Synthesis of β-Benzo[*b*]thienyldehydrophenylalanine Derivatives by One-Pot Palladium-Catalyzed Borylation and Suzuki Coupling (BSC) and Metal-Assisted Intramolecular Cyclization – Studies of Fluorescence and Antimicrobial Activity

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Keywords: Dehydrophenylalanines / Benzothiophenes / Borylation / Suzuki coupling / Cyclization / Indoles / Fluorescence / Antimicrobial activity

Palladium-catalyzed borylation and Suzuki coupling (BSC) in a one-pot procedure was successfully applied to the synthesis of several β-substituted dehydrophenylalanines in the benzo[*b*]thiophene series, with the stereochemistry of the starting materials being maintained. Bromobenzo[*b*]thiophenes bearing an *ortho* EDG (OMe or Me) were used as the components to be borylated with pinacolborane, whilst pure stereoisomers of β-bromodehydrophenylalanines were used as the other Suzuki coupling components. Treatment of the obtained methyl ester of (*Z*)-*N*-(*tert*-butoxycarbonyl)-β-(2,3,5-trimethylbenzo[*b*]thien-6-yl)dehydrophenylalanine with $Pd(OAc)_2$ and $Cu(OAc)_2$ in DMF at 160 °C gave two indole derivatives (1:3), the major product resulting from isomerization and cyclization and the minor product resulting from direct cyclization (thienoindole). Carrying out the reaction at 100 °C gave the same products in similar amounts. Use of the methyl ester of (*Z*)-*N*-(*tert*-butoxycarbonyl)-β-(2,3,7-trimethylbenzo[*b*]thien-6-yl)dehydrophenylalanine as starting material gave only one product, resulting from isomerization and cyclization at 100 °C. Two of the cyclized compounds were subjected to fluorescence studies; the thienoindole could be useful as a fluorescent probe. Preliminary studies of antimicrobial activity were performed on the precursors and on the cyclized products.

Introduction

In recent years we have been interested in the linkage of dehydroamino acid derivatives to benzo[*b*]thiophenes and in the study of the fluorescence and antimicrobial activity of the resulting products. By Suzuki coupling we were able to synthesize either β-benzo[*b*]thienyl or β,β-bis(benzo[*b*]-thienyl)dehydroamino acid derivatives.[1,2] The latter have been cyclized to the corresponding dehydroprolines by metal-assisted intramolecular cyclization and the fluorescence properties of one of them have been studied.[2b]

Here we describe the application of one-pot borylation/Suzuki coupling (BSC) to the synthesis of new benzo[*b*]thienyldehydrophenylalanine derivatives from methylated or methoxylated benzo[*b*]thiophenes and brominated dehydrophenylalanines. This method was first applied by Baudoin et al.[3] to the synthesis of 2,2′-disubstituted biaryl compounds. Substituted halobenzenes were subjected to borylation with pinacolborane, followed by in situ Suzuki cross-coupling with a second 2-substituted halobenzene. The authors postulated that the borylation should be performed on a component bearing an *ortho*-EDG (electron-donating group), with the other coupling component having an *ortho*-EWG (electron-withdrawing group). The method is also based on the use of the electron-rich sterically hindered 2-(dicyclohexylphosphanyl)biphenyl as ligand.[4] Recently we applied this reaction to the synthesis of 2-methyl-2′-nitrobiaryl compounds in the benzo[*b*]thiophene series to obtain thienocarbazoles.[5] Another application of the BSC to the synthesis of substituted biaryl compounds, with use of DPEphos as ligand and CsF as base in the Suzuki coupling, has appeared more recently.[6]

In this work the brominated Suzuki coupling component is not aromatic, which constitutes a novel application of the BSC reaction as reported by us previously.[7] The C–N metal-assisted cyclization[2] of differently *ortho*-methylated *Z*-β-benzo[*b*]thienyldehydrophenylalanine derivatives was studied, and a thienoindole obtained in this way was shown to be fluorescent. Preliminary antimicrobial studies were performed on the precursors or on the cyclized products.

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Results and Discussion

Synthesis

Several benzo[*b*]thienyldehydrophenylalanines were prepared by palladium-catalyzed BSC reactions between bro-

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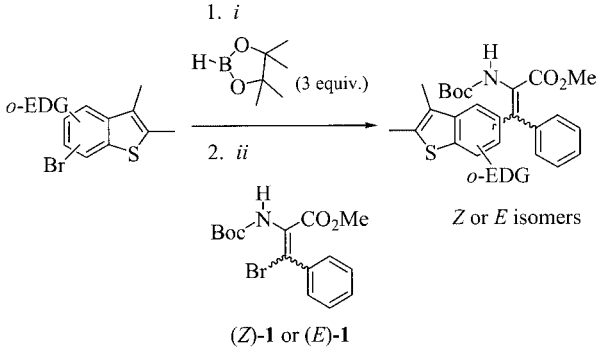
mobenzo[*b*]thiophenes *ortho*-substituted with EDG groups (OMe or Me) and pure stereoisomers of the methyl esters of *N*-(*tert*-butoxycarbonyl)-β-bromodehydrophenylalanine: (*Z*)-Boc-ΔPhe-(β-Br)-OMe) [(*Z*)-**1**] and (*E*)-Boc-ΔPhe-(β-Br)-OMe ((*E*)-**1**), already prepared by us.[1] The bromobenzo[*b*]thiophenes were borylated with pinacolborane with subsequent Suzuki coupling with (*Z*)-**1** or (*E*)-**1** in a one-pot procedure (Scheme 1, Table 1).



