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Drug design and synthesis of a novel κ opioid receptor agonist with an oxabicyclo[2.2.2]octane skeleton and its pharmacology

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ABSTRACT

A conformational analysis of κ opioid receptor agonists, TRK-820 and U-50,488H indicated an active conformation of TRK-820 in which the C-ring was in the boat form with the 14-OH interacting with the amide nitrogen. Based on the obtained active conformation of TRK-820, we designed and synthesized a novel κ agonist KNT-63 with oxabicyclo[2.2.2]octane skeleton. KNT-63 showed profound antinociceptive effects via the κ receptor which were as potent as that of TRK-820.

Three types of opioid receptors (μ, δ, κ) are now well established not only by pharmacological studies but by molecular biological studies.[1] Narcotic addiction is believed to be derived from μ receptor type, and therefore δ and κ types are promising drug targets for analgesics without addiction. A putative ε receptor, which has not yet been cloned, has also been proposed as another opioid receptor type and much pharmacological data support its existence.[2] To obtain ideal analgesics without addiction and other side effects derived from the μ receptor, we have synthesized various kinds of naltrexone derivatives and have reported selective ligands for κ, δ, and putative ε receptors.[3–5] Quite recently, one of our designed κ selective agonists, TRK-820 (Fig. 1),[3a,b] was launched in Japan as an antipruritic for patients undergoing dialysis. Although many arylacetamide derivatives such as U-50,488H[6] (Fig. 1) and U-69,593[7] were synthesized and developed as κ agonists, all of these derivatives were eliminated from clinical trials as not only analgesics but also as antipruritics because of their serious side effects like psychotomimetic[8] and aversive[8,9] reactions. On the other hand, TRK-820 has neither aversive nor addictive effects. We were interested in the differences in the pharmacological effects between TRK-820 and the arylacetamide derivatives, and carried out the conformational analyses of them and detailed SAR investigations of TRK-820 derivatives to develop the working hypothesis that the C-ring in TRK-820 would be in the boat form in its active conformation.[3c] Based on this hypothesis, we designed and synthesized 4,5-epoxymorphinan derivative **1** (Fig. 1) having an oxabicyclo[2.2.2]octane skeleton. Herein, we report the drug

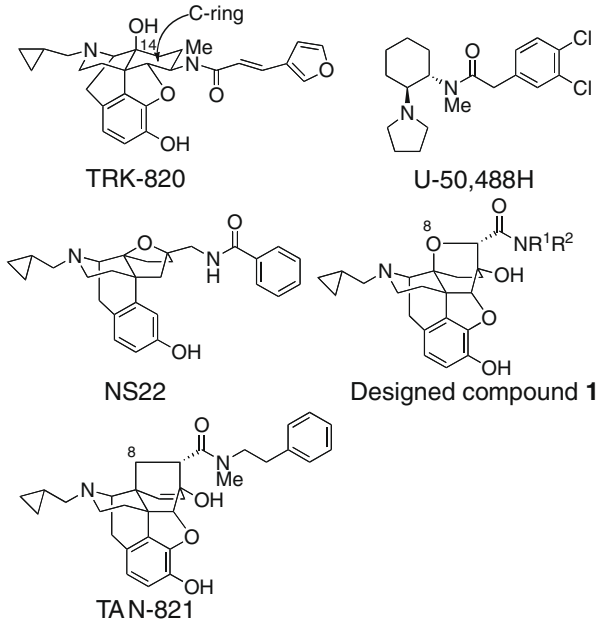

Figure 1. Structures of TRK-820, U-50,488H, NS22, TAN-821, and designed compound **1**.

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design and synthesis of the target compounds possessing the oxabicyclo[2.2.2]octane skeleton and their pharmacological properties.

