The Asymmetric Total Synthesis of (+)- and (-)-Trypargine via Noyori Asymmetric Transfer Hydrogenation

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A concise and efficient total synthesis of (+)- and (-)-trypargine (6 steps and 38% overall yield), a 1-substituted \( \beta \)-carboline guanidine alkaloid isolated from the skin of the African frog \( K. \) senegalensis, was developed based on the construction of the \( \beta \)-carboline moiety via Bischler-Napieralski reaction and the enantioselective reduction of the dihydro-\( \beta \)-carboline intermediate via an asymmetric transfer hydrogenation reaction using Noyori’s protocol.

Keywords: trypargine, tetrahydro \( \beta \)-carboline alkaloid, Bischler-Napieralski reaction, Noyori asymmetric transfer hydrogenation

Introduction

The \( \beta \)-carboline skeleton is widely distributed among natural products including yohimbine (yohimbine and alloyohimbine), corynantheidine (corynantheidine), \textit{Rauwolfia} (reserpine) and \textit{Vinca} (vincamine) alkaloids.\(^1\)

(-)-Trypargine (1) is a \( \beta \)-carboline alkaloid which has been isolated from the skin of the African racophorid frog, \textit{Kassina senegalensis}, by Akizawa et al.\(^2\) in 1982 and more recently has also been isolated in nearly racemic form from the methanol extract of the ground ascidian \textit{Eudistoma sp}.\(^3\)

The absolute configuration of natural (-)-trypargine (1) was initially assigned via resolution of its racemate, prepared via the Pictet-Spengler reaction between tryptamine and 4-guanidino butyraldehyde ethylenecetal, followed by optical rotatory dispersion and circular dichroism studies.\(^4\) Ishikawa and co-workers\(^4,5\) established its absolute configuration by total synthesis via the Pictet-Spengler reaction between \((R)\)-N-benzyl tryptophan methyl ester and 2-ketoglutaric acid, followed by X-ray diffraction analysis of the corresponding methyl ester of the major diastereoisomer. Synthetic (-)-(S)-1-(3’-guanidinopropyl)-1,2,3,4-tetrahydro-\( \beta \)-carboline was shown to be identical to natural (-)-trypargine (1) thus establishing its absolute configuration.

The presence of a \( \beta \)-carboline and a guanidine moieties in the structure of (-)-trypargine (1) has led to preliminary evaluation of its biological profile. Neither a full account of the biological properties nor its biosynthetic origin have been reported although one may envision trypargine (1) to be formed from tryptophan and the 2-keto carboxylic acid derived from arginine. It has been reported to be toxic to mice (LD\(_{50}\) = 16.9 mg kg\(^{-1}\), intravenous administration) although complete details of this study are lacking.\(^5,6\) Ireland and co-workers\(^3\) have failed to observe significant cytotoxicity in fractions containing trypargine alkaloids against human colon tumor cells. Additionally, (-)-trypargine (1) has been reported to block voltage gated sodium channels in squid axon membrane.\(^7\)

Our interest to further explore the biological profile of trypargine and analogues led us to consider the available methodologies for chirality transfer in order to develop a short and asymmetric route to these compounds which would secure enough quantities to carry out biological assessment. Our approach to a catalytic and asymmetric synthesis of (+)-trypargine (1) relied on our previous results on the enantioselective total syntheses of arborescidines when we successfully employed the Bischler-Napieralski protocol to assembly the 3,4-dihydro-\( \beta \)-carboline moiety, followed by the Noyori catalytic asymmetric hydrogen transfer reaction to reduce the intermediate prochiral...
Recently, Drabowicz and co-workers disclosed the total synthesis of (+)-trypargine (1) via the latter approach which prompted us to disclose our results in this topic.

**Results and Discussion**

Our approach to (R)-trypargine (1) was based on the construction of the dihydro-β-carboline moiety via the Bischler-Napieralski reaction of tryptamine derivative 4 (Scheme 1). Among the routes evaluated to secure the preparation of intermediate 4, two emerged as the most efficient: introduction of the 3-aminopropionyl group via Curtius rearrangement on 5-ketopentanoyl tryptamine (3, Scheme 1, step b) or via N-acylation of tryptamine (2) with N-allyloxy carbonyl-4-aminobutyric acid (6, Scheme 1, step d).

