# The Preparation and Evaluation of 1-Substituted 1,2,3,4-Tetrahydro- and 3,4-Dihydro-β-carboline Derivatives as Potential Antitumor Agents

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Received June 17, 2004; accepted September 22, 2004

**A series of 1-substituted 1,2,3,4-tetrahydro- and 3,4-dihydro-β-carboline derivatives have been synthesized and evaluated for antitumor activity against murine P-388 and human tumor cell lines, KB-16, A-549 and HT-29. All of the compounds prepared, except for 19, showed significant cytotoxicity. Among them, compound 29 exhibited the most potent activity against all tested tumor cell lines. There was an apparent lack of correlation regarding cytotoxicity between 1,2,3,4-tetrahydro- and 3,4-dihydro-β-carbolines. This study is the first to discover compound 29 as a potential lead for the development of future anticancer agents. The mode of inhibition for compound 29 was proposed.**

**Key words** 1,2,3,4-tetrahydro-β-carboline; 3,4-dihydro-β-carboline; antitumor agent

Marine natural products which contain a β-carboline skeleton are widely distributed in marine invertebrates.[1—3] The discovery of natural β-carboline metabolites as potent antitumor and antiviral agents has stimulated great interest in the synthetic and pharmacological studies of β-carboline derivatives.[4—6] Particularly interesting targets include such compounds as eudistomins[7,8] and manzamines,[9,10] which were isolated from marine tunicates and sponges, respectively. As a class, the oxathiazepine containing eudistomines exhibited potent inhibitory activity toward DNA virus *HSV*-1. In addition, the antiviral eudistomines C (**1**) and E (**2**) were also found active against *HSV*-2, the *Vaccinia* virus and RNA viruses.[11] The novel structures of manzamines, however, were reported to possess potent antitumor,[12,13] antibacterial, antifungal, and anti-HIV activities.[14] The most active was manzamine A (**3**), a principal metabolite from several species of sponges, which showed cytotoxicity against murine P-388 cells at 0.07 μg/ml.[15] Our previous report has revealed that manzamine A—D and H exhibited potent cytotoxicities against human KB-16, A-549 and HT-29 tumor cell lines.[16] The common β-carboline or 1,2,3,4-tetrahydro-β-carboline moieties appear to be important for the biological activity. In order to investigate the essential structural features for the antitumor activity of manzamine A (**3**), the synthesis of 1-substituted 1,2,3,4-tetrahydro- and 3,4-dihydro-β-carboline derivatives were initiated. The difficult isolation of manzamine A (**3**) from sponge and its limited source make this current program more important and significant. A facile synthetic method by the application of Pictet–Spengler reaction[17—20] and DDQ oxidation[21] allowed the preparation of compounds **4**—**29**. Herein, we wish to report the preparation, characterization and biological evaluation of 1-substituted 1,2,3,4-tetrahydro- and 3,4-dihydro-β-carboline derivatives.

## Results and Discussion

As summarized in Chart 1, compounds **4**—**16** were synthesized from tryptamine (I) and substituted benzaldehydes *via* Pictet–Spengler cyclization. Subsequent oxidation of 1,2,3,4-tetrahydro-β-carbolines (**4**—**16**, A compounds) by DDQ furnished 3,4-dihydro-β-carboline derivatives (**17**—**29**, B compounds). The spectral data for **4**—**29** are illustrated in Experimental.

Table 1 shows the biological results for compounds **4**—**29** and manzamine A (**3**) as tested against murine P-388 (leukemia) and human tumor cell lines, including KB-16 (nasopharyngeal carcinoma), A-549 (lung adenocarcinoma) and HT-29 (colon adenocarcinoma) *in vitro*. All of the compounds exhibited promising activity except for **19**, which had no activity toward the four tumor cell lines. Among them, compound **29** exhibited the most potent activity against all tested tumor cells. For P-388 inhibitory activity, compounds **8** and **29** were more potent than or comparable to **3**. Compounds **4**, **12**, **14**, **15**, **25** and **28** showed potent to moderate activity. As for KB-16 cell cytotoxicity, compound **29** was equal in potency to manzamine A (**3**). Compounds **8** and **21** have lower $ID_{50}$ values, while the others were less active. Compounds **12**, **22** and **24** possessed specific inhibition against A-549 and HT-29 tumor cells.

There was an apparent lack of correlation between 1,2,3,4-tetrahydro-β-carbolines and 3,4-dihydro-β-carbolines in the preliminary cytotoxicity tests. For example, comparison of the biological data of compounds **6** and **19**, as well as compounds **16** and **29**, revealed that B compounds (**17**—**29**) are not always superior to A compounds (**4**—**16**). Both compounds **29** and **19**, which contain a 3,4-dihydro-β-carboline skeleton showed notable difference in activity. The reason for

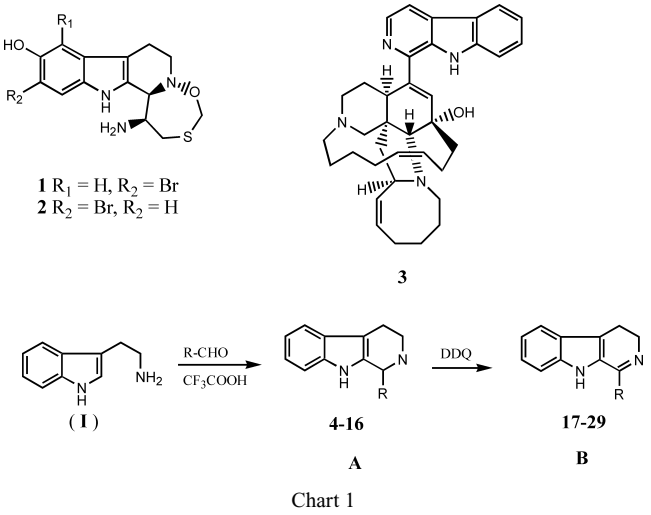


Chart 1

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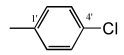
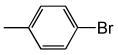
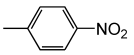
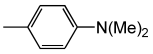
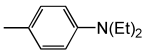
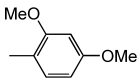
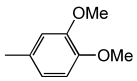
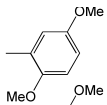
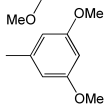
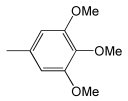
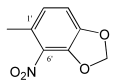
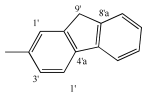
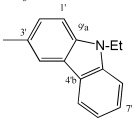
Table 1. Biological Evaluation[a)] of 1-Substituted 1,2,3,4-Tetrahydro-$\beta$-carboline and 3,4-Dihydro-$\beta$-carbolines

