Pergamon

SCIENCE DIRECT®

Tetrahedron Letters 44 (2003) 1487–1489

TETRAHEDRON LETTERS

Solid-phase synthesis of novel hybrid opiates

Ming-Lei Wang and Bahman E. Nassim*

Department of Chemistry, Indiana University Southeast, 4201 Grant Line Road, New Albany, IN 47150, USA

Received 7 November 2002; revised 11 December 2002; accepted 12 December 2002

Abstract—An efficient solid-phase synthesis of some novel hybrid opiates, consisting of part peptide and part alkaloid, connected together through a hydrazone linkage is reported. NMR studies show the *E* to *Z* isomerization of the hydrazone bond.

Opioid analgesics exert their actions through three types of opioid receptors: μ, κ, and δ. Research in the area of opioid receptors has focused on the development of selective ligands, aimed to clarify their anatomical distributions and functions.[1] Over the past two decades, a large number of endogenous peptides such as enkephalins[2] have been characterized and classified as 'opioid peptides'.[3] Most of these opioid peptides contain an N-terminal tetrapeptide fragment (Tyr-Gly-Gly-Phe). Biological studies of these peptides have suggested that the N-terminal tetrapeptide is the key segment for their analgesic activity. It has been proposed that the N-terminal tetrapeptide sequence carries the 'message,' which triggers the opioid effects. The C-terminal segments of these endogenous peptides, which vary in sequence, seem to play an 'address' role, which is responsible for the receptors selectivity.[4] While endogenous peptides are potentially useful as therapeutic agents, their use in clinical studies is limited both by their tendency to undergo enzymatic cleavage and by their general inability to cross the blood–brain barrier (BBB). To overcome their lack of resistance to enzymatic cleavage, many opioid peptide derivatives containing unnatural amino acids have been synthesized, showing interesting properties. To assist in crossing the BBB, a number of options, including development of chimeric peptides, have been explored.[5] Some of these delivery methods seem promising, though their actual employment may be rather complicated. Our work constitutes the synthesis of hybrid opiates which contain the N-terminal tetrapeptide (Tyr-Gly-Gly-Phe) as the message moiety and an alkaloid opiate as the address portion. The latter part is expected to deliver and dock the peptide at the appropriate opiate sites, allowing the peptide message to be expressed. This idea is consistent with other biologically active synthetic opiates that seem to follow the message–address concept,[6] and it offers the opportunity to modify either part towards more desired properties. Alkaloid opiates are both easily transportable through BBB and stable under physiological conditions. Therefore, it is anticipated that the peptide attachment, in the hybrid structures, can be carried along while being sheltered from enzymatic attacks.

