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SYNTHESIS, CHARACTERIZATION, AND BIOLOGICAL ACTIVITIES OF NEW BENZOFURAN DERIVATIVES

Kifah S. M. Salih,[a] Mikdad T. Ayoub,[b] Haythem A. Saadeh,[a] Najim A. Al-Masoudi,[c] and Mohammad S. Mubarak*[a]

[a]Chemistry Department, Faculty of Science, University of Jordan, Amman 11942, Jordan
[b]Chemistry Department, Faculty of Sciences, The Hashemite University, Zarqa, Jordan
[c] Fachbereich Chemie, Universität Konstanz, Postfach 5560, D-78457 Konstanz, Germany (*Formerly*)
* E-mail: mmubarak@ju.edu.jo

Abstract- A number of new benzofuran derivatives, ethyl 3-[(alkylamino)methyl]-6-methoxy-1-benzofuran-2-carboxylates (**5a-i**), were obtained *via* the reaction between ethyl 3-(bromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (**3**) and amines or amino acid ethyl esters. In addition, 1,4-bis[(ethyl 6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]piperazine (**9**), *N*,*N*'-diethyl-*N*,*N*'-bis[(6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]but-2-ene-1,4-diamine (**10**) and 1,2-bis[(ethyl 6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]-1,2-dimethyl-hydrazine (**11**) were also obtained from the reaction of **3** with diamines. Their *in vitro* anti-HIV-1 (strain III_B) and HIV-2 (strain ROD) activities of the synthesized compounds in human T-lymphocyte were tested; ethyl 3-bromomethyl-6-methoxycoumarlate displayed an ability to inhibit HIV-1 and HIV-2 replication in cell culture at non-toxic concentrations.

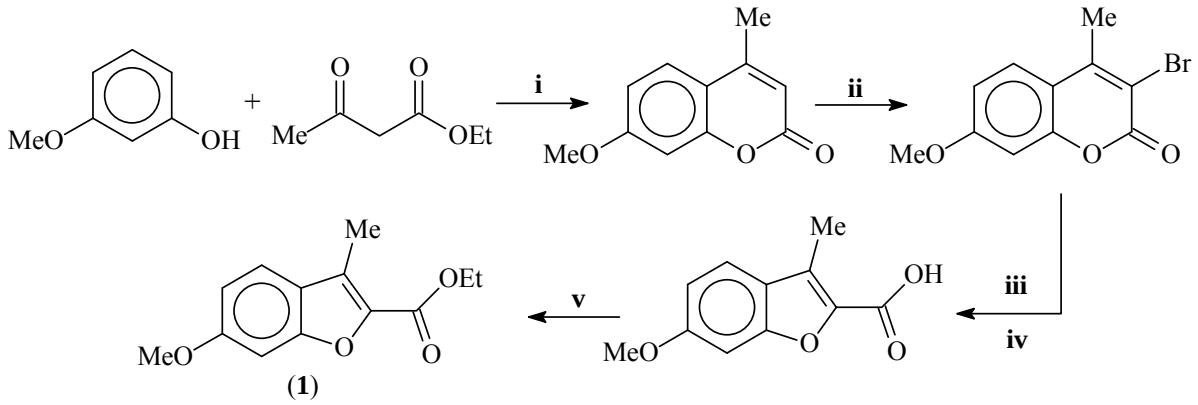
INTRODUCTION

Benzofurans have been found to exist in nature since the late 19th century, and at that time their investigations have revealed many avenues of understanding their properties.[1] Recently, isolation of benzofuran derivatives was reported along with other natural subunit derivatives, from the roots of *Leontopodium alpinum* and *L. leontopodioides*[2] and from seeds of *styrax officinalis*.[3] A wide variety of pharmacological properties have been shown to be associated with benzofuran derivatives.[4,5] New cytotoxic of cyclopenta[*b*]benzofuran derivatives, exhibited potent cytotoxicity against some cell lines.[6] The insecticidal activity of 2-carboxylbenzofuran and their coumarin precursors were demonstrated.[7] In continuation of our research on the synthesis of new heterocyclic compounds of pharmacological

interest,[8,9] we report, herein, on the synthesis, characterization, and anti HIV activities of new compounds containing a benzofuran skeleton combined with amines, amino acid ethyl esters, and diamines.

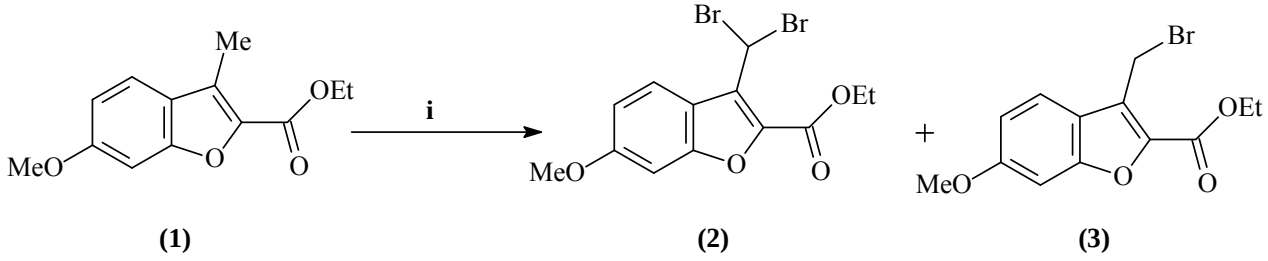
RESULTS AND DISCUSSION

The starting material, ethyl 6-methoxy-3-methyl-1-benzofuran-2-carboxylate (**1**) was synthesized through the steps depicted in Scheme 1.[9]



