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Synthesis and biological evaluation of peptidyl organotrifluoroborates and their corresponding boron analogs

Chia-Hua Tsai · Chia-Hung Lin · Ching-Tien Hsieh · Chih-Cheng Cai · Ting-Ju Lin · Pin-Yi Liu · Meng-Hsuan Lin · Meng-Ju Wu · Chia-Chieh Fu · Yang-Chang Wu · Fang-Rong Chang · Po-Shen Pan

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Abstract Six peptidyl organotrifluoroborates and their corresponding boronate esters and/or boronic acid analogs were designed and synthesized. Their anti-proliferative activity against hepatocellular carcinoma cells (HepG2) and human metastatic breast cancer cells (MDA-MB231) were evaluated by use of an MTT assay. Potassium {4-[(3S,6S,9S)-3,6-dibenzyl-9-isopropyl-4,7,10-trioxo-11-oxa-2,5,8-triazadodecyl]phenyl}trifluoroborate (**B6**) was potent ($IC_{50} = 29.9$ μM) against MDA-MB231, and {4-[(3S,6S,9S)-6-benzyl-3-((benzyloxy)methyl)-9-isopropyl-4,7,10-trioxo-11-oxa-2,5,8-triazadodecyl]phenyl}boronic acid (**B9**) and Potassium {4-[(3S,6S,9S)-6-benzyl-3-((benzyloxy)methyl)-9-isopropyl-4,7,10-trioxo-11-oxa-2,5,8-triazadodecyl]phenyl}trifluoroborate (**B10**) had broad anti-proliferative activity against HepG2 ($IC_{50} = 24.7$ and 21.8 μM, respectively) and MDA-MB231 ($IC_{50} = 24.5$ and 18.9 μM, respectively).

Keywords Peptide · Boronic acid · Organotrifluoroborate · Potassium trifluoroborate · HepG2 · MDA-MB231

C.-H. Tsai · C.-H. Lin · C.-T. Hsieh · C.-C. Cai · T.-J. Lin · P.-Y. Liu · M.-H. Lin · M.-J. Wu · C.-C. Fu · P.-S. Pan (✉)
Department of Chemistry, Tamkang University, No. 151 Yingzhuan Rd., Tamsui Dist., New Taipei City 25137, Taiwan, ROC
e-mail: popan@mail.tku.edu.tw

Y.-C. Wu · F.-R. Chang
Graduate Institute of Natural Products, Kaohsiung Medical University, 100 Shi-Chuan 1st Rd., Kaohsiung 80708, Taiwan, ROC

Y.-C. Wu
Graduate Institute of Integrated Medicine, College of Chinese Medicine, China Medical University, Taichung 404, Taiwan, ROC

F.-R. Chang
Cancer Center, Kaohsiung Medical University Hospital, Kaohsiung 807, Taiwan, ROC

Introduction

Among boron-containing compounds, organotrifluoroborates are seldom considered in biological applications because of their lack of an empty *p*-orbital. Although the value of organofluoroborates in organic synthesis is well recognized [1], studies of their biologic applications are limited [2, 3]. Herein, we report the synthesis of two series of peptidyl organotrifluoroborates and their corresponding boronate esters and/or boronic acids, and their anti-proliferative activity against hepatocellular carcinoma cells (HepG2) and human metastatic breast cancer cells (MDA-MB231), determined by use of MTT assays [4].

Experimental

General procedure for conversion of potassium organotrifluoroborate to boronic acid [5]

Potassium organotrifluoroborate (1.0 equivalent) and silica gel (1.0 equivalent) were weighed into a 20-ml vial, and H_2O–ethyl acetate (1:1) was added. The reaction was stirred at room temperature for 3–6 h. The reaction mixture was filtered to remove silica gel, and the filter cake was thoroughly washed with ethyl acetate–acetone (9:1). The aqueous and organic layers were separated, the aqueous layer was extracted with ethyl acetate, and the combined organic layers were dried, filtered, and concentrated in vacuo. The crude product was then purified by reversed-phase HPLC to afford the pure desired product.

General procedure for conversion of potassium organotrifluoroborate to boronate ester [5]

Potassium organotrifluoroborate (1.0 equivalent), pinacol (1.1 equivalents), and silica gel (1.0 equivalent) were weighed into a 20-ml vial and H_2O–ethyl acetate (1:1) was added in one portion. The reaction was stirred at room temperature for 3–5 h. The reaction mixture was then filtered, and the filter cake was thoroughly rinsed with ethyl acetate–acetone (9:1). The aqueous and organic layers were separated, the aqueous layer was extracted with ethyl acetate, and the combined organic layers were dried, filtered, and concentrated in vacuo. The crude ester analogs were purified by flash column chromatography to afford the desire product.

