Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2014, 6(9):376-380

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Design, synthesis and biological activity evaluation of novel anticancer agent 5-(2-carboxyethenyl)indole derivatives

Kailin Han[1,2,3], Haomeng Wang[1,2,3], Binbin Song[1,2,3], Yashan Li[1,2,3], Wei Na Ding[1,2,3], Hongye Zhao[1,2,3], Hua Sun[1,2,3], Yuou Teng[1,2,3*] and Peng Yu[1,2,3,i*]

[1]Key Lab of Industrial Fermentation Microbiology (Tianjin University of Science and Technology), Ministry of Education, Tianjin, P. R. China
[2]Tianjin Key Lab of Industrial Microbiology, Tianjin University of Science and Technology, Tianjin, P. R. China
[3]Sino-French Joint Lab of Food Nutrition/Safety and Medicinal Chemistry, Tianjin University of Science and Technology, Tianjin, P. R. China

---

ABSTRACT

*A series of novel 5-(2-Carboxyethenyl) indole derivatives were designed and synthesized about 38-48% overall yields. Two of the seven newly synthesized compounds, compound **5** and **7**, have not been reported before. Their structures were characterized on the basis of ^{1}H, ^{13}C NMR, and those compounds were tested for their anticancer activities against K562 and HT-29 cell lines. Results indicated that compounds **5**, **6** and **7** containing 5-(2-Carboxyethenyl)indole derivatives demonstrated significant anticancer activity against HT-29 cell, their potency reached 4.67, 8.24 and 6.73 μM, respectively.*

Keywords: indole derivatives, anticancer, Heck-Coupling reaction

---

INTRODUCTION

The indole ring has been considered as an important moiety found in many pharmacologically active compounds possessing certain biological activities in which some studies have been attributed to its anticancer effectiveness as described in the literature [1-3]. In the last several decades, increasing numbers of researchers from both industry and academia have embarked on the development of new indole-based anticancer agents [4-6].

Recently, Lai's group synthesized several N-arylsulfonyl and C-5 N-hydroxyacrylamide indole analogues as potent histone deacetylase inhibitors[7]. N-1 and C-5 substituted indoles have been referred to as "privileged structures" for plenty of pharmacologically active lead compounds in the drug research and development since they are capable of binding to many receptors with high affinity [8]. In this paper, we developed of new 1, 5-disubstituted indole derivatives to the synthesis and in vitro antitumor evaluation of N-benzyl and C-(5-(2-Carboxyethenyl) substituted indole derivatives.

EXPERIMENTAL SECTION

2.1 Materials and measurements

All reagents and solvents used in this article were of reagent grade. Reaction temperatures were controlled using oil bath temperature modulator. Heck coupling reaction was performed using Biotage microwave reactor. Thin layer chromatography (TLC) was performed using E. Merck silica gel 60 GF254 precoated plates (0.25 mm) and visualized using a combination of UV and ninhydrin solution exposing. Silica gel (particle size 200-400 mesh) was used for flash chromatography. ^{1}H and ^{13}C spectra were recorded on Bruker AM-400 NMR spectrometers in deuterated chloroform and deuterated DMSO. The chemical shifts are reported in δ (ppm) relative to

tetramethylsilane as internal standard.

2.2 Synthesis route of indole derivatives

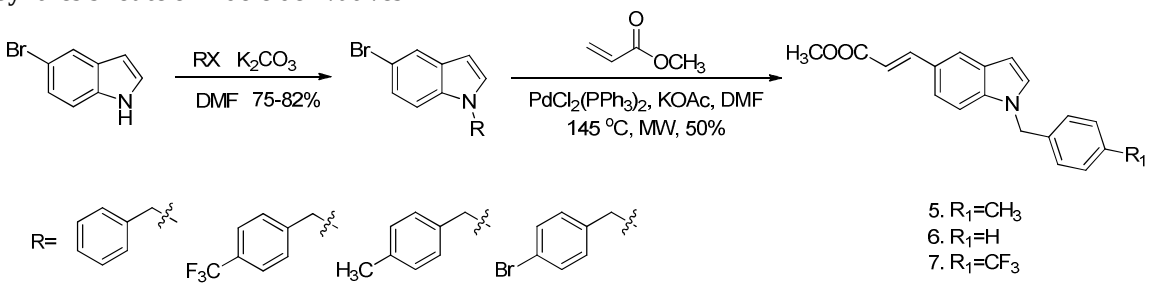

Fig. 1 Synthesis route of indole derivatives

2.2.1 1-benzyl-5-bromo-1*H*-indole (1) [9]

