Biomimetic synthesis of Nitraramine

Wanner, M.J.; Koomen, G.J.

DOI
10.1021/jo00122a052

Publication date
1995

Published in
Journal of Organic Chemistry

Citation for published version (APA):

General rights
It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations
If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: https://uba.uva.nl/en/contact, or a letter to: Library of the University of Amsterdam, Secretariat, Singel 425, 1012 WP Amsterdam, The Netherlands. You will be contacted as soon as possible.
Biomimetic Synthesis of Nitraramine

Martin J. Wanner and Gerrit-Jan Koomen

Amsterdam Institute of Molecular Studies, Laboratory of Organic Chemistry, University of Amsterdam,
Nieuwe Achtergracht 129, 1018 WS Amsterdam, The Netherlands

Received March 7, 1995

The synthesis of a possible biosynthetic precursor (9) of nitraramine (1) is described, utilizing N-Boc-piperidone as an equivalent of didehydropiperidine 4. Heating this reactive, achiral intermediate 9 in aqueous solution results in the stereoselective formation of natural nitraramine via three successive cyclization reactions. Since nitraramine and several other Nitraria alkaloids are obtained from Nitraria species in racemic form, this synthesis provides additional support for our hypothesis of non-enzyme-catalyzed formation of these alkaloids.

Alkaloids containing a 3-spiropiperidine structural unit are exclusively obtained from plants of the Nitraria genus. The Nitraria spiroalkaloids that contain 10 carbon atoms such as nitraramine (2) and isonitramine (3) are most likely biogenetically derived from two piperidine synthons.\(^\text{1,2}\) Nitraramine (1) was isolated in 1975 from Nitraria schoberi\(^\text{2}\) and the unusual structure of this considerably more complicated alkaloid was established by X-ray analysis in 1985.\(^\text{4}\) Nitraramine is a 15 carbon-atom-containing spiroalkaloid and is probably derived from three piperidine equivalents. Three boat-formed rings in the center of the molecule form an oxaza bicyclooctane ring system, which is surrounded by three chair-formed six-membered rings, thus creating a completely rigid structure.

![Diagram of Nitraramine]

Remarkable is the racemic form in which nitraramine and several other Nitraria alkaloids occur in nature. In a recent publication\(^\text{5}\) Heathcock and co-workers synthesized petrosin, a racemic bisquinolizidone sponge alkaloid containing eight stereocenters. They nicely demonstrated that postbiosynthetic equilibriations are responsible for the racemic form in which petrosin is isolated from its natural source. Nitraramine (1), however, is a configurationally stable molecule\(^\text{6}\) and racemization via retroreactions is unlikely. Racemization experiments with enantiomerically pure material should give additional information in this respect; however, separation of nitraramine into enantiomers was unsuccessful until now.\(^\text{7}\) After some earlier synthetic approaches\(^\text{8}\) we describe here a synthesis that is based on the retrosynthetic analysis we recently proposed\(^\text{1}\) and we will show that three successive cyclization reactions smoothly take place, once the reactive precursor anticipated is obtained. The ease with which this process occurs chemically (water, reflux) combined with the racemic form of natural nitraramine give an indication that no enzymes are involved in the stereochemically important spirocyclization reaction (Scheme 4). Another example of imine/enamine based cyclization reactions of achiral precursors in aqueous solution was recently provided by our biomimetic synthesis\(^\text{9}\) of the indole alkaloids nitrarine and nitramidine, also obtained as racemates form from Nitraria plants.

**Biogenetic Hypothesis.** Distinction should be made between the biosynthetic origin of these Nitraria alkaloids and for instance histrionicotoxine, which contains a 2-spiropiperidine ring system. A clear relationship exists between the Lupine alkaloids and those of the Nitraria family\(^\text{1}\) (Scheme 1). Labeling studies\(^\text{6}\) in Lupine species have established dipiperidine 5 (tetrahydroabasine) as the biogenetic precursor for piperidine alkaloids such as lupinine and sparteine. Although lysine, via cadaverine, is shown to be the precursor of 5, the existence of the unstable didehydropiperidine 4\(^\text{10}\) as an intermediate has not been confirmed. An alternative ring opening of the saturated piperidine in 5 leads to 6, providing aldehyde 7 after oxidative deamination. An indication for this ring opening reaction is found in aqueous solutions of synthetic 5\(^\text{11}\) (dihydrobromide), showing the presence of about 30% of the open form 6 according to \(^1\)H-NMR spectroscopy. Condensation of 7 with the enamine form of didehydropiperidine 4 gives precursor 8, which is probably in equilibrium with the


\(^{\text{7}}\) We attempted resolution of nitraramine by crystallization with 1 and 2 equivs of the following reagents: (S)-(+) mandelic acid; (1R)-(-)-10-camphorsulfonic acid; (R)-(+)-1,1'-binaphthalene-2,2'-diyl hydrogen phosphate.