The thermolysis of acyl azides to the corresponding isocyanates is known as the thermal Curtius rearrangement and the reaction mechanism probably involves the concerted migration of the alkyl or aryl substituent in the carbonyl group to the nitrogen atom of the azido group with concomitant loss of dinitrogen. The isocyanate intermediate can be isolated when the reaction is carried out in non-nucleophilic solvent or can be intercepted by water, amines or alcohols to give the corresponding amines, ureas or carbamates.

Starting from commercially available tryptamine (2), reaction with glutaric anhydride in acetone at room temperature provided the corresponding amide 3 in quantitative yield (Scheme 2). The Curtius rearrangement was carried out by treating a solution of amide 3 in acetone with methyl chloroformate in order to give rise to the corresponding mixed anhydride at 0 °C which was then converted to the corresponding acyl azide 7 upon treatment with aqueous sodium azide at rt. Acyl azide 7 was heated in toluene for 2 h and the reaction was quenched by the addition of allylic alcohol to provide, after chromatography on silica gel, allyl carbamate 4 in 73% overall yield from tryptamine (2).

Alternatively, allyl carbamate 4 was prepared in 88% yield via acylation of tryptamine (2) with N-carboxyalkoxy-4-aminocarbonyl acid 6 and catalytic amount of N-hydroxybenzotriazol (HOBt) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDC). Despite the slight increase in the overall yield of allyl carbamate 4, the use of expensive reagents in the method employing 4-amino butyric acid makes the Curtius rearrangement the method of choice for large scale preparation.

With the preparation of allyl carbamate 4 secured, the formation of dihydro-β-carboline 9 was successfully carried out upon treating 4 with 3 equivalents of POCl₃ in toluene/nitromethane (7:3, v/v) under reflux, to give 9 in 76% yield after column chromatography. However, it proved to be more efficient to filter the crude reaction mixture through a pad of silica gel in order to remove most of the contaminants and proceed to the next step, the asymmetric transfer hydrogenation under the conditions described by Noyori and coworkers. Using the protocol, (R)-(−)-10 was obtained in 75% overall yield from allyl carbamate 4 (Scheme 3).
The Bischler-Napieralski reaction is a cyclodehydration which takes place when an acyl derivative of a β-arylethylamine is treated with a dehydrating agent. Mechanistic studies carried out by Fodor and coworkers have shown that the formation of imidoyl chloride and nitrilium intermediates is promoted by several dehydrating agents such as POCl₃, PCl₅ and SOCl₂. In our case, the formation of the imidoyl chloride or nitrilium salt from the indole amine would be followed by an aromatic electrophilic substitution to afford dihydro-β-carboline (Scheme 4).

The asymmetric reduction of the prochiral imine 9 was efficiently carried out using the asymmetric transfer hydrogenation as devised by Noyori and co-workers. The reaction is catalyzed by chiral N-sulfonated diamine-Ru(II)-η⁶ arene complexes and has become the method of choice for the enantioselective reduction of cyclic imines due to the high yield and enantiomeric excess usually attained and the simplicity of the experimental protocol. Despite the advantages of this methodology, relatively few examples of asymmetric synthesis of natural products containing the tetrahydro-β-carboline core have appeared since our first disclosure of the application of Noyori asymmetric transfer hydrogenation in the total synthesis of arborescindines A, B and C.

The 16 electron catalytic active species II was generated upon treatment of pre-catalyst RuCl(S,S)-H_NCPPhCHPhNTs(η-p-cymene) (I) in DMF at 80 °C in the presence of Et₃N. Then, a solution of prochiral imine 9 in DMF was added, followed by a 5:2 formic acid-triethylamine azeotropic mixture and the reaction mixture was kept at room temperature for 8 h. Under these conditions tetrahydro-β-carboline 10 was isolated in 75% overall yield from allyl carbamate 4. However, when longer reaction time was employed the hydrogenation of the allyloxy group present in 10 was also observed.