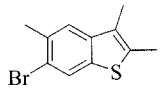
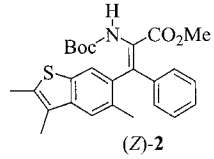
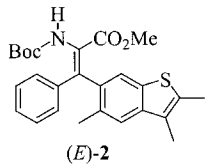
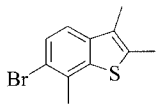
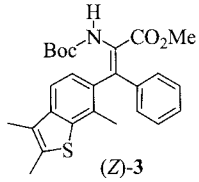
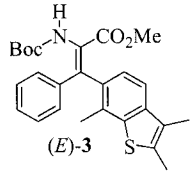
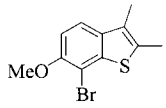
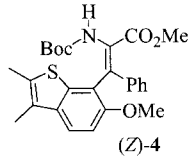
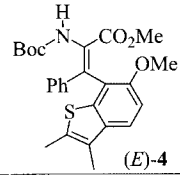
Scheme 1. *i*) $Pd(OAc)_2$ (5 mol%), 2-(dicyclohexylphosphanyl)biphenyl (20 mol%), NEt_3 (4 equiv.), dry dioxane, 80 °C, 1 h, Ar. *ii*) $Ba(OH)_2{\cdot}8H_2O$ (3 equiv.), H_2O, 100 °C, 1 h 30 min.

Debrominated benzo[*b*]thiophenes were isolated as by-products in all cases (25–50%). If the two components are aromatic the component to be borylated needs an EDG and the other Suzuki coupling component requires an EWG.[3,5] In our case, however, the carbamate group of the dehydroamino acid derivative has a slight electron-donating effect and this can affect the product yields, which were moderate to good (Table 1). The products maintained the stereochemistry of the starting materials, as observed by NOE difference experiments. Enhancements on the phenyl protons for the (*E*) isomers and on the *ortho*-methyl or -methoxy and on the benzo[*b*]thienyl protons for the (*Z*) isomers were observed on irradiation of the α-NH, as shown as examples in Figure 1 and 2 for compounds (*Z*)-**3** and (*E*)-**3**. In Figure 1 we observe an enhancement of the 7-Me (δ = 2.39 ppm) and of the doublet of 5-H (δ = 7.06 ppm). In Figure 2 the NOE enhancement is only observed for the phenyl protons.

In order to obtain cyclic amino acids, compounds (*Z*)-**2** and (*Z*)-**3** were subjected to our cyclization conditions[2] (Scheme 2 and Scheme 3). Compound (*Z*)-**2** gave two cyclized products resulting either from isomerization followed by cyclization to give indole **5** (major product), or from direct cyclization to give thienoindole **6**. The reaction was also performed at 100 °C for 6 h, but the same compounds were obtained in similar amounts.

Compound (*Z*)-**3** was also subjected to the same conditions (heating at 100 °C for 2 h; Scheme 3), but a cyclized intermediate **7**, resulting from isomerization and cyclization without *N*-deprotection was formed (this compound was isolated from an independent experiment, stopping the reaction at this point). Heating at 130 °C for an additional hour gave the corresponding *N*-deprotected dehydroproline derivative in high yield. In this case no product resulting from direct cyclization of the starting material was isolated. This could be due to the cyclization on the phenyl ring being much more favorable than on position 5 of the benzo[*b*]-thiophene moiety.

Table 1. Starting materials and product yields of BSC reaction.

Benzo[*b*]thiophene	Dehydrophenyl-alanine	Products	Yield (%)
	(*Z*)-**1**	 (*Z*)-**2**	61
	(*E*)-**1**	 (*E*)-**2**	30
	(*Z*)-**1**	 (*Z*)-**3**	52
	(*E*)-**1**	 (*E*)-**3**	43
	(*Z*)-**1**	 (*Z*)-**4**	54
	(*E*)-**1**	 (*E*)-**4**	40

We believe that the mechanism of cyclization involves an electrophilic attack of Pd^{II} on the aromatic ring and a nucleophilic attack of nitrogen, forming a palladacycle, the indole being formed after Pd^0 extrusion. The role of the $Cu(OAc)_2$ may be the reoxidation of Pd^0, avoiding the use of stoichiometric amounts of $Pd(OAc)_2$ (Scheme 4). In our case *N*-deprotection also occurs depending on the reaction conditions (temperature and/or time).

Antimicrobial Activity in vitro

A screening of antibacterial activities with two Gram negative (*Escherichia coli* and *Pseudomonas aeruginosa*) and two Gram positive bacteria (*Bacillus subtilis* and *Bacillus*

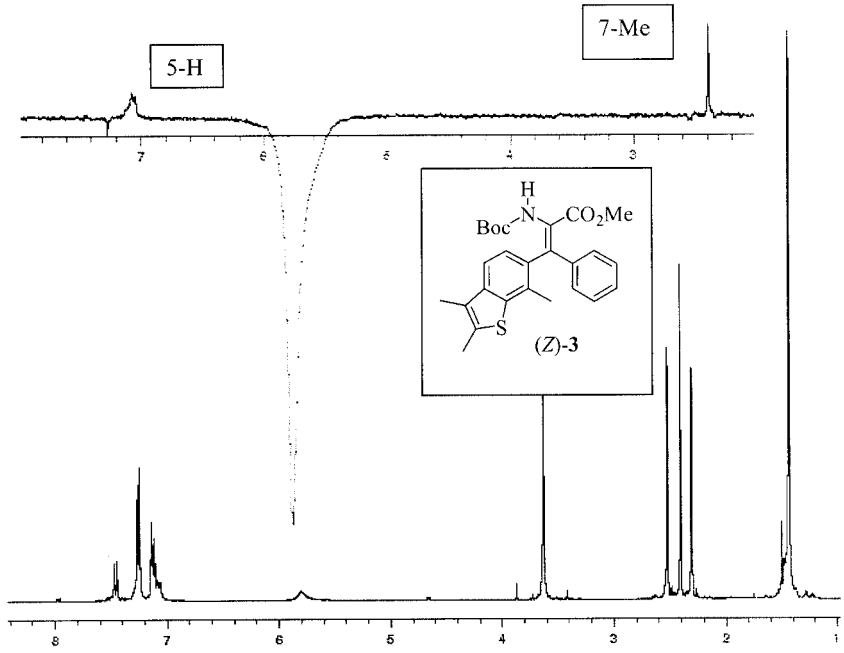


Figure 1. ^{1}H NMR ($CDCl_3$) spectrum and NOE difference experiment with irradiation of the α-NH of compound (*Z*)-**3**.