Scheme 1. *Reagents and conditions*: (i) H_2SO_4, 18 h, 80%; (ii) NBS, DBP, $CHCl_3$, reflux 5 h, 91%; (iii) KOH, EtOH, reflux 2 h; (iv) dil. HCl, 85%; (v) EtBr, K_2CO_3, acetone, reflux 12 h, 70%.

Following our previous literature route;[9] bromination of compound **1** by NBS in carbon tetrachloride and a few milligrams of dibenzoyl peroxide (DBP), as an initiator, afforded two products, ethyl 3-(dibromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (**2**) and ethyl 3-(bromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (**3**) in 17% and 57% yield, respectively (Scheme 2). However, when **1** was treated with portionwise addition of NBS,[9] and illumination with 100 W bulb,[10] or by refluxing the mixture of ethyl coumarlate and NBS in CCl_4 for 48 h without using dibenzoyl peroxide (DBP), the two products were obtained in nearly the same percentages.

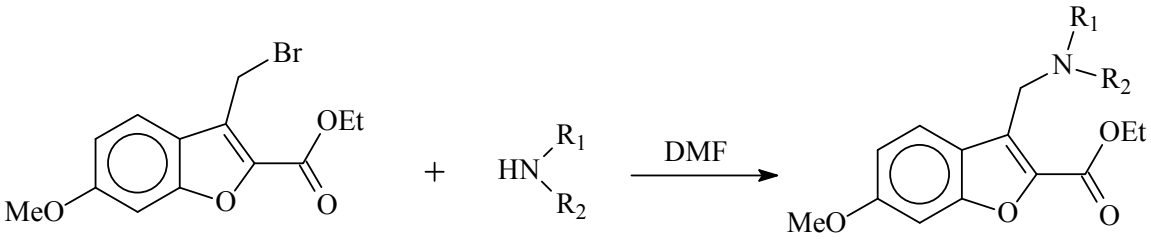


i) NBS, DBP, CCl4, reflux 10 h, 74 % (17 %) (57 %)

Scheme 2. *Reagents and conditions*: (i) NBS, DBP, CCl_4, reflux 10 h, 74%.

Reaction of **3** with amines and amino acid ethyl ester hydrochlorides (cyclopropylamine (**4a**), L-alanine (**4b**), L-leucine (**4c**), S-proline (**4d**), pyrrolidine (**4e**), piperidine (**4f**), morpholine (**4g**), thiomorpholine

(**4h**), and *N*-methylpiperazine (**4i**)) yielded the corresponding ethyl 3-[(alkylamino)methyl]-6-methoxy-1-benzofuran-2-carboxylates (**5a-i**) (Scheme 3). The prepared compounds were separated, purified, and their structures were confirmed by means of ^{1}H NMR, ^{13}C NMR, mass spectrometry, IR, and elemental analysis. Moreover, the newly synthesized heterocycles are expected to display some biological activities, due to the important functional subunits used in the synthesis, which were found in many naturally occurring compounds.[11]