The proposed mechanism involves the six-membered arrangement IV which displays an hydrogen bond between the N-H in the chiral ligand and the nitrogen in the imine and hydride transfer from the ruthenium hydride species to the imine via an out-of-plane interaction between Ru-H and the C=N bonds. According to the working model proposed by Noyori and co-workers, the use of (S,S)-DPEN as the chiral ligand would direct the asymmetric transfer hydrogenation to take place at the Si face of prochiral imine 9 leading to (R)-10 as the major enantiomer as depicted in Scheme 5.

Considering that the sense of chirality in the Noyori reduction would be validated by comparing the specific
optical rotation of synthetic trypargine (1) with that described for trypargine isolated from natural sources, we focused on the determination of the enantiomeric ratio of tetrahydro-β-carboline 11 which was shown to be 98:2 after 19F-NMR spectroscopic analyses of the Mosher amide prepared from (R)-(-)-α-methoxy-α-(trifluoromethyl) phenyl acetic acid (MTPA).

Our choice for the allyloxycarbonyl group in our synthetic scheme evolved after screening several different protecting groups (carbomethoxy, tert-butoxycarbonyl, benzyloxycarbonyl, 9-fluorenylmethoxycarbonyl and trifluorocarbonyl groups) as it proved to be compatible with the acidic conditions during the Bischler-Napieralski reaction using POCl3, the Noyori protocol for the transfer hydrogenation reaction and eventually it was easily removed under very mild conditions. In fact, by employing the conditions described by Mandal and McMurry, amine (R)-11 was isolated in 90% yield after 30 min at rt in the presence of Et3SiH and catalytic amount of Pd/C in ethanol.

The final step toward (R)-trypargine (1) was the introduction of the guanidine moiety which was carried out with the method described by Bernatowicz and co-workers4,9 which makes use of 1H-pyrazol-1-carboxyamidine hydrochloride (12) in DMF and diisopropylethylamine. After column chromatography on basic alumina and treatment with methanolic HCl, (R)-trypargine hydrochloride (1•HCl) was isolated in 77% yield. Its physical (melting point and specific optical rotation) and spectroscopic data (1H- and 13C-NMR data) nicely matched those described in the literature.

Natural (S)-(-)-trypargine (1) was also prepared in 6 steps and 38% overall yield from tryptamine according to the same reaction sequence.

The approach described herein is robust enough to provide trypargine and its derivatives in both enantiomeric forms in few steps, good overall yield and very high enantiomeric ratios from tryptamine and analogues and should be of value for the determination of the absolute configuration of others dihydro-β-carbolines such as the novel toxin isolated from the venom of the *Parawixia bistriata* spider endemic in the brazilian cerrado27 and pharmacological screening of related compounds.

**Experimental**

Commercially available reagents and solvents were previously purified. THF was distilled from calcium hydride and redistilled from sodium/benzophenone immediately prior to use. Dichloromethane, acetonitrile and triethylamine were distilled from calcium hydride immediately prior to use. Dimethylformamide (DMF) was distilled from calcium hydride under reduced pressure and
temperature lower than 70 °C. Methanol was distilled from magnesium containing catalytic amount of iodine. POCI₃ was distilled immediately prior to use.

The reactions were monitored with TLC plates (aluminum foils covered with silica gel) and exposed to UV radiation, followed by treatment with fosfomimolibdic acid (25% ethanolic solution) or aq. KMnO₄ and heating on a hot plate. Chromatographic separations were carried out in silica gel (70-230 or 230-400 Mesh) or basic alumina (5-200 μ).

Specific optical rotations were measured at 25 °C at 589 nm (sodium D line) and IR spectra were measured as film in NaCl cells and frequencies are reported as cm⁻¹. Melting points are not corrected.

