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Total Syntheses of Makaluvamines A, B, C, D and E, Cytotoxic Pyrroloiminoquinone Alkaloids Isolated from Marine Sponge Bearing Inhibitory Activities against Topoisomerase II

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Abstract: Syntheses of makaluvamines A, B, C, D and E (1 – 5), new members of tetrahydropyrroloiminoquinone alkaloids, have been successfully carried out. Particularly, olefin introduction for makaluvamines B and E could be achieved by Pd - mediated and E_2 type methodologies.

Among a number of bioactive marine natural products, pyrroloquinoline alkaloids isolated from marine sponges[1] share an indole-based tricyclic structure, and particularly such members bearing iminoquinone moieties as prianosins (discorhabdins) were found to exhibit potent cytotoxicities. Recently, makaluvamines, new members of the alkaloids were isolated from the Fijian sponge *Zyzza* cf. *marsailis* (A – F)[2a] and the Indonesian sponge *Histodermella* sp. (G).[2b] In addition to cytotoxicities, the Fijian makaluvamines are potent topoisomerase II inhibitors, whereas the other exhibits a moderate inhibitory effect against topoisomerase I and not against topoisomerase II. The unique highly fused skeletons carrying the interesting biological character have served additional targets related to the pyrroloquinoline alkaloids, which have been synthetically challenged

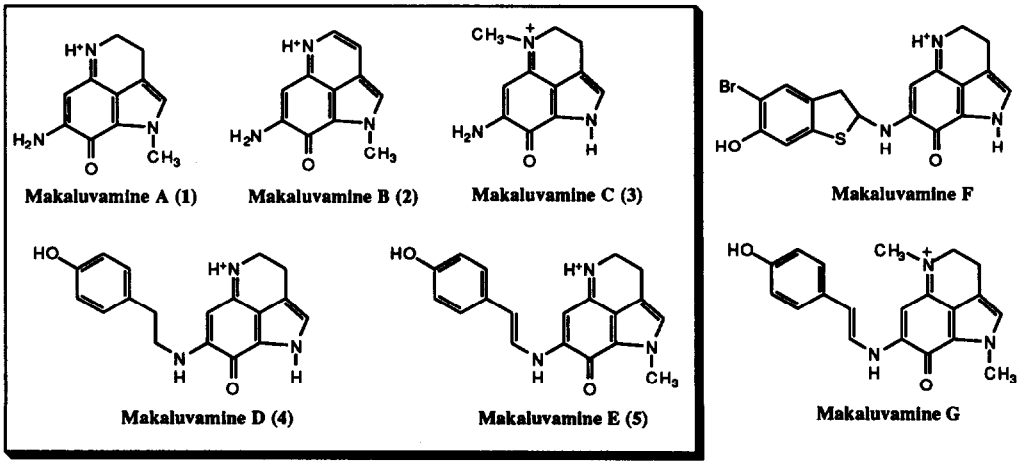
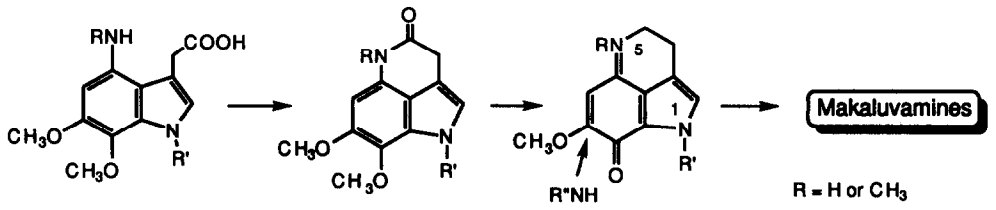


Figure 1. Structures of Makaluvamines.

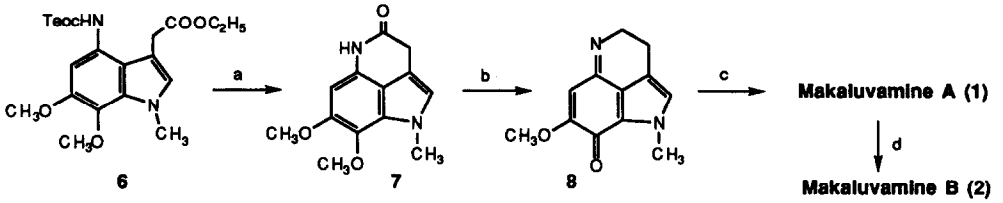


Scheme 1. General Synthetic Route to Makaluvamines

by diverse approaches[3] including the first total syntheses of discorhabdin C, batzelline C and isobatzelline C by us.[4] Based on a synthetic potential obtained by our own total syntheses, we initiated syntheses of makaluvamines. In the synthetic process, the pyrroloquinoline core was composed by cyclization of indole derivatives followed by reduction, oxidation and final attachment of amino functions (Scheme 1). This sequence facilitated introduction of N-methyl groups located at 1 and/or 5 positions, which might be a factor to control biological activities. We describe herein details of our investigation.[5]