(3) **(4a-f)** **(5a-f)**

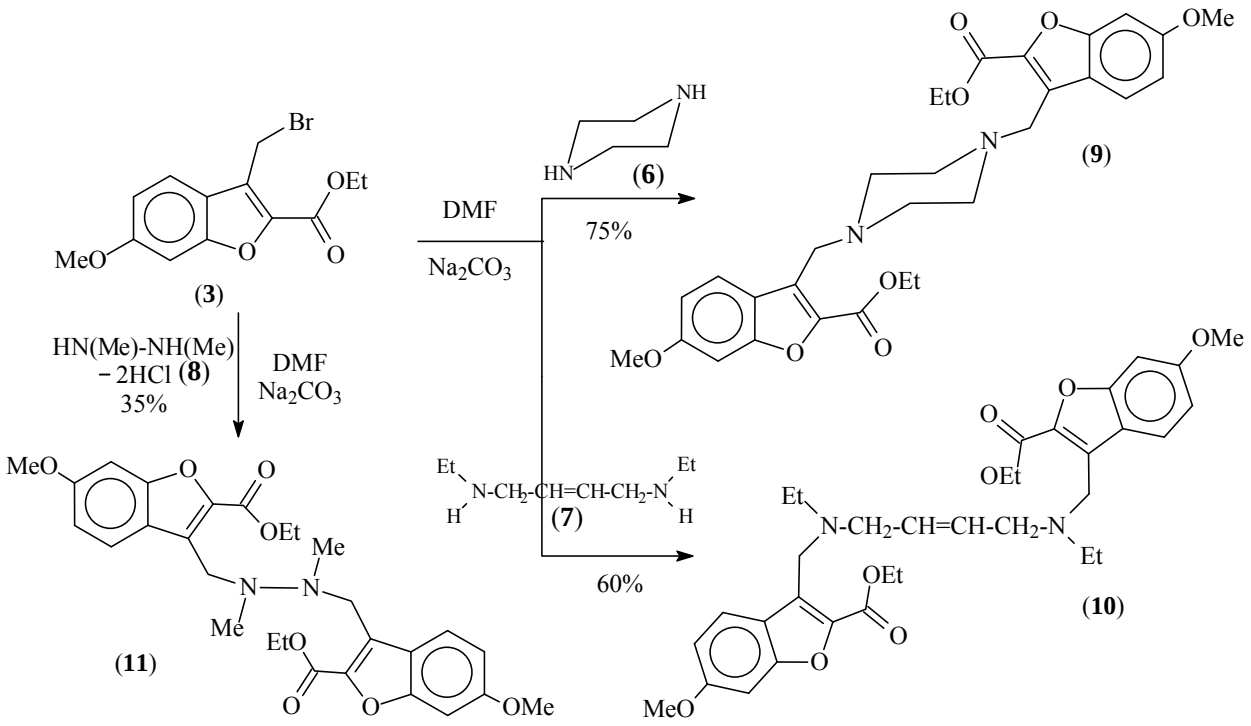
Scheme 3

Table 1. Yields percentages of the reaction of **3** with different amines

Amines	Products	Yield	Substituents
4a	**5a**	47 %	R_1 = H, R_2 = $CH(CH_2)_2$
4b	**5b**	51 %	R_1 = H, R_2 = $CH(CH_3)CO_2Et$
4c	**5c**	53 %	R_1 = H, R_2 = $CH[CH_2CH(CH_3)_2]CO_2Et$
4d	**5d**	68 %	R_1, R_2 = $CH(CO_2Et)(CH_2)_3$
4e	**5e**	72 %	R_1, R_2 = $(CH_2)_4$
4f	**5f**	86 %	R_1, R_2 = $(CH_2)_5$
4g	**5g**	81 %	R_1, R_2 = $[(CH_2)_2]_2O$
4h	**5h**	87 %	R_1, R_2 = $[(CH_2)_2]_2S$
4i	**5i**	83 %	R_1, R_2 = $[(CH_2)_2]_2N$-CH_3

In case of the primary amines and amino acid ethyl ester hydrochlorides (these were synthesized by a standard procedure from amino acids *via* the acid chloride),[12] the products were obtained in moderate yield. However, high yields were obtained with secondary amines.

Additionally, some known naturally occurring bis(coumarinyl)ethers were reported to display some biological activities; daphnoretin for example was found to have antinocoplastic activity and inhibition of DNA-producing enzymes or proteins and nucleic acid synthesis *in vivo*.[13,14] This has attracted our attention towards the synthesis of dimers of substituted coumarins[9] and the dimeric analogues of benzofurans. When compound **3** was treated with the diamines, piperazine (**6**), *N*,*N*'-diethylbut-2-ene-1,4-diamine (**7**), and 1,2-dimethylhydrazine dihydrochloride (**8**), 1,4-bis[(ethyl 6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]piperazine (**9**), *N*,*N*'-diethyl-*N*,*N*'-bis[(6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]but-2-ene-1,4-diamine **(10)** and 1,2-bis[(ethyl 6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]-1,2-dimethylhydrazine **(11)**, were produced, respectively (Scheme 4).



Scheme 4

The ^{1}H-NMR and ^{13}C-NMR spectra of all prepared compounds are in total agreement with the suggested structures. DEPT experiments were employed to differentiate secondary and quaternary carbons from primary and tertiary carbons. Additional support of the proposed structures comes from mass spectral data; mass spectra of the prepared compounds showed the correct molecular ions, ($M^{+\cdot}$), as suggested by their molecular formulas. Analyses of the molecular ions and the fragmentation pattern are used in the identification and characterization of these compounds.

In vitro *anti-HIV assay*

Compounds **3**, **5d**, **5e**, **5f, 5g, 5h, 5i, 9, 10, and 11** were tested for their *in vitro* anti-HIV-1 (strain III_B) and HIV-2 (strain ROD) activity in human T-lymphocyte (MT-4) cells using the MT-4/MTT assay.[15] The results are summarized in Table 1. The antiviral activity was compared with that of known and approved antiviral drugs efavirenz[16] and capravirine.[17] The gp41 subunit of the human immunodeficiency virus type 1 (HIV-1) envelope glycoprotein plays an important role in HIV-1 entry and serves as an attractive target for the development of HIV-1 entry inhibitors. A new class of anti-HIV drugs,[18] triggered by gp120 binding to CD4 and a coreceptor, gp41 undergoes a conformation shift from a native prefusogenic state to a fusogenic state, in which the N-terminal heptad repeat (NHR) and C-terminal heptad repeat (CHR) associate to form a six-helix bundle, representing the fusion-active gp41 core. Any compound that disrupts the gp41 sixhelix bundle formation may inhibit the gp41-mediated membrane fusion, thereby blocking HIV-1 entry into target cells.[19] Our target was the distruption of the gp41 sixhelix bundle

formation by the newly synthesized compounds, leading to inhibition of HIV. None of the *in vitro* tested compounds **5d**, **5e**, **5f, 5g, 5h, 5i, 9, 10,** and **11** were found to inhibit HIV-replication, at EC_{50} lower than the CC_{50} compared to the antiviral agents efavirenz (EFV) and azidothymidine capravirine.[17] However **3** was found to be the only compound in the series to inhibit HIV-1 and HIV-2 replication in cell culture with EC_{50} = >6.60 μg/ml at non-toxic concentration, with no selectivity witnessed. Results are displayed in Table 2.

