Bioorganic & Medicinal Chemistry xxx (2014) xxx–xxx

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry

journal homepage: www.elsevier.com/locate/bmc

Combination of cyclohexane and piperazine based κ-opioid receptor agonists: Synthesis and pharmacological evaluation of *trans,trans*-configured perhydroquinoxalines

Christian Bourgeois, Elena Werfel, Dirk Schepmann, Bernhard Wünsch *

Institut für Pharmazeutische und Medizinische Chemie der Universität Münster, Corrensstraße 48, D-48149 Münster, Germany

ARTICLE INFO

Article history:
Received 6 March 2014
Revised 23 April 2014
Accepted 26 April 2014
Available online xxxx

Keywords:
κ Agonists
Perhydroquinoxalines
Desymmetrization
Nitroaldol
Conformational restriction
Pseudochiral center
Structure–affinity relationships
Selectivity

ABSTRACT

Desymmetrization of the pseudochiral (2*r*)-configured cyclohexane-1,2,3-triamines **8** with dimethyl oxalate led to racemic aminoquinoxaline-2,3-diones **9**. Selective introduction of the κ pharmacophoric structural elements pyrrolidine and 3,4-dichlorophenylacetamide with a two-carbon distance afforded conformationally restricted κ agonists **13**–**15** based on the quinoxaline ring system. In competitive radioligand receptor binding studies the benzylamine **13b**, the secondary amine **14b**, and the carbamate **15** displayed high κ receptor affinity. The K_i value of the lead compound derived methoxycarbonyl derivative **15** is 9.7 nM. However, the κ affinity of **15** is exceeded by **13b** and **14b** with a basic functional group instead of the methoxycarbonyl group in 1-position of the quinoxaline system. The chlorine atoms of the dichlorophenylacetyl residue are essential, since the corresponding phenylacetyl analogs show considerably reduced κ affinity. The potent κ ligands **13b**, **14b** and **15** are selective over the related μ- and δ-opioid receptors, σ_1, σ_2 and NMDA receptors. In the [^{35}S]GTPγS-binding assay **13b** behaved as partial agonist with lower activity than U-69,593.

1. Introduction

κ receptors belonging to the class of opioid receptors are widely distributed throughout the central nervous system and the periphery. Activation of centrally localized κ opioid receptors leads to strong analgesia.[1] Compared to clinically used μ receptor agonists (e.g. morphine, fentanyl, pethidine, levomethadone), κ receptor agonists show a quite different side effect profile. In particular dangerous μ receptor mediated side effects like respiratory depression and addiction are not caused by activation of κ opioid receptors. However κ receptor agonists are not devoid of side effects: dysphoria, sedation and strong diuresis are the most common side effects associated with activation of κ receptors.[2,3] Since κ receptors are also found in the periphery, κ agonists which are restricted to the periphery can be used for the treatment of visceral pain and inflammatory and itching skin diseases.[4–6]

The known κ agonists can be classified into four compound classes: peptides including the physiological agonist dynorphin A,[2,7] morphinoids with the prototypical ligand ketocyclazocine giving name to this opioid receptor subtype,[2,7] the natural product salvinorin A without a basic functional group[8,9] and ethylenediamines (arylacetamides) with the first synthetic κ agonist U-50,488 (**1**)[10] (Fig. 1). The pharmacophore of the fourth class of κ agonists is represented by an ethylenediamine, whose terminal N-atoms are incorporated into a pyrrolidine ring and an arylacetamide moiety, respectively. In U-50,488 (**1**, K_i = 0.34 nM) this ethylenediamine structural element is part of a *trans*-configured cyclohexane ring. In the potent κ agonist **2** (K_i = 0.31 nM) the ethylenediamine pharmacophore is found as part of a piperazinylmethylamine system.[11]

Since we are interested in conformationally restricted κ agonists with novel scaffolds, it was planned to combine the structural features of the potent κ agonists **1** and **2** containing the ethylenediamine substructure (Fig. 1). A superposition of the ethylenediamine substructures of **1** and **2** is only possible by annulation of the cyclohexane and piperazine rings to afford the perhydroquinoxalines **3**, which also result from addition of an ethylene moiety between the cyclohexane ring and the *N*-methyl moiety of **1** or between the piperazine ring and the methyl group of **2** (see arrows at compounds **1** and **2**). Whereas the configuration of the centers of chirality in 8- and 8a-position are defined by the lead compounds **1** and **2**, the configuration of the third center of chirality (C-4a) is not defined by the lead compounds **1** and **2**.