General procedure for conversion of boronate ester to potassium organotrifluoroborate

Boronate ester (1.0 equivalent) was added 6.0 equivalents of KHF_2 in H_2O–methanol (1:1), and the mixture was stirred at room temperature for 3 h. The solvent was then removed in vacuo and the crude solid was obtained by Soxhlet extraction (3 h) with acetone. The collected solution was concentrated and the desired product was precipitated by addition of 10 mL of acetone–hexane (1:3).

General procedure for synthesis of series **A** analogs

Tripeptide (1.1 equivalent) and 4-carboxyphenylboronic acid pinacol ester (**3**, 1.0 equivalent) were weighed into a dry flask with DIPEA (6.0 equivalents) and TBTU (1.1 equivalents). Anhydrous dichloromethane was added to generate a 0.1 M solution. On completion of the reaction the crude material was washed with aqueous HCl (pH 1) and saturated sodium bicarbonate. The resulting organic solutions were collected, dried by $MgSO_4$, filtered, and concentrated to yield the crude product. This crude product was then purified by a short flash column chromatography to afford the desire product.

General procedure for synthesis of series **B** analogs

To a vial containing potassium 4-formylphenyltrifluoroborate (**4**, 1.0 equivalent) in MeOH, tripeptide (1.2 equivalents) was added to generate 0.5 M solution. The reaction mixture was stirred for 3 h at room temperature. 5-Ethyl-2-methylpyridine borane (PEMB, 1.0 equivalent) was then added, and stirring was continued for 3–5 h. The solvent was removed in vacuo, and the resulting crude material was washed with hexane. The crude solid was obtained by Soxhlet extraction (3 h) with acetone. The collected solvent was concentrated and the desired pure product was then precipitated by addition of 10 mL of acetone–hexane (1:3).

*Compound **A1***

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.83 (s, 4H), 7.74 (br, 1H), 7.51 (br, 1H), 7.45 (br, 1H), 7.07–6.91 (m, 5H), 5.00 (q, $J = 8.0$ Hz, 1H), 4.70 (t, $J = 8.7$ Hz, 1H), 4.44 (dd, $J = 8.8$, 5.9 Hz, 1H), 3.66 (s, 3H), 3.03–2.88 (m, 2H), 2.17–1.99 (m, 2H), 1.34 (s, 12H), 0.90 (dd, $J = 13.8$, 6.7 Hz, 6H), 0.78 (dd, $J = 12.9$, 6.8 Hz, 6H). ^{13}C NMR (75.5 MHz, $CDCl_3$) δ 172.1, 171.8, 171.6, 167.6, 136.6, 134.7, 129.5, 128.3, 127.1, 126.6, 84.1, 59.1, 57.4, 54.3, 52.1, 38.9, 31.6, 31.2, 25.0, 19.3, 19.2, 19.0, 18.2. ^{11}B NMR (192.5 MHz, acetone-d_6) δ 29.4. M.p. 164 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 608.3499$, *m/z* found 608.3526.

*Compound **A2***

^{1}H NMR (300 MHz, CD_3OD) δ 7.77 (br, 4H), 7.22–7.03 (m, 5H), 4.77 (dd, $J = 8.4$, 6.3 Hz, 1H), 4.39 (d, $J = 5.7$ Hz, 1H), 4.30 (d, $J = 6.3$ Hz, 1H), 3.66 (s, 3H), 3.13–3.08 (m, 1H), 2.94–2.85 (m, 1H), 2.09 (oct, $J = 6.6$ Hz, 2H), 0.97–0.89 (m, 12H). ^{13}C NMR (75.5 MHz, CD_3OD) δ 173.6, 173.3, 170.3, 138.2, 135.0, 130.5, 129.5, 127.8, 127.6, 61.0, 59.3, 55.8, 52.6, 39.2, 32.1, 20.0, 19.6, 19.3, 18.7. ^{11}B NMR (192.5 MHz, CD_3OD) δ 28.8. M.p. 327 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 526.2717$, *m/z* found 526.2763.