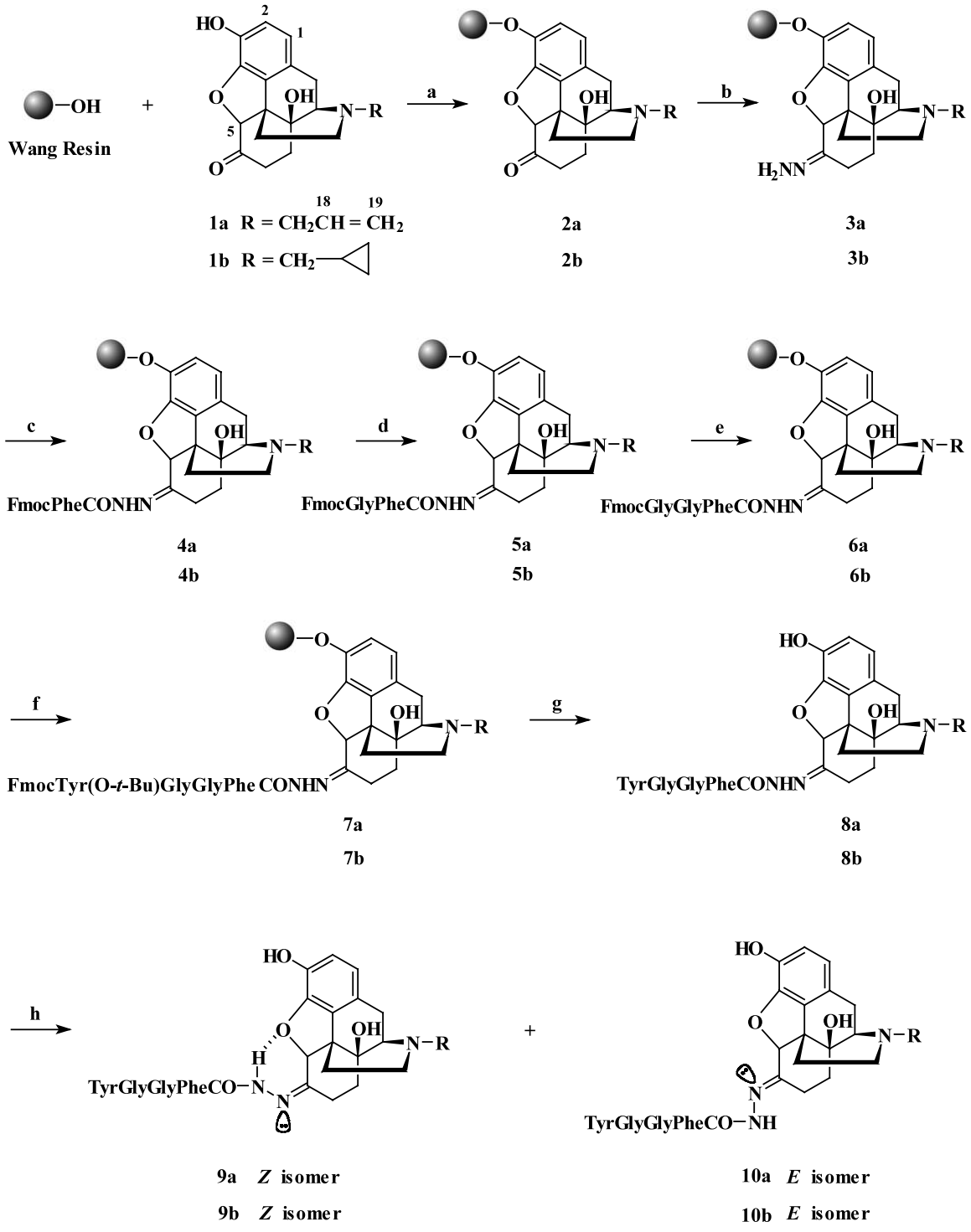
Synthesis on solid support, originally developed in peptide chemistry, is now widely accepted as a powerful tool for rapidly synthesizing a variety of organic molecules without tedious purification process. This paper presents the synthesis of some novel hybrid opiates on the solid support.

As illustrated in Scheme 1, our synthesis started with immobilization of the opiate alkaloids (Naloxone or Naltrexone) onto Wang resin via Mitsunobu reaction.[7,8] The 6-ketone functional group was converted to hydrazone by treating with excess anhydrous hydrazine in dimethyl formamide (DMF) at room temperature.[9] This reaction was monitored by IR for the disappearance of the 6-ketone stretch at 1720 and 1715 cm^{-1} corresponding to naloxone and naltrexone respectively. Subsequent coupling of various Fmoc-protected amino acids onto the resin-bound hydrazone **3** proceeded smoothly in the presence of diisopropylcarbodiimide (DIC) and 1-hydroxybenzotriazole (HOBt) in DMF.[10,11] The Fmoc-protecting group was removed by the standard piperidine treatment. Cleavage of the Wang resin with concentrated trifluoroacetic acid (TFA) in the presence of scavengers (generally Reagent K[12]) gave the hybrid opiates **8a** and **8b**. Preparative thin

Keywords: hybrid opiates; solid-phase synthesis; isomerization.