**A** **B**

| Compound | R | P-388 | | KB-16 | | A-549 | | HT-29 | |
|---|---|---|---|---|---|---|---|---|---|
| | | **A** | **B** | **A** | **B** | **A** | **B** | **A** | **B** |
| **4** |  | 0.2 | | 0.7 | | 0.6 | | 0.9 | |
| **17** | | | 0.5 | | 0.9 | | 1.2 | | 1.3 |
| **5** |  | 1.0 | | 2.1 | | 3.0 | | 1.1 | |
| **18** | | | 1.1 | | 2.4 | | 2.0 | | 1.2 |
| **6** |  | 0.8 | | 1.1 | | 0.9 | | 1.5 | |
| **19** | | | 7.7 | | >50 | | >50 | | >50 |
| **7** |  | 2.2 | | 1.7 | | 1.6 | | 0.7 | |
| **20** | | | 2.9 | | 4.8 | | 50 | | 3.0 |
| **8** |  | 0.07 | | 0.2 | | 1.1 | | 1.1 | |
| **21** | | | 0.7 | | 0.3 | | 1.8 | | 1.3 |
| **9** |  | 0.6 | | 0.7 | | 1.1 | | 0.8 | |
| **22** | | | 0.6 | | 2.7 | | 0.5 | | 0.5 |
| **10** |  | 2.8 | | >50 | | >50 | | >50 | |
| **23** | | | 2.7 | | 15 | | 12 | | 2.7 |
| **11** |  | 2.6 | | 3.7 | | 2.0 | | 2.2 | |
| **24** | | | 1.1 | | 0.7 | | 0.2 | | 0.4 |
| **12** |  | 0.2 | | 1.4 | | 0.6 | | 0.4 | |
| **25** | | | 0.4 | | 1.0 | | 0.6 | | 0.6 |
| **13** |  | 1.2 | | 8.3 | | 2.0 | | 1.6 | |
| **26** | | | 1.2 | | 1.4 | | 0.5 | | 0.7 |
| **14** |  | 0.3 | | 6.0 | | 1.7 | | 1.4 | |
| **27** | | | 0.3 | | 0.8 | | 0.6 | | 1.0 |
| **15** |  | 0.1 | | 0.4 | | 0.6 | | 0.5 | |
| **28** | | | 0.1 | | 1.4 | | 0.6 | | 0.7 |
| **16** |  | 0.7 | | 0.8 | | 0.8 | | 0.5 | |
| **29** | | | <0.001 | | <0.001 | | <0.001 | | <0.001 |
| **30** (paclitaxel) | | | <0.001 | | <0.001 | | <0.001 | | <0.001 |
| **3** (manzamineA) | | | 0.07 | | <0.001 | | 0.03 | | 0.1 |

*a*) The concentration of compound inhibiting 50% ($IC_{50}$, $\mu$g/ml) of the growth of tumor cell lines, P-388 (murine leukemia), KB-16 (human nasopharyngeal carcinoma), A-549 (human lung adenocarcinoma) and HT-29 (human colon adenocarcinoma), after 3, 3, 6 and 6 d drug exposure. (— Not tested).

different degrees of activity might be explained by the differences in bioavailability such as drug uptake and fate of metabolism. Compound **29** showed much better activity than that of **16**, suggesting that the double bond between C-1 and C-2 in **29** is critical in cytotoxicity. This double bond makes compound **29** into a planar structure, which may allow it to intercalate more easily with the DNA molecule, although there is no evidence to support this proposal.[22)] However, recently the cytotoxic mechanism of **29** was first reported and characterized as chromosome missegregation.[23)]

Introducing the moiety of *N*-ethyl carbazole onto 3,4-dihydro-$\beta$-carboline to give **29** seems to be a correct route to

reach the lead compound. However, stronger cytotoxicity may cause increased side effects and thus result in a lower therapeutic index. Further biological evaluation of 1-substituted 1,2,3,4-tetrahydro- and 3,4-dihydro-$\beta$-carboline derivatives as potential antitumor agents by application of mechanism-based bioassay systems, such as the inhibition of human topoisomerase I and II and animal antitumor tests, is currently under investigation.

## Experimental

**General Methods** All mp's were taken on a Buchi mp B-540 appratus and are uncorrected. UV and IR spectra were taken on a Hitachi V-3210 and JASCO A-100 IR spectrophotometers, respectively. EI-MS spectra were obtained on a MAT 112S-JMS D300 spectrometer using direct inlet systems. $^1$H- and $^{13}$C-NMR spectra were recorded on a Varian FT-300 spectrometer. Analytical thin-layer chromatography (TLC) was carried out on Kiesel gel $GF_{254}$ plates, and detection was made under UV light. EM Kieselgel 60 (230—400 mesh ASTM) was used for column chromatography.

**Synthesis of Compounds 4—16** To a stirred solution of tryptamine (1.6 g, 1 mmol) and the appropriate substituted aldehyde (1 mmol) in toluene (30 ml) at room temperature was slowly added trifloroacetic acid (TFA, 2 ml). The reaction mixture was stirred at room temperature for 2 d. After evaporation of the solvent under vacuum, the residue was chromatographed on a Si gel column (60 g) and eluted with a solvent mixture of $CHCl_3$/MeOH by the following ratios and volumes (99 : 1, 98 : 2, 97 : 3, 96 : 4, 95 : 5; each 100 ml), to afford compounds **4**—**16** with a yield which varied in the range of 30—50%.