¹H- and ¹³C-NMR spectra were obtained in CDCl₃ or CD₂OD solutions at 250 and 62.5 MHz, respectively, or at 300 and 75 MHz, respectively. Chemical shifts (δ) were expressed in ppm and coupling constants in Hz. Multiplicity is reported as singlet (s), doublet (d), double dublets (dd), triplet (t), double triplet (dt), quartet (q), quintuplet (quint), multiplet (m) and broad singlet (br s).

5-{[2-(1H-IIndol-3-yl)ethyl]amino}-5-oxopentanoic acid (3)

To a solution of glutaric anhydride (0.10 g, 0.88 mmol) in acetonitrile (3 mL), was added tryptamine (0.15 g, 0.93 mmol) dissolved in acetone (0.5 mL). The reaction mixture was stirred 2 h at rt and then a solution of sodium azide (0.35 mL, 4.5 mmol). The reaction mixture was stirred for 45 min at rt and then a solution of sodium azide (0.35 mL, 4.5 mmol) in water (1 mL) was added. The reaction mixture was stirred for 2 h at rt and then concentrated under reduced pressure (Caution! Do not concentrate to dryness due to the risk of explosion). The residue was dissolved in toluene (20 mL) and the solution was refluxed for 2 h under nitrogen atmosphere when allylic alcohol (0.38 mL, 5.6 mmol) was added and the reaction was stirred for 3 h. The reaction mixture was purified by column chromatography on silica gel and ethyl acetate-hexanes (1:1, v/v) as eluent to yield carbamate 4 a pale brown solid in 73% yield (0.24 g, 0.70 mmol). mp 84-86 °C. IR (KBr) νmax/cm⁻¹: 3305, 1701, 1643, 1535, 1257.

4-{[(Allyloxy)carbonyl]amino}butanoic acid (6)

To a suspension of 4-aminobutyric acid (0.46 g, 4.5 mmol) in THF (10 mL) containing sat. aq. NaHCO₃ (10 mL) at 0 °C, was added dropwise allyl chloroformate (0.5 mL, 4.7 mmol). The reaction mixture was stirred for 24 h at rt when conc. HCl was added dropwise to adjust to pH 2. The reaction mixture was extracted with CH₂Cl₂ (3 × 10 mL) and the organic phase was dried under anhydrous magnesium sulfate. The carbamate 6 was obtained in quantitative yield as viscous oil which was used in the next step without further purification. IR (KBr) νmax/cm⁻¹: 3332, 2974, 1709, 1538, 1255. ¹H-NMR (250 MHz, CDCl₃) δ 1.85 (quint, J 7.0 Hz, 2H); 2.38 (t, J 7.2 Hz, 2H); 3.15-3.39 (m, 5H).
5.0 mmol) in THF (10 mL). The reaction mixture was stirred 16 h at rt, quenched with brine (25 mL) and extracted with ethyl acetate (3 × 25 mL). The organic phase was dried over magnesium sulfate and evaporated under reduced pressure. After purification over a pad of silica gel with ethyl acetate as eluent, carbamate 4 was obtained in 88% yield (1.25 g, 3.78 mmol).