TRK-820 contains a tyrosine–glycine moiety that is a common structural unit for endogenous opioid peptides. The representative exogenous opioid morphine also has the same functionality, but the arylacetamide derivatives like U-50,488H have no tyrosine–glycine moiety. To clarify the common overlapping structural units between TRK-820 and a typical arylacetamide U-50,488H, we carried out computer analyses.[10] The analyses indicated that both ligands had a basic nitrogen and an amide side chain as common overlapping pharmacophores (Fig. 2). The former would interact with an aspartic acid residue in the third transmembrane region in the receptor,[11] while the latter would play a role in recognition of κ receptor (so-called κ address site).[12] However, U-50,488H did not contain a phenolic structure, which is generally thought to be an important pharmacophore in opioid ligands.[13] It is worthwhile noting that the C-ring in TRK-820 was in the boat conformation in its active conformation, thereby elevating the amide side chain, that is, the κ address.[14] Until we proposed the active hypothetical conformation, we were unable to define any common overlapping structural units between the two κ agonists. The obtained active TRK-820 conformation was also supported by detailed SAR investigations of TRK-820 derivatives and used for a drug design of new κ agonist NS22 (Fig. 1).[3c] Although NS22 showed dose-dependent antinociceptive effects via the κ receptor in hot-plate and tail-flick tests, the analgesic effects induced by NS22 were less potent than for TRK-820. A careful comparison of structures between TRK-820 and NS22 led us to find that the position of the amide side chain in TRK-820 (red color), which was held by an interaction between the 14-OH and the amide nitrogen, was oriented a bit upper than that of NS22 (blue color, Fig. 3). The spatial difference of the amide side chain's orientation may have significant influence on κ potency and selectivity. Based on this consideration, we designed an oxabicyclo[2.2.2]octane derivative **1** whose amide side chain seemed to be located at almost the same position as that of the active conformation of TRK-820 (Fig. 4).

We attempted to synthesize an intermediate **3a** to afford designed compound **1** using Darzens reaction (Scheme 1). Naltrexone

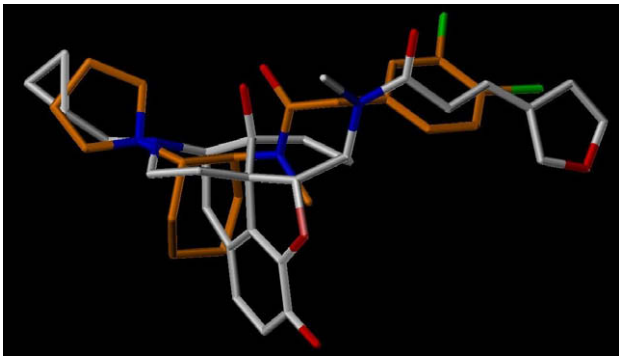


Figure 2. Superimposition of active conformations of TRK-820 (white) and U-50,488H (orange).

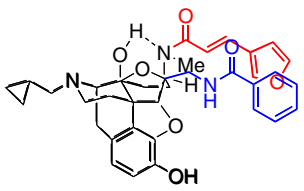


Figure 3. Comparison between active conformation of TRK-820 and NS22. The side chain in TRK-820 or NS22 was indicated by red or blue color, respectively.

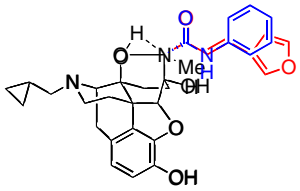
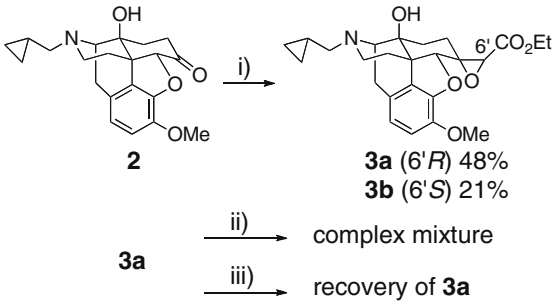
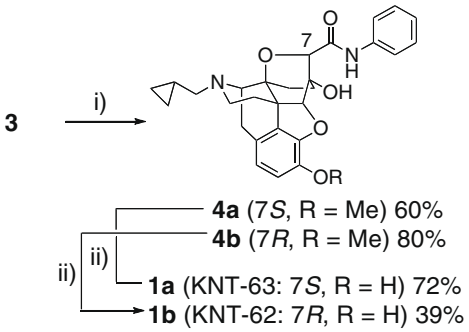


Figure 4. Comparison between active conformation of TRK-820 and designed compound (R^1 = Ph, R^2 = H; KNT-63 (**1a**)). The side chain in TRK-820 or the designed compound was indicated by red or blue color, respectively.



Scheme 1. Reagents and conditions: (i) NaH, $ClCH_2CO_2Et$, THF, rt; (ii) NaH, DMF, 90 °C; (iii) CSA, DMF, 110 °C.



