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产品名称：MRK-016
产品别名：MRK-016

生物活性：

Description	MRK-016 is a selective, orally bioavailable inverse agonist of $GABA_A$ α5 receptor, with an EC_{50} of 3 nM for $GABA_A$α5, and K_is of 0.83, 0.85, 0.77 and 1.4 nM for human $GABA_A$ α1β3γ2, $GABA_A$ α2β3γ2, $GABA_A$ α3β3γ2, and $GABA_A$ α5β3γ2, respectively; MRK-016 also readily penetrates the CNS.
IC_{50} & Target	EC50: 3 nM ($GABA_A$ α5)[1] Ki: 0.83 nM (Human $GABA_A$ α1β3γ2), 0.85 nM (Human $GABA_A$ α2β3γ2), 0.77 nM (Human $GABA_A$ α3β3γ2), 1.4 nM (Human $GABA_A$ α5β3γ2)[1]
In Vitro	MRK-016 is a selective, orally bioavailable inverse agonist of $GABA_A$ α5 receptor, with an EC_{50} of 3 nM for $GABA_A$α5, and K_is of 0.83, 0.85, 0.77 and 1.4 nM for human $GABA_A$ α1β3γ2, $GABA_A$ α2β3γ2, $GABA_A$ α3β3γ2, and $GABA_A$ α5β3γ2, respectively[1][2]. MRK-016 is a full inverse agonist at the α5-subtype, shows very weak affinity at the $GABA_A$α4β3γ2-subtype (K_i 395 ± 173 nM) and is essentially inactive at the $GABA_A$ α6β3γ2 receptor (K_i > 4000 nM)[1]. MRK-016 shows a weak effect on $GABA_A$ α4β3γ2 with a K_i of 400 nM. MRK-016 (100 nM) alao increases long-term potentiation in mouse hippocampal slices[2].
In Vivo	MRK-016 does not enhance pentylenetetrazole-induced convulsions at 10 mg/kg via ip, or cause seizures at 30 mg/kg, via po for 20 days in mice. MRK-016 shows no obvious anxiogenic-like effects in rats at doses that occupy >95% of benzodiazepine (BZ) binding sites. MRK-016 (0.3, 1, and 3 mg/kg, p.o.) dose-dependently improves performance of rats hippocampal-dependent memory task[1]. MRK-016 (0.3-30 mg/kg, p.o.) causes good receptor occupancy in rats. MRK-016 (0.3, 1, or 3 mg/kg p.o.) shows cognition-enhancing activity in the delayed matching-to-position version of the Morris water maze. MRK-016 (1, 3, or 10 mg/kg i.p.) does not produce kindling in mice[2]. MRK-016 (3 mg/kg, i.p.) protects against LPS-induced learning/memory decrements in mice[3].
Solvent&Solubility	***In Vitro:*** **DMSO : 50 mg/mL (135.73 mM;** Need ultrasonic)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.7145 mL	13.5726 mL	27.1451 mL
	5 mM	0.5429 mL	2.7145 mL	5.4290 mL
	10 mM	0.2715 mL	1.3573 mL	2.7145 mL

*请根据产品在不同溶剂中的溶解度选择合适的溶剂配制储备液；一旦配成溶液，请分装保存，避免反复冻融造成的产品失效。

储备液的保存方式和期限：-80°C, 6 months; -20°C, 1 month。-80°C 储存时，请在 6 个月内使用，-20°C 储存时，请在 1 个月内使用。

In Vivo:

请根据您的实验动物和给药方式选择适当的溶解方案。以下溶解方案都请先按照 In Vitro 方式配制澄清的储备液，再依次添加助溶剂：

——为保证实验结果的可靠性，澄清的储备液可以根据储存条件，适当保存；体内实验的工作液，建议您现用现配，当天使用； 以下溶剂前显示的百分比是指该溶剂在您配制终溶液中的体积占比；如在配制过程中出