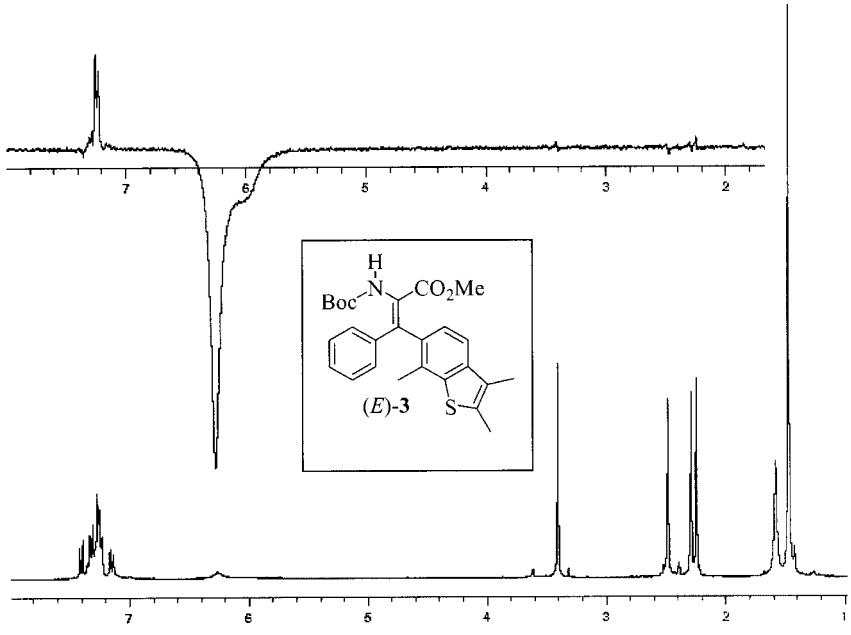
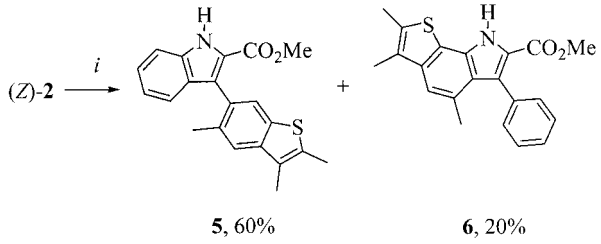
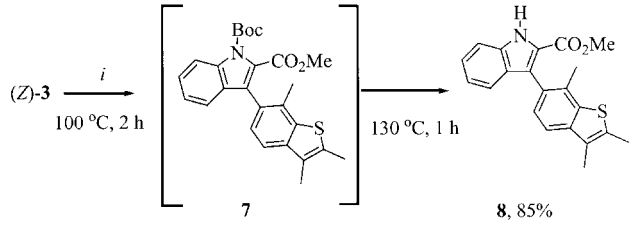


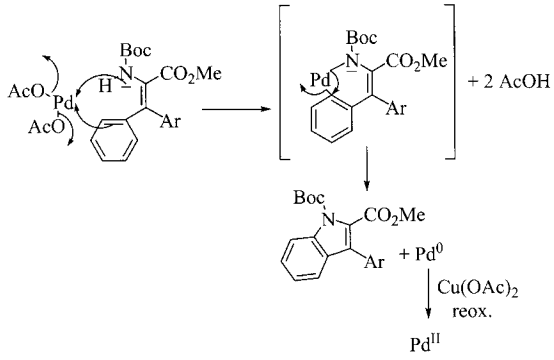
Figure 2. ^{1}H NMR ($CDCl_3$) spectrum and NOE difference experiment with irradiation of the αNH of compound (*E*)-**3**.



Scheme 2. *i*) $Pd(OAc)_2$ (50 mol%), $Cu(OAc)_2{\cdot}H_2O$ (3 equiv.), DMF, 160 °C, 2 h 30 min.



Scheme 3. *i*) $Pd(OAc)_2$ (50 mol%), $Cu(OAc)_2{\cdot}H_2O$ (3 equiv.), DMF.



Scheme 4.

cereus) and of antifungal activity with *Candida albicans* as a representative of fungi was assessed for compounds (*Z*)-**2**, (*Z*)-**3**, **5**, **6**, **7**, and **8**. The minimal inhibitory concentrations (MICs in $\mu g \cdot mL^{-1}$) were determined (Table 2) by an adaptation of the agar streak dilution method based on radial diffusion.[8] Under the same conditions, different concentrated solutions of ampicillin (antibacterial) and cyclohexamide (antifungal) were used as standards. The MIC was considered to be the lowest concentration of the tested compound able to inhibit growth of bacteria or fungi on the plate. The diameters of the inhibition zones corresponding to the MICs are presented in Table 2. The compounds tested are not active against *Pseudomonas aeruginosa*, starting from DMSO solutions of 6000 $\mu g \cdot mL^{-1}$ of each compound. Compounds (*Z*)-**2** and (*Z*)-**3** are also not active against *Escherichia coli*, but are the only ones active against *Candida albicans*, (*Z*)-**2** (MIC = 6 $\mu g \cdot mL^{-1}$) being more active than (*Z*)-**3** (MIC = 600 $\mu g \cdot mL^{-1}$) and than cyclohexamide (MIC = 12.5 $\mu g \cdot mL^{-1}$). Against Gram-positive bacteria (*Z*)-**2** is more active against *B. cereus*, but (*Z*)-**3** shows a lower MIC, even lower than ampicillin, against *B. subtilis*.

Against *Escherichia coli*, indoles **7** and **8** are more active (MIC = 0.06 $\mu g \cdot mL^{-1}$) than thienoindole **6** (MIC = 0.6 $\mu g \cdot mL^{-1}$). All the cyclized products are active against Gram-positive bacteria, presenting lower MICs than their precursors (*Z*)-**2**, (*Z*)-**3** and ampicillin, indole **7** being the most active (MIC = 0.006 $\mu g \cdot mL^{-1}$).