*Compound **A3***

^{1}H NMR (300 MHz, CD_3OD) δ 7.62 (dd, $J = 11.6$, 8.0 Hz, 4H), 7.23–7.06 (m, 5H), 4.74 (dd, $J = 8.6$, 6.3 Hz, 1H), 4.31 (dd, $J = 13.9$, 8.4 Hz, 2H), 3.66 (s, 3H),

3.15–3.08 (m, 1H), 2.95–2.87 (m, 1H), 2.08 (oct, $J = 6.8$ Hz, 2H), 0.95 (d, $J = 6.7$ Hz, 3H), 0.90 (dd, $J = 6.8$, 2.6 Hz, 9H). ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 171.6, 171.3, 170.9, 167.2, 137.5, 131.0, 129.1, 127.9, 126.2, 125.4, 59.0, 57.4, 53.4, 51.6, 37.5, 30.2, 29.9, 19.2, 18.8, 18.7, 18.2. ^{11}B NMR (192.5 MHz, DMSO-d_6) δ 2.6. M.p. 275 °C. HRMS (ESI, negative ion) m/z calcd for $[M - K]^-$ = 548.2546, *m/z* found 548.2560.

Compound ***A4***

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.82 (d, $J = 8.1$ Hz, 2H), 7.63 (d, $J = 8.1$ Hz, 2H), 7.24–7.01 (m, 10H), 6.90 (br, 2H), 6.70 (br, 1H), 4.93 (q, $J = 6.9$ Hz,1H), 4.73 (q, $J = 9.0$ Hz, 1H), 4.44–4.39 (m, 1H), 3.68 (s,3H), 3.13 (d, $J = 6.9$ Hz, 2H), 2.99 (d, $J = 7.2$ Hz, 2H), 2.08–2.04 (m, 1H), 1.35 (s, 12H), 0.84 (dd, $J = 12.9$ Hz, 6H). ^{13}C NMR (75.5 MHz, $CDCl_3$) δ 172.0, 171.5, 171.0, 167.5, 136.9, 136.3, 134.9, 129.5, 128.7, 128.5, 127.1, 126.9, 126.7, 84.2, 57.6, 54.8, 54.7, 52.2, 38.8, 38.4, 31.3, 25.0, 19.0, 18.3. ^{11}B NMR (192.5 MHz, $CDCl_3$) δ 29.3. M.p. 137 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+$ = 656.3499, *m/z* found 656.3498.

Compound ***A5***

^{1}H NMR (300 MHz, D_2O) δ 7.91 (d, $J = 8.1$ Hz, 2H), 7.78 (d, $J = 7.8$ Hz, 2H), 7.38–7.24 (m, 10H), 4.86–4.78 (m, 2H), 4.34 (d, $J = 6.3$ Hz, 1H), 3,75 (s, 3H), 3.20–3.17 (m, 2H), 3.09–2.96 (m, 2H), 2.21–2.14 (m, 1H), 1.02–0.98 (m, 6H). ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 171.7, 171.4, 171.2, 166.3, 138.3, 137.5, 135.2, 134.1, 133.7, 129.2, 127.9, 126.7, 57.4, 54.7, 53.5, 51.6, 37.5, 36.9, 30.0, 18.9, 18.2. ^{11}B NMR (192.5 MHz, CD_3OD) δ 28.2. M.p. 267 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+$ = 574.2717, *m/z* found 574.2744.

Compound ***A6***

^{1}H NMR (300 MHz, DMSO-d_6) δ 8.29 (d, 1H), 8.21 (d, 1H), 7.54–7.51 (m, 2H), 7.36–7.33 (m, 4H), 7.26 (t, $J = 7.3$ Hz, 2H), 7.18–7.13 (m, 1H), 4.81–4.73 (m, 1H), 4.23 (dd, $J = 8.1$, 6.2 Hz, 1H), 3.64 (s, 3H), 3.11–2.96 (m, 2H), 2.07 (sext, $J = 6.7$ Hz, 1H), 0.90 (dd, $J = 8.6$, 6.9 Hz, 6H). ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 172.0, 171.8, 167.3, 138.3, 130.9, 130.8, 129.2, 128.0, 126.1, 125.3, 57.4, 54.4, 51.7, 36.8, 30.6, 30.0, 18.9, 18.1. ^{11}B NMR (192.5 MHz, CD_3OD) δ 3.8. M.p. 268 °C. HRMS (ESI, negative ion) *m/z* calcd for $[M - K]^-$ = 596.2546, *m/z* found 596.2546.