\(^{\text{10}}\) Didehydropiperidine 4 can be prepared in situ from its crystalline, symmetrical trimer of piperidine: Schöpf, C.; Konzak, A.; Braun, F.; Jacobi, E.; Chem. Ber. 1951, 84, 699.

Biomimetic Synthesis of Nitraramine

**Scheme 1. Biogenetic Hypothesis**

Lysine →

\[
\begin{align*}
\text{didehydropiperidine} & \quad 4 \\
\text{tetrahydroanabasine} & \quad 5 \\
\text{Nitraramine} & \quad 9
\end{align*}
\]

dimerization

**Scheme 2**

\[
\begin{align*}
\text{1. LDA} \\
\text{2. MsCl} \\
\text{3. TEA, } & \Delta \\
\text{70%}
\end{align*}
\]

\[
\begin{align*}
\text{1. LiEt}_3\text{BH} \\
\text{2. TFA, DCM} \\
\text{hydrolyzed with PPTS as catalyst, and the aldehyde 14 was alkylated with a second equivalent of the lithium enolate of 12 to give 15, representing the carbon framework that is required for the cyclization reactions. Water elimination as described for 13 produced the symmetrical dimer 19 (Scheme 3), the lactam analogue of 9. Adjusting the oxidation state of both piperidone carbonyls in 15 turned out to be problematic. Our initial goal, protection of both hydroxyls and the double bond in the form of pyran 20 (Scheme 3) was not possible since base-catalyzed conjugate addition of the alcohol function in}
\end{align*}
\[
\begin{align*}
\text{Scheme 3}
\end{align*}
\]

\[
\begin{align*}
\text{1. MsCl} \\
\text{2. TEA, } & \Delta \\
\text{78%}
\end{align*}
\]

**Chemistry.** Initially our biomimetic synthesis was performed by condensation reactions of glutaric aldehyde 10b with in situ-generated didehydropiperidine 4 in aqueous solution. Although these reactants have the correct oxidation state to obtain nitraramine, the capricious reactivity of glutaric aldehyde caused several undesired side reactions. Protection of one of the aldehyde functionalities in glutaric aldehyde gave acetal 10a, which was submitted to condensation reactions with didehydropiperidine 4 according to a literature procedure, describing the synthesis of 3-alkylidene substituted piperidines from aromatic and aliphatic aldehydes. None of the anticipated ene-imine product 11a could be obtained, however, due to limited stability of the imine functionality. Next we switched to N-Boc-piperidinone 12, a stable piperidine equivalent that can be converted efficiently into 4 via a reduction/protection sequence. The lithium enolate of N-Boc-piperidinone 12 was alkylated with aldehyde 10b and without isolation, the resulting mixture of diastereomeric alcohols was converted into \( \alpha,\beta \)-unsaturated lactam 13 (\( E:Z = 15:1 \)) via mesylation and triethylamine-catalyzed elimination. This elimination effectively "protects" the OH during the next alkylation step. The acetal functionality of 13 was hydrolyzed with PPTS as catalyst, and the aldehyde 14 was alkylated with a second equivalent of the lithium enolate of 12 to give 15, representing the carbon framework that is required for the cyclization reactions. Water elimination as described for 13 produced the symmetrical dimer 19 (Scheme 3), the lactam analogue of 9. Adjusting the oxidation state of both piperidone carbonyls in 15 turned out to be problematic. Our initial goal, protection of both hydroxyls and the double bond in the form of pyran 20 (Scheme 3) was not possible since base-catalyzed conjugate addition of the alcohol function in
Among the more selective reducing agents lithium triethylborohydride is superior for reductions of N-Boc-piperidone. Workup the unsaturated hydroxypiperidine ring followed by 1,3-reduction of the unsaturated lactam moiety. In our situation both lactam carbonyls of the proposed precursors, which were reduced, leading to the unstable precursor 16. It should be noted that after aqueous workup the unsaturated hydroxy-piperidine ring preferred the conjugated aldehyde form. Reduction of the unsaturated lactam moiety followed by 1,2-reduction to form 18 lowered the yield. Acid-catalyzed removal of the Boc-substituents in 16 was performed with TFA in dichloromethane, leading to symmetrical dimer probably via elimination of water from 8 as is shown in Scheme 1.