RESULTS AND DISCUSSIONS

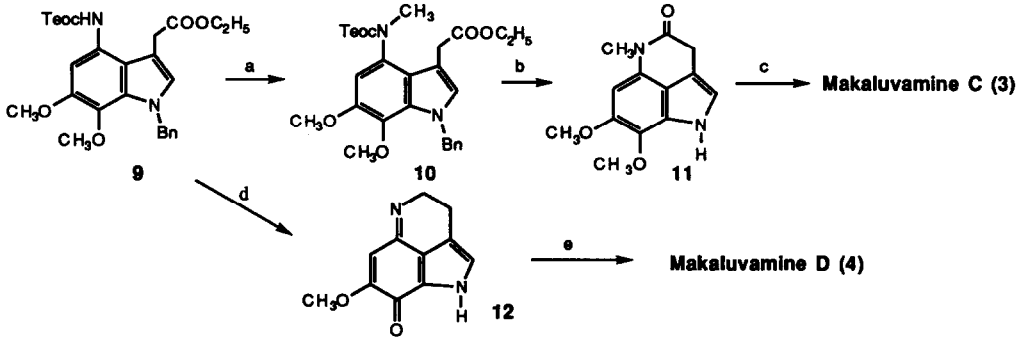
According to the above-mentioned methodology, the synthesis was initiated by conversion of the previously reported indole (**6**)[4] into lactam **7** which was effected by the successive deprotection followed by DCC-mediated lactamization. Treatment of **7** with BH_3•SMe_2 provided the corresponding amine in good yield, which on CAN oxidation yielded the desired iminoquinone (**8**). Upon reaction with NH_4Cl, a methoxy group of **8** could be smoothly transformed into an amino group, leading to makaluvamine A (**1**). This conversion required slightly acidic conditions to activate the C_7 position, and actually such basic conditions as NH_3/MeOH and NH_4OH did not provide the desired product.[4d] Makaluvamine B (**2**) possesses the same framework as **1**, except for a double bond between C_3 and C_4 positions. Accordingly, introduction of a double bond to **1** was elaborated. Among several methodologies, Pd-mediated dehydration protocol could realize our expectation to produce **2**.



Scheme 2. *reagents*: a. i) 60%$HClO_4$; ii) KOH; iii) DCC (73%). b. i) BH_3•SMe_2; ii) CAN / 60% aq. $HClO_4$ (60%). c. NH_4Cl (98%). d. 10% Pd-C, Δ (41%).

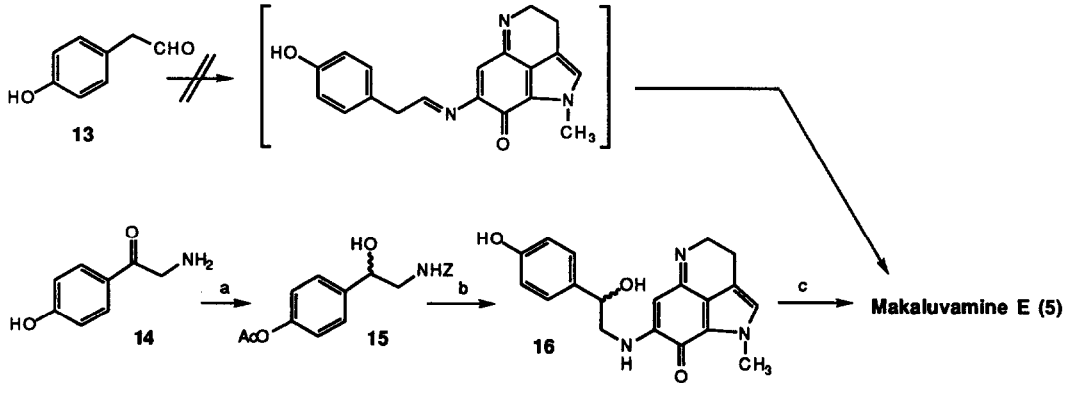
Makaluvamine C (**3**) is regarded as a regio-isomer of **1**, and introduction of a methyl group was performed in relatively early stage; appropriately protected indole **9**[4d] was methylated in a usual manner to give **10**. Hydrogenation under acidic conditions simultaneously to remove benzyl and Teoc groups, followed by alkaline hydrolysis afforded an unstable amino acid, which was directly treated with DCC to yield lactam **11** in 41% from **10**. After BH_3•SMe_2 reduction, CAN oxidation yielded an unstable imine, which was immediately submitted to silica gel supported amination employing column chromatography ($CHCl_3$ – MeOH containing NH_4OH), from

which **3** was obtained in 26% yield. The amination procedure used in the case of **1** was unsuccessful, probably due to an unstable character of a methylated iminoquinone structure. Up to now, the chromatographic approach might be the only procedure to achieve the purpose. Additionally, makaluvamine D (**4**) was synthesized in 92% yield by coupling of the known iminoquinone (**12**)[4d] obtained from **9** with tyramine hydrochloride.