1-(4′-Chlorophenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**4**): White solid; mp 165 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.09 (1H, s, H-1), 3.11 (1H, m, H-3a), 3.28 (1H, m, H-3b), 2.85 (2H, m, H-4), 7.17 (3H, overlap, H-5, 6, 7), 7.57 (1H, d, $J$=8.4 Hz, H-8), 7.87 (1H, s, NH-9), 7.20 (2H, d, $J$=7.8 Hz, H-2′, 6′), 7.31 (2H, d, $J$=7.8 Hz, H-3′, 5′), $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 57.1 (d, C-1), 42.4 (t, C-3), 22.3 (t, C-4), 110.2 (s, C-4a), 133.7 (s, C-4b), 121.8 (d, C-5), 119.4 (d, C-6), 118.2 (d, C-7), 110.8 (d, C-8), 140.3 (s, C-8a), 135.9 (s, C-9a), 127.2 (s, C-1′), 129.8 (d, C-2′, 6′), 128.9 (d, C-3′, 5′), 133.9 (s, C-4′); EI-MS $m/z$ 282 (100, $M^+$), 281 (72), 255 (13), 254 (17), 253 (39), 252 (30), 219 (25), 218 (92), 217 (80), 189 (9), 171 (81), 169 (18), 154 (9), 144 (21), 143 (20), 130 (10), 123 (15), 115 (22), 109 (51); HR-EI-MS $m/z$ 282.0926 (M+, Calcd for $C_{17}H_{15}N_2Cl$, 282.0924).

1-(4′-Bromophenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**5**): Pale yellow solid, mp 226—228 °C; $C_{17}H_{15}N_2Br$; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.10 (1H, s, H-1), 3.12 (1H, m, H-3a), 3.30 (1H, m, H-3b), 2.87 (2H, m, H-4), 7.18 (3H, overlap, H-5, 6, 7), 7.65 (1H, d, $J$=6.9 Hz, H-8), 7.74 (1H, s, NH-9), 7.17 (2H, d, $J$=8.4 Hz, H-2′, 6′), 7.47 (2H, d, $J$=8.4 Hz, H-3′, 5′); $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 57.3 (d, C-1), 42.5 (t, C-3), 22.4 (t, C-4), 110.3 (s, C-4a), 133.6 (s, C-4b), 121.9 (d, C-5), 119.4 (d, C-6), 118.2 (d, C-7), 110.8 (d, C-8), 140.8 (s, C-8a), 135.9 (s, C-9a), 127.2 (s, C-1′), 130.2 (d, C-2′, 6′), 131.8 (d, C-3′, 5′), 122.0 (s, C-4′); EI-MS $m/z$ 328 (43), 327 (37, $M^+$), 326 (48), 325 (34), 299 (17), 297 (18), 219 (28), 218 (100), 217 (93), 216 (30), 189 (9), 171 (64), 169 (17), 144 (19), 143 (21), 130 (15), 123 (27), 109 (55); HR-EI-MS $m/z$ 326.0418 (M+, Calcd for $C_{17}H_{15}N_2Br$, 326.0420).

1-(4′-Nitrophenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**6**): Yellow solid, mp 79 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$: 5.28 (1H, s, H-1), 3.17 (1H, m, H-3a), 3.28 (1H, m, H-3b), 2.87 (2H, m, H-4), 7.27 (1H, d, $J$=7.5 Hz, H-5), 7.17 (2H, t, $J$=7.5 Hz, H-6, 7), 7.57 (1H, d, $J$=7.5 Hz, H-8), 7.87 (1H, s, NH-9), 7.50 (2H, d, $J$=8.7 Hz, H-2′, 6′), 8.18 (2H, d, $J$=8.7 Hz, H-3′, 5′), $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 57.1 (d, C-1), 42.2 (t, C-3), 22.4 (t, C-4), 110.8 (s, C-4a), 127.1 (s, C-4b), 122.2 (d, C-5), 119.7 (d, C-6), 118.4 (d, C-7), 110.9 (d, C-8), 147.6 (s, C-8a), 135.6 (s, C-9a), 136.0 (s, C-1′), 129.4 (d, C-2′, 6′), 123.9 (d, C-3′, 5′), 149.3 (s, C-4′); EI-MS $m/z$ 293 (82, $M^+$), 292 (45), 264 (31), 247 (20), 218 (80), 217 (100), 216 (38), 204 (11), 189 (10), 171 (68), 169 (13), 154 (8), 144 (17), 143 (20), 130 (11), 115 (21), 109 (23); HR-EI-MS $m/z$ 293.1168 (M+, Calcd for $C_{17}H_{15}N_3O_2$, 293.1166).

1-(4′-*N*-Dimethylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**7**): Pale yellow solid, mp 235—237 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.11 (1H, s, H-1), 3.14 (1H, m, H-3a), 3.44 (1H, m, H-3b), 2.84 (2H, m, H-4), 7.56 (1H, d, $J$=7.0 Hz, H-5), 7.14 (2H, overlap, H-6, 7), 7.75 (1H, d, $J$=7.5 Hz, H-8), 7.17 (2H, d, $J$=8.4 Hz, H-2′, 6′), 6.69 (2H, d, $J$=8.4 Hz, H-3′, 5′), 2.95 (6H, s, Me); $^{13}$C-NMR (75 MHz, $CDCl_3$) $\delta$ 53.0 (d, C-1), 41.2 (t, C-3), 25.9 (t, C-4), 113.2 (s, C-4a), 127.2 (s, C-4b), 122.1 (d, C-5), 119.5 (d, C-6), 118.8 (d, C-7), 112.0 (d, C-8), 136.4 (s, C-8a), 132.9 (s, C-9a), 122.1 (s, C-1′), 127.4 (d, C-2′, 6′), 112.9 (d, C-3′, 5′), 149.4 (s, C-4′), 40.7 (q, Me); HR-EI-MS $m/z$ 291.1738 (M+, Calcd for $C_{19}H_{21}N_3$, 291.1735).