To a flask (25 mL) which contained the solution of 5-bromo-1*H*-indole (1.00 g, 5.10 mmol) in dry N,N-Dimethylformamide (5 mL). The reaction temperature was maintained at 0 ℃ followed by the dropwise addition sodium hydride (0.24 g, 10.20 mmol). Stirring 5 min, was added benzyl chloride (0.77 g, 6.12 mmol) and the mixture allowed warm to room temperature. The reaction mixture was stirred at room temperature for 4 h. The orange solution was poured water (25 mL) and extracted with dichloromethane (3 × 100 mL). The combined extracts were dried over anhydrous magnesium sulfate, filtered and evaporated. Chromatography (petroleum ether/ethyl acetate 8:1) afforded the title compound as white solid (1.16 g, 80%).

2.2.2 1-(4-(trifluoromethyl) benzyl)-5-bromo-1*H*-indole (2)

To a flask (25 mL) which contained the solution of 5-bromo-1*H*-indole (1.00 g, 5.10 mmol) in dry N,N-Dimethylformamide (5 mL). The reaction temperature was maintained at 0 ℃ followed by the dropwise addition sodium hydride (0.24 g, 10.20 mmol). Stirring 5 min, was added 4-Trifluoromethylbenzyl chloride (1.19 g, 6.12 mmol) and the mixture allowed warm to room temperature. The reaction mixture was stirred at room temperature for 4 h. The orange solution was poured water (25 mL) and extracted with dichloromethane (3 × 100 mL). The combined extracts were dried over anhydrous magnesium sulfate, filtered and evaporated. Chromatography (petroleum ether/ethyl acetate 6:1) afforded the title compound as white solid (1.37 g, 75%).

2.2.3 1-(4-methylbenzyl)- 5-bromo-1*H*-indole (3)

To a flask (25 mL) which contained the solution of 5-bromo-1*H*-indole (1.00 g, 5.10 mmol) in dry N,N-Dimethylformamide (5 mL). The reaction temperature was maintained at 0 ℃ followed by the dropwise addition sodium hydride (0.24 g, 10.20 mmol). Stirring 5 min, was added 4-Methylbenzyl chloride (0.86 g, 6.12 mmol) and the mixture allowed warm to room temperature. The reaction mixture was stirred at room temperature for 4 h. The orange solution was poured water (25 mL) and extracted with dichloromethane (3 × 100 mL). The combined extracts were dried over anhydrous magnesium sulfate, filtered and evaporated. Chromatography (petroleum ether/ethyl acetate 10:1) afforded the title compound as white solid (1.17 g, 77%).

2.2.4 1-(4-bromobenzyl)-5-bromo-1*H*-indole (4)

To a flask (25 mL) which contained the solution of 5-bromo-1*H*-indole (1.00 g, 5.10 mmol) in dry N,N-Dimethylformamide (5 mL). The reaction temperature was maintained at 0 ℃ followed by the dropwise addition sodium hydride (0.24 g, 10.20 mmol). Stirring 5 min, was added 4-Bromobenzyl bromide (1.53 g, 6.12 mmol) and the mixture allowed warm to room temperature. The reaction mixture was stirred at room temperature for 4 h. The orange solution was poured water (25 mL) and extracted with dichloromethane (3 × 100 mL). The combined extracts were dried over anhydrous magnesium sulfate, filtered and evaporated. Chromatography (petroleum ether/ethyl acetate 10:1) afforded the title compound as white solid (1.45 g, 78%).

2.2.5 (E)-methyl 3-(1-(4-methylbenzyl)-1*H*-indol-5-yl) acrylate (5) [10]

To a microwave reactor vial (5 mL) which contained the solution of 1-(4-methylbenzyl)- 5-bromo-1H-indole (0.50 g, 1.67 mmol) in DMF (5 mL) were added Bis(triphenylphosphine)palladium(II) chloride (61 mg, 0.09 mmol), CH_3COOK (0.23 g, 2.33 mmol) and Methyl acrylate (0.18 g, 2.00 mmol) under the atmosphere of Ar. The microwave reactor vial was caped and placed into the microwave cavity. The reaction mixture was irradiated at high level for 1 h at 150 ℃. The reaction mixture was cooled to room temperature, then poured onto 100 mL ice-water and then extracted with dichloromethane (3×100 mL). The combined organic layers were washed with water (2×100 mL), brine (100 mL) and dried over anhydrous magnesium sulfate. The solvent was removed under vacuum to afford the crude which was purified by flash column chromatography (silica gel, petroleum ether/ethyl acetate 5:1) to yield the desired compound (E)-methyl 3-(1-benzyl-1H-indol-5-yl)acrylate (5) (0.25 g, 50%).