**Allyl [3-(4,9-dihydro-3H-carbolin-1-yl)propyl]carbamate (9)**

A solution of 4 (1.18 g, 3.56 mmol) in a mixture (7:3 v/v) of toluene/acetonitrile (30 mL) was added POCl₃ (1 mL, 10 mmol) dropwise and the reaction mixture was refluxed for 5 h. The reaction mixture was cooled to rt and concentrated under vacuum. The crude mixture was dissolved in CH₂Cl₂ (40 mL) and washed with aq. 10% NH₄OH (25 mL) and brine (2 × 25 mL), successively. The organic phase was dried over anhydrous magnesium sulfate and concentrated under reduced pressure. After purification over a pad of silica gel with ethyl acetate/methanol (10%, v/v) as eluent, carbamate 9 was obtained in quantitative yield (1.11 g, 3.56 mmol). ¹H-NMR (250 MHz, CDCl₃) δ 1.98 (quint, J 7.0 Hz, 2H); 2.74 (t, J 7.4 Hz, 2H); 2.85 (t, J 8.6 Hz, 2H); 3.26 (q, J 6.4 Hz, 2H); 3.84 (t, J 8.6 Hz, 1H); 4.53 (d, J 5.5 Hz, 2H); 5.16 (dd, J 1.4 and 10.4 Hz, 1H); 5.25 (dd, J 1.4 and 17.2 Hz, 1H); 5.28 (dd, J 1.4 and 10.4 Hz, 1H); 5.92 (m, 1H); 7.00-7.14 (m, 2H); 7.23-7.29 (m, 1H); 7.40 (d, J 8.2 Hz, 1H); 7.57 (d, J 8.2 Hz, 1H). ¹³C-NMR (62.5 MHz, CDCl₃) δ 19.3 (CH₃); 27.0 (CH₃); 32.3 (CH₂); 40.7 (CH); 47.8 (CH₂); 65.6 (CH); 112.2 (CH); 117.0 (C²); 117.5 (C²⁺); 119.9 (CH); 120.1 (CH); 124.5 (CH); 125.3 (C₁); 128.5 (C₁); 132.7 (CH); 137.0 (C₁); 156.9 (C₀); 161.3 (C₉). HRMS (ESI+) Found 312.1722 [M+H]+; calc. for C₁₉H₂₅N₂O₂+: 312.1707 (+3.2 ppm error).

**Allyl [3-[(1R)-2,3,4,9-tetrahydro-1H-β-carbolin-1-yl]propyl]carbamate (10)**

To a previously dried round-bottomed flask was added RuCl₃(η-p-cymene) (5.4 mg, 8.8 µmol), (S,S)-TsDPEN (6.4 mg, 17.5 µmol), triethylamine (1.3 µL, 17.6 µmol) and dry DMF (2 mL) was heated 1 h at 80 °C. The mixture was cooled to rt and a solution of 9 (0.59 g, 1.9 mmol) in DMF (2 mL) was added followed by the addition of an azeotropic mixture of formic acid and triethylamine (1.0 mL, 5.2 v/v). The reaction mixture was kept 8 h at rt and then DMF was distilled under reduced pressure and the crude product was purified by column chromatography on silica gel (20% CHCl₃/MeOH, v/v) to afford (0.45 g, 1.42 mmol) in 75% yield as a viscous yellow oil. [α]D²⁰ +27.0 (c 1.0, MeOH). ¹H-NMR (250 MHz, CD₃OD) δ 1.14-1.34 (m, 2H); 1.69-2.01 (m, 3H); 2.13-2.23 (m, 1H); 2.85-3.02 (m, 2H); 3.20-3.25 (m, 2H); 4.51-4.53 (m, 2H); 5.16 (dd, J 1.4 and 10.4 Hz, 1H); 5.28 (dd, J 1.4 and 17.2 Hz, 1H); 5.92 (m, 1H); 7.00-7.14 (m, 2H); 7.34 (d, J 8.0 Hz, 1H); 7.44 (d, J 8.0 Hz, 1H). ¹³C-NMR (62.5 MHz, CD₃OD) δ 20.5 (CH₂); 27.0 (CH₂); 31.2 (CH₂); 41.4 (CH₂); 43.2 (CH₂); 54.4 (CH); 66.5 (CH₂); 107.0 (C₁); 112.4 (CH); 117.6 (=CH₂); 119.0 (CH); 120.4 (CH); 123.2 (CH); 127.8 (C₁); 131.9 (CH); 134.6 (C₁); 138.2 (C₁); 159.0 (C₀). HRMS (ESI+) Found 314.1835 [M+H]+; calc. for C₁₈H₂₅N₂O₂+: 314.1869 (-10.8 ppm error).

**Allyl [3-[(1S)-2,3,4,9-tetrahydro-1H-β-carbolin-1-yl]propyl]carbamate (11)**

The reaction was carried out as described above for (R)-10, except that the catalyst was prepared using (S)-TsDPEN instead of (R)-TsDPEN. [α]D²⁰ –28.0 (c 1.0, MeOH).

