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Synthesis of 6- or 7-substituted 1,2,3,4-tetrahydroisoquinoline-3-carboxylic acids

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Summary

A straightforward approach for the synthesis of several new, aryl-substituted derivatives of the conformationally constrained phenylalanine analogue 1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid (Tic) is described. Tic, nitro-substituted at the 6 or 7 position, was prepared by base-catalyzed cyclization of diethyl acetamidomalonate with α,α-dibromo-4-nitro-*o*-xylene followed by decarboxylation and deacylation under refluxing conditions in aqueous HCl. Catalytic hydrogenation of nitro-Tic in the presence of 10% Pd/C afforded amino-Tic, which was then converted to iodo-Tic by a modified Sandmeyer reaction. Both amino-Tic and iodo-Tic can easily be transformed to other substituents.

Introduction

The use of conformational restrictions for the development of topographically well-defined peptide analogues with improved potency and/or selectivity is an established approach in peptide-based drug design. Toward this end, 1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid (Tic) has seen increased use as a conformationally constrained phenylalanine analogue in the synthesis of peptide analogues [1–5] with well-defined side-chain orientations. Although the tetrahydroisoquinoline ring system structurally provides several positions for further modification of Tic in order to probe features of the ligand binding site, relatively few Tic derivatives have been reported, largely due to limitations in the standard approach for the synthesis of Tic. We describe here an alternate approach more suitable for the synthesis of a wide variety of modified Tic analogues.

Tic is generally synthesized by a Pictet-Spengler reaction [6] using phenylalanine with formaldehyde in the presence of concentrated hydrochloric acid under refluxing conditions. However, this method is not widely suitable for ring-substituted phenylalanine analogues since the Pictet-Spengler reaction is usually limited to analogues in which the ring-substituents increase electron density at the point of ring closure to facilitate cyclization [7]. For example, 3-hydroxy-phenylalanine [8], 3-methoxy-phenylalanine [9], 3,4-dihydroxy-phenylalanine [10,11], 3,4-dimethoxy-phenylalanine [11,12], and 3,4-methanediyldioxy-phenylalanine [13] have been cyclized successfully to their corresponding Tic analogues directly through this reaction or via a modification to this process. However, tyrosine failed to yield 7-hydroxy-Tic due to a polymerization reaction to a phenol-formaldehyde polymer. Conversely, 3′,5′-diiodo-L-tyrosine or 3′,5′-dibromo-L-tyrosine, in which the positions *ortho* to the phenolic OH are blocked by halogen atoms, were successfully cyclized by a modified procedure [14]. Phenylalanine derivatives lacking activating groups such as alkoxy or hydroxy functions have rarely been cyclized. One exception is 4-fluoro-phenylalanine, from which 7-fluoro-Tic was prepared [15]. However, the use of similar conditions for the attempted synthesis of 7-chloro-Tic was unsuccessful in our hands. Tic analogues with even more electron-withdrawing substituents, such as a nitro group, which could readily

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be converted to other moieties, are inaccessible via the Pictet-Spengler reaction. Consequently, we undertook the alternative pathway [16,17] depicted in Schemes 1 and 2 for the synthesis of Tic derivatives with several different possible substituents, each derived from a nitro group introduced at the initial stage of the reaction route, at the 6 or 7 positions.

Materials and Methods

Chemicals were purchased from Aldrich Chemical Company, Inc. Thin-layer chromatography (TLC) was performed on glass plates precoated with silica gel (Analtech). Products were visualized by UV (254 nm), and purified by flash chromatography (silica gel, 70–230 mesh, 60 Å, Aldrich). Melting points were determined on a Haake Buchler Melting Point Apparatus and were not corrected. ^{1}H NMR spectra were recorded on Bruker DPX300 and DRX500 spectrometers. Elemental analysis was carried out at the University of Michigan, Department of Chemistry. Analytical RP-HPLC was performed on a C18 Vydac column (0.46 × 25 cm) using the solvent system: 0.1% TFA (w/v) in water (solvent A)/0.1% TFA (w/v) in acetonitrile (solvent B), at a flow rate of 1 ml/min. The HPLC column eluates were monitored by their UV absorbance at 230 nm.