2.2.6 (E)-methyl 3-(1-benzyl-1H-indol-5-yl) acrylate (6)

To a microwave reactor vial (5 mL) which contained the solution of 1-benzyl- 5-bromo- 1H-indole (0.50 g, 1.75 mmol) in DMF (5 mL) were added Bis(triphenylphosphine)palladium(II) chloride (60 mg, 0.09 mmol), CH_3COOK (0.24 g, 2.45 mmol) and Methyl acrylate (0.18 g, 2.10 mmol) under the atmosphere of Ar. The microwave reactor vial was caped and placed into the microwave cavity. The reaction mixture was irradiated at high level for 1 h at 150 ℃. The reaction mixture was cooled to room temperature, poured onto 100 mL ice-water and then extracted with dichloromethane (3×100 mL). The combined organic layers were washed with water (2×100 mL), brine (100 mL) and dried over anhydrous magnesium sulfate. The solvent was removed under vacuum to afford the crude which was purified by flash column chromatography (silica gel, petroleum ether/ethyl acetate 6:1) to yield the desired compound (E)-methyl 3-(1-benzyl-1H-indol-5-yl)acrylate (6) (0.32 g, 61%).

2.2.7 (E)-methyl 3-(1-(4-(trifluoromethyl) benzyl)-1H-indol-5-yl)acrylate (7)

To a microwave reactor vial (5 mL) which contained the solution of 5-bromo-1-(4-(trifluoromethyl) benzyl)-1H-indole (0.50 g, 1.41 mmol) in DMF (5 mL) were added Bis(triphenylphosphine)palladium(II) chloride (50 mg, 0.07 mmol), CH_3COOK (0.19 g, 1.98 mmol) and Methyl acrylate (0.15 g, 1.69 mmol) under the atmosphere of Ar. The microwave reactor vial was caped and placed into the microwave cavity. The reaction mixture was irradiated at high level for 1 h at 150 ℃. The reaction mixture was cooled to room temperature, poured onto 100 mL ice-water and then extracted with dichloromethane (3×100 mL). The combined organic layers were washed with water (2×100 mL), brine (100 mL) and dried over anhydrous magnesium sulfate. The solvent was removed under vacuum to afford the crude which was purified by flash column chromatography (silica gel, petroleum ether/ethyl acetate 5:1) to yield the desired compound (E)-methyl 3-(1-(4-(trifluoromethyl)benzyl)-1H-indol-5-yl)acrylate (7) (0.27 g, 53%).

2.3 Biological assay [11].

Cells (100 μL) were cultured in 96-well plates at a density of 5×10^4 cells/mL for 2 hours(K562) or overnight (HT-29). Compounds (DMSO solution of 0.5 μL) were added to each well to culture for another 48 h. MTT assay was performed using Thermo microplate reader. The DMSO-treated controls were calculated as a cell viability value of 100%. The IC_{50} values were obtained by nonlinear regression using GraphPad Prism 4.0. IC_{50} measurements for each compound were done three times.

RESULTS AND DISCUSSION

3.1 Characterize isatin derivatives by ^{1}H and ^{13}C NMR.

3.1.1 1-benzyl-5-bromo-1*H*-indole (1).

^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 5.35 (s, 2H), 6.51 (s, 1H), 7.05-7.07 (d, 1H, *J* =8.4 Hz); 7.13-7.14 (d, 3H, *J* = 7.6 Hz), 7.24 (m, 2H), 7.53-7.55 (d, 2H, *J* =8.0 Hz); 7.81 (s, 1H).; ^{13}C NMR (100 MHz, $CDCl_3$) δ 51.12, 110.23, 112.05, 112.05, 118.57, 120.12, 121.28, 123.44, 125.66, 127.83, 127.83, 129.31, 129.58, 129.58, 135.41.

3.1.2 1-(4-(trifluoromethyl)benzyl)-5-bromo-1*H*-indole (2).