Table 2. *In vitro* anti-HIV-1[a] and HIV-2[b] of some new benzofuran derivative

Compound	Virus strai	EC_{50} (μg/ml)[c]	CC_{50} (μg/ml)[d]	SI[e]
3	III_B	>6.60	6.60 ± 5.59	<1
	ROD	>6.60	6.60 ± 5.59	<1
5d	III_B	>66.88	66.88 ± 10.49	<1
	ROD	>66.88	66.88 ± 10.49	<1
5f	III_B	>63.20	63.20 ± 12.77	<1
	ROD	>63.20	63.20 ± 12.77	<1
5g	III_B	>83.38	83.83 ± 7.62	<1
	ROD	>83.38	83.83 ± 7.62	<1
5h	III_B	>97.17	97.17 ± 11.12	<1
	ROD	>97.17	97.17 ± 11.12	<1
5i	III_B	>73.58	73.58 ± 7.94	<1
	ROD	>73.58	73.58 ± 7.94	<1
5e	III_B	>83.15	83.15± 13.70	<1
	ROD	>83.15	83.15± 13.70	<1
10	III_B	>87.0	≥87.0	≤1
	ROD	>87.0	≥87.0	≤1
9	III_B	>70.95	70.95± 26.88	<1
	ROD	>70.95	70.95± 26.88	<1
11	III_B	≥93.70	≥93.70	≤1
	ROD	≥93.70	≥93.70	≤1
Efavirenz	III_B	0.003	40	13,333
Capravirine	III_B	0.0014	11	7,857

[a] Anti-HIV-1 activity measured with strain III_B. [b] Anti-HIV-2 activity measured with strain ROD. [c] Compound concentration required to achieve 50% protection of MT-4 cells from the HIV-1- and 2-induced cytopathogenic effect. [d] Compound concentration that reduces the viability of mock-infected MT-4 cells by 50%. [e] SI: Selectivity index (CC_{50}/EC_{50})

CONCLUSION

The structure activity relationship showed that the substituted benzofuran ring by haloalkyl groups having more HIV inhibition than the corresponding nitrogen analogues. This result might lead us to optimize the anti-HIV activity through the modification of our new target molecules by introducing various potent substituted alkyl halide groups.

EXPERIMENTAL

General consideration

Melting points were measured on an electrothermal melting point apparatus and are uncorrected. The IR spectra were acquired, as KBr discs, with the aid of a Thermo Nicolet Nexus 670 FT-IR instrument. The ^{1}H-NMR and ^{13}C-NMR spectra were recorded with a Bruker DPX-300 spectrometers and are reported in ppm (δ) relative to tetramethylsilane as internal reference and deuterated chloroform as a solvent. EIMS spectra were obtained using a Finnegan MAT TSQ-70 spectrometer at 70 eV. ESI-MS spectra were acquired with an AP15000 LC-MS instrument from Applied Biosystems. Elemental analyses were acquired with the aid of Eurovector Euro EA3000, CHNS-O elemental analyzer. TLC was carried out using pre-coated silica gel (E. Merck Kiesegel 60 F_{254} layer thickness 0.25 mm).

Ethyl 6-methoxy-3-methyl-1-benzofuran-2-carboxylate (1)

A mixture of of 6-methoxy-3-methylcoumarlic acid[9] (2.06 g, 10.0 mmol), anhyd. K_2CO_3 (2.07 g, 15.0 mmol) and EtBr (2.5 mL, 30.0 mmol) in dry acetone (100 mL) was refluxed for 12 h. Acetone was removed under vacuum and the solid product was treated with 100 mL of water to remove the excess of potassium carbonate and the formed potassium bromide. The product was filtered, washed, and recrystallized from aqueous EtOH. Yield of **1** = 1.64 g (70%); mp 71-72 ^{o}C (lit.,[15] 74-75 ^{o}C); IR (cm^{-1}): 1705 (C=O), 1624 (C=C); ^{1}H-NMR: δ 1.39 (t, 3H, J = 7.0 Hz), 2.50 (s, 3H), 3.81 (s, 3H), 4.39 (q, 2H, J = 7.0 Hz), 6.88 (dd, 1H, J = 2, 9 Hz), 6.97 (d, 1H, J = 2.0 Hz), 7.42 (d, 1H, J = 9.0 Hz); ^{13}C-NMR: δ 9.5, 14.5, 55.7, 60.9, 95.5, 113.2, 121.4, 122.5, 126.2, 140.3, 155.7, 160.6, 160.7; MS ($C_{13}H_{14}O_4$), *m*/*z* (% rel. int.): 234 ($M^{+\cdot}$, 100), 190 (100), 147 (26), 118 (23), 88 (34).