* Corresponding author. Tel.: +1-(812)-941-2305; fax: +1-(812)-941-2637; e-mail: bnassim@ius.edu

0040-4039/03/$ - see front matter
PII: S0040-4039(02)02854-X



Scheme 1. *Reagents and conditions*: (a) Ph_3P/DIAD/THF/48 h; (b) H_2NNH_2/DMF; (c) Fmoc-Phe/HOBt/DIC/DMF; (d) 20% piperidine/DMF, Fmoc-Gly/HOBt/DIC/DMF; (e) 20% piperidine/DMF, Fmoc-Gly/HOBt/DIC/DMF; (f) 20% piperidine/DMF, Fmoc-Tyr (*O*-*t*-Bu)/HOBt/DIC/DMF; (g) 20% piperidine/DMF, Reagent K; (h) preparative TLC, CH_2Cl_2/MeOH (10:1).

layer chromatography (PTLC) of **8a** and **8b** led to the separation of the corresponding *Z* and *E* isomers of the hybrid opiates. The isomers were identified by their proton NMR spectra. The hydrazone proton of the *Z* isomer absorbed at lower field strength, relative to the hydrazone proton of the *E* isomer. Molecular modeling, using Dreiding models, showed an approximate distance of 1.4 Å between the *Z* hydrazone proton and the ether oxygen of the alkaloid, indicating a possible intramolecular hydrogen bond.[13] Variable temperature NMR studies supported this conclusion (Table 1). Furthermore, the *Z* isomer is a stable product but the *E* isomer is unstable and undergoes spontaneous isomerization to the *Z* configuration at room temperature in solution, as observed from ^{1}H NMR.

Table 1. ^{1}H NMR data of hydrazone proton of the *syn* isomers **9a** and **9b** under different temperatures (300 MHz, $CDCl_3$)

Temperature (°C)	δ **9a** (ppm)	δ **9b** (ppm)
20	10.61	10.87
0	10.68	10.95
−20	10.89	10.99
−40	11.06	11.11
−60	11.10	11.15

In a typical procedure to prepare hybrid opiates **9a** and **10a**, diisopropyl azodicarboxylate (DIAD, 3.0 equiv.) was added dropwise to a mixture of Naloxone (3.0 equiv.), triphenylphosphine (3.0 equiv.) and Wang resin (1.0 equiv., 1.0 mmol/g) in 10 ml dry THF, and the mixture was shaken at room temperature for 48 h. The reaction solvent was removed by filtration and the resin was washed with DMF, MeOH and CH_2Cl_2. The resin was collected and dried in vacuo to afford **2a**. To a suspension of resin **2a** in DMF was added anhydrous hydrazine (4 ml). The mixture was shaken at room temperature for 2 h. The solvent was drained, and the solid was washed with DMF, MeOH and CH_2Cl_2 to give the resin-bound hydrazone. The dried resin-bound material was suspended in DMF with Fmoc-L-phenylalanine (2.5 equiv.), HOBt (2.5 equiv.) and DIC (2.5 equiv.) to react for 2 h. After filtration, the solid was washed as described above. Kaiser ninhydrin test was used on the beads of the resin to determine the progress of the coupling. Following a coupling reaction the Fmoc group was removed by treatment with 20% piperidine in DMF for 15 min. This deprotection was repeated twice. Fmoc-glycine and Fmoc-L-tyrosine (*O*-*t*-butyl) were attached using the same conditions. After completion of the peptide assembly and removal of the Fmoc group from the N-terminal residue, the resin was washed successively with DMF, MeOH, CH_2Cl_2 and reacted at room temperature with Reagent K[12] (82.5% TFA, 5% H_2O, 5% phenol, 5% thioanisole, and 2.5% ethanedithiol) under nitrogen for 2 h to give hybrid opiate **8a**. Preparative TLC of **8a** in CH_2Cl_2/MeOH (10/1) gave the *Z* and *E* isomers of hybrid opiates **9a** and **10a**. Compound **9a**: ^{1}H NMR (300 MHz, DMSO): δ 10.61 (1H, s, hydrazone hydrogen), 7.15 (1H, d, $J=8.1$ Hz, H-1), 6.80 (1H, d, $J=8.1$ Hz, H-2), 5.98 (1H, m, H-18), 5.66 (2H, m, H-19), 5.06 (1H, s, H-5). HRFABMS m/z 766.3549 $[MH]^+$ (calcd for $C_{41}H_{48}N_7O_8$, 766.3553). Compound **10a**: ^{1}H NMR (300 MHz, DMSO): δ 10.33 (1H, s, hydrazone hydrogen), 7.14 (1H, d, $J=8.4$ Hz, H-1), 6.79 (1H, d, $J=8.4$ Hz, H-2), 5.99 (1H, m, H-18), 5.64 (2H, m, H-19), 5.03 (1H, s, H-5). HRFABMS m/z 766.3589 $[MH]^+$ (calcd for $C_{41}H_{48}N_7O_8$, 766.3553). Compound **9b**: ^{1}H NMR (300 MHz, DMSO): δ 10.87 (1H, s, hydrazone hydrogen), 6.89 (1H, d, $J=8.4$ Hz, H-1), 6.50 (1H, d, $J=8.4$ Hz, H-2), 5.21 (1H, s, H-5). HRFABMS m/z 780.3713 $[MH]^+$ (calcd for $C_{42}H_{50}N_7O_8$, 780.3709). Compound **10b**: ^{1}H NMR (300 MHz, DMSO): δ 10.28 (1H, s, hydrazone hydrogen), 6.76 (1H, d, $J=8.7$ Hz, H-1), 6.38 (1H, d, $J=8.7$ Hz, H-2), 4.68 (1H, s, H-5). HRFABMS m/z 780.3688 $[MH]^+$ (calcd for $C_{42}H_{50}N_7O_8$, 780.3709).