^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 5.35 (s, 2H), 6.52 (s, 1H), 7.05-7.07 (d, 1H, *J* =8.4 Hz); 7.13-7.15 (d, 3H, *J* = 7.6 Hz), 7.24 (m, 1H), 7.53-7.55 (d, 2H, *J* =8.0 Hz); 7.78 (s, 1H).; ^{13}C NMR (100 MHz, $CDCl_3$) δ 50.68, 110.01, 111.86, 111.86, 117.86, 120.02, 122.12, 124.13, 124.48, 124.96, 127.52, 127.52, 128.79, 129.64, 129.64, 135.22.

3.1.3 1-(4-methylbenzyl)- 5-bromo-1*H*-indole (3).

^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 2.32 (s, 2H), 5.36 (s, 2H), 6.53 (s, 1H), 7.02-7.04 (d, 1H, *J* =8.4 Hz); 7.10-7.13 (m, 3H), 7.24 (m, 1H), 7.57-7.59 (d, 2H, *J* =8.4 Hz); 7.78 (s, 1H).; ^{13}C NMR (100 MHz, $CDCl_3$) δ 26.28, 50.87, 110.11, 110.98, 112.32, 118.56, 121.47, 122.55, 124.78, 125.61, 128.04, 128.04, 128.82, 129.30, 129.30, 134.85.

3.1.4 1-(4-bromobenzyl)-5-bromo-1*H*-indole (4).

^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 5.24 (s, 2H), 6.49 (s, 1H), 6.91-6.93 (d, 2H, *J* =8.4 Hz); 7.06-7.11 (m, 2H), 7.24 (m, 1H), 7.40-7.42 (d, 2H, *J* =8.4 Hz); 7.76 (s, 1H).; ^{13}C NMR (100 MHz, $CDCl_3$) δ 50.87, 111.12, 112.56, 112.56, 119.03, 120.82, 121.75, 124.01, 125.43, 127.96, 127.96, 130.01, 130.15, 130.15, 135.88.

3.1.5 (E)-methyl 3-(1-(4-methylbenzyl)-1*H*-indol-5-yl)acrylate (5).

^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 2.30 (s, 3H), 3.80 (s, 3H), 5.32 (s, 2H), 6.37-6.41 (d, 1H, J =16.0 Hz), 6.58 (s, 1H), 7.09-7.11 (d, 1H, J = 7.6 Hz), 7.15 (s, 1H), 7.25-7.32 (m, 4H), 7.38-7.41 (d, 1H, J = 8.8 Hz), 7.81 (m, 1H), 7.83 (d, 1H, J =16.0 Hz).; ^{13}C NMR (100 MHz, $CDCl_3$) δ 26.80, 50.87, 52.24, 108.35, 112.30, 117.86, 119.21, 122.53, 124.35, 127.56, 127.56, 130.23, 130.36, 130.36, 137.58, 139.64, 140.11, 142.26, 143.68, 169.54.

3.1.6 (E)-methyl 3-(1-benzyl-1H-indol-5-yl)acrylate (6).
^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 3.78 (s, 3H), 5.39 (s, 2H), 6.36-6.40 (d, 1H, J =16.0 Hz), 6.45 (s, 1H), 7.07-7.09 (d, 1H, J = 8.0 Hz), 7.08 (s, 1H), 7.20-7.28 (m, 5H), 7.36-7.38 (d, 1H, J = 8.0 Hz), 7.78 (d, 1H, J = 8.0 Hz), 7.80 (d, 1H, J =16.0 Hz).; ^{13}C NMR (100 MHz, $CDCl_3$) δ 51.10, 52.23, 107.44, 111.80, 118.02, 119.43, 121.76, 125.16, 127.89, 127.89, 130.78, 131.04, 131.04, 138.07, 140.12, 141.36, 142.87, 145.17, 170.04.

3.1.7 (E)-methyl 3-(1-(4-(trifluoromethyl)benzyl)-1H-indol-5-yl)acrylate (7).
^{1}H NMR ($CDCl_3$ 400 MHz): δ/ppm 3.75 (s, 3H), 5.28 (s, 2H), 6.32-6.36 (d, 1H, J =16.0 Hz), 6.41 (s, 1H), 6.97-6.99 (d, 1H, J = 8.0 Hz), 7.01 (s, 1H), 7.18-7.25 (m, 4H), 7.28-7.30 (d, 1H, J = 8.0 Hz), 7.65 (d, 1H, J = 8.0 Hz), 7.67 (d, 1H, J =16.0 Hz). ; ^{13}C NMR (100 MHz, $CDCl_3$) δ 52.03, 52.45, 108.22, 112.03, 119.14, 120.25, 122.61, 124.87, 125.78, 128.43, 128.43, 131.02, 131.75, 131.75, 137.89, 139.38, 140.89, 141.54, 143.64, 168.99.