Compound ***A8***

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.77 (dd, $J = 48.8$, 8.1 Hz, 4H), 7.36–7.28 (m, 5H), 7.18–7.10 (m, 5H), 7.03–7.0 (m, 2H), 6.51 (d, 1H), 4.72–4.67 (m, 2H), 4.56 (d, $J = 4.0$ Hz, 2H), 4.44 (dd, $J = 8.5$, 5.3 Hz, 1H), 4.02–3.97 (m, 1H), 3.71 (s, 3H), 3.66–3.60 (m, 1H), 3.11–3.06 (m, 2H), 2.06 (oct, $J = 5.4$ Hz, 1H), 1.36 (s, 12H), 0.81 (dd, $J = 13.3$, 6.9 Hz, 6H). ^{13}C NMR (75.5 MHz, $CDCl_3$) δ 172.0, 170.5, 170.2, 167.5, 137.4, 136.3, 135.7, 135.2, 129.5, 128.9,128.8, 128.3, 128.2, 127.3,

126.4, 84.4, 73.8, 69.1, 57.7, 54.8, 53.2, 52.3, 37.7, 31.3, 25.1, 19.0, 18.1. ^{11}B NMR (192.5 MHz, Acetone-d_6) δ 31.0. M.p. 107 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 686.3605$, *m/z* found 686.3576.

Compound ***A9***

^{1}H NMR (300 MHz, CD_3OD) δ 8.21 (d, $J = 8.6$ Hz, 1H), 7.73 (br, 4H), 7.32–7.25 (m, 5H), 7.20–7.11 (m, 5H), 4.82–4.73 (m, 2H), 4.54 (s, 2H), 4.31–4.26 (m, 1H), 3.77 (d, $J = 5.7$ Hz, 1H), 3.67 (s, 3H), 3.13 (dd, $J = 13.9$, 6.0 Hz, 1H), 2.95 (dd, $J = 13.9$, 8.1 Hz, 1H), 2.07 (sext, $J = 6.7$ Hz, 1H), 0.88 (d, $J = 6.8$ Hz, 6H). ^{13}C NMR (75.5 MHz, CD_3OD) δ 173.4, 173.3, 172.0, 170.3, 139.3, 138.1, 136.4, 135.0, 130.8, 130.5, 129.5, 129.0, 128.9, 127.8, 127.7, 74.3, 70.7, 59.5, 55.9, 55.3, 55.2, 52.6, 39.0, 32.0, 19.5, 18.8. ^{11}B NMR (192.5 MHz, CD_3OD) δ 28.9. M.p. 254 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 604.2822$, *m/z* found 604.2802.

Compound ***A10***

^{1}H NMR (300 MHz, CD_3OD) δ 8.16 (br, 1H), 7.61 (dd, $J = 15.3$, 8.3 Hz, 4H), 7.33–7.25 (m, 5H), 7.17–7.11 (m, 5H), 4.76–4.70 (m, 2H), 4.54 (s, 2H), 4.26 (dd, $J = 11.3$, 4.9 Hz, 1H), 3.77 (d, $J = 5.7$ Hz, 2H), 3.68 (s, 3H), 3.16–3.10 (m, 1H), 3.0–2.92 (m, 1H), 2.08 (oct, $J = 6.8$ Hz, 1H), 0.89 (dd, $J = 6.8$, 2.4 Hz, 6H). ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 171.7, 171.0, 169.5, 167.3, 138.2, 137.4, 131.0, 130.7, 129.2, 128.1, 127.9, 127.5, 127.4, 126.2, 125.4, 72.0, 69.4, 57.5, 53.4, 51.6, 37.4, 29.8, 18.8, 18.2. ^{11}B NMR (192.5 MHz, DMSO-d_6) δ 3.1. M.p. 221 °C. HRMS (ESI, negative ion) *m/z* calcd for $[M - K]^- = 626.2652$, *m/z* found 626.2657.