Fluorescence Studies

The fluorescence of compounds **5** and **6** was studied (Figure 3). Thienoindole **6** showed a λ_{em} = 393 nm and a relative quantum yield of fluorescence in dichloromethane Φ_{dcm} = 0.33 by the standard method[9] with 9,10-diphenylanthracene in EtOH (2×10^{-5} M) as reference (Φ_{EtOH} = 0.95)[10] at λ_{exc} = 333 nm. Indole **5** showed a λ_{em} = 408 nm and a relative quantum yield of fluorescence in dichloromethane Φ_{dcm} = 0.0067 by the same method and at λ_{exc} = 296 nm. The excitation wavelengths (λ_{exc}) were chosen from the UV spectra of the compounds (see Experimental Section). From the results obtained it can be concluded that thienoindole **6** may be useful as a fluorescent probe.

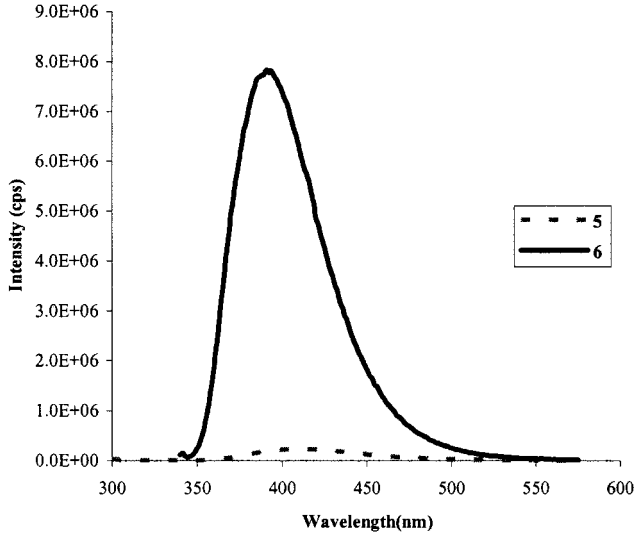


Figure 3. Fluorescence spectra of compounds **5** and **6** in dichloromethane (1.6×10^{-6} M).

Conclusions

Several new β-benzo[*b*]thienyldehydrophenylalanines were synthesized in moderate to good yields from *ortho*-substituted bromobenzo[*b*]thiophenes and pure stereoisomers of β-brominated dehydrophenylalanines by one-pot borylation and Suzuki coupling (BSC), with the stereochemistry of the starting materials being maintained. Two of the (*Z*) isomers obtained, (*Z*)-**2** and (*Z*)-**3**, were subjected to C–N metal-assisted cyclization to give dehydroprolines (indoles and a thienoindole). From preliminary antimicrobial studies on the precursors and on the cyclized products it is possible to conclude that all the compounds were active

Table 2. Antimicrobial activity of compounds (*Z*)-**2,** (*Z*)-**3**, **5**, **6**, **7**, and **8**.

Compounds	MIC in $\mu g \cdot mL^{-1}$ (Zone of inhibition in mm)			
	E. coli CECT 101[a]	*B. cereus CECT148*[a]	*B. subtilis CECT498*[a]	*C. albicans CECT 1394*[a]
(*Z*)-**2**	not active[b]	6 (10)	60 (11)	6 (8)
(*Z*)-**3**	not active[b]	60 (12)	6 (8)	600 (9)
Indole **5**	600 (11)	0.6 (13)	0.6 (9)	not active[b]
Thienoindole **6**	0.6 (7)	0.06 (10)	0.06 (9)	not active[b]
Indole **7**	0.06 (8)	0.006 (10)	0.006 (13)	not active[b]
Indole **8**	0.06 (11)	0.06 (9)	0.06 (8)	not active[b]
Ampicillin	6.25 (15)	3.13 (13)	12.5 (10)	–
Cyclohexamide	–	–	–	12.5 (5)

[a] CECT-Spanish type culture collection from Valencia University. [b] Not active starting from 6000 $\mu g \cdot mL^{-1}$.

against the Gram positive bacteria (*B. cereus* and *B. subtilis*), the cyclized products being more active (lower MICs). The latter were also active against *E. coli*, despite presenting very different MICs, and the precursors were in turn the only compounds active against *Candida albicans*. Fluorescence studies on compounds **5** and **6** show that the thienoindole **6** may be useful as a fluorescent probe.

Experimental Section

General Remarks: Melting points were determined on a Gallenkamp apparatus and are uncorrected. The ^{1}H NMR spectra were measured on a Varian Unity Plus instrument at 300 MHz. Spin–spin decoupling techniques were used to assign the signals. NOE difference experiments were performed to determine the stereochemistries of the products. The ^{13}C NMR spectra were measured in the same instrument at 75.4 MHz (with DEPT θ 45°). The UV spectra were recorded on a Schimadzu UV-250 1PC, UV/Vis recording spectrophotometer.

Elemental analyses were determined on a LECO CHNS 932 elemental analyser. Mass spectra (EI) and HRMS were measured by the mass spectrometry service of the University of Vigo, Spain.

Column chromatography was performed on Macherey–Nagel silica gel (230–400 mesh). "Petroleum ether" had a boiling range of 40–60 °C. "Ether" refers to diethyl ether. When a solvent gradient was used the increase of polarity was done gradually from neat petroleum ether to mixtures of ether/petroleum ether, increasing by 10% of ether until the isolation of the product.

For the determination of in vitro antimicrobial activity, suspensions of the microorganism were prepared to contain approximately 10^8 cfu·mL^{-1} and the plates were inoculated. Stock solutions of the synthesized compound (6000 μg·mL^{-1}) in DMSO were prepared and graded dilutions (10×) of the tested compounds were incorporated in a cavity (depth 3 mm, diameter 4 mm) made in the center of the Petri dish (nutrient agar for antibacterial activity and sabouraud dextrose agar medium for antifungal activity). The plates were incubated in duplicate at 37 °C (for bacteria) and at 30 °C (for fungi) for 24 h. Positive control using only inoculation and negative control using only DMSO in the cavity were carried out.

The fluorescence studies were performed on a spectrofluorimeter Spex Fluorolog 1680 Double Spectrometer.