1-(4′-*N*-Diethylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**8**): Pale yellow solid, mp 282—284 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.04 (1H, s, H-1), 3.12 (1H, m, H-3a), 3.40 (1H, m, H-3b), 2.83 (2H, m, H-4), 7.15 (3H, overlap, H-5, 6, 7), 7.55 (1H, d, $J$=8.4 Hz, H-8), 8.05 (1H, s, H-9), 7.15 (2H, d, $J$=9 Hz, H-2′, 6′), 6.63 (2H, d, $J$=9 Hz, H-3′, 5′), 1.17 (6H, t, $J$=7.1 Hz, $CH_2\underline{C}H_3$), 3.35 (4H, q, $J$=7.1 Hz, $\underline{C}H_2CH_3$); $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 57.3 (d, C-1), 42.7 (t, C-3), 22.4 (t, C-4), 109.3 (s, C-4a), 127.8 (s, C-4b), 121.3 (d, C-5), 119.0 (d, C-6), 118.0 (d, C-7), 110.8 (d, C-8), 1335.7 (s, C-8a), 135.2 (s, C-9a), 127.4 (s, C-1′), 129.5 (d, C-2′, 6′), 111.6 (d, C-3′, 5′), 147.6 (s, C-4′), 12.5 (q, $CH_2\underline{C}H_3$), 44.3 (t, $\underline{C}H_2CH_3$); EI-MS $m/z$ 319 (96, $M^+$), 318 (100), 290 (28), 289 (29), 276 (19), 275 (17), 274 (12), 260 (5), 245 (9), 219 (11), 218 (39), 217 (41), 216 (12), 204 (5), 189 (5), 171 (28), 169 (14), 160 (10), 144 (13), 143 (21), 137 (18), 130 (15), 123 (14), 109 (14); HR-EI-MS $m/z$ 319.2045 (M+, Calcd for $C_{21}H_{25}N_3$, 319.2048).

1-(2′,4′-Dimethoxylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**9**): Pale yellow solid; mp 210—213 °C; IR (KBr) $\nu_{max}$ 3410, 2950, 1615, 1505, 1465, 1300, 1160, 1040, 835, 715 cm$^{-1}$; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.54 (1H, s, H-1), 3.11 (1H, m, H-3a), 3.27 (1H, m, H-3b), 2.84 (2H, m, H-4), 7.23 (1H, d, $J$=6.8 Hz, H-5), 7.13 (2H, overlap, H-6, 7), 7.53 (1H, d, $J$=6.8 Hz, H-8), 7.78 (1H, s, H-9), 6.53 (1H, d, $J$=2.4 Hz, H-3′), 6.37 (1H, d, $J$=8.7, 2.4 Hz, H-5′), 6.93 (1H, d, $J$=8.7 Hz, H-6′), 3.79, 3.87 (6H, s, OMe); $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 50.6 (d, C-1), 42.0 (t, C-3), 22.5 (t, C-4), 110.1 (s, C-4a), 127.4 (s, C-4b), 121.4 (d, C-5), 119.2 (d, C-6), 118.0 (d, C-7), 110.7 (d, C-8), 135.7 (s, C-8a), 134.6 (s, C-9a), 122.3 (s, C-1′), 98.8 (d, C-3′), 104.0 (d, C-5′), 110.7 (d, C-6′), 55.4, 55.6 (q, OMe); EI-MS $m/z$ 308 (100, $M^+$), 307 (73), 279 (31), 278 (43), 264 (12), 249 (13), 248 (30), 233 (9), 217 (7), 204 (18), 191 (8.4), 171 (38), 154 (17), 143 (21), 130 (13), 115 (15), 102 (13), 95 (9), 77 (13), 69 (22); HR-EI-MS $m/z$ 308.1523 (M+, Calcd for $C_{19}H_{20}N_2O_2$, 308.1526).

1-(3′,4′-Dimethoxylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**10**): Pale yellow solid; mp >300 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.04 (1H, s, H-1), 3.10 (1H, m, H-3a), 3.32 (1H, m, H-3b), 2.89 (2H, m, H-4), 7.12 (3H, overlap, H-5, 6, 7), 7.56 (1H, d, $J$=5.7 Hz, H-8), 8.32 (1H, s, H-9), 6.80 (1H, H-2′), 6.75 (1H, d, $J$=8.1 Hz, H-5′), 6.78 (1H, d, $J$=8.1 Hz, H-6′), 3.70 (3H, s, 3′-OMe), 3.84 (3H, s, 4′-OMe); $^{13}$C-NMR (75 MHz, $CDCl_3$) $\delta$ 57.9 (d, C-1), 42.9 (t, C-3), 22.3 (t, C-4), 109.7 (s, C-4a), 134.2 (s, C-4b), 121.4 (d, C-5), 120.5 (d, C-6), 119.0 (d, C-7), 111.2 (d, C-8), 135.8 (s, C-8a), 134.5 (s, C-9a), 127.2 (s, C-1′), 110.8 (d, C-2′), 148.6 (s, C-3′), 149.0 (s, C-4′), 110.8 (d, C-5′), 118.0 (d, C-6′), 55.7, 55.6 (q, OMe); EI-MS $m/z$ 308 (100, $M^+$), 307 (82), 291 (9.4), 279 (19), 264 (8), 249 (15), 248 (43), 233 (9), 217 (9), 204 (20), 191 (11), 171 (64), 154 (20), 143 (16), 130 (9), 115 (12), 102 (14), 95 (9), 77 (8); HR-EI-MS $m/z$ 308.1528 (M+, Calcd for $C_{19}H_{20}N_2O_2$, 308.1525).