Compound ***B1***

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.81 (br, 1H), 7.73 (d, $J = 6$ Hz, 2H), 7.26–7.08 (m, 8H), 4.85 (q, $J = 6.3$ Hz, 1H), 4.44 (dd, $J = 8.4$, 5.1 Hz, 1H), 3.68–3.64 (m, 4H), 3.47 (d, $J = 12.9$ Hz, 1H), 3.12–3.05 (m, 2H), 2.91 (d, $J = 4.8$ Hz, 1H), 2.09–1.70 (m, 2H), 1.27 (s, 12H), 0.82 (dd, $J = 6.9$, 5.1 Hz, 6H), 0.72 (dd, $J = 28.5$, 6.9 Hz, 6H). ^{13}C NMR (75.5 MHz, $CDCl_3$) δ 174.2, 172.0, 171.3, 142.3, 137.0, 136.9, 135.2, 129.5, 128.9, 128.7, 127.8, 127.1, 126.9, 84.0, 67.6, 57.6, 54.5, 53.4, 52.3, 37.7, 31.4, 31.3, 31.1, 30.9, 25.0, 19.5, 19.1, 17.9, 17.7. ^{11}B NMR (192.5 MHz, $CDCl_3$) δ 31.1. M.p. 96 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 594.3706$, *m/z* found 594.3726.

Compound ***B2***

^{1}H NMR (300 MHz, DMSO-d_6) δ 8.48 (br, 1H), 7.74 (br, 3H), 7.39–7.05 (m, 8H), 5.20–5.00 (m, 1H), 4.33 (dd, $J = 8.4$, 6.0 Hz, 1H), 3.69–3.61 (m, 4H), 3.54–3.49 (m, 2H), 3.22–3.15 (m, 1H), 2.82 (dd, $J = 14.1$, 10.5 Hz, 1H), 2.13 (oct, $J = 6.6$ Hz, 2H), 1.05–0.88 (m, 12H). ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 173.7,

173.2, 167.5, 138.3, 135.6, 134.7, 133.1, 129.7, 128.1, 127.0, 104.7, 66.3, 59.4, 55.6, 52.7, 51.7, 39.3, 32.0, 31.4, 19.6, 19.2, 18.6, 18.4. ^{11}B NMR (192.5 MHz, CD_3OD) δ 28.2. M.p. 223 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 512.2924$, *m/z* found 512.2948.

Compound ***B3***

^{1}H NMR (300 MHz, CD_3OD) δ 7.48–7.46 (m, 2H), 7.34–7.30 (m, 4H), 7.18–7.16 (m, 1H), 6.99–6.97 (m, 2H), 4.95 (m, 2H), 4.35 (d, $J = 6$ Hz, 1H), 3.69 (s, 3H), 3.58 (d, $J = 12.9$ Hz, 1H), 3.37–3.30 (m, 1H), 3.16–3.09 (m, 2H), 2.93–2.85 (m, 1H), 2.18–2.11 (m, 1H), 1.93–1.91 (m, 1H), 0.96–0.82 (m, 12H). ^{13}C NMR (75.5 MHz, CD_3OD) δ 173.6, 173.4, 138.4, 133.6, 133.1, 130.5, 129.7, 129.0, 128.1, 66.7, 59.4, 55.5, 52.7, 39.2, 32.1, 19.6, 19.1, 18.6. ^{11}B NMR (192.5 MHz, CD_3OD) δ 4.2. M.p. 215 °C. HRMS (ESI, negative ion) *m/z* calcd for $[M - K]^- = 534.2753$, *m/z* found 534.2777.

Compound ***B4***

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.96 (br, 1H), 7.66 (d, $J = 7.5$ Hz, 2H), 7.39–7.18 (m, 9H), 7.05–6.97 (m, 4H), 6.72 (br, 1H), 4.71 (q, $J = 7.5$ Hz, 1H), 4.47 (dd, $J = 8.4$, 4.8 Hz, 1H), 3.70 (s, 3H), 3.66–3.48 (m, 3H), 3.16–3.02 (m, 3H), 2.56 (br, 1H), 2.11 (oct, $J = 5.4$ Hz, 1H), 1.33 (s, 12H), 0.86 (t, $J = 7.5$ Hz, 6H). ^{13}C NMR (75.5 MHz, $CDCl_3$) δ 172.1, 171.1, 136.9, 136.5, 135.3, 129.6, 129.3, 129.0, 128.9, 127.8, 127.4, 127.2, 84.0, 62.9, 57.6, 54.7, 52.3, 38.7, 37.8, 31.4, 25.1, 19.1, 18.0. ^{11}B NMR (192.5 MHz, $CDCl_3$) δ 31.0. M.p. 105 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 642.3706$, *m/z* found 642.3733.