Samples quantum yields are given by $\Phi_s = [(A_r F_s n_s^2)/(A_s F_r n_r^2)]\Phi_r$, where A is the absorbance at the excitation wavelength, F the integrated emission area and n the refraction index of the solvents used. Subscripts refer to the reference (r) or sample (s) compound.

Compounds (*Z*)-**1** and (*E*)-**1** were already described by us.[1] The 6-bromotrimethylbenzo[*b*]thiophenes were prepared by a procedure described by others[11a] and previously used by us.[11b]

7-Bromo-6-methoxy-2,3-dimethylbenzo[*b*]thiophene and 5-Bromo-6-methoxy-2,3-dimethylbenzo[*b*]thiophene: Br_2 (3.00 mmol, 0.154 mL, 1 equiv.) and a small amount of Fe were added to a cooled solution (0 °C) of 6-methoxy-2,3-dimethylbenzo[*b*]thiophene[12] (3.00 mmol, 0.500 g) in dry ether (30 mL), protected against light. The reaction was followed by ^{1}H NMR and was left stirring for 1 h. Iced water (10 mL) was added, and the mixture was stirred for 10 min. The phases were separated and the aqueous phase was extracted with more ether (2 × 30 mL). The organic phases were collected, washed with a 5% solution of sodium sulfite and a 10% solution of sodium carbonate, and dried ($MgSO_4$). Removal of the solvent gave a solid that was subjected to column chromatography with petroleum ether.

5-Bromo-6-methoxy-2,3-dimethylbenzo[*b*]thiophene: This compound (62.0 mg, 8%) was isolated as the less polar product as a white solid, m.p. 119–120 °C. ^{1}H NMR ($CDCl_3$): δ = 2.24 (s, 3 H, $ArCH_3$), 2.45 (s, 3 H, $ArCH_3$), 3.95 (s, 3 H, OCH_3), 7.25 (s, 1 H, 7-H), 7.74 (s, 1 H, 4-H) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.29 (CH_3), 13.65 (CH_3), 56.44 (OCH_3), 104.53 (CH), 109.34 (C), 125.27 (CH), 125.94 (C), 132.40 (C), 136.01 (C), 138.08 (C), 152.54 (C) ppm. $C_{11}H_{11}BrOS$ (271.17): calcd. C 48.72, H 4.09, S 11.82; found C 48.87, H 4.23, S 11.60.

7-Bromo-6-methoxy-2,3-dimethylbenzo[*b*]thiophene: This compound (484 mg, 60%) was isolated as a white solid, m.p. 120–122 °C. ^{1}H NMR ($CDCl_3$): δ = 2.26 (s, 3 H, $ArCH_3$), 2.46 (s, 3 H, $ArCH_3$), 3.97 (s, 3 H, OCH_3), 7.00 (d, J = 8.4 Hz, 1 H, 5-H), 7.48 (d, J = 8.4 Hz, 1 H, 4-H) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.60 (CH_3), 13.67 (CH_3), 57.05 (OCH_3), 103.97 (C), 109.73 (CH), 120.39 (CH), 127.39 (C), 132.55 (C), 135.85 (C), 141.55 (C), 152.74 (C) ppm. $C_{11}H_{11}BrOS$ (271.17): calcd. C 48.72, H 4.09, S 11.82; found C 49.00, H 4.27, S 11.74.

General Procedure for *One-pot* BSC Reactions: A dried Schlenk tube was charged under Ar with dry dioxane (2 mL), the *ortho*-methylated or -methoxylated bromobenzo[*b*]thiophene (0.5 mmol) was added, and the mixture was heated for 5 min at 80 °C. Triethylamine (4 equiv.), $Pd(OAc)_2$ (5 mol%), 2-(dicyclohexylphosphanyl) biphenyl (20 mol%), and pinacolborane (3 equiv.) were added, and the mixture was left rapidly stirring at 80 °C for 1 h. After cooling, a pure stereoisomer of β-bromodehydrophenylalanine derivative (1 equiv.) and $Ba(OH)_2{\cdot}8H_2O$ (3 equiv.) were added, and the mixture was heated at 100 °C for 1 h 30 min. Water and ethyl acetate were added, the phases were separated, and the aqueous phase was then extracted with more ethyl acetate. The organic phases were collected, dried ($MgSO_4$), and filtered, and the solvent was then evaporated at reduced pressure to give a brown solid, which was subjected to column chromatography.

(*Z*)-Boc-ΔPhe[β-(2,3,5-trimethylbenzo[*b*]thien-6-yl)]-OMe (*Z*)-2: The procedure described above was followed, with 6-bromo-2,3,5-trimethylbenzo[*b*]thiophene (0.500 mmol, 128 mg) and (*Z*)-Boc-ΔPhe(β-Br)-OMe (0.410 mmol, 147 mg), but with addition of water (200 μL) in the second step. Column chromatography with a solvent gradient from neat petroleum ether to 30% diethyl ether/petroleum ether gave product (*Z*)-**2** (112 mg, 61%) as an oil. Crystallization from diethyl ether/*n*-hexane gave a light yellow solid, m.p. 154–156 °C. ^{1}H NMR ($CDCl_3$): δ = 1.44 (s, 9 H, CH_3 Boc), 2.31 (s, 3 H, 5-CH_3), 2.35 (s, 3 H, $ArCH_3$), 2.49 (s, 3 H, $ArCH_3$), 3.64 (s, 3 H, OCH_3), 5.80 (s, 1 H, NH), 7.13–7.16 (m, 2 H, ArH), 7.24–7.29 (m, 3 H, ArH), 7.48 (broad s, 1 H, ArH), 7.51 (s, 1 H, ArH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.25 (CH_3), 13.76 (CH_3), 19.57 (CH_3), 27.99 ($C(CH_3)_3$), 52.03 (OCH_3), 81.22 ($OC(CH_3)_3$), 122.87 (CH), 123.20 (CH), 126.50 (C), 126.73 (C), 127.56 (CH), 128.00 (CH), 128.41 (CH), 132.28 (C), 133.32 (C), 135.21 (C), 135.91 (C), 139.17 (C), 141.22 (C), 152.48 (C=O), 166.30 (C=O) ppm. MS: *m*/*z* (%) = 451 (22) $[M]^+$, 351 (100) $[M - Boc]^+$, 291 (34) $[(M - Boc - CO_2CH_3]^+$. HRMS: calcd. for $C_{26}H_{29}NO_4S$ $[M]^+$ 451.1817; found 451.1801.