1-(2′,5′-Dimethoxylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**11**): Pale yellow solid; mp >300 °C; IR (KBr) $\nu_{max}$ 3410, 2950, 1680, 1505, 1465, 1135, 1045, 750 cm$^{-1}$; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.64 (1H, s, H-1), 3.20 (1H, m, H-3a), 3.32 (1H, m, H-3b), 2.88 (2H, m, H-4), 7.23 (1H, d, $J$=6.8 Hz, H-5), 7.12 (2H, overlap, H-6, 7), 7.52 (1H, d, $J$=6.8 Hz, H-8), 7.85 (1H, s, H-9), 6.90 (1H, d, $J$=8 Hz, H-3′), 6.83 (1H, dd, $J$=8, 2 Hz, H-4′), 6.71 (1H, d, $J$=2 Hz, H-6′), 3.67, 3.81 (6H, s, OMe); $^{13}$C-NMR (75 MHz, $CDCl_3$) $\delta$ 51.2 (d, C-1), 42.0 (t, C-3), 21.8 (t, C-4), 109.8 (s, C-4a), 129.7 (s, C-4b), 121.7 (d, C-5), 119.3 (d, C-6), 118.1 (d, C-7), 110.8 (d, C-8), 127.1 (s, C-1′), 111.8 (d, C-3′), 113.6 (d, C-4′), 115.5 (d, C-6′), 55.7, 56.2 (q, OMe); EI-MS $m/z$ 308 (100, $M^+$), 307 (56), 292 (4), 279 (21), 264 (6), 249 (23), 248 (67), 233 (15), 217 (12), 204 (24), 191 (7.7), 171 (79), 169 (20), 154 (17), 144 (30), 143 (27), 130 (18), 115 (18), 102 (15), 69 (39); HR-EI-MS $m/z$ 308.1527 (M+, Calcd for $C_{19}H_{20}N_2O_2$, 308.1525).

1-(3′,5′-Dimethoxylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**12**): Pale yellow solid; mp 215—218 °C; IR (KBr) $\nu_{max}$ 3420, 2910, 1605, 1465, 1350, 1305, 1155, 1065, 845, 785 cm$^{-1}$; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.03 (1H, s, H-1), 3.11 (1H, m, H-3a), 3.35 (1H, m, H-3b), 2.91 (2H, m, H-4), 7.15 (3H, overlap, H-5, 6, 7), 7.56 (1H, d, $J$=7 Hz, H-8), 8.13 (1H, s, H-9), 6.47 (2H, d, $J$=2.1 Hz, H-2′, 6′), 6.44 (1H, d, $J$=2.1 Hz, H-4′), 3.71 (6H, s, OMe); $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 58.3 (d, C-1), 43.0 (t, C-3), 22.3 (t, C-4), 109.8 (s, C-4a), 127.3 (s, C-4b), 121.6 (d, C-5), 119.2 (d, C-6), 118.1 (d, C-7), 110.8 (d, C-8), 144.1 (s, C-8a), 135.8 (s, C-9a), 134.2 (s, C-1′), 106.5 (d, C-2′, 6′), 161.1 (s, C-3′, 5′), 100.0 (d, C-4′), 55.3 (q, OMe); EI-MS $m/z$ 308 (100, $M^+$), 307 (64), 279 (38), 278 (37), 264 (11), 249 (17), 248 (30), 233 (10), 220 (9), 217 (9), 204 (21), 191 (10), 171 (85), 154 (13), 144 (17), 130 (10), 115 (16), 102 (13), 95 (9), 77 (10); HR-EI-MS $m/z$ 308.1526 (M+, Calcd for $C_{19}H_{20}N_2O_2$, 308.1525).

1-(3′,4′,5′-Trimethoxylphenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**13**): Pale

yellow solid, mp 161 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.85 (1H, s, H-1), 3.59 (1H, m, H-3a), 3.68 (1H, m, H-3b), 3.29 (2H, m, H-4), 7.32 (1H, d, $J$=8 Hz, H-5), 7.16 (1H, t, $J$=8 Hz, H-6), 7.11 (1H, $J$=8 Hz, H-7), 7.55 (1H, d, $J$=8 Hz, H-8), 6.73 (2H, s, H-2′, 6′), [3.71 (9H, s, OMe)]; $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 56.1 (d, C-1), 41.7 (t, C-3), 51.5 (t, C-4), 109.4 (s, C-4a), 126.7 (s, C-4b), 122.4 (d, C-5), 119.7 (d, C-6), 118.4 (d, C-7), 111.1 (d, C-8), 136.2 (s, C-8a), 133.3 (s, C-9a), 128.6 (s, C-1′), 106.1 (d, C-2′, 6′), 153.4 (s, C-3′, 5′), 153.8 (s, C-4′), 56.1 (q, 3′, 5′-OMe), 60.7 (q, 4′-OMe); EI-MS $m/z$ 338 (100, $M^+$), 337 (67), 309 (17), 279 (14), 278 (39), 262 (6.7), 247 (9), 234 (7.8), 219 (6.9), 204 (6.7), 191 (10), 180 (12), 171 (69), 169 (24), 154 (13), 144 (20), 130 (11), 115 (15), 109 (12), 77 (11), 69 (54); HR-EI-MS $m/z$ 338.1629 (M+, Calcd for $C_{20}H_{22}N_2O_3$, 338.1630).