Compound ***B5***

^{1}H NMR (300 MHz, CD_3OD) δ 8.49 (br, 1H), 7.74 (br, 2H), 7.34–7.05 (m, 13H), 4.99–4.90 (m, 1H), 4.27 (dd, $J = 8.1$, 6.0 Hz, 1H), 3.95 (t, $J = 6.6$ Hz, 1H), 3.71–3.61 (m, 3H), 3.28 (s, 1H), 3.19–3.07 (m, 3H), 2.77 (dd, $J = 13.8$, 10.5 Hz, 1H), 2.16 (oct, $J = 6.6$ Hz, 1H), 0.99 (dd, $J = 6.6$, 4.8 Hz, 6H). ^{13}C NMR (75.5 MHz, CD_3OD) δ 173.4, 172.9, 172.8, 168.0, 138.3, 135.6, 135.3, 130.7, 130.6130.4, 130.1, 129.7, 128.9, 128.2, 62.6, 59.5, 55.5, 55.1, 52.7, 51.3, 39.7, 37.9, 31.9, 19.7, 18.9. ^{11}B NMR (192.5 MHz, CD_3OD) δ 28.2. M.p. 265 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+ = 560.2942$, *m/z* found 560.2926.

Compound ***B6***

^{1}H NMR (300 MHz, CD_3OD) δ 7.51 (d, $J = 7.8$ Hz, 2H),7.34–7.23 (m, 8H), 7.14–7.11 (m, 3H), 6.95 (d, $J = 7.8$ Hz, 2H), 5.00–4.95 (m, 1H), 4.29–4.27 (m, 1H), 3.87 (t, $J = 6.9$ Hz, 1H), 3.70 (s, 3H), 3.43 (q, $J = 17.1$ Hz, 2H), 3.17 (dd, $J = 5.1$, 4.8 Hz, 1H), 3.06–3.02 (m, 2H), 2.78 (dd, $J = 10.8$,10.5 Hz,1H), 2.18–2.15 (m, 1H) 1.01–0.97 (m, 6H). ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 171.8, 170.8, 166.4, 137.3, 134.6, 131.7, 129.4, 128.4, 128.1, 127.6, 127.1, 126.6, 59.9,

57.5, 53.2, 51.8, 49.7, 36.2, 29.9, 19.0, 18.2. ^{11}B NMR (192.5 MHz, DMSO-d_6) δ 3.2. M.p. 210 °C. HRMS (ESI, negative ion) *m/z* calcd for $[M - K]^-$ = 582.2753, *m/z* found 582.2779.

Compound ***B8***

^{1}H NMR (300 MHz, CD_3OD) δ 7.67 (d, J = 8.0 Hz, 2H), 7.35–7.14 (m, 12H), 4.82 (dd, J = 8.9, 5.5 Hz, 1H), 4.44 (s, 2H), 4.33 (d, J = 6.1 Hz, 1H), 3.70 (s, 3H), 3.57 (d, J = 14.6 Hz, 1H), 3.53–3.47 (m, 4H), 3.16 (dd, J = 13.9, 5.5 Hz, 1H), 2.90 (dd, J = 13.9, 8.8 Hz, 1H), 2.10 (sext, J = 6.5 Hz, 1H), 1.34 (s, 12H), 0.92 (dd, J = 6.8, 1.7 Hz, 6H). ^{13}C NMR (75.5 MHz, CD_3OD) δ 173.8, 173.5, 173.4, 143.4, 139.4, 138.2, 136.1, 130.7, 129.6, 129.5, 129.1, 129.0, 128.9, 128.0, 85.2, 76.0, 74.3, 71.3, 62.5, 59.4, 55.3, 52.8, 39.2, 32.1, 25.4, 19.6, 18.7. ^{11}B NMR (192.5 MHz, CD_3OD) δ 30.9. M.p. 95 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+$ = 672.3812, *m/z* found 672.3818.