Bromination of ethyl 6-methoxy-3-methyl-1-benzofuran-2-carboxylate

A mixture of **1** (2.34 g, 10 mmol), NBS (2.31 g, 13.0 mmol) and dibenzoyl peroxide (DBP) (0.01 g) in CCl_4 (75 mL) was refluxed for 12 h.[20] The hot mixture was filtered, concentrated under vacuum and was chromatographed on silica gel plates using CH_2Cl_2/hexane (2:1) as eluent. The earlier fractions afforded ethyl 3-(dibromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (**2**) as the minor product, while the later fractions gave ethyl 3-(bromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (**3**) as the major product. The products were pure enough to be used in subsequent steps without further purification.

Ethyl 3-(dibromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (2)

Yield of **2** = 0.67 g (17%), mp 132-133 °C; IR (cm^{-1}): 1703 (C=O), 1590 (C=C); ^{1}H-NMR: δ 1.23 (s, 1H), 1.43 (t, 3H, *J* = 7.0 Hz), 3.85 (s, 3H), 4.45 (q, 2H, *J* = 7.0 Hz), 7.04 (dd, 1H, *J* = 2.0, 9.0 Hz), 7.72 (s, 1H), 8.08 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.4, 28.6, 55.8, 61.9, 95.7, 114.1, 118.9, 123.4, 129.0, 135.4, 156.3, 159.4, 161.2; MS ($C_{13}H_{12}Br_2O_4$), *m*/*z* (% rel. int.): 392 ($M^{+\cdot}$, 7), 390 ($M^{+\cdot}$, 12), 388 ($M^{+\cdot}$, 6), 312 (48), 202 (100). Anal. Calcd for $C_{13}H_{12}O_4Br_2$: C, 39.83; H, 3.09. Found: C, 40.13; H, 3.37.

Ethyl 3-(bromomethyl)-6-methoxy-1-benzofuran-2-carboxylate (3)

Yield of **3** = 1.78 g (57%), mp 97-98 °C; IR (cm^{-1}): 1705 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.40 (t, 3H, *J* = 7.0 Hz), 3.80 (s, 3H), 4.42 (q, 2H, *J* = 7.0 Hz), 4.92 (s, 2H), 6.93 (dd, 1H, *J* = 2.0, 9.0 Hz), 6.98 (d, 1H, *J* = 2.0 Hz), 7.58 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.4, 20.8, 55.7, 61.5, 95.7, 114.0, 120.0, 121.6, 125.5, 140.4, 155.9, 159.6, 161.1; MS ($C_{13}H_{13}BrO_4$), *m*/*z* (% rel. int.): 313 ($M^{+\cdot}$, 37), 311 ($M^{+\cdot}$, 35), 232 (85), 204 (100), 189 (13). Anal. Calcd for $C_{13}H_{13}O_4Br$: C, 49.86; H, 4.18. Found: C, 50.02; H, 3.91.

Ethyl 3-[(alkylamino)methyl]-6-methoxy-1-benzofuran-2-carboxylates (5a-f), general procedure

By a modified procedure,[21] the appropriate amine or amino acid ethyl ester hydrochloride[22] (**4a-i**) (1.5 mmol) was added to a stirred solution of **3** (172 mg, 0.55 mmol) in dry DMF (3 mL). After stirring for 30 min, 25 mL of water (25 mL) and of 5% aqueous $NaHCO_3$ (5 mL) were added, followed by extraction with Et_2O (50 mL x 3). The combined organic extract was washed with water (50 mL x 2), dried, and the solvent was removed under vacuum. The crude product was purified by TLC plates with EtOAc: hexane (1:1.5) as eluent and was recrystallized from hexane-EtOH.

Presented below are the yields, physical states, and the spectral properties of the different ethyl 3-(*N*-aminomethyl)-6-methoxycoumarlates prepared using this method.

Ethyl 3-(cyclopropylaminomethyl)-6-methoxy-1-benzofuran-2-carboxylate (5a)

Yield = 75 mg (47%), oily; IR (cm^{-1}): 1703 (C=O), 1624 (C=C); ^{1}H-NMR ($CDCl_3$): δ 0.41 (d, 4H, *J* = 5.0 Hz), 1.42 (t, 3H, *J* = 7.0 Hz), 2.09 (m, 1H, *J* = 5.0 Hz), 2.23 (br s, 1H), 3.83 (s, 3H), 4.20 (s, 2H), 4.43 (q, 2H, *J* = 7.0 Hz), 6.92 (dd, 1H, *J* = 2.0, 9.0 Hz), 7.01 (d, 1H, *J* = 2.0 Hz), 7.62 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR ($CDCl_3$): δ 6.6, 14.5, 30.0, 42.2, 55.7, 61.3, 95.6, 113.7, 121.8, 121.9, 128.8, 140.8, 155.8, 160.2, 160.8; MS ($C_{16}H_{19}NO_4$), *m*/*z*: 290 $[M+1]^+$. Anal. Calcd for $C_{16}H_{19}NO_4$: C, 66.42; H, 6.62; N, 4.84. Found: C, 66.39; H, 6.60; N, 4.79.