1-(6′-Nitro-1′-piperoyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**14**): Yellow solid, mp 267 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.66 (1H, s, H-1), 3.17 (1H, m, H-3a), 3.25 (1H, m, H-3b), 2.89 (2H, m, H-4), 7.23 (1H, d, $J$=7.6 Hz, H-5), 7.16 (2H, overlap, H-6, 7), 7.56 (1H, d, $J$=7.6 Hz, H-8), 8.04 (1H, s, H-9), 6.77 (1H, s, H-2′), 7.39 (1H, s, H-5′), 6.01 (2H, d, $J$=4.2 Hz, $OCH_2O$); $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 52.4 (d, C-1), 42.0 (t, C-3), 22.1 (t, C-4), 111.1 (s, C-4a), 127.0 (s, C-4b), 122.0 (d, C-5), 119.5 (d, C-6), 118.3 (d, C-7), 110.9 (d, C-8), 136.0 (s, C-8a), 134.1 (s, C-9a), 132.6 (s, C-1′), 109.9 (d, C-2′), 151.7 (s, C-3′), 147.3 (s, C-4′), 104.9 (s, C-5′), 143.7 (s, C-6′), 102.9 (t, $OCH_2O$); EI-MS $m/z$ 337 (6, $M^+$), 335 (6.7), 320 (9), 319 (70), 303 (56), 290 (93), 289 (100), 275 (6), 261 (24), 244 (11), 232 (13), 216 (9), 204 (33), 203 (19), 191 (18), 171 (20), 151 (18), 144 (32), 130 (53), 115 (50), 102 (52), 101 (22), 89 (20), 77 (25); HR-EI-MS $m/z$ 337.1066 (M+, Calcd for $C_{18}H_{15}N_3O_4$, 337.1064).

1-(2′-Fluorenyl)-1,2,3,4-tetrahydro-$\beta$-carboline (**15**): Yellow solid; mp >300 °C; IR (KBr) $\nu_{max}$ 3540, 2990, 1680, 1710, 1465, 745 cm$^{-1}$; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 5.23 (1H, s, H-1), 3.17 (1H, m, H-3a), 3.41 (1H, m, H-3b), 2.94 (2H, m, H-4); $^{13}$C-NMR (75 MHz, $CDCl_3$): $\delta$ 58.3 (d, C-1), 43.0 (t, C-3), 22.6 (t, C-4), 110.2 (s, C-4a), 127.5 (s, C-4b), 121.7 (d, C-5), 119.4 (d, C-6), 118.3 (d, C-7), 110.9 (d, C-8), 143.9 (s, C-8a), 134.8 (s, C-9a), 125.1 (s, C-1′), 135.9 (s, C-2′), 125.1 (d, C-3′), 120.0 (d, C-4′), 141.2 (s, C-4′a), 141.8 (s, C-4′b), 120 (d, C-5′), 126.9 (d, C-6′), 126.8 (d, C-7′), 125.1 (d, C-8′), 143.5 (s, C-8′a), 36.9 (t, C-9′); EI-MS $m/z$ 336 (100, $M^+$), 335 (87), 334 (60), 307 (73), 306 (86), 304 (50), 292 (14), 171 (52), 152 (55), 143 (26), 115 (28); HR-EI-MS $m/z$ 336.1626 (M+, Calcd for $C_{24}H_{20}N_2$, 336.1625).

1-(9′-Ethyl-3′-carbazole)-1,2,3,4-tetrahydro-$\beta$-carboline (**16**): Yellow solid; mp 242 °C; IR (KBr) $\nu_{max}$ 3470, 2930, 1465, 1390, 750 cm$^{-1}$; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 6.04 (1H, s, H-1), 3.56 (1H, m, H-3a), 3.66 (1H, m, H-3b), 3.18 (1H, m, H-4a), 3.28 (1H, m, H-4b); $^{13}$C-NMR ($CDCl_3$) $\delta$ 59.2 (d, C-1), 42.3 (t, C-3), 19.8 (t, C-4), [110.2 (s, C-4a), 127.5 (s, C-4b), 121.7 (d, C-5), 119.4 (d, C-6), 118.3 (d, C-7), 110.9 (d, C-8), 143.9 (s, C-8a), 125.1 (s, C-1′), 135.9 (s, C-2′), 125.1 (d, C-3′), 120.0 (d, C-4′), 141.2 (s, C-4′a), 141.8 (s, C-4′b), 120 (d, C-5′), 126.9 (d, C-6′), 126.8 (d, C-7′), 125.1 (d, C-8′), 143.5 (s, C-8′a), 36.9 (t, C-9′)]; EI-MS $m/z$ 365 (100, $M^+$), 364 (8), 336 (46), 335 (45), 319 (8), 306 (18), 223 (5.7), 208 (7.3), 183 (18), 171 (39), 160 (51), 153 (18), 144 (15), 130 (5), 115 (11); HR-EI-MS $m/z$ 365.1897 (M+, Calcd for $C_{25}H_{23}N_3$, 365.1894).

**Synthesis of Compounds 17—29** To a stirred solution of compound A (**4**—**16**, 0.03 mmol) in EtOH (1 ml) and $CHCl_3$ (3 ml) at room temperature was added 2,3-dichloro-5,6-dicyanobezoquinone (DDQ, 40 mg). The reaction mixture was stirred for 30 min. After concentration, the residue was subjected to prep. TLC and developed with $CHCl_3$/MeOH (10 : 1) to yield compound B (**17**—**29**, 30—75%).

1-(4′-Chlorophenyl)-3,4-dihydro-$\beta$-carboline (**17**): Yellow solid, mp 102—104 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.06 (2H, t, $J$=8.3 Hz, H-3), 2.99 (2H, t, $J$=8.3 Hz, H-4), 7.39 (1H, d, $J$=7.7 Hz, H-5), 7.32 (1H, dd, $J$=7.7, 7.2 Hz, H-6), 7.20 (1H, dd, $J$=7.8, 7.2 Hz, H-7), 7.67 (1H, d, $J$=7.8 Hz, H- 8), 8.04 (1H, s, NH-9), 7.49 (2H, d, $J$=8.6 Hz, H-2′, 6′), 7.71 (2H, d, $J$=8.6 Hz, H-3′, 5′); HR-EI-MS $m/z$ 280.0764 (M+, Calcd for $C_{17}H_{13}N_2Cl$, 280.0767).