3.2 Anticancer activity assay.
Seven synthesized compounds which were dissolved in dimethylsulphoxide were subjected to anticancer activity by MTT assay[11]. The observed IC_{50} value is presented in **Table 1**.

As shown in **Table 1**, compounds **1**, **2**, **3** and **4** did exhibit certain inhibitory activities against both K562 and HT-29 cell lines, which might come from the N-benzyl groups of its mother nuclear structure. In order to improving the inhibitory activities, compounds **5**, **6** and **7** were designed and tested. Compounds **5**, **6** and **7** exhibited much better inhibitory activities against HT-29 cell lines. For HT-29, their potency reached 4.67, 8.24 and 6.73 μM, respectively, which mean the lipophilic enhancement of the N-benzyl group and C-(5-(2-Carboxyethenyl) group did improve their inhibitory capability.

Table 1 *In vitro* anticancer activity of the compounds 1-7

compound	IC_{50}(μM)						
	1	2	3	4	5	6	7
K562	>10	>10	>10	>10	>10	>10	>10
HT-29	>10	>10	>10	>10	4.67	8.24	6.73

CONCLUSION

In summary, we have described synthetic approach to prepare the novel anticancer compounds **5**, **6** and **7** via microwave-assistant Heck coupling reaction as the key steps in 38-48% overall yield. The newly synthesized compounds were also tested for the antitumor activities against K562 and HT-29 cell lines. Among them, compounds **5**, **6** and **7** demonstrated even better anticancer activity against HT-29 cells than that of compounds **1**, **2**, **3**, and **4**. Compound **5**, as highly potent anticancer compound with IC_{50} = 4.67 μM, against HT-29. Further modification and SAR studies based on the lead compound **7** are ongoing in this lab.

Acknowledgments
The authors sincerely thank the financial support from the National Natural Science Foundation of China (31301142), the International Science & Technology Cooperation Program of China (2013DFA31160) and the Ministry of Education Changjiang Scholars and Innovative Research Team Development Plan (IRT1166).

REFERENCES

[1] A Andreani; M Granaiola; A Leoni; A Locatelli; R Morigi; M Rambaldi; V Garaliene, *J. Med. Chem*., **2002**, 45(12), 2666-2669.
[2] K Swathi; A Srinivas; M Sarangapani, *J. Chem. Pharm. Res.*, **2010**, 2(2), 220-225.
[3] A Andreani; S Burnelli; M Granaiola; A Leoni; A Locatelli; R Morigi; M Rambaldi; L Varoli; L Landi; C Prata; MV Berridge; C Grasso; HH Fiebig; G Kelter; AM Burgere; MW Kunkelf, *J. Med. Chem*., **2008**, 51(15), 4563-4570.
[4] JP Perchellet; AM Waters; EM Perchellet; PD Thornton; N Brown; D Hill; B Neuenswander; GH Lushington; C Santini; N Chandrasoma; KR Buszek, *Anticancer Res*., **2012**, 32(11), 4671-4684.
[5] RS Dave; RJ Odedara; RI Kalaria; JJ Upadhyay, *J. Chem. Pharm. Res.*, **2012**, 4(11), 4864-4869.
[6] J Azimvand, *J. Chem. Pharm. Res*., **2012**, 4(8), 3909-3913.
[7] MJ Lai; HL Huang; SL Pan; YM Liu; CY Peng; HY Lee; TK Yeh; PH Huang; CM Teng; CS Chen; HY Chuang; JP Liou, *J.Med.Chem*., **2012**, 55(8), 3777-3791.
[8] BE Evans; KE Rittle; MG Bock; RM DiPardo; RM Freidinger; WL Whitter; GF Lundell; DF Veber; PS Anderson, *J.Med.Chem.*, **1988**, 31(12), 2235-2246.
[9] DY Zhang; D Stephens; G Hernandez; R Mendoza; OV Larionov, *Chem-Eur. J*., **2012**, 18(52), 16612-16615.

[10] F Luiz; Jr Silva; MV Craveiro, *Organic Letters*., **2008**, 10(23), 5417-5420.
[11] JS Biradar; BS Sasidhar, *Eur. J. Med. Chem*., **2011**, 46(12), 6112-6118.