(*E*)-Boc-ΔPhe[β-(2,3,5-trimethylbenzo[*b*]thien-6-yl)]-OMe (*E*)-2: The procedure described above was followed, with 6-bromo-2,3,5-trimethylbenzo[*b*]thiophene (0.460 mmol, 117 mg) and (*E*)-Boc-ΔPhe(β-Br)-OMe (0.380 mmol, 136 mg), but with addition of water (200 μL) in the second step. Column chromatography with a solvent gradient from neat petroleum ether to 30% diethyl ether/petroleum ether gave product (*E*)-**2** (51.0 mg, 30%) as an oil. Crystalli-

zation from petroleum ether gave white crystals, m.p. 156–157 °C. ^{1}H NMR ($CDCl_3$): δ = 1.48 (s, 9 H, CH_3 Boc), 2.13 (s, 3 H, $ArCH_3$), 2.26 (s, 3 H, $ArCH_3$), 2.48 (s, 3 H, $ArCH_3$), 3.39 (s, 3 H, OCH_3), 6.26 (s, 1 H, NH), 7.24–7.34 (m, 6 H, ArH), 7.51 (s, 1 H, ArH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.35 (CH_3), 13.82 (CH_3), 20.25 (CH_3), 28.18 (C(CH_3)$_3$), 52.00 (OCH_3), 81.21 (OC(CH_3)$_3$), 122.45 (CH), 122.54 (CH), 126.40 (C), 126.67 (C), 128.09 (CH), 128.63 (CH), 129.38 (CH), 132.98 (C), 134.29 (C), 135.06 (C), 137.52 (C), 140.98 (C), 152.99 (C=O), 166.24 (C=O) ppm. $C_{26}H_{29}NO_4S$ (451.58): calcd. C 69.15, H 6.47, N 3.10, S 7.10; found C 68.90, H 6.54, N 3.14, S 7.03.

(*Z*)-Boc-ΔPhe[β-(2,3,7-trimethylbenzo[*b*]thien-6-yl)]-OMe (*Z*)-3: The procedure described above was followed, with 6-bromo-2,3,7-trimethylbenzo[*b*]thiophene (0.500 mmol, 128 mg) and (*Z*)-Boc-ΔPhe(β-Br)-OMe (0.500 mmol, 178 mg), but with addition of water (200 μL) in the second step. Column chromatography with a solvent gradient from neat petroleum ether to 30% diethyl ether/petroleum ether gave product (*Z*)-**3** (122 mg, 52%) as an oil. Crystallization from petroleum ether gave white crystals, m.p. 149–150 °C. ^{1}H NMR ($CDCl_3$): δ = 1.42 (s, 9 H, CH_3 Boc), 2.31 (s, 3 H, $ArCH_3$), 2.39 (s, 3 H, 7-CH_3), 2.53 (s, 3 H, $ArCH_3$), 3.62 (s, 3 H, OCH_3), 5.80 (s, 1 H, NH), 7.06 (d, J = 8.4 Hz, 1 H, 5-H), 7.09–7.13 (m, 2 H, ArH), 7.22–7.28 (m, 3 H, ArH), 7.45 (d, J = 8.4 Hz, 1 H, 4-H) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.42 (CH_3), 13.84 (CH_3), 17.59 (CH_3), 28.08 (C(CH_3)$_3$), 52.07 (OCH_3), 81.33 (OC(CH_3)$_3$), 119.44 (CH), 126.53 (C), 126.93 (CH), 127.60 (CH), 127.90 (C), 128.06 (CH), 128.52 (CH), 130.04 (C), 133.02 (C), 134.78 (C), 139.45 (C), 139.55 (C), 140.63 (C), 152.57 (C=O), 166.38 (C=O) ppm. $C_{26}H_{29}NO_5S$ (467.58): calcd. C 69.12, H 6.47, N 3.10, S 7.10; found C 69.15, H 6.63, N 3.11, S 6.91.

(*E*)-Boc-ΔPhe[β-(2,3,7-trimethylbenzo[*b*]thien-6-yl)]-OMe (*E*)-3: The procedure described above was followed, with 6-bromo-2,3,7-trimethylbenzo[*b*]thiophene (0.310 mmol, 79.0 mg) and (*E*)-Boc-ΔPhe(β-Br)-OMe (0.310 mmol, 111 mg), but with addition of water (125 μL) in the second step. Column chromatography with a solvent gradient from neat petroleum ether to 30% diethyl ether/petroleum ether gave product (*E*)-**3** (41 mg, 43%) as an oil. Crystallization from petroleum ether gave light yellow crystals, m.p. 172–173 °C. ^{1}H NMR ($CDCl_3$): δ = 1.47 (s, 9 H, CH_3 Boc), 2.24 (s, 3 H, $ArCH_3$), 2.29 (s, 3 H, $ArCH_3$), 2.48 (s, 3 H, $ArCH_3$), 3.40 (s, 3 H, OCH_3), 6.24 (s, 1 H, NH), 7.15 (d, J = 8.1 Hz, 1 H, ArH), 7.23–7.33 (m, 5 H, ArH), 7.40 (d, J = 8.1 Hz, 1 H, ArH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.48 (CH_3), 13.84 (CH_3), 18.16 (CH_3), 28.17 (C(CH_3)$_3$), 52.01 (OCH_3), 81.21 (OC(CH_3)$_3$), 118.31 (CH), 126.43 (CH), 126.59 (C), 127.81 (C), 128.04 (CH), 128.52 (C), 128.65 (CH), 129.28 (CH), 130.18 (C), 133.76 (C), 133.94 (C), 138.17 (C), 139.17 (C), 140.52 (C), 152.97 (C=O), 166.27 (C=O) ppm. $C_{26}H_{29}NO_4S$ (451.58): calcd. C 69.15, H 6.47, N 3.10, S 7.10; found C 69.19, H 6.60, N 3.14, S 7.11.