In this communication we report on the synthesis of perhydroquinoxalines **3** with the relative *trans,trans*-configuration of the three N-functionalities attached to the cyclohexane ring (Fig. 1). The additional N-4-atom of the perhydroquinoxaline ring, which

* Corresponding author. Tel.: +49 251 8333311; fax: +49 251 8332144.
E-mail address: wuensch@uni-muenster.de (B. Wünsch).

http://dx.doi.org/10.1016/j.bmc.2014.04.054
0968-0896/

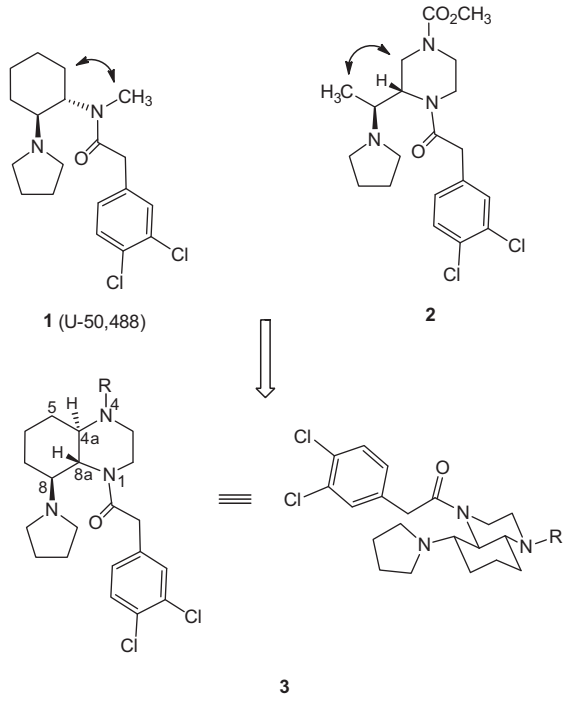

Figure 1. Development of the novel κ agonists **3** by combining the cyclohexane and piperazine rings of the lead compounds **1** and **2** to give a perhydroquinoxaline ring system.

is located outside of the κ pharmacophore, will allow fine tuning of the pharmacodynamic and more importantly the pharmacokinetic properties including the passage of the blood brain barrier of the new compounds by introducing various substituents.

2. Synthesis

According to literature,[12,13] the one-pot reaction of glutaraldehyde (**4**), nitromethane (**5**) and an excess of benzylamine led to the nitrocyclohexanediamine **7a** in 60% yield (Scheme 1). However, due to the formation of several side products and subsequent purification problems the stepwise synthesis was preferred. Thus, a double *Henry* reaction (nitroaldol reaction) of nitromethane (**5**) with glutaraldehyde (**4**) in the presence of NaOH[13,14] afforded the nitrocyclohexanediol **6** in 80% yield. Subsequent treatment of **6** with benzylamine in an aqueous solution provided the nitrodiamine **7a**.[13] Reduction of the amount of benzylamine and increasing the amount of H_2O resulted in an optimized yield of 92%, exceeding the previously reported yield of 75%.[13] The nitrodiamines **7b**, **7d**, and **7e** were obtained under the same conditions in 86–93% yields. Due to purification problems the *p*-chlorobenzylamine **7c** could only be isolated in 49% yield.

The exchange of the OH moieties of the nitrodiol **6** with amino groups is explained either by a base induced β-elimination of water followed by conjugated addition of amine or by a retro-nitroaldol reaction followed by imine formation and subsequent addition of the nitroalkane to the intermediate imine. The formation of the nitrodiol **6** and its transformation into nitrodiamines **7** occurred with high diastereoselectivity giving only the thermodynamically most stable *trans*,*trans*-configured diastereomers with all substituents in the favorable equatorial orientation. The *trans*,*trans*-configuration of **6** and **7** is confirmed by the triplet for the proton adjacent to the nitro moiety (CH-NO_2) with large coupling constants of 10.3–10.6 Hz indicating the axial orientation of three adjacent protons.