Scheme 2. Reagents and conditions: (i) *n*-BuLi or NaH, $PhNH_2$, THF, reflux; (ii) BBr_3, CH_2Cl_2, rt.

methyl ether **2**[14c,15] was treated with NaH and ethyl chloroacetate in THF to give epoxyesters **3** as an *R* and *S* mixture (2:1) at the 6′ position.[16] We attempted to convert the resulting 6′*R*-epoxyester **3a** into the oxabicyclo[2.2.2]octane derivative by intramolecular cyclization. However, this approach gave either a complex product mixture under basic conditions (NaH in DMF) or led to recovery of starting epoxyester **3a** under acidic conditions (camphor sulfonic acid (CSA) in DMF). These results were probably derived from the activation of the ester group by the inductive effect of the epoxy ring under basic conditions or were due to difficulty of forming a carbonium ion at 6′ position by the inductive effect of the ester group under acidic conditions. We next tried to transform the ester group into a more inactive amide group before cyclization. Fortunately, the amidation of **3a** with aniline and *n*-BuLi in THF under reflux directly afforded the objective cyclized amide **4a** in 60% yield. 6′*S*-Isomer **3b** was also transformed to the corresponding amide **4b** under the same reaction conditions in 80% yield (except NaH was used instead of *n*-BuLi). In these transformations, no epimerizations were observed.[16] Amides **4a** and **4b** were demethylated with BBr_3 in CH_2Cl_2 to provide **1a** (KNT-63) and **1b** (KNT-62), respectively (Scheme 2).

The affinities of both compounds KNT-62 (**1b**) and KNT-63 (**1a**) for κ and μ opioid receptors were stronger than that of naltrexone

Table 1
Binding affinity of KNT-62 (**1b**), KNT-63 (**1a**), TRK-820, and naltrexone for opioid receptors[a]

Compound	K_i (μ)[b] (nM)	K_i (κ)[c] (nM)	K_i (δ)[d] (nM)
KNT-62 (**1b**)	0.205	0.152	3.23
KNT-63 (**1a**)	0.212	0.111	2.73
TRK-820	0.582	0.225	96.5
Naltrexone	0.335	0.373	20.7

[a] Binding assay was carried out in duplicate using homogenate of guinea-pig brain (κ: cerebellum, μ and δ: forebrain).
[b] [^{3}H]DAMGO was used.
[c] [^{3}H]U-69,593 was used.
[d] [^{3}H]NTI was used.

(Table 1). KNT-63 (**1a**) showed more κ selective affinity (K_i = 0.111 nM for κ and K_i = 0.212 nM for μ) than did KNT-62 (**1b**) (K_i = 0.152 nM for κ and K_i = 0.205 nM for μ). We then evaluated the antinociceptive effect induced by sc-administered KNT-63 (**1a**) using the acetic acid writhing test (AAW test). A sc-administration of KNT-63 (**1a**) produced a strong antinociceptive effect (ED_{50} = 0.0063 mg/kg) in a dose-dependent manner (Fig. 5A). This analgesia was antagonized by κ antagonist nor-BNI, but not by μ antagonist naloxone (NLX) and δ antagonist NTI (Fig. 5B). NS22 showed the same affinity for μ (K_i = 0.134 nM) and κ receptors (K_i = 0.135 nM) and antinociceptive effect (ED_{50} = 0.1 mg/kg) through κ receptor.[3c] As a result, KNT-63 (**1a**) showed slightly higher selectivity for the κ receptor and a 16-fold improved analgesic activity in comparison to NS22.

The analgesic activity of KNT-63 (**1a**) was almost the same as that of TRK-820 (ED_{50} = 0.0033 mg/kg) in AAW test (100-fold potent than morphine).[3a,b] These results support our working hypothesis that the position of the amide side chain in KNT-63 (**1a**) would play an important role for κ agonist activity and selectivity. The pharmacological effects of KNT-63 (**1a**) support our hypothesis that the C-ring in an active TRK-820 conformation may be in the boat form with an interaction between the 14-OH and the amide nitrogen.