1-(4′-Bromophenyl)-3,4-dihydro-$\beta$-carboline (**18**): Yellow solid, mp 102—104 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.05 (2H, t, $J$=7.6 Hz, H-3), 2.98 (2H, t, $J$=7.6 Hz, H-4), 7.39 (1H, d, $J$=8.0 Hz, H-5), 7.31 (1H, d, $J$=8.0 Hz, H-6), 7.20 (1H, t, $J$=7.2 Hz, H-7), 7.60 (1H, d, $J$=7.2 Hz, H-8), 8.00 (1H, s, NH-9), 7.47 (4H, s, H-2′, 3′, 5′, 6′); HR-EI-MS $m/z$ 325.0342 (M+, Calcd for $C_{17}H_{13}N_2Br$, 325.0340).

1-(4′-Nitrophenyl)-3,4-dihydro-$\beta$-carboline (**19**): Yellow solid, mp 211—213 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.12 (2H, t, $J$=8.6 Hz, H-3), 2.99 (2H, t, $J$=8.6 Hz, H-4), 7.41 (1H, d, $J$=7.7 Hz, H-5), 7.34 (1H, t, $J$=7.8 Hz, H-6), 7.22 (1H, t, $J$=7.8 Hz, H-7), 7.68 (1H, d, $J$=7.8 Hz, H-8), 7.95 (2H, d, $J$=8.4 Hz, H-2′, 6′), 8.37 (2H, d, $J$=8.4 Hz, H-3′, 5′); HR-ESI-MS $m/z$ 292.1085 (M+H+, Calcd for $C_{17}H_{14}N_3O_2$, 292.1086).

1-(4′-*N*-Dimethylphenyl)-3,4-dihydro-$\beta$-carboline (**20**): Yellow solid, mp 150 °C; $^1$H-NMR (300 MHz, $CD_3OD$): $\delta$ 3.94 (2H, t, $J$=7.8 Hz, H-3), 3.31 (2H, overlap, H-4), 7.55 (1H, d, $J$=8.4 Hz, H-5), 7.21 (1H, t, $J$=7.5 Hz, H-6), 7.40 (1H, t, $J$=7.5 Hz, H-7), 7.75 (1H, d, $J$=8.1 Hz, H-8), 7.81 (2H, d, $J$=9.0 Hz, H-2′, 6′), 6.94 (2H, d, $J$=9.0 Hz, H-3′, 5′), 3.16 (6H, s, NMe2); HR-EI-MS $m/z$ 289.1581 (M+, Calcd for $C_{19}H_{19}N_3$, 289.1578).

1-(4′-*N*-Diethylphenyl)-3,4-dihydro-$\beta$-carboline (**21**): Yellow solid, mp 244—246 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 3.83 (2H, dd, $J$=7.8, 15.3 Hz, H-3), 3.09 (2H, t, $J$=7.2, 15.0 Hz, H-4), 7.62 (1H, d, $J$=8.4 Hz, H-5), 7.20 (1H, t, $J$=7.8 Hz, H-6), 7.38 (1H, t, $J$=7.8 Hz, H-7), 7.68 (1H, d, $J$=7.8 Hz, H-8), 10.6 (1H, br s, NH-9), 7.94 (2H, d, $J$=9.0 Hz, H-2′, 6′), 6.55 (2H, d, $J$=9.0 Hz, H-3′, 5′), 1.08 (6H, t, $J$=6.9 Hz, $CH_2\underline{CH}_3$), 3.23 (4H, q, $J$=6.9 Hz, $CH_2CH_3$); HR-EI-MS $m/z$ 317.1889 (M+, Calcd for $C_{19}H_{18}N_3$, 317.1892).

1-(2′,4′-Dimethoxylphenyl)-3,4-dihydro-$\beta$-carboline (**22**): Yellow solid, mp 165—168 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.07 (2H, t, $J$=8.4 Hz, H-3), 3.10 (2H, t, $J$=8.4 Hz, H-4), 7.54 (1H, d, $J$=8.4 Hz, H-5), 7.41 (1H, dd, $J$=8.4 Hz, H-6), 7.34 (1H, t, $J$=8.4 Hz, H-7), 7.63 (1H, d, $J$=8.4 Hz, H-8), 6.53 (1H, d, $J$=1.8 Hz, H-3′), 6.57 (1H, d, $J$=7.2, 1.8 Hz, H-5′), 7.19 (1H, d, $J$=7.2 Hz, H-6′), 3.82, 3.84 (6H, s, OMe); HR-EI-MS $m/z$ 306.1365 (M+, Calcd for $C_{19}H_{18}N_2O_2$, 306.1368).

1-(3′,4′-Dimethoxylphenyl)-3,4-dihydro-$\beta$-carboline (**23**): Yellow solid, mp 288—290 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 3.98 (2H, t, $J$=7.8 Hz, H-3), 2.96 (2H, t, $J$=7.8 Hz, H-4), 7.40 (1H, d, $J$=8.4 Hz, H-5), 7.20 (1H, d, $J$=8.4 Hz, H-6), 7.66 (1H, d, $J$=8.4 Hz, H-8), 7.30 (1H, d, $J$=1.8 Hz, H-2′), 6.86 (1H, d, $J$=7.8 Hz, H-5′), 7.15 (1H, d, $J$=7.8 Hz, H-6′), 3.77, 3.82, 3.84 (6H, s, OMe); HR-EI-MS $m/z$ 306.1366 (M+, Calcd for $C_{19}H_{18}N_2O_2$, 306.1368).