Compound ***B9***

^{1}H NMR (300 MHz, Acetone-d_6) δ 8.44 (br, 1H), 7.85 (d, J = 7.8 Hz, 2H), 7.73 (br, 1H), 7.33–7.15 (m, 12H), 7.16 (br, 1H), 5.04 (m, 1H), 4.50 (s, 2H), 4.41 (dd, J = 8.5, 5.9 Hz, 1H), 4.18 (m, 1H), 4.09 (d, J = 12.9 Hz, 1H), 3.99–3.86 (m, 3H), 3.69 (s, 3H), 3.26 (dd, J = 14.0, 5.0 Hz, 1H), 2.94 (dd, J = 13.9, 9.3 Hz, 1H), 0.91 (d, J = 6.8 Hz, 6H). ^{13}C NMR (75.5 MHz, Acetone-d_6) δ 172.5, 171.8, 167.1, 138.9, 138.5, 136.0, 135.0, 130.8, 130.3, 129.6, 129.5, 129.0, 128.9, 128.0, 74.4, 69.4,60.7, 58.9, 55.7, 52.6, 51.7, 39.5, 32.0, 19.8, 19.0. ^{11}B NMR (192.5 MHz, $CDCl_3$) δ 29.6. M.p. 121 °C. HRMS (ESI, positive ion) *m/z* calcd for $[M + H]^+$ = 590.3030, *m/z* found 590.3029.

Compound ***B10***

^{1}H NMR (300 MHz, CD_3OD) δ 7.53 (d, J = 7.7 Hz, 2H), 7.34–7.13 (m, 10H), 7.04 (d, J = 7.6 Hz, 2H), 4.96–4.92 (m, 1H), 4.47 (s, 2H), 4.33 (d, J = 3.7 Hz, 1H), 3.79–3.62 (m, 7H), 3.24–3.17 (m, 1H), 2.89 (dd, J = 13.9, 9.9 Hz, 1H), 2.08 (sext, J = 6.8 Hz, 1H), 0.91 (dd, J = 6.8, 3.7 Hz, 6H). ^{13}C NMR (75.5 MHz, CD_3OD) δ 173.3, 173.2, 167.4, 138.5, 138.1, 133.9, 133.5, 130.5, 129.7, 129.5, 129.0, 128.1, 74.5, 69.0, 59.9, 59.4, 55.8, 52.7, 51.6, 39.2, 31.9, 19.6, 18.7. ^{11}B NMR (192.5 MHz, CD_3OD) δ 4.6. M.p. 139 °C. HRMS (ESI, negative ion) *m/z* calcd for $[M - K]^-$ = 612.2859, *m/z* found 612.2850.

Results and discussion

Building blocks used for constructing the desired products are shown in Fig. 1. Six potassium peptidyl organotriboroborates and their corresponding boron-containing analogs were successfully synthesized (Fig. 2). The general synthetic strategies are outlined in Schemes 1 and 2.

Compounds **B3**, **B6**, and **B10** were successfully obtained by formation of imines from the free amine building blocks, by use of potassium 4-formylphenyltrifluoroborate (**4**), followed by reductive amination [6]. The resulting organotrifluoroborates were then converted into boronate esters (**B1**, **B4**, and **B8**) or boronic acids (**B2**, **B5**, and **B9**). Compounds **B7** and **B11** were also synthesized and used as positive controls. The structures of the synthesized compounds are summarized in Fig. 2.

The anti-proliferative activity of the synthesized compounds against HepG2 and MDA-MB231 were evaluated by use of an MTT assay; the IC_{50} values are listed in Table 1. Doxorubicin was used as positive control because it is currently used to treat different types of breast cancer and other cancers [7, 8].