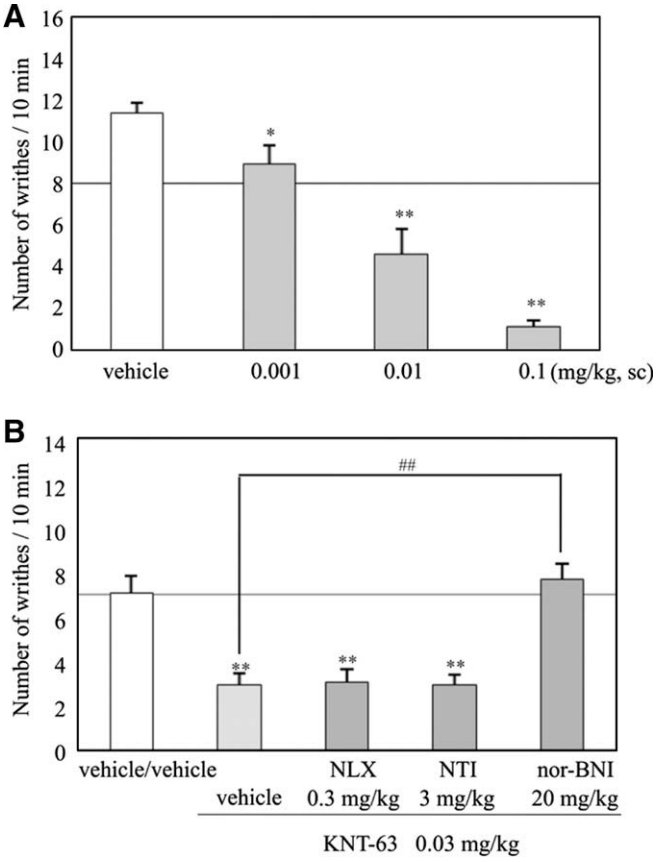


Figure 5. (A) Antinociceptive effect induced by KNT-63 (**1a**) in the acetic acid writhing (AAW) assay. *: p <0.05 versus vehicle, **: p <0.01 versus vehicle. (B) Effects of opioid receptor antagonists on the antinociception induced by KNT-63 (**1a**) in AAW test. **: p <0.01 versus vehicle/vehicle, ##: p <0.01 versus vehicle/KNT-63 (**1a**).

The location of the amide side chain in KNT-63 (**1a**), a bit upper than that of NS22, would lead to more κ selectivity and more potent analgesic activity for KNT-63 (**1a**). Detailed SARs of KNT-63 (**1a**) derivatives are now under investigation.

We also reported that putative ε agonist TAN-821 (Fig. 1) with the bicyclo[2.2.2]octene skeleton showed no κ selectivity.[5] The antinociceptive effect induced by TAN-821 was not antagonized by either nor-BNI, μ antagonist β-FNA, or NTI. Only β-endorphin [1–27], which is a prototypical putative ε antagonist, antagonized the analgesia produced by TAN-821, which means TAN-821 would be a putative ε agonist not a κ, μ, and δ agonist. Other TAN-821 derivatives having the bicyclo[2.2.2]octene skeleton also showed putative ε selectivity. The structures of KNT-63 (**1a**) and TAN-821 differ by the type of the atom at the 8 position, that is, O in KNT-63 (**1a**) and C in TAN-821. The 14-OH in TRK-820 also played an important role for κ selectivity.[3b,c] We are now investigating the effect of the atom (C, O, or N) at the 8 position on κ selectivity.

In conclusion, a novel 4,5-epoxymorphinan derivative with an oxabicyclo[2.2.2]octane skeleton, KNT-63 (**1a**) was designed and synthesized on the basis of an active TRK-820 conformation, the boat conformation, with interaction between the 14-OH and the amide nitrogen. The designed KNT-63 (**1a**) showed stronger analgesia than that of the previously reported κ agonist NS22 whose amide side chain was positioned a bit lower than that of KNT-63 (**1a**). The antinociceptive effect of KNT-63 (**1a**) (ED_{50} = 0.0063 mg/kg) was almost as potent as that of TRK-820.

Acknowledgments

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14. The C-ring in 4,5-epoxymorphinan derivatives would be usually in chair-like form. Therefore, the 6β-substituent may be in quasi-equatorial position to extend horizontally. In the active conformation of TRK-820, the C-ring would be in boat-like form to lead the 6β-substituent to be in quasi-axial position. The X-ray analysis of some 4,5-epoxymorphinan derivatives was reported: (a) de Costa, B. R.; Iadarola, M. J.; Rothman, R. B.; Berman, K. F.; George, C.; Newman, A. H.; Mahboubi, A.; Jacobson, A. E.; Rice, K. C. *J. Med. Chem.* **1992**, *35*, 2826; (b) Bolognesi, M.; Ojala, W. H.; Gleason, W. B.; Griffin, J. F.; Farouz-Grant, F.; Larson, D. L.; Takemori, A. E.; Portoghese, P. S. *J. Med. Chem.* **1996**, *39*, 1816; (c) Nagase, H.; Watanabe, A.; Harada, M.; Nakajima, M.; Hasebe, K.; Mochizuki, H.; Yoza, K.; Fujii, H. *Org. Lett.* **2009**, *11*, 539.
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