Syntheses

*Diethyl 2-acetyl-6-nitro-1,2,3,4-tetrahydroisoquinoline-3,3-dicarboxylate (**3a**) and diethyl 2-acetyl-7-nitro-1,2,3,4-tetrahydroisoquinoline-3,3-dicarboxylate (**3b**)*

To a solution of diethyl acetamidomalonate (1.44 g, 6.50 mmol) in dry acetone was added sodium hydride (60% dispersion in mineral oil, 0.51 g, 13 mmol), followed by α,α-dibromo-4-nitro-*o*-xylene (2.01 g, 6.50 mmol), which had been prepared from 4-nitro-*o*-xylene as described by Kleinschmidt and Braeuniger [18] (see Scheme 1). The flask was placed in a preheated oil bath and gently refluxed for 30 min (until the starting material, α,α-dibromo-4-nitro-*o*-xylene, was no longer visible by TLC). The reaction mixture was then cooled to room temperature, evaporated, and partitioned between H_2O and EtOAc. The organic layer was washed with brine, dried ($MgSO_4$), and concentrated. Analysis by TLC (hexane:EtOAc/1:1) showed two major components with R_f values of 0.11 and 0.24. These compounds were separated by flash column chromatography (silica gel). The more polar component of the flash chromatography separation was determined to be the 6-nitro product, **3a** (0.69 g, 29% yield). ^{1}H NMR ($CDCl_3$): δ (ppm) 1.19 (m, 6H, $CH_2C\underline{H_3}$), 2.31 (s, 3H, $COC\underline{H_3}$), 3.54 (s, 2H, $C\underline{H_2}C$), 4.18 (m, 4H, $C\underline{H_2}CH_3$), 4.79 (s, 2H, $C\underline{H_2}N$), 7.36 (d, J = 8.3 Hz, 1H, aromatic), 8.08 (s, 1H, aromatic), 8.13 (d, J = 8.3 Hz, 1H, aromatic); mp 126–127 °C (recrystallized from EtOAc/hexane). Analysis by HPLC (0–60% B/60 min) indicated a purity in excess of 95%, with r_t = 43.0 min. Elemental analysis: calculated for $C_{17}H_{20}N_2O_7$: C, 56.04; H, 5.53; N, 7.69; found: C, 56.11; H, 5.53; N, 7.69.

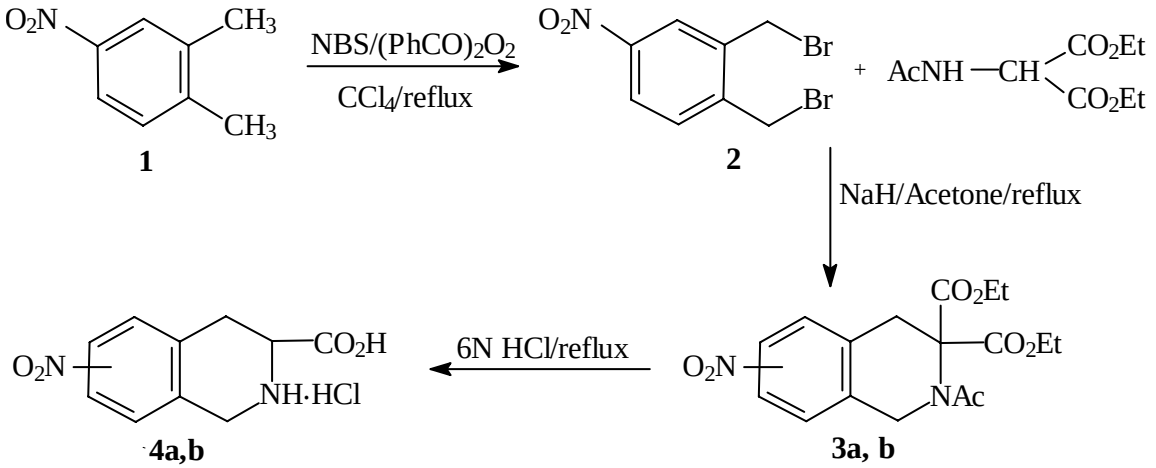
The less polar component proved to be the 7-nitro product, **3b** (0.95 g, 40% yield). ^{1}H NMR ($CDCl_3$): δ (ppm) 1.19 (m, 6H, $CH_2C\underline{H_3}$), 2.31 (s, 3H, $COC\underline{H_3}$), 3.54 (s, 2H, $C\underline{H_2}C$), 4.17 (m, 4H, $C\underline{H_2}CH_3$), 4.79 (s, 2H, $C\underline{H_2}N$), 7.38 (d, J = 8.2 Hz, 1H, aromatic), 8.10 (s, 1H, aromatic), 8.12 (d, J = 8.2 Hz, 1H, aromatic); mp 102–103 °C (recrystallized from EtOAc/hexane). Analysis by HPLC (0–60% B/60 min) indicated a purity in excess of 90% with r_t = 43.2 min. Elemental analysis: calculated for $C_{17}H_{20}N_2O_7$: C, 56.04; H, 5.53; N, 7.69; found: C, 56.13; H, 5.65; N, 7.80.