Cyclization reactions with 9 (Scheme 4) were performed at reflux temperature in buffered aqueous solutions at pH 7, leading directly to nitramine as the only isomer. Spirocyclization of 8 completes the carbon framework, and via cyclohexane ring-inversion of 21 the required 1,3-dialixal conformation for the two final acetal-forming steps is obtained. Although stereoisomerism is possible, especially at C1, none of these compounds was observed in H NMR spectra of crude reaction mixtures. It should be noted that 1-epi-nitraramine was recently isolated from Nitraria billardieri by Quirion et al.

Identification of nitramine was possible by comparison of H and C NMR spectra with data from the literature. Extensive 2D NMR-spectroscopy was performed on both nitramine and its mono-hydrochloride, resulting in a partial revision of the literature chemical shift assignments.

In summary, it was demonstrated that water at neutral pH is an effective catalyst for biomimetic cyclization reactions with the imines and enamines that might be present in Nitraria species. Synthesis of the appropriate precursors, which in vivo would require several redox-enzymes, was accomplished with N-Boc-piperidone as a dihydro-piperidine equivalent.

### Experimental Section

**General Information.** NMR spectra were obtained from a 400 MHz Bruker spectrometer. Thin layer chromatography (TLC) was performed on silica gel-coated plastic sheets. Chromatography refers to flash chromatography on silica gel (0.030–0.075 mm). When ammonia-containing eluents were used, the silica gel was pretreated with this eluent.

3-(2-Piperidinyl)-2,3-dihydro-piperidine Dihydropyridazine (5) and 1-Amino-5,5-diethoxypentane (6) were prepared as described by Schöpf et al. H NMR (DMSO-d6 + 2 drops of D2O) 5 δ 4.8 (m), 3.46 (m, 4H), 3.1 (m, 2H), 2.15–1.4 (m, 10H); 6: δ 8.58 (s, 1H), 7.0 (m, J = 7.4 Hz, 1H), 1.62 (t, J = 5.3 Hz, 2H), 2.49 (J = 6.2 Hz, 2H), 2.39 (J = 7.1 Hz, 2H), 1.9–1.3 (m, 4H).

5.5-Diethoxy-pentanal (10a). A solution of glutaric aldehyde (25% in water, 150 mL, 0.385 mol) in EtOH (1.5 L, 99%) was stirred with Dowex 50WX8 (H+ form, 2.0 g) at 20 °C during 4 days. The catalyst was removed by filtration, and the resulting solution was stirred with solid NaHCO3 during 30 min, filtered, and evaporated in vacuo. The residue was coevaporated with ethyl acetate (2 × 100 mL) and with PE 60/80 (100 mL), dissolved in PE 60/80 (200 mL), and separated by chromatography on silica (Φ 10 cm, PE 60/80/ETOAc 3/1; TLC-spots were made visible with anisaldehyde/sulfuric acid). A mixture of diacets (open and cyclic) was eluted first, followed by monocet 10a, which was obtained in pure form by distillation (8.9 g, 51.11 mol, 13%): Bp 42–45 °C/0.6 mbar; H NMR (CDCl3) δ 8.76 (t, J = 1.4 Hz, 1H), 4.48 (t, J = 5.2 Hz, 1H), 3.7–3.4 (m, 4H), 2.47 (dt, J = 1.4 Hz, J = 7.0 Hz, 2H), 1.74–1.61 (m, 4H), 1.19 (t, J = 7.0 Hz, 6H); 13C NMR (CDCl3) A 202.28, 102.51, 61.15, 43.52, 32.91, 17.34, 15.26; IR (CHCl3) 1720 cm⁻¹.

1,1-Diethoxy-5-[N-(tert-butyloxy-carbonyl)-2-oxo-piperidin-3-yldene]pentane (13). N-(tert-Butyloxy-carbonyl)-2-piperidione (12) 7.96 g, 40 mmol, crystallized from hexanes at −20 °C was added to a solution of LDA (44 mmol) in THF (120 mL) at −78 °C. The reaction mixture was allowed to warm to −20 °C, stirred at this temperature during 1 h, and cooled to −78 °C. Aldehyde 10a (6.96 g, 40 mmol) was added dropwise, and the mixture was stirred at this temperature for...
1 h and quenched with saturated NH₄Cl solution. Extractive workup (ether) yielded the crude alcohol, which was isolated in a mixture of dry toluene (50 mL) and triethylamine (8.3 mL, 60 mmol). Methanesulfonyl chloride (3.5 mL, 45 mmol) was added dropwise at 0 °C, and the mixture was allowed to warm to room temperature. Additional triethylamine (13.8 mL, 100 mmol) was added and the mixture was refluxed during 6 h. Aqueous workup and chromatographic purification (PE 60/80/EtOAc 3/1) gave first impure Z-isomer (0.6 g, 4%) and next 13 E-isomer (8.75 g, 62%) both as a syrup. Z-isomer: 1H NMR (CDCl₃) δ 5.82 (t, J = 7.1 Hz), 3.90 (m, 2H). E-isomer: 1H NMR (CDCl₃) δ 6.89 (m, J = 7.5 Hz), 4.41 (t, J = 5.4 Hz, 1H), 3.64–3.39 (m, 6H), 2.40 (m, 2H), 2.10 (q, J = 6.4 Hz, 2H), 1.80 (m, J = 5.4 Hz, 2H), 1.56 (m, 2H), 1.48 (s, 9H), 1.14 (t, J = 7.0 Hz, 3H, 11C NMR (CDCl₃)) δ 167.1, 153.2, 142.2, 142.1, 130.1, 102.6, 82.65, 61.02, 45.77, 33.31, 28.06, 27.79, 24.30, 23.60, 22.22, 15.27; IR (CHCl₃) 3300 (br), 1757, 1680–1720, 1620 cm⁻¹; HRMS obs mass 378.2184 (M⁻Na⁺), calcd for C₁₃H₂₃NO₅Na 378.2383; obs mass 535.2400 (MH⁺), calcd for C₁₃H₂₃NO₅Na 535.2503.