{3-[(1R)-2,3,4,9-tetrahydro-1H-β-carbolin-1-yl]propyl}amine (11)

To a previously dried round-bottomed flask containing (R)-10 (0.30 g, 0.95 mmol), 5% Pd/C (0.03 g) and methanol (15 mL) at rt was added dropwise triethylsilane (0.75 mL, 4.7 mmol). The reaction mixture was stirred 30 min at rt and filtered to remove the catalyst. The solvent was removed under reduced pressure and the crude product was purified on basic alumina (ethyl acetate and 20% ethyl acetate/methanol as eluents) to afford pale brown oil (0.20 g, 0.85 mmol) in 90% yield. [α]D²⁰ +75.0 (c 1.0, MeOH), IR (KBr) ν/cm⁻¹: 3453 and 3245. ¹H-NMR (250 MHz, CD₃OD) δ 1.60-1.76 (m, 2H); 1.96-2.07 (m, 2H); 2.65-2.78 (m, 4H); 2.90-2.98 (m, 2H); 4.01-4.05 (m, 1H); 6.95-7.10 (m, 2H); 7.29-7.41 (m, 2H). ¹³C-NMR (62.5 MHz, CD₃OD) δ 22.4 (CH₂); 25.6 (CH₂); 32.9 (CH₂); 40.9 (CH₂); 43.2 (CH₂); 53.5 (CH); 109.2 (C₁); 112.7 (CH); 118.8 (CH); 120.0 (CH); 122.5 (CH); 128.5 (C₀); 135.0 (C₁); 138.0 (C₀).

{3-[(1S)-2,3,4,9-tetrahydro-1H-β-carbolin-1-yl]propyl}amine (11)

The same procedure as described above was employed to afford (S)-11 in 87% yield [α]D²⁰ -74.0 (c 1.0, MeOH).

(+)-(R)-Trypargine Hydrochloride (1a)

To a solution of (R)-11 (0.110 g, 0.5 mmol) in DMF (1 mL) was added pyrazole 12 (0.075 g, 0.5 mmol) and disopropylethylamine (0.25 mL, 1.4 mmol). The reaction
mixture was stirred 24 h at rt when DMF was removed under reduced pressure. The crude product was purified by column chromatography on basic alumina (chloroform/methanol 30%, v/v) to afford (+)-(R)-trypargine (0.104 g, 0.38 mmol) in 77% yield as a yellow solid which was treated with methanolic HCl to obtain (+)-(R)-trypargine hydrochloride as a yellow solid. mp 213-214 °C; [α]D 20 +32 (c 1.0, MeOH); [lit.9]: 210-213 °C; [α]D 20 +34.3 (c 1.0, MeOH)). IR (KBr) νmax/cm⁻¹: 3350, 2934, 1660. ¹H-RM N (250 MHz, CD3OD) δ 1.87-1.96 (m, 2H); 2.01-2.10 (m, 3H); 3.67-3.71 (m, 1H); 4.70-4.78 (m, 1H); 7.04 (t, J 7.2 Hz, 1H); 7.14 (t, J 7.2 Hz, 1H); 7.38 (d, J 8.0 Hz, 1H); 7.47 (d, J 8.0 Hz, 1H). ¹³C-NMR (62.5 MHz, CD3OD) δ 19.4 (CH3); 25.8 (CH3); 30.5 (CH3); 42.0 (CH3); 43.0 (CH3); 54.4 (CH); 107.4 (Cg); 112.4 (CH); 119.1 (CH); 120.6 (CH); 123.5 (CH2); 127.3 (C6); 129.7 (C6); 138.3 (C6); 158.6 (Cg); HRMS (ESI+) Found 272.1884 [M+H]+; calc. for C15H22N5+: 272.1875 (+3.3 ppm error).

(-)-(S)-Trypargine Hydrochloride (1b)

The same procedure as above was employed starting from (S)-3-(2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indol-1-yl)propan-1-amine (1b) to afford (-)-(S)-trypargine hydrochloride (1) in 78% yield, [α]D 20 +34 (1.0, MeOH).

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Supplementary Information

Supplementary data are available free of charge at jbcs.sbq.org.br, as pdf file.

References


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