	现沉淀、析出现象，可以通过加热和/或超声的方式助溶 1.请依序添加每种溶剂： 10% DMSO→40% PEG300 →5% Tween-80 → 45% saline Solubility: ≥ 2.75 mg/mL (7.46 mM); Clear solution 此方案可获得 ≥ 2.75 mg/mL (7.46 mM，饱和度未知) 的澄清溶液。 以 1 mL 工作液为例，取 100 μL 27.5 mg/mL 的澄清 DMSO 储备液加到 400 μL PEG300 中，混合均匀向上述体系中加入 50 μL Tween-80，混合均匀；然后继续加入 450 μL 生理盐水定容至 1 mL。 2.请依序添加每种溶剂： 10% DMSO→ 90% (20% SBE-β-CD in saline) Solubility: ≥ 2.75 mg/mL (7.46 mM); Clear solution 此方案可获得 ≥ 2.75 mg/mL (7.46 mM，饱和度未知) 的澄清溶液。 以 1 mL 工作液为例，取 100 μL 27.5 mg/mL 的澄清 DMSO 储备液加到 900 μL 20% 的 SBE-β-CD 生理盐水水溶液中，混合均匀。 3.请依序添加每种溶剂： 10% DMSO →90% corn oil Solubility: ≥ 2.75 mg/mL (7.46 mM); Clear solution 此方案可获得 ≥ 2.75 mg/mL (7.46 mM，饱和度未知) 的澄清溶液，此方案不适用于实验周期在半个月以上的实验。 以 1 mL 工作液为例，取 100 μL 27.5 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。
References	[1]. Chambers MS, et al. An orally bioavailable, functionally selective inverse agonist at the benzodiazepine site of GABAA alpha5 receptors with cognition enhancing properties. J Med Chem. 2004 Nov 18;47(24):5829-32. [2]. Atack JR, et al. In vitro and in vivo properties of 3-tert-butyl-7-(5-methylisoxazol-3-yl)-2-(1-methyl-1H-1,2,4-triazol-5-ylmethoxy)-pyrazolo[1,5-d]-[1,2,4]triazine (MRK-016), a GABAA receptor alpha5 subtype-selective inverse agonist. J Pharmacol Exp Ther. 2009 Nov;331(2):470-84. [3]. Eimerbrink MJ, et al. Administration of the inverse benzodiazepine agonist MRK-016 rescues acquisition and memory consolidation following peripheral administration of bacterial endotoxin. Behav Brain Res. 2015 Jul 15;288:50-3.

实验参考：

Animal Administration	Mice[1] The proconvulsant potential MRK-016 is determined in this assay. The convulsant is pentylenetetrazole which is dosed at 15 mg/mL with an infusion rate of 0.2 mL/min. This rate is chosen to ensure that drug-naive mince reach the terminal convulsion sign within 1 min. MRK-016 is dosed intraperitoneally (ip) at a concentration of 1, 3, and 10 mg/kg in 70% PEG 300[1].
Kinase Assay	L(tk$^-$) cells expressing human recombinant $GABA_A$ receptors containing β3- and γ2-subunits in combinatioon with various α-subunits are harvested and binding performed. The displacement of [^{3}H]Ro 15-1788 binding by the test compounds (MRK-016, etc.) is measured in $GABA_A$ receptors containing eigher an α1-, α2-, α3-, and α5-subunit and from the IC_{50} and the K_i is calculated assuming respective K_d values of [^{3}H]Ro 15-1788 binding of 0.92, 1.05, 0.58 and 0.45 nM at the α1-, α2-, α3-, and α5-subtypes. Nonspecific binding is defined by the inclusion of 10 μM flunitrazepam for

	the α1-, α2-, α3-, and α5-subtypes. The percentage inhibition of [^{3}H]Ro 15-1788, the IC_{50} and the K_i values are calculated using ActivityBase[1].
References	[1]. Chambers MS, et al. An orally bioavailable, functionally selective inverse agonist at the benzodiazepine site of GABAA alpha5 receptors with cognition enhancing properties. J Med Chem. 2004 Nov 18;47(24):5829-32. [2]. Atack JR, et al. In vitro and in vivo properties of 3-tert-butyl-7-(5-methylisoxazol-3-yl)-2-(1-methyl-1H-1,2,4-triazol-5-ylmethoxy)-pyrazolo[1,5-d]-[1,2,4]triazine (MRK-016), a GABAA receptor alpha5 subtype-selective inverse agonist. J Pharmacol Exp Ther. 2009 Nov;331(2):470-84. [3]. Eimerbrink MJ, et al. Administration of the inverse benzodiazepine agonist MRK-016 rescues acquisition and memory consolidation following peripheral administration of bacterial endotoxin. Behav Brain Res. 2015 Jul 15;288:50-3.