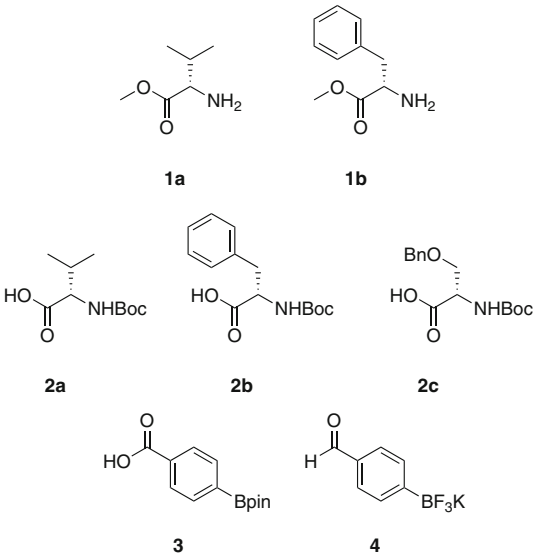


Fig. 1 Amino acid and boron building blocks

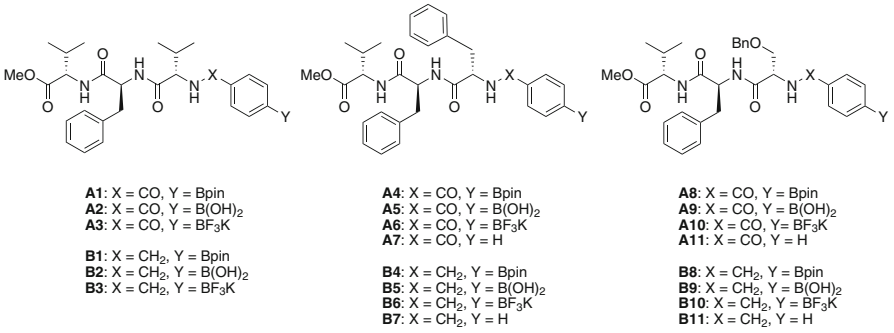
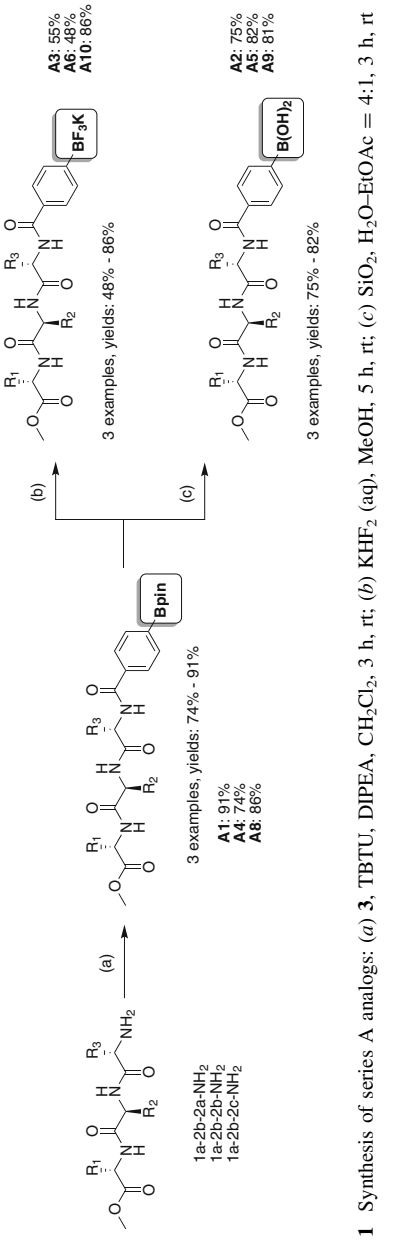


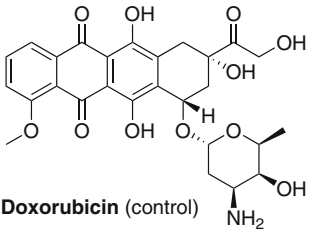
Fig. 2 Summary of synthesized boron-containing peptidyl analogs



Scheme 1 Synthesis of series A analogs: (*a*) **3**, TBTU, DIPEA, CH_2Cl_2, 3 h, rt; (*b*) KHF_2 (aq), MeOH, 5 h, rt; (*c*) SiO_2, H_2O–EtOAc = 4:1, 3 h, rt

Scheme 2 Synthesis of series B analogs: (*a*) 4, PEMB, MeOH, 3–12 h, rt; (*b*) SiO_2, pinacol, H_2O–EtOAc = 4:1, 4 h, rt; (*c*) SiO_2, H_2O–EtOAc = 4:1, 3 h, rt

Table 1 SAR study of series **A** and series **B** analogs

Entry	Compound	IC_{50} (μM)[a]	
		HepG2	MDA-MB231
1	**B6**	> 100	29.9
2	**B7** (control)	> 100	> 100
3	**B9**	24.7	24.5
4	**B10**	21.8	18.9
5	**B11** (control)	> 100	> 100
6	**KF** (control)	> 100	> 100
7	**KBF_4** (control)	> 100	> 100
8	 **Doxorubicin** (control)	0.4	2.3

Conclusions

Preliminary structure activity relationships (SAR) analysis revealed that the presence of potassium trifluoroborate is vital for a compound's biological activity; replacing it with boronic acid or boronate ester greatly diminished or eliminated this activity. The presence of a large hydrophobic and/or aromatic residue, for example a benzyl or benzyloxymethyl group, on the tripeptidyl compounds significantly enhanced the compounds' activity. Compared with the full peptide backbones, an aminomethyl linking group was preferred for the tri-peptidyl compounds. These results provide insight into the structural optimization necessary for future syntheses. Further work is in progress and the results will be presented in due course.

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