(*Z*)-Boc-ΔPhe[β-(6-methoxy-2,3-dimethylbenzo[*b*]thien-7-yl)]-OMe (*Z*)-4: The procedure described above was followed, with 7-bromo-6-methoxy-2,3-dimethylbenzo[*b*]thiophene (0.400 mmol, 108 mg) and (*Z*)-Boc-ΔPhe(β-Br)-OMe (0.400 mmol, 142 mg), but with addition of water (160 μL) in the second step. Column chromatography with a solvent gradient from neat petroleum ether to 40% diethyl ether/petroleum ether gave product (*Z*)-**4** (100 mg, 54%) as a yellow solid. Recrystallization from petroleum ether gave yellow crystals, m.p. 104–106 °C. ^{1}H NMR ($CDCl_3$): δ = 1.41 (s, 9 H, CH_3 Boc), 2.26 (s, 3 H, $ArCH_3$), 2.37 (s, 3 H, $ArCH_3$), 3.59 (s, 3 H, OCH_3), 3.74 (s, 3 H, 6-OCH_3), 5.86 (s, 1 H, NH), 7.04 (d, J = 8.4 Hz, 1 H, ArH), 7.20–7.24 (m, 5 H, ArH), 7.53 (d, J = 8.4 Hz, 1 H, ArH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.33 (CH_3), 13.57 (CH_3), 28.03 (C(CH_3)$_3$), 52.08 (OCH_3), 56.48 (OCH_3), 80.86 (OC(CH_3)$_3$), 109.79 (CH), 119.50 (C), 121.93 (CH), 126.38 (C), 127.48 (CH), 127.77 (CH), 127.82 (C), 128.39 (CH), 132.46 (C), 135.62 (C), 138.18 (C), 139.75 (C), 152.70 (C), 153.77 (C=O), 166.25 (C=O) ppm. $C_{26}H_{29}NO_5S$ (467.58): calcd. C 66.79, H 6.25, N 3.00, S 6.86; found C 66.93, H 6.62, N 2.96, S 6.63.

(*E*)-Boc-ΔPhe[β-(6-methoxy-2,3-dimethylbenzo[*b*]thien-7-yl)]-OMe (*E*)-4: The procedure described above was followed, with 7-bromo-6-methoxy-2,3-dimethylbenzo[*b*]thiophene (0.340 mmol, 92.0 mg) and (*E*)-Boc-ΔPhe(β-Br)-OMe (0.340 mmol, 119 mg), but with addition of water (134 μL) in the second step. Column chromatography with a solvent gradient from neat petroleum ether to 40% diethyl ether/petroleum ether gave product (*E*)-**4** (63.0 mg, 40%) as a yellow solid. Recrystallization from petroleum ether gave yellow crystals, m.p. 135–136 °C. ^{1}H NMR ($CDCl_3$): δ = 1.46 (s, 9 H, CH_3 Boc), 2.22 (s, 3 H, $ArCH_3$), 2.36 (s, 3 H, $ArCH_3$), 3.44 (s, 3 H, OCH_3), 3.78 (s, 3 H, OCH_3), 6.22 (s, 1 H, NH), 6.99 (d, J = 8.7 Hz, 1 H, ArH), 7.26–7.37 (m, 5 H, ArH), 7.45 (d, J = 8.7 Hz, 1 H, ArH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.34 (CH_3), 13.66 (CH_3), 28.15 (C(CH_3)$_3$), 51.79 (OCH_3), 56.64 (OCH_3), 80.92 (OC(CH_3)$_3$), 109.56 (CH), 121.12 (CH), 122.29 (C), 126.26 (C), 127.68 (C), 127.80 (C) 128.07 (C), 128.48 (CH), 129.22 (CH), 132.04 (CH), 135.37 (C), 137.26 (C), 140.02 (C), 152.96 (C), 153.57 (C=O), 165.33 (C=O) ppm. $C_{26}H_{29}NO_5S$ (467.58): calcd. C 66.79, H 6.25, N 3.00, S 6.86; found C 66.54, H 6.33, N 3.07, S 6.91.

General Procedure for the Synthesis of Dehydroprolines: $Pd(OAc)_2$ (50 mol%) and $Cu(OAc)_2{\cdot}H_2O$ (3 equiv.) were added to a solution of the (*Z*)-benzo[*b*]thienyldehydrophenylalanines (0.1 M) in DMF, and the mixture was heated at 100 °C, 130 °C or 160 °C, the reaction being monitored by TLC. Ethyl acetate (50 mL) was then added and the organic layer was washed with water and brine (2 × 25 mL) and dried with $MgSO_4$, and the solvents were evaporated at reduce pressure to give an oil that was subjected to column chromatography.

Methyl 2,3,5-Trimethyl-6-phenyl-8*H*-thieno[3,2-*g*]indole-7-carboxylate (6) and Methyl 3-(2,3,5-Trimethylbenzo[*b*]thien-6-yl)indole-2-carboxylate (5): The procedure described above was applied with compound (*Z*)-**2** (0.177 mmol, 80.0 mg) and heating for 2 h 30 min at 160 °C. Column chromatography with a solvent gradient from neat petroleum ether to 30% diethyl ether/petroleum ether gave product **6** (12.0 mg, 20%) as a white solid, m.p. 215–217 °C, as the less polar product. ^{1}H NMR ($CDCl_3$): δ = 2.10 (s, 3 H, Ar–CH_3), 2.32 (s, 3 H, Ar–CH_3), 2.54 (s, 3 H, Ar–CH_3), 3.74 (s, 3 H, OCH_3), 7.07 (s, 1 H, ArH), 7.41 (s, 5 H, ArH), 9.07 (broad s, 1 H, NH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.80 (CH_3), 13.84 (CH_3), 20.62 (CH_3), 51.64 (OCH_3), 116.40 (CH), 118.53 (C), 121.87 (C), 123.47 (C), 126.82 (C), 127.14 (CH), 127.28 (CH), 128.41 (C), 129.83 (C), 130.50 (CH), 132.09 (C), 135.88 (C), 139.94 (C), 162.23 (C=O). UV(CH_2Cl_2): λ_{max} (ε $mol^{-1}{\cdot}dm^3{\cdot}cm^{-1}$) = 333 (14750), 266 (30938) nm. MS: m/z (%) = 349 (85) $[M]^+$, 318 (26) $[M - OCH_3]^+$, 317 (100), 288 (28). HRMS: calcd. for $C_{21}H_{19}NO_2S$ $[M]^+$ 349.1137; found 349.1130.

