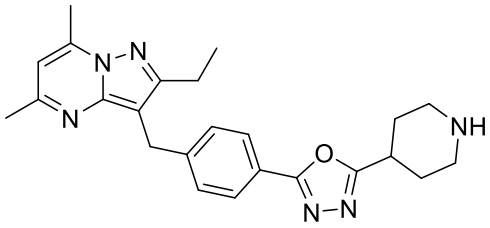
Product data sheet

MedKoo Cat#: 555805 Name: NE 52-QQ57 CAS#: 1401728-56-0 Chemical Formula: $C_{24}H_{28}N_6O$ Exact Mass: 416.2325 Molecular Weight: 416.53	
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:
NE52-QQ57 is an orally available GPR4 antagonist with an IC50 of 70 nM. NE 52-QQ57 significantly inhibited the AGE-induced increased expression of several key inflammatory cytokines and signaling molecules, including tumor necrosis factor-α (TNF-α), interleukin (IL)-1β, IL-6, inducible nitric oxide synthase (iNOS), nitric oxide (NO), cyclooxygenase 2 (COX2), and prostaglandin E2 (PGE2).

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	10	24.1
Ethanol	45	108

4. Stock solution preparation table:

Concentration / Solvent Volume / Mass	**1 mg**	**5 mg**	**10 mg**
1 mM	2.40 mL	12.00 mL	24.01 mL
5 mM	0.48 mL	2.40 mL	4.80 mL
10 mM	0.24 mL	1.20 mL	2.40 mL
50 mM	0.05 mL	0.24 mL	0.48 mL

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. Hosford PS, Mosienko V, Kishi K, Jurisic G, Seuwen K, Kinzel B, Ludwig MG, Wells JA, Christie IN, Koolen L, Abdala AP, Liu BH, Gourine AV, Teschemacher AG, Kasparov S. CNS distribution, signalling properties and central effects of G-protein coupled receptor 4. Neuropharmacology. 2018 Aug;138:381-392. doi: 10.1016/j.neuropharm.2018.06.007. Epub 2018 Jun 9. PMID: 29894771; PMCID: PMC6063991.

In vivo study

1. Hosford PS, Mosienko V, Kishi K, Jurisic G, Seuwen K, Kinzel B, Ludwig MG, Wells JA, Christie IN, Koolen L, Abdala AP, Liu BH, Gourine AV, Teschemacher AG, Kasparov S. CNS distribution, signalling properties and central effects of G-protein coupled receptor 4. Neuropharmacology. 2018 Aug;138:381-392. doi: 10.1016/j.neuropharm.2018.06.007. Epub 2018 Jun 9. PMID: 29894771; PMCID: PMC6063991.

2. Velcicky J, Miltz W, Oberhauser B, Orain D, Vaupel A, Weigand K, Dawson King J, Littlewood-Evans A, Nash M, Feifel R, Loetscher P. Development of Selective, Orally Active GPR4 Antagonists with Modulatory Effects on Nociception, Inflammation, and

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Angiogenesis. J Med Chem. 2017 May 11;60(9):3672-3683. doi: 10.1021/acs.jmedchem.6b01703. Epub 2017 Apr 26. PMID: 28445047.

7. Bioactivity

Biological target:

NE 52-QQ57 is a selective, and orally available GPR4 antagonist with an IC50 of 70 nM. NE 52-QQ57 has anti-inflammatory activity.

In vitro activity

The effect of NE 52-QQ57 on proton-mediated cAMP accumulation in cells over-expressing GPR4 was dependent on pH (Fig. 3A) and this dependence in the acidic range could be explained by competition with protons. NE 52-QQ57 (100nM) was maximally effective at pH 7.7 but approximately equally effective at pH 8.0 and 7.4. At pH 7.1, the potency of NE 52-QQ57 was reduced and at pH 6.8 NE 52-QQ57 (100nM) was ineffective. At pH 7.4, NE 52-QQ57 acts as a highly potent antagonist of GPR4 mediated cAMP accumulation with IC50 of 26.8nM (Fig. 3B).

Reference: Neuropharmacology. 2018 Aug;138:381-392. https://www.ncbi.nlm.nih.gov/pmc/articles/pmid/29894771/

In vivo activity

Minute ventilation (VE) which is the cumulative of RR and VT was not significantly affected at baseline by 20 mg kg−1 of NE-52-QQ57 in either rats (n = 8) or mice (n = 10). Relative to the vehicle (25% DMSO), NE-52-QQ57 significantly attenuated VE during hypercapnia at 5 and 10% CO2 in both species. For example, with 10% CO2 VE increased to 216 ± 11 a.u. in the group treated with vehicle, but only to 164 ± 18 a.u. in mice treated with NE-52-QQ57 ($p < 0.001$). Similarly, in rats at 10% CO2 VE increased to 662 ± 40 a.u. in the vehicle group, but to 568 ± 22 a.u. in the treated group ($p < 0.001$). Thus, in freely behaving mice and rats NE22-QQ57 blunted hypercapnic response to CO2. In anaesthetised rats, hypercapnia (10% CO2 in the inspired air) increased respiratory rate from 77 ± 9 to 94 ± 13 bursts/min (Fig. 6A), and this was not affected following systemic administration of NE 52-QQ57 (20 mg kg−1; n = 8, 76 ± 9 to 95 ± 12 bursts min−1; $p = 0.92$). NE 52-QQ57 had no effect on CO2-evoked increases in diaphragm EMG amplitude (0.25 ± 0.03 to 0.33 ± 0.04V cf. 0.25 ± 0.02 to 0.34 ± 0.03V; $p = 0.94$) and minute ventilation (19 ± 3 to 32±7A.U cf. 20 ± 3 to 33±6A.U; $p = 0.88$). Systemic NE 52-QQ57 also had no effect on basal respiratory rate (Fig. 6B). To additionally ensure that NE 52-QQ57 reaches the central GPR4 targets, the drug was then delivered directly on the ventral surface of the brainstem, where the RTN is located. Hypercapnia (10% CO2 in the inspired air) in 7 animals with denervated peripheral chemoreceptors increased respiratory frequency, phrenic nerve amplitude and minute ventilation (Fig. 6C). Hypercapnia increased the respiratory rate from 18 ± 6 to 40 ± 5 bursts min−1 following application of the vehicle on the ventral brainstem surface. CO2 had a similar effect in the presence of NE-52-QQ57 (1 mM) on the ventral brainstem surface (18 ± 4 to 41 ± 5 bursts min−1; $p = 0.86$, Fig. 6C). NE 52-QQ57 had no effect on other components of the hypercapnic respiratory response including increases in phrenic nerve burst amplitude (0.04 ± 0.01 to 0.06 ± 0.02V vs. 0.04 ± 0.02 to 0.06 ± 0.02V; $p = 0.92$) and minute ventilation (11 ± 4 to 34±8A.U vs. 11 ± 2 to 33±7A.U; $p = 0.97$). Direct application of NE 52-QQ57 to the ventral medulla had no effect on baseline respiratory activity (Fig. 6D).

Reference: Neuropharmacology. 2018 Aug;138:381-392. https://www.ncbi.nlm.nih.gov/pmc/articles/pmid/29894771/