Ethyl *N*-[(6-methoxy-2-carbethoxy-1-benzofuran-3-yl)methyl]alaninate (5b)

Yield = 98 mg (51%), oily; IR (cm^{-1}): 1703 (C=O), 1624 (C=C); ^{1}H-NMR ($CDCl_3$): δ 1.21 (m, 3H), 1.30 (d, 3H, *J* = 7.0 Hz), 1.40 (t, 3H, *J* = 2 Hz), 2.36 (br s, 1H), 3.38 (q, 1H, *J* = 7.0 Hz), 3.82 (s, 3H), 4.11 (q, 2H, *J* = 7.0 Hz), 4.18 (s, 2H), 4.41 (q, 2H, *J* = 7.0 Hz), 6.91 (dd, 1H, *J* = 3.0, 9.0 Hz), 6.99 (d, 1H, *J* = 3.0

Hz), 7.68 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR ($CDCl_3$): δ 14.2, 14.4, 19.1, 41.2, 55.7, 56.2, 60.8, 61.2, 95.6, 113.7, 121.4, 122.2, 127.7, 140.9, 155.9, 160.1, 160.8, 175.3; MS ($C_{18}H_{23}NO_6$), *m*/*z* (% rel. int.): 348 [M-1]$^{+\cdot}$ (10), 275 (72), 232 (85), 204 (100), 189 (17). Anal. Calcd for $C_{18}H_{23}NO_6$: C, 61.88; H, 6.64; N, 4.01. Found: C, 62.20; H, 6.15; N, 4.32.

Ethyl *N*-[(6-methoxy-2-carbethoxy-1-benzofuran-3-yl)methyl]leucinate (5c)

Yield = 114 mg (53%), oily; IR (cm^{-1}): 1705 (C=O), 1615 (C=C); ^{1}H-NMR ($CDCl_3$): δ 0.78 (d, 3H, *J* = 7.0 Hz), 0.86 (d, 3H, *J* = 7.0 Hz), 1.19-1.46 (m, 8H), 1.69 (sept, 1H, *J* = 7.0 Hz), 2.04 (br s, 1H), 3.2 (t, 1H, *J* = 7.0 Hz), 3.82 (s, 3H), 4.06-4.16 (m, 4H), 4.41 (q, 2H, *J* = 7.0 Hz), 6.91 (dd, 1H, *J* = 2.0, 9.0 Hz), 6.99 (d, 1H, *J* = 2.0 Hz), 7.68 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR ($CDCl_3$): δ 14.2, 14.4, 22.1, 22.4, 24.9, 41.6, 42.8, 55.7, 59.5, 60.6, 61.2, 95.5, 113.6, 121.4, 122.4, 127.9, 140.9, 155.9, 160.1, 160.7, 175.8; MS ($C_{18}H_{23}NO_6$), *m*/*z* (% rel. int.): 390 [M-1]$^{+\cdot}$ (5), 317 (100), 232 (53), 204 (86), 189 (11). Anal. Calcd for $C_{21}H_{29}NO_6$: C, 64.43; H, 7.47; N, 3.58. Found: C, 65.12; H, 7.73; N, 3.18.

Ethyl 1-{[2-(ethoxycarbonyl)-6-methoxy-1-benzofuran-3-yl]methyl}prolinate (5d)

Yield = 140 mg (68%), oily; IR (cm^{-1}): 1705 (C=O), 1615 (C=C); ^{1}H-NMR ($CDCl_3$): δ 1.23-1.43 (m, 6H), 1.70-3.33 (m, 7H), 3.83 (s, 3H), 4.15 (q, 2H, *J* = 7.0 Hz), 4.26 (s, 2H), 4.41 (q, 2H, *J* = 7.0 Hz), 6.91 (dd, 1H, *J* = 2.0, 9.0 Hz), 6.99 (d, 1H, *J* = 2.0 Hz), 7.95 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR ($CDCl_3$): δ 14.3, 14.4, 23.1, 29.5, 47.4, 53.3, 55.7, 60.6, 61.1, 65.3, 95.2, 113.6, 122.0, 123.7, 124.0, 140.7, 155.8, 160.3, 160.7, 174.3; MS ($C_{20}H_{25}NO_6$), *m*/*z* (% rel. int.): 375 ($M^{+\cdot}$, 5), 301 (100), 232 (45), 204 (74), 189 (13). Anal. Calcd for $C_{20}H_{25}NO_6$: C, 63.99; H, 6.71; N, 3.73. Found: C, 63.61; H, 6.41; N, 3.41.

