doi: 10.5056/jnm20064. PMID: 32321198; PMCID: PMC7547197.
2. Alom F, Miyakawa M, Matsuyama H, Nagano H, Tanahashi Y, Unno T. Possible antagonistic effects of the TRPC4 channel blocker ML204 on M2 and M3 muscarinic receptors in mouse ileal and detrusor smooth muscles and atrial myocardium. J Vet Med Sci. 2018 Sep 13;80(9):1407-1415. doi: 10.1292/jvms.18-0197. Epub 2018 Jul 5. PMID: 29973432; PMCID: PMC6160885.

In vivo study

1. Lee SH, Tonello R, Choi Y, Jung SJ, Berta T. Sensory Neuron-Expressed TRPC4 Is a Target for the Relief of Psoriasiform Itch and Skin Inflammation in Mice. J Invest Dermatol. 2020 Nov;140(11):2221-2229.e6. doi: 10.1016/j.jid.2020.03.959. Epub 2020 Apr 11. PMID: 32289348.
2. Pereira DMS, Mendes SJF, Alawi K, Thakore P, Aubdool A, Sousa NCF, da Silva JFR, Castro JA Jr, P Pereira IC, Silva LCN, Grisotto MAG, Monteiro-Neto V, Costa SKP, da Costa R, Calixto JB, Brain SD, Fernandes ES. Transient Receptor Potential

Product data sheet

Canonical Channels 4 and 5 Mediate Escherichia coli-Derived Thioredoxin Effects in Lipopolysaccharide-Injected Mice. Oxid Med Cell Longev. 2018 Jun 10;2018:4904696. doi: 10.1155/2018/4904696. PMID: 29983857; PMCID: PMC6015690.

7. Bioactivity

Biological target:

ML204 is a potent, selective TRPC4/TRPC5 channel inhibitor.

In vitro activity

The purpose of this study was to investigate an effect of ML204 (an inhibitor of transient receptor potential canonical 4 and 5 [TRPC4/5] channels) on interstitial cells of Cajal (ICCs) and therefore determine whether TRPC4/5 channels act on ICC-generated pacemaker activity. Whole cell patch clamp analysis, measurements of the intracellular Ca2+ concentration, and reverse transcription polymerase chain reaction were enforced to determine the effect of ML204 (10 μM) or englerin A (a selective activator of TRPC4/5 channeles, 10 μM) and the existence of TRPC4/5 in mouse small intestinal ICC. Treatment of ICCs with ML204 or englerin A caused the membrane potentials to depolarize. This depolarization effect of membrane potentials by ML204 in ICCs was observed to be concentration-dependent. After treating Ca2+- and Na+-free solutions or flufenamic acid (a non-selective cation channel blocker), the pacemaker potentials in the ICCs were abolished. A specific anoctamin 1 channel blocker did not have any effect on the pacemaker activity in ML204-untreated control cells; however, they blocked ML204-induced pacemaker activity in ICCs. Specific primers designed against TRPC4 and TRPC5 detected the presence of TRPC4/5 in small intestinal ICCs, and the application of ML204 increased raise the frequency of Ca2+ oscillations in ICCs, as assessed using Fluo-4 AM. The results implied that ML204 could not inhibit the pacemaker activity but depolarized the membrane potential of ICCs by regulating intracellular Ca2+ oscillations and anoctamin 1 channels.

J Neurogastroenterol Motil. 2020 Sep 30;26(4):521-528. https://pubmed.ncbi.nlm.nih.gov/32321198/

In vivo activity

It is still not known whether other pruritogens are dependent on TRPC4. Common pruritogens were then tested in Trpc4 KO mice and in mice treated with the specific TRPC4 antagonist ML204 (Alom et al., 2018, Miller et al., 2011).It was found that 5-HT–evoked acute itch (assessed by the number of scratch bouts of the mice hind paw over 30 minutes) was significantly attenuated in Trpc4 KO and ML204-treated mice (Figure 2a and b). Similarly, histamine-evoked itch was also attenuated in Trpc4 KO and ML204-treated mice (Figure 2c and d). However, chloroquine-evoked acute itch was independent of TRPC4 (Figure 2e and f), and ML204 did not change acute itch induced either by the protease-activated receptor-2 peptide SLIGRL-NH2 or toll-like receptor 7 agonist imiquimod (IMQ) (see Supplementary Figure S2a and b). To increase the clinical relevance of our study, it was then enquired whether targeting TRPC4 locally by a small-molecule inhibitor (i.e., ML204) can also promote the resolution of established psoriasiform itch and skin inflammation (Figure 6a). Mice intradermally treated with ML204 for 3 days showed reduced spontaneous scratch and alloknesis at day 7 after the initial application of IMQ (Figure 6b). However, mice treated with ML204 exhibited a significant reversal at day 7 in both erythema and scaling compared with mice treated with a CTRL vehicle (Figure 6c). Histological staining also indicated a decrease in psoriasiform skin inflammation as there were reduced thickness of epidermis and number of immune cells in the skin in mice treated with ML204 (Supplementary Figure S5a–c). In particular, there was a significant decrease in the skin tissue of protein levels for the pro-inflammatory cytokines IL-17F and CCL2 and neuropeptide CGRP (Figure 6d); these pro-inflammatory cytokines and neuropeptide play crucial roles in the local inflammation and pathogenesis of psoriasis

J Invest Dermatol. 2020 Nov;140(11):2221-2229.e6. https://www.jidonline.org/article/S0022-202X(20)31372-5/fulltext