*6-Nitro-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid hydrochloride (**4a**)*

Compound **3a** (0.64 g, 1.74 mmol) was suspended in 10 ml 6 N HCl and held at reflux for 3–4 h. The resulting cloudy solution was cooled to room temperature, filtered, and dried to give **4a** as an off-white solid (0.33 g, 73%). Since ^{1}H NMR showed no other side products present, **4a** was used (see below) without further purification. ^{1}H NMR (D_2O): δ (ppm) 2.86 (dd, J = 10.4 Hz, J = 16.9 Hz, 1H, $C\underline{H_2}CH$), 3.10 (dd, J = 4.7 Hz, J = 16.9 Hz, 1H, $C\underline{H_2}CH$), 3.45 (dd, J = 4.7 Hz, J = 10.4 Hz, 1H, $CH_2C\underline{H}$), 4.02 (d, J = 17.1 Hz, 1H, $C\underline{H_2}N$), 4.11 (d, J = 17.1 Hz, 1H, $C\underline{H_2}N$), 7.24 (d, J = 8.5 Hz, 1H, aromatic), 7.94 (d, J = 8.5 Hz, 1H, aromatic), 8.01 (s, 1H, aromatic); mp >220 °C (decomp.); HPLC r_t = 16.2 min (0–60% B/60 min); ES-MS ($M+H^+$): 223.1. Elemental analysis: calculated for $C_{10}H_{11}N_2O_4Cl$: C, 46.44; H, 4.28; N, 10.83; found: C, 46.62; H, 4.42; N, 10.64.

*7-Nitro-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid hydrochloride (**4b**)*

7-Nitro-1, 2, 3, 4- tetrahydroisoquinoline -3-carboxylic acid hydrochloride (**4b**) was synthesized in 90% yield in a similar manner as described above for **4a**. ^{1}H NMR (D_2O): δ (ppm) 2.83 (dd, J = 10.4 Hz, J =



Scheme 1. Synthesis of 6-nitro or 7-nitro-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid hydrochloride.

17.4 Hz, 1H, $C\underline{H_2}CH$), 3.08 (dd, J = 4.6 Hz, J = 17.4 Hz, 1H, $C\underline{H_2}CH$), 3.43 (dd, J = 4.6 Hz, J = 10.4 Hz, 1H, $C\underline{H_2}C\underline{H}$), 3.88 (d, J = 16.2 Hz, 1H, $C\underline{H_2}N$), 4.07 (d, J = 16.2 Hz, 1H, $C\underline{H_2}N$), 7.29 (d, J = 8.4 Hz, 1H, aromatic), 7.92 (m, 2H, aromatic); mp >220 °C (decomp.); HPLC r_t = 16.5 min (0–60% B/60 min); ES-MS ($M+H^+$): 223.0. Elemental analysis: calculated for $C_{10}H_{11}N_2O_4Cl$: C, 46.44; H, 4.28; N, 10.83; found: C, 46.74; H, 4.47; N, 10.44.