Ethyl 6-methoxy-3-(pyrrolidin-1-ylmethyl)-1-benzofuran-2-carboxylate (5e)

Yield = 120 mg (72%), oily; IR (cm^{-1}): 1703 (C=O), 1590 (C=C); ^{1}H-NMR: δ 1.40 (t, 3H, *J* = 7.0 Hz), 1.74 (quint, 4H, *J* = 3.0 Hz), 2.56 (t, 4H, *J* = 7.0 Hz), 3.82 (s, 3H), 4.12 (s, 2H), 4.40 (q, 2H, *J* = 7.0 Hz), 6.88 (dd, 1H, *J* = 2.0, 9.0 Hz), 6.99 (d, 1H, *J* = 2.0 Hz), 7.78 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.5, 23.6, 49.0, 54.2, 55.7, 61.1, 95.3, 113.5, 122.0, 123.3, 127.4, 140.7, 155.8, 160.3, 160.6; ESI-MS ($C_{17}H_{21}NO_4$), *m*/*z* 304 [M+1]$^+$. Anal. Calcd for $C_{17}H_{21}NO_4$: C, 67.31; H, 6.98; N, 4.62. Found: C, 67.28; H, 7.05; N, 4.59.

Ethyl 6-methoxy-3-(piperidin-1-ylmethyl)-1-benzofuran-2-carboxylate (5f)

Yield = 150 mg (86%), oily; IR (cm^{-1}): 1705 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.40 (m, 5H), 1.56 (quint, 4H, *J* = 5.0 Hz), 2.47 (br s, 4H), 3.84 (s, 3H), 4.01 (s, 2H), 4.41 (q, 2H, *J* = 7.0 Hz), 6.90 (dd, 1H, *J* = 2.0, 9.0 Hz), 7.00 (d, 1H, *J* = 2.0 Hz), 7.90 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.5, 24.1, 26.6, 52.8, 54.6, 55.7, 61.1, 95.3, 113.4, 122.0, 123.9, 124.5, 141.0, 155.9, 160.4, 160.6; ESI-MS ($C_{18}H_{23}NO_4$), *m*/*z* 318 [M+1]$^+$. Anal. Calcd for $C_{18}H_{23}NO_4$: C, 68.12; H, 7.30; N, 4.41. Found: C, 68.10; H, 7.37; N, 4.35.

Ethyl 6-methoxy-3-(morpholin-4-ylmethyl)-1-benzofuran-2-carboxylate (5g)

Yield = 142 mg (81%), mp 106-107 °C; IR (cm^{-1}): 1692 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.41 (t, 3H, *J* = 7.0 Hz), 2.50 (t, 4H, *J* = 5.0 Hz), 3.68 (t, 4H, *J* = 5.0 Hz), 3.84 (s, 3H), 4.01 (s, 2H), 4.41 (q, 2H, *J* = 7.0 Hz), 6.90 (dd, 1H, *J* = 2.0, 9.0 Hz), 7.00 (d, 1H, *J* = 2.0 Hz), 7.85 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.5, 52.7, 53.8, 55.7, 61.2, 67.1, 95.4, 113.6, 121.7, 123.5, 126.4, 141.1, 155.9, 160.3, 160.7; ESI-MS ($C_{17}H_{21}NO_5$), *m*/*z* 320 $[M+1]^+$. Anal. Calcd for $C_{17}H_{21}NO_5$: C, 63.94; H, 6.63; N, 4.39. Found: C, 63.90; H, 6.58; N, 4.43.

Ethyl 6-methoxy-3-(piperazin-1-ylmethyl)-1-benzofuran-2-carboxylate (5h)

Yield = 154 mg (84%), mp 103 °C; IR (cm^{-1}): 1703 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.40 (t, 3H, *J* = 7.0 Hz), 2.63 (t, 4H, *J* = 5.0 Hz), 2.76 (t, 4H, *J* = 5.0 Hz), 3.83 (s, 3H), 4.01 (s, 2H), 4.40 (q, 2H, *J* = 7.0 Hz), 6.89 (dd, 1H, *J* = 2.0, 9.0 Hz), 7.99 (d, 1H, *J* = 2.0 Hz), 7.82 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ: 14.5, 28.1, 53.2, 55.1, 55.7, 61.2, 95.4, 113.5, 121.7, 123.6, 126.7, 141.1, 155.9, 160.3, 160.7; ESI-MS ($C_{17}H_{21}NO_4S$), *m*/*z* 336 $[M]^+$. Anal. Calcd for $C_{17}H_{21}NO_4S$: C, 60.88; H, 6.31; N, 4.18; S, 9.56. Found: C, 61.01; H, 5.98; N, 4.28; S, 9.59.

Ethyl 6-methoxy-3-[(4-methylpiperazin-1-yl)methyl]-1-benzofuran-2-carboxylate (5i)

Yield = 152 mg (83%), mp 66-67 °C; IR (cm^{-1}): 1730 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.38 (t, 3H, *J* = 7.0 Hz), 2.25 (s, 3H), 2.47 (br s, 8H), 3.81 (s, 3H), 3.99 (s, 2H), 4.38 (q, 2H, *J* = 7.0 Hz), 6.87 (dd, 1H, *J* = 2.0, 9.0 Hz), 6.97 (d, 1H, *J* = 2.0 Hz), 7.82 (d, 1H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.4, 46.0, 52.3, 53.1, 55.2, 55.7, 61.1, 95.3, 113.5, 121.8, 123.6, 126.7, 141.0, 155.9, 160.3, 160.6; ESI-MS ($C_{18}H_{24}N_2O_4$), *m*/*z* 333 $[M+]^+$. Anal. Calcd for $C_{18}H_{24}N_2O_4$: C, 65.04; H, 7.28; N, 8.43. Found: C, 65.31; H, 7.18; N, 8.81.