In summary, an efficient and straightforward solid-phase synthesis of hybrid opiates has been developed. This protocol is amenable to the synthesis of large combinatorial libraries. Synthesis of other hybrid opiates and the results of the biological test will be reported in due course.

Acknowledgements

This work was supported by Indiana University Office of Research & Graduate School, IU Southeast Office of Research, and the James Y. McCullough Memorial Endowment.

References

1. Dhawan, B. N.; Cesselin, F.; Raghubir, R.; Reisine, T.; Bradley, P. B.; Portoghese, P. S.; Hamon, M. *Pharmacol. Rev.* **1996**, *48*, 567–592.
2. Hughes, J.; Smith, T. W.; Kosterlitz, H. W.; Fothergill, L. A.; Morgan, B. A.; Morris, H. R. *Nature (London)* **1975**, *258*, 577–579.
3. Morley, J. C. *Br. Med. Bull.* **1983**, *39*, 5–10.
4. Chavkin, C.; Goldstein, A. *Proc. Natl. Acad. Sci. USA* **1981**, *78*, 6543–6547.
5. Bickel, U.; Pardridge, W. M. In *Vector Mediated Delivery of Opioid Peptides to the Brain*; Membranes and Barriers: Targeted Drug Delivery. Rapaka, R. S., Ed.; NIDA Research Monogaph 154, 1995; pp. 28–46.
6. Metzger, T. G.; Paterlini, M. G.; Portoghese, P. S.; Ferguson, D. M. *Neurochem. Res.* **1996**, *21*, 1287–1294.
7. Richter, L. S.; Gadek, T. R. *Tetrahedron Lett.* **1994**, *35*, 4705–4706.
8. Zuzka, V. K.; Flegelova, Z.; Weichsel, A. S.; Lebl, M. *Tetrahedron Lett.* **1995**, *36*, 6193–6196.
9. Wang, S. S. *J. Am. Chem. Soc.* **1973**, *243*, 1328–1333.
10. Choi, H.; Aldrich, J. V. *Int. J. Peptide Protein Res.* **1993**, *42*, 58–63.
11. Synder, K. R.; Story, S. C.; Heidt, M. E.; Murray, T. F.; DeLander, G. E.; Aldrich, J. V. *J. Med. Chem.* **1992**, *35*, 4330–4333.
12. King, D. S.; Fields, C. G.; Fields, G. B. *Int. J. Peptide Protein Res.* **1990**, *36*, 255–266.
13. Palla, G.; Predieri, G.; Domiano, P. *Tetrahedron* **1986**, *42*, 3649–3654.