Product 5: This compound was eluted next, with 40% diethyl ether/petroleum ether and was isolated as a white solid (37.0 mg, 60%), m.p. 267–269 °C. ^{1}H NMR ($CDCl_3$): δ = 2.23 (s, 3 H, Ar–CH_3), 2.35 (s, 3 H, Ar–CH_3), 2.52 (s, 3 H, Ar–CH_3), 3.75 (s, 3 H, OCH_3), 7.12 (ap.t, 1 H, ArH), 7.33–7.49 (m, 3 H, ArH), 7.54 (s, 1 H, ArH), 7.63 (s, 1 H, ArH), 9.04 (broad s, 1 H, NH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.43 (CH_3), 13.84 (CH_3), 20.40 (CH_3), 51.84 (OCH_3), 111.64 (CH), 120.72 (CH), 121.78 (CH), 121.93 (CH), 123.43 (C), 123.71 (CH), 123.78 (C), 125.78 (CH), 126.69 (C), 128.54 (C), 129.33 (C), 133.38 (C), 133.84 (C), 135.07 (C), 135.70

(C), 140.80 (C), 162.46 (C=O). UV (CH_2Cl_2): λ_{max} (ε $mol^{-1}\cdot dm^3\cdot cm^{-1}$) = 296 (20875) 240 (48125) nm. MS: *m*/*z* (%) = 349 (100) $[M]^+$, 318 (15) $[M - OCH_3]^+$, 317 (41). HRMS: calcd. for $C_{21}H_{19}NO_2S$ $[M]^+$: 349.1137; found 349.1126.

Methyl *N*-(*tert*-Butoxycarbonyl)-3-(2,3,7-trimethylbenzo[*b*]thien-6-yl)indole-2-carboxylate (7): The procedure described above was applied with compound (*Z*)-**3** (0.310 mmol, 140 mg) and heating for 2 h at 100 °C. Column chromatography with a solvent gradient from neat petroleum ether to 10% diethyl ether/petroleum ether gave the compound (70.0 mg, 50%) as an oil. ^1H NMR ($CDCl_3$): δ = 1.70 (s, 9 H, CH_3 Boc), 2.37 (s, 3 H, Ar–CH_3), 2.38 (s, 3 H, Ar–CH_3), 2.56 (s, 3 H, Ar–CH_3), 3.72 (s, 3 H, OCH_3), 7.25–7.27 (m, 2 H, ArH), 7.32 (d, *J* = 8.1 Hz, 1 H, ArH), 7.42–7.49 (m, 1 H, ArH), 7.53 (d, *J* = 8.1 Hz, 1 H, ArH), 8.23 (d, *J* = 8.4 Hz, 1 H, ArH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.47 (CH_3), 13.82 (CH_3), 18.38 (CH_3), 27.91 ($C(CH_3)_3$), 52.19 (OCH_3), 84.67 ($OC(CH_3)_3$), 115.10 (CH), 118.40 (CH), 121.31 (CH), 123.33 (CH), 125.72 (C), 125.84 (C), 126.53 (CH), 127.03 (CH), 127.54 (C), 127.89 (C), 129.25 (C), 130.78 (C), 134.00 (C), 135.93 (C), 138.91 (C), 140.72 (C), 149.30 (C=O), 163.03 (C=O) ppm. MS: *m*/*z* (%) = 449 (22) $[M]^+$, 349 (100) $[M - Boc]^+$, 317 (43). HRMS: calcd. for $C_{26}H_{27}NO_4S$ $[M]^+$ 449.1650; found 449.1661.

Methyl 3-(2,3,7-Trimethylbenzo[*b*]thien-6-yl)indole-2-carboxylate (8): The procedure described above was applied with compound (*Z*)-**3** (0.137 mmol, 62.0 mg) and heating for 1 h at 130 °C. Column chromatography with a solvent gradient from neat petroleum ether to 20% diethyl ether/petroleum ether, gave compound **8** (41.0 mg, 85%) as a yellow solid, m.p. 230 –232 °C. ^1H NMR ($CDCl_3$): δ = 2.34 (s, 3 H, Ar–CH_3), 2.38 (s, 3 H, Ar–CH_3), 2.56 (s, 3 H, Ar–CH_3), 3.77 (s, 3 H, OCH_3), 7.14 (broad t, *J* = 6.9 Hz, 1 H, ArH), 7.33–7.41 (m, 3 H, ArH), 7.48 (d, *J* = 9.0 Hz, 1 H, ArH), 7.54 (d, *J* = 8.1 Hz, 1 H, ArH), 9.21 (s largo, 1 H, NH) ppm. ^{13}C NMR ($CDCl_3$): δ = 11.52 (CH_3), 13.84 (CH_3), 18.33 (CH_3), 51.84 (OCH_3), 111.70 (CH), 118.09 (CH), 120.70 (CH), 122.04 (CH), 123.50 (C), 125.73 (CH), 127.55 (CH), 127.90 (C), 128.62 (C), 130.54 (C), 133.49 (C), 135.70 (C), 138.75 (C), 140.20 (C), 162.56 (C=O) ppm. MS: *m*/*z* (%) = 349 (100) $[M]^+$, 318 (17) $[M - OCH_3]^+$, 317 (46), 288 (22). HRMS: calcd. for $C_{21}H_{19}NO_2S$ $[M]^+$ 349.1137; found 349.1142.

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