***N,N`*-Di[ethyl 6-methoxy-3-coumarlatemethyl]-1,4-diamines (9-11), general procedure**

A mixture of **3** (235 mg, 0.75 mmol), K_2CO_3 (100 mg, 0.90 mmol) and the appropriate diamine: piperazine (32 mg), *N,N`*-diethyl-2-butene-1,4-diamine (53 mg) and *N,N`*-dimethylhydrazine dihydrochloride (99 mg) in DMF (5 mL) was stirred at rt and left overnight. Water (40 mL) was added and the product was extracted with Et_2O (50 mL x 5). The combined organic extracts were washed with water (50 mL x 2), dried and the solvent was removed under vacuum. The crude was chromatographed on TLC plates with EtOAc : hexane (2 : 1) as eluent and was then recrystallized from EtOH.

1,4-Bis[(ethyl 6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]piperazine (9)

Yield = 155 mg (75%), mp 188 °C; IR (cm^{-1}): 1711 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.40 (t, 6H, *J* = 7.0 Hz), 2.53 (br s, 8H), 3.83 (s, 6H), 4.01 (s, 4H), 4.40 (q, 4H, *J* = 7.0 Hz), 6.86 (dd, 2H, *J* = 2.0, 9.0 Hz), 6.99 (d, 2H, *J* = 2.0 Hz), 7.84 (d, 2H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.5, 52.3, 53.4, 55.7, 61.1, 95.3, 113.5, 121.8, 123.7, 126.9, 141.0, 155.9, 160.3, 160.6; MS ($C_{30}H_{34}N_2O_8$), *m*/*z* (% rel. int.): 550 $[M]^+$ (10), 275

(72), 232 (85), 204 (100), 189 (17). Anal. Calcd for $C_{30}H_{34}N_2O_8$: C, 65.44; H, 6.22; N, 5.09. Found: C, 65.28; H, 5.93; N, 5.13.

_N,N'_-Diethyl-_N,N'_-bis[(6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]but-2-ene-1,4-diamine (10)

Yield = 136 mg (60%), mp 90-91 ^oC; IR (cm^{-1}): 1705 (C=O), 1589 (C=C); ^{1}H-NMR: δ 1.01 (t, 6H, *J* = 7.0 Hz), 1.39 (t, 6H, *J* = 7.0 Hz), 2.49 (q, 4H, *J* = 7.0 Hz), 3.07 (d, 4H, *J* = 4.0 Hz), 3.82 (s, 6H), 4.01 (s, 4H), 4.38 (q, 4H, *J* = 7.0 Hz), 5.66 (br s, 2H), 6.84 (dd, 2H, *J* = 2.0, 9.0 Hz), 6.97 (d, 2H, *J* = 2.0 Hz), 7.87 (d, 2H, *J* = 9.0 Hz); ^{13}C-NMR: δ 11.8, 14.5, 47.6, 47.9, 55.6, 55.7, 61.1, 95.2, 113.2, 121.9, 124.0, 128.8, 130.8, 140.8, 155.9, 160.4, 160.6; ESI-MS ($C_{34}H_{42}N_2O_8$), 607 $[M+1]^+$. Anal. Calcd for $C_{34}H_{42}N_2O_8$: C, 67.31; H, 6.98; N, 4.62. Found: C, 67.69; H, 7.13; N, 4.88.

1,2-Bis[(ethyl-6-methoxy-1-benzofuran-3-yl-2-carboxylate)methyl]-1,2-dimethylhydrazine (11)

Yield = 155 mg (35%), oily; IR (cm^{-1}): 1703 (C=O), 1590 (C=C); ^{1}H-NMR: δ 1.41 (t, 6H, *J* = 7.0 Hz), 2.43 (s, 6H), 3.83 (s, 6H), 4.21 (s, 4H), 4.40 (q, 4H, *J* = 7.0 Hz), 6.70 (dd, 2H, *J* = 2.0, 9.0 Hz), 6.90 (d, 2H, *J* = 2.0 Hz), 7.27 (d, 2H, *J* = 9.0 Hz); ^{13}C-NMR: δ 14.4, 34.0, 48.6, 55.6, 61.0, 95.0, 113.0, 121.8, 123.0, 127.9, 140.7, 155.6, 160.3, 160.4; ESI-MS ($C_{28}H_{32}N_2O_8$), *m/z* 525 $[M+1]^+$. Anal. Calcd for $C_{28}H_{32}N_2O_8$: C, 64.11; H, 6.15; N, 5.34. Found: C, 63.92; H, 6.22; N, 5.30.

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20. The reaction was followed by TLC and small quantities of NBS (~ 0.1 g) were added if the starting material was still in the reaction mixture.
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