*6-Amino-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid hydrochloride (**5a**)*

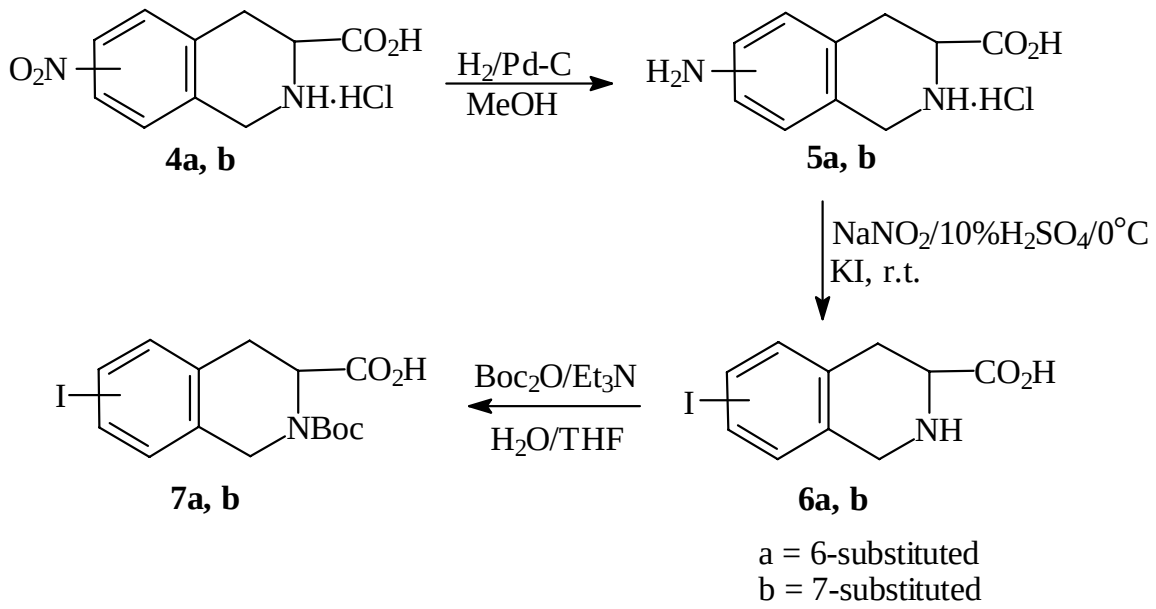
A hydrogenation bottle was charged with compound **4a** (0.26 g, 1.01 mmol) in 20 ml MeOH and to it was added 10% Pd/C (0.03 g). The suspension was hydrogenated at 30 psi for 1 h (until no further uptake of hydrogen was observed). The suspension was filtrated through Celite® and evaporated to give **5a** as a light yellow solid (0.22 g, 96%). This product was sufficiently pure, as indicated by ^{1}H NMR, to be used directly in the next step. ^{1}H NMR (D_2O): δ (ppm) 3.20 (dd, J = 10.9 Hz, J = 17.4 Hz, 1H, $C\underline{H_2}CH$), 3.43 (dd, J = 5.1 Hz, J = 17.4 Hz, 1H, $C\underline{H_2}CH$), 4.05 (dd, J = 5.1 Hz, J = 10.9 Hz, 1H, $CH_2C\underline{H}$), 4.36 (d, J = 15.9 Hz, 1H, $C\underline{H_2}N$), 4.53 (d, J = 15.9 Hz, 1H, $C\underline{H_2}N$), 7.27 (m, 2H, aromatic), 7.40 (d, J = 8.0 Hz, 1H, aromatic); mp >230 ° C (decomp.); HPLC r_t = 3.0 min (0–20% B/20 min); ES-MS ($M+H^+$): 193.1.