5-N[(tert-Butyloxy)carbonyl]-2-oxopiperidin-3-ylidenepentan-14 (14). Acetal 13 (3.55 g, 10 mmol) was stirred with PPTS (0.12 g) in a mixture of water (5 mL) and acetonitrile (150 mL) at 20 °C during 4 h. The reaction mixture was concentrated to half PE 60/80 (0.5mL) and diluted with CHCl₃ (100 mL) to remove the catalyst. Extraction and drying yielded 14 (2.6 g, 100%), which was pure enough for further transformations. 1H NMR (CDCl₃) δ 9.72 (t, J = 1.5 Hz, 1H), 6.86 (m, J = 5.5 Hz, 1H), 3.64 (m, 2H), 2.43 (m, 4H), 2.13 (q, J = 7.5 Hz, 2H), 1.88–1.71 (m, 4H), 1.49 (s, 9H); 13C NMR (CDCl₃) δ 201.8, 165.1, 153.1, 140.8, 130.9, 82.79, 45.77, 28.04, 27.46, 27.08, 24.41, 22.29; HRMS (FAB) obs mass 378.2184 (M⁻Na⁺), calcd for C₁₃H₂₃NO₅Na 378.2383; obs mass 535.2400 (MH⁺), calcd for C₁₃H₂₃NO₅Na 535.2503.

Acidic Hydrolysis of 5-Benzyl-2,2-dimethyl-1,5-bis[(tert-butyloxy)carbonyl]-3-oxopiperidin-3-ylidenepentan-14 and Cyclization to Nitraramine. Trichloroacetic acid (0.5 mL) was added in one portion to an ice-cold solution of 16 (0.11 g, 0.22 mmol) in CHCl₃ (1 mL). After 15 min at 0 °C the mixture was stirred at room temperature for 30 min and evaporated in vacuo (bath T < 25 °C). The oily residue (7.9 TFA salt) was dissolved in CDCl₃ by the addition of CD₃COCD₃ (2 drops); 1H NMR (CDCl₃) δ 6.43 (bs, 2H), 6.87 (bs, 2H), 3.85 (m, 4H), 2.52 (m, 4H), 1.98 (m, 4H), 1.74 (m, 2H); 13C NMR (CDCl₃) δ 162.2, 129.5, 49.2, 43.10, 39.53, 25.53, 20.68, 18.78. This bis-iminium salt was not stable enough for further analysis and was evaporated directly after the acid-treatment and dissolved in a pH 7 phosphate buffer (20 mL). The resulting solution was refluxed under nitrogen during 20 h, made alkaline with excess solid sodium carbonate, and extracted three times with ethyl acetate. The organic layer was dried (Na₂SO₄), and removal of the solvents gave a crude alkaloid mixture (15–20 mg) containing ca. 50% nitraramine. Chromatography (CH₃Cl/ methanol/NH₄OH 90/10/1) gave nitraramine 1 (17–22%) as a glass. An analytical pure sample was prepared from the easily crystallizing monohydrochloride. A solution of nitraramine in ethanol was treated with conc HCl. The solvents and the excess of HCl were removed by coevaporation with methanol, and the dihydrochloride was dissolved in ethanol and treated with a small excess of triethylamine. Evaporation and crystallization from CH₃Cl/ethyl acetate produced triethylamine hydrochloride, which was removed by filtration. The filtrate was evaporated to dryness and nitraramine hydrochloride crystallized from a small amount of ethanol by precipitation with ethyl acetate. The free base was obtained in pure form by treatment of nitraramine hydrochloride with aqueous K₂CO₃ followed by ether extraction.