1-(2′,5′-Dimethoxylphenyl)-1,2,3,4-dihydro-$\beta$-carboline (**24**): Yellow solid, mp 175—176 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.11 (2H, t, $J$=8.6 Hz, H-3), 2.99 (2H, t, $J$=8.6 Hz, H-4), 7.34 (1H, d, $J$=8.1 Hz, H-5), 7.27 (1H, t, $J$=7.4 Hz, H-6), 7.15 (1H, t, $J$=7.4 Hz, H-7), 7.63 (1H, d, $J$=8.1 Hz, H-8), 8.50 (1H, br s, H-9), 7.00 (2H, s, H-3′, 4′), 7.06 (1H, s, H-6′), 3.81 (6H, s, OMe); HR-EI-MS $m/z$ 306.1370 (M+, Calcd for $C_{19}H_{18}N_2O_2$, 306.1368).

1-(3′,5′-Dimethoxylphenyl)-3,4-dihydro-$\beta$-carboline (**25**): Yellow solid, mp 111—113 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.05 (2H, t, $J$=8.4 Hz, H-3), 2.98 (2H, t, $J$=8.4 Hz, H-4), 7.37 (1H, d, $J$=8.1 Hz, H-5), 7.18 (1H, t, $J$=7.5 Hz, H-6), 7.31 (1H, t, $J$=7.5 Hz, H-7), 7.66 (1H, d, $J$=8.1 Hz, H-8), 8.38 (1H, s, H-9), 6.88 (2H, d, $J$=2.1 Hz, H-2′, 6′), 6.58 (1H, dd, $J$=2.4, 2.1 Hz, H-4′), 3.83 (6H, s, OMe); HR-ESI-MS $m/z$ 307.1450 (M+H+, Calcd for $C_{19}H_{19}N_2O_2$, 307.1447).

1-(3′,4′,5′-Trimethoxylphenyl)-3,4-tetrahydro-$\beta$-carboline (**26**): Yellow solid, mp 251 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.06 (2H, dd, $J$=8.1, 16.5 Hz, H-3), 3.01 (2H, dd, $J$=8.1, 16.5 Hz, H-4), 7.43 (1H, d, $J$=8.1 Hz, H-5), 7.22 (1H, dd, $J$=8.1, 7.8 Hz, H-6), 7.36 (1H, t, $J$=7.8 Hz, H-7), 7.69 (1H, d, $J$=7.8 Hz, H-8), 8.28 (1H, br s, H-9), 6.99 (2H, s, H-2′, 6′), 3.92 (9H, s, OMe); HR-EI-MS $m/z$ 336.1476 (M+, Calcd for $C_{20}H_{20}N_2O_3$, 336.1473).

1-(6′-Nitro-1′-piperoyl)-3,4-dihydro-$\beta$-carboline (**27**): Yellow solid, mp 191—193 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 4.08 (2H, t, $J$=8.6 Hz, H-3), 3.06 (2H, t, $J$=8.6 Hz, H-4), 7.30 (1H, overlap, H-5), 7.20 (2H, overlap, H-6, 7), 7.65 (1H, d, $J$=7.8 Hz, H-8), 8.11 (1H, s, H-9), 6.94 (1H, s, H-2′), 7.63 (1H, s, H-5′), 6.19 (2H, s, $OCH_2O$); HR-EI-MS $m/z$ 335.0903 (M+, Calcd for $C_{18}H_{13}N_3O_4$, 335.0907).

1-(2′-Fluorenyl)-3,4-dihydro-$\beta$-carboline (**28**): Yellow solid, mp 121—123 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 3.03 (2H, t, $J$=7.8 Hz, H-3), 4.09 (2H, t, $J$=7.8 Hz, H-4), 3.99 (2H, s, H-9′), 7.20 (1H, t), 7.35 (1H, t), 7.40 (3—4H, m), 7.58 (1H, d), 7.66 (1H, d, $J$=7.8 Hz), 7.80 (1H, d), 7.83 (1H, d), 7.88 (1H, d, $J$=8 Hz), 8.00 (1H, s); HR-EI-MS $m/z$ 334.1468 (M+, Calcd for $C_{24}H_{18}N_2$, 334.1471).

1-(9′-Ethyl-3′-carbazole)-3,4-dihydro-$\beta$-carboline (**29**): Yellow solid, mp 175—177 °C; $^1$H-NMR (300 MHz, $CDCl_3$): $\delta$ 3.01 (2H, m, H-3), 2.60 (2H, m, H-4), 1.41 (3H, t, $J$=7.2 Hz, $CH_2C\underline{H}_3$), 4.32 (2H, q, $J$=7.2 Hz, $C\underline{H}_2CH_3$), 7.0—7.6 (m), 8.00 (1H, d), 8.11 (1H, s); HR-EI-MS $m/z$ 363.1724 (M+, Calcd for $C_{25}H_{21}N_3$, 363.1735).

**Biological Assay** The cytotoxic activities of compounds against P-388, KB-16, A-549, and HT-29 cells were assayed by the MTT {3-(4,5-dimethylthiazole-2-yl)-2,5-diphenyltetrazolium bromide} colorimetric assay with some modifications.[24] Each cell line was plated at 1000 cells/well in 96-well microtiter plates. Two-fold serial dilutions of tested compounds were added to the cells, and P-388, KB-16, A549, and HT29 cells were enumerated using MTT after 3, 3, 6 and 6 d, respectively. MTT (50 $\mu$l, 1 mg/ml)

was added to each well, and the plates were incubated at 37 °C for 5 h. Formazan crystals were redissolved in DMSO (Merck) for 10 min with shaking, and plates were then immediately read on a microtiter plate reader (Dynatech) at 540 nm. The $ID_{50}$ was defined as the concentration of test compound resulting in a 50% reduction of absorbance compared to untreated cells in the MTT assay. Manzamine A (**3**) and Paclitaxel (**30**) were used as standard compounds. Results are given in Table 1.

**Acknowledgments** We thank Ms. Ho Chao Lein and Yu Shiu Ching of the NSC Southern NMR and MS Instrument Center for measurement of NMR an MS spectral data. This research was supported by grants from the National Science Council, Republic of China (NSC 85-2321-B110-003 BH) and by the National Institute of Health, Republic of China ( DOH 86-HR-509) awarded to Y.-C.S.

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