*7-Amino-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid hydrochloride (**5b**)*

7-Amino-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid hydrochloride (**5b**) was synthesized in 98% yield using the procedure described above for **5a**. ^{1}H NMR (D_2O): δ (ppm) 3.16 (dd, J = 10.8 Hz, J = 17.4 Hz, 1H, $C\underline{H_2}CH$), 3.42 (dd, J = 5.3 Hz, J = 17.4 Hz, 1H, $C\underline{H_2}CH$), 4.09 (dd, J = 5.3 Hz, J = 10.8 Hz, 1H, $C\underline{H_2}C\underline{H}$), 4.39 (d, J = 16.0 Hz, 1H, $C\underline{H_2}N$), 4.51 (d, J = 16.0 Hz, 1H, $C\underline{H_2}N$), 7.26 (s, 1H, aromatic), 7.31 (d, J = 8.2 Hz, 1H, aromatic), 7.42 (d, J = 8.24 Hz, 1H, aromatic); mp >210 °C (decomp.); HPLC r_t = 3.0 min (0–20% B/20 min); ES-MS ($M+H^+$): 193.1.

*6-Iodo-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid (**6a**)*

To a solution of **5a** (0.22 g, 0.96 mmol) in 3 ml 10% H_2SO_4 at 0 °C was added, dropwise, a precooled aqueous solution of $NaNO_2$ (0.07 g, 1.01 mmol, 0.5 ml H_2O). The resulting diazonium salt solution was stirred at 0 °C for 30 min to 1 h, followed by the addition of a few crystals of urea, to decompose any extra nitrous acid, and KI (1.60 g, 9.64 mmol). The dark mixture was stirred at room temperature for another 2 h and extracted with EtOAc. The organic layer was then washed with an aqueous solution of Na_2SO_3 resulting in the precipitation of an off-white solid, which was filtered and dried to give the title product (0.22 g, 75% yield). ^{1}H NMR (D_2O/NaOD): δ (ppm) 2.67 (dd, J = 10.9 Hz, J = 16.5 Hz, 1H, $C\underline{H_2}CH$), 2.88 (dd, J = 4.7 Hz, J = 16.5 Hz, 1H, $C\underline{H_2}CH$), 3.31 (dd, J = 4.7 Hz, J = 10.9 Hz, 1H, $CH_2C\underline{H}$), 3.73 (d, J = 16.1 Hz, 1H, $C\underline{H_2}N$), 3.87 (d, J = 16.1 Hz, 1H, $C\underline{H_2}N$), 6.79 (d, J = 8.0 Hz, 1H, aromatic), 7.42 (d, J = 8.0 Hz, 1H, aromatic), 7.48 (s, 1H, aromatic); mp >200 °C (decomp.); HPLC r_t = 28.2 min (0–35% B/35 min); ES-MS ($M+H^+$): 303.8.



Scheme 2. Synthesis of compounds **7a** and **7b**.

*7-Iodo-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid (***6b***)*

7-Iodo-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid (**6b**) was synthesized in 75% yield using the procedure described above for **6a**. ^{1}H NMR (D_2O/NaOD): δ (ppm) 2.71 (dd, J = 10.6 Hz, J = 16.7 Hz, 1H, $C\underline{H_2}CH$), 2.94 (dd, J = 4.4 Hz, J = 16.7 Hz, 1H, $C\underline{H_2}CH$), 3.37 (dd, J = 4.5 Hz, J = 10.6 Hz, 1H, $CH_2C\underline{H}$), 3.81 (d, J = 16.2 Hz, 1H, $C\underline{H_2}N$), 3.93 (d, J = 16.2 Hz, 1H, $C\underline{H_2}N$), 6.92 (d, J = 8.2 Hz, 1H, aromatic), 7.47 (s, 1H, aromatic) 7.48 (d, J = 8.2 Hz, 1H, aromatic); mp >230 °C (decomp.); HPLC r_t = 28.0 min (0–35% B/35 min); ES-MS ($M+H^+$): 303.9.