Although the reduction of the nitrodiamine **7a** to form the triamine **8a** has been reported in literature using H_2 and Raney Ni,[12,13] this reaction step required special attention and careful selection of the reaction conditions. Reproducible yields of **8a** could only be achieved by a certain combination of reaction conditions (H_2 pressure, temperature) and amount and quality of Raney Ni. Alternative reduction methods (Zn/HCl, Sn/HCl, SmI_2, $LiAlH_4$) did not improve the reproducibility and yields. Finally, the triamines **8a–c** and **8e** were prepared by reduction of the nitrodiamines **7a–c** and **7e** with H_2 (1 bar) in the presence of Raney Ni as catalyst.

For the establishment of the quinoxaline ring two amino moieties of the triamines **8** had to be connected by a C_2-building block. For this purpose the benzyl derivative **8a** was reacted with oxalyl chloride at −78 °C. According to tlc several products were formed, but the desired quinoxalinedione **9a** could not be isolated. Therefore the triamines **8a–c** were treated with the less reactive dimethyl oxalate in boiling methanol for 48 h (Scheme 1). After complete conversion the quinoxalinediones **9a–c** were isolated by recrystallization in yields of 33–64%. The corresponding 3,4-dichlorobenzyl derivative **9d** was not obtained due to side reactions during the reduction of the nitro moiety of nitrodiamine **7d**.

The nitrodiol **6**, the nitrodiamines **7** and the triamines **8** represent achiral compounds with a pseudochiral center in 2-position having (2*r*)-configuration. Treatment of the triamines **8** with dimethyl oxalate led to desymmetrization since both enantiotopic benzylamino moieties reacted with the same probability with dimethyl oxalate. However the original *trans*,*trans*-configuration of the triamines **8** is retained in the quinoxalinediones **9**. Although the quinoxalinediones **9** contain three centers of chirality, this synthetic strategy led to only one out of four possible diastereomers. In this manuscript only one enantiomer of the racemic mixtures is shown in the Schemes.

The construction of the quinoxalinedione **9a** led to a differentiation of the benzylamino moieties; one benzylamine is part of an amine, whereas the other one is part of an amide allowing their chemoselective conversion. Treatment of **9a** with ammonium formate in the presence of Pd/C as catalyst[15] cleaved chemoselectively the benzyl moiety of the benzylamine affording the primary amine **10** in 97% yield (Scheme 2). Reaction of the primary amine **10** with 1,4-diiodobutane provided the pyrrolidine **11** in 76% yield. Several reducing agents were tried for the reduction of the quinoxalinedione **11**. A 3:1 mixture of $LiAlH_4$ and $AlCl_3$ forming AlH_3 in situ[16] turned out to give the highest yield (96%) of the quinoxaline **12**. The reduced quinoxaline **12** contains a secondary (N-4) and a tertiary amine (N-1) within the heterocycle. Acylation of the secondary amino moiety (N-4) with phenylacetyl chloride and 3,4-dichlorophenylacetyl chloride led to the phenylacetamides **13a** and **13b**, respectively.

In order to introduce the methoxycarbonyl moiety of the lead compound **2** the remaining benzyl moiety should be removed by conventional hydrogenolysis. However, treatment of the benzylamines **13** with H_2 and Pd/C in a THF/H_2O mixture led to incomplete transformation and provided considerable amounts (approx. 30%) of dechlorinated products from **13b**. Therefore concd HCl (10%) was added to the solvent, which should increase the hydrogenolysis rate. After limitation of the reaction time to 30 min, the secondary amines **14a** and **14b** were isolated in 90% and 94% yield, respectively. The amount of dechlorinated products found after hydrogenolysis of **13b** was less than 1%. Finally, acylation of the secondary amine **14b** with methyl chloroformate afforded the carbamate **15** in 59% yield.

3. Pharmacological evaluation

The κ receptor affinity of the quinoxalines **9–15** was determined in competition receptor binding studies. Guinea pig brain