*2-Tert-butoxycarbonyl-1,2,3,4-tetrahydro-6-iodo-isoquinoline-3-carboxylic acid (***7a***)*

A solution of **6a** (0.19 g, 0.63 mmol) in a mixture of THF (10 ml), water (10 ml), Et_3N (0.13 g, 1.23 mmol) and di-*tert*-butyldicarbonate (0.16 g, 0.75 mmol) was stirred at 25 °C overnight. The solution was concentrated to about half the original volume, cooled in an ice bath, covered with a layer of EtOAc, and acidified to pH 2–3 with 5% aqueous citric acid. The aqueous layer was extracted with EtOAc and the combined organic phase was dried ($MgSO_4$) and evaporated. The resulting residue was purified by flash column chromatography (silica gel, EtOAc:acetic acid/200:0.4) to give 0.2 g of **7a** (79%). ^{1}H NMR ($CDCl_3$): δ (ppm) 1.44 and 1.53 (*cis* and *trans* conformers) (s, 9H, Boc), 3.1 (m, 2H, $C\underline{H_2}CH$), 4.4–4.7 (m, 2H, $C\underline{H_2}N$), 4.81 and 5.12 (m, 1H, $CH_2C\underline{H}N$), 6.88 (d, J = 7.8 Hz, 1H, aromatic), 7.54 (m, 2H, aromatic). ES-MS ($M+H^+$): 403.5. HPLC r_t = 50.5 min (0–60% B/60 min).

*2-Tert-Butoxycarbonyl-1,2,3,4-tetrahydro-7-iodo-isoquinoline-3-carboxylic acid (***7b***)*

2-*tert*-Butoxycarbonyl-1,2,3,4-tetrahydro-7-iodoisoquinoline-3-carboxylic acid (**7b**) was synthesized in 86% yield using the procedure described above for **7a**. ^{1}H NMR ($CDCl_3$): δ (ppm) 1.44 and 1.53 (*cis* and *trans* conformers) (s, 9H, Boc), 3.1 (m, 2H, $C\underline{H_2}CH$), 4.66 (m, 2H, $C\underline{H_2}N$), 4.80 and 5.13 (m, 1H, $CH_2C\underline{H}N$), 6.92 (d, J = 7.8 Hz, 1H, aromatic), 7.51 (m, 2H, aromatic). ES-MS ($M+H^+$): 403.5. HPLC r_t = 51.0 min (0–60% B/60 min).

Results and Discussion

Schemes 1 and 2 represent a convenient route for the synthesis of aryl-substituted analogues of Tic in reasonable yields. In Scheme 1, K_2CO_3 or Cs_2CO_3 can also be used as the base to promote the condensation of α,α-dibromo-4-nitro-*o*-xylene (**2**) with diethyl acetamidomalonate, but require longer reaction times

and give rise to side products, as indicated by smears on the TLC plates. Acetone was found to be superior to other polar solvents such as THF, MeOH, or DMF for this step, resulting in decreased reaction times and improved purity. The two nitro regioisomers, **3a** and **3b**, were assigned based on a one-dimensional difference nuclear Overhauser experiment, wherein the irradiation of H-1 (δ = 4.79 ppm) in **3a** led to increased intensity of a doublet (H-8, δ = 7.36 ppm) in the aromatic proton region, while irradiation of H-1 (δ = 4.79 ppm) in **3b** led to increased intensity of a singlet (H-8, δ = 8.10 ppm).

The amino-Tic compounds **5a** and **5b** might have minor contamination as suggested by the presence of a light yellow color; however, these proved to be difficult to purify by either crystallization or chromatography and were therefore carried on to the next step in Scheme 2. The iodo-Tic derivatives, **6a** and **6b**, which are only slightly soluble in water and other common organic solvents, were purified by first converting to their corresponding Boc-derivatives **7a** and **7b** using standard conditions, then purified by flash chromatography (silica gel, AcOEt:AcOH/150:1) in 39 and 42% yield, respectively.

Conclusions

Tic analogues substituted at the 6 or 7 position with nitro, amino, or iodo groups have been synthesized in reasonable yields in three to five steps. The ready conversion of amino-Tic to iodo-Tic without the need of protecting groups as well as the ease of isolating iodo-Tic are advantages of this synthetic route. Further conversions of the functionally versatile amino-Tic or iodo-Tic to other Tic analogues are in progress and the extension of this pathway to functionalize Tic at the 5 or 8 position is also planned.

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