Scheme 3. *reagents*: a. MeI, NaH (77%). b. i) H_2, Pd-black / AcOH-60% $HClO_4$; ii) KOH; iii) DCC (41%). c. i) BH_3•SMe_2; ii) CAN; iii) NH_4OH, silica gel (26%). d. ref. 4d. e. tyramine hydrochloride (92%).

At the outset, a synthesis of makaluvamine E (**5**) was attempted by condensation of **1** with the known aldehyde **13**[6] followed by a 1,3-proton shift of the corresponding imine. However, no desired reactions were observed under heating conditions (80 ~ 100 °C in DMF, Molecular Sieves, with or without such catalysts as TFA). Therefore, elaboration was focused on an assembly of an amino substituent containing an olefinic bond. Ultimately, Lewis acid promoted removal of an leaving group at the benzylic position was a method of choice. Thus, the known amino ketone **14**[7] was protected with benzyloxycarbonyl (Z) and acetyl groups, followed by reaction with $NaBH_4$ to give **15** in 80% yield. Compound **15** was submitted to catalytic hydrogenation to remove a Z group, and coupling with **8** furnished the desired **16** in 97% yield based on **8**. Fortunately, a phenolic acetyl group was removed during chromatographic purification involving small amounts of NH_4OH. Heating of **16**



Scheme 4. *reagents*: a. i) ZCl, $NaHCO_3$; ii) K_2CO_3 / MeOH; iii) Ac_2O, pyr. (98%); iv) $NaBH_4$ (81%). b. i) H_2, 10% Pd-C; ii) **8** (97%). c. BF_3•OEt_2, 100 °C (60%).

with BF_3•OEt_2 took place successful olefin introduction to produce makaluvamine E (**5**) in 60% yield. To our knowledge, this conversion might be the first example in the presence of a tetrahydropyrroloquinone residue.

All of the TFA salts of makaluvamines A – E (**1** – **5**) synthesized here were nicely matched to the reported data. Further synthetic investigation of this class of natural products and biological evaluation of related derivatives are in progress.

EXPERIMENTAL

IR spectra were recorded on a JASCO Model A-202 spectrophotometer. ^1H NMR and ^{13}C NMR spectra were obtained on a JEOL JNM EX-270, a JEOL JNM GX-400 NMR or a JEOL JNM ALPHA-400 spectrometer in a deuteriochloroform ($CDCl_3$) solution using tetramethylsilane as an internal standard, unless otherwise stated. High resolution mass spectra were obtained on a Hitachi M-80 GC-MS spectrometer operating at the ionization energy (70 eV). Preparative and analytical TLC were carried out on silica gel plates (Kieselgel 60 F_{254}, E. Merck A. G., Germany) using UV light and/or 5% molybdophosphoric acid in ethanol for detection. Katayama silica gel (K 070) was used for column chromatography.

Preparation of TFA salts: Brown solids of synthetic makaluvamines (0.5 mmol) were dissolved in MeOH (10 mL). After the addition of 1M TFA in MeOH (0.5 mL, 0.5 mmol), the solution was evaporated to dryness to give dark green solids of the corresponding TFA salts.

7,8-Dimethoxy-1-methyl-1,3,4,5-tetrahydropyrrolo[4,3,2-*de*]quinolin-4-one (7).

To a solution of **6** (2.10 g, 4.75 mmol) in AcOH (20 mL) was added 60% $HClO_4$ (1 mL); the mixture was stirred at room temperature for 1 h, and poured into ice-cooled sat. aq. $NaHCO_3$. The mixture was extracted with EtOAc (100 mL x 2), and the organic extracts were washed with H_2O and brine, dried (Na_2SO_4), then evaporated to dryness. The residue was dissolved in EtOH (20 mL), and 10M KOH (1.9 mL) was added. After being stirred at room temperature for 30 min, the reaction mixture was acidified to pH ~3 by the addition of 4M HCl, then the solvent was removed in vacuo. To a solution of the residue in THF (25 mL) was added DCC (1.48 g, 7.1 mmol) below -15°C. After being stirred at the same temperature for 1.5 h, the mixture was gradually warmed up to room temperature during overnight. The resulting mixture was filtered, and the filtrate was concentrated in vacuo to give a crude product, which on purification by silica gel column chromatography ($CHCl_3$ / MeOH = 20:1) provided **7** (847 mg, 73%) as an oil: IR (film) 1665, 1645, 1615, 1460 cm^{-1}; δ_H 3.89 (3H, s), 3.90 (3H, s), 3.93 (3H, s), 3.94 (2H, d, J= 1 Hz), 6.13 (1H, s), 6.55 (1H, t, J= 1 Hz), 7.80 (1H, broad s); δ_C (DMSO-d_6) 31.0 (t), 34.4 (q), 57.8 (q), 61.7 (q), 91.8 (d), 106.2 (s), 111.8 (s), 112.3 (s), 123.4 (d), 127.7 (s), 149.0 (s), 169.0 (s). Found: *m/z* 246.0995. Calcd for $C_{13}H_{14}N_2O_3$: M, 246.1003.

7-Methoxy-1-methyl-1,3,4,8-tetrahydropyrrolo[4,3,2-*de*]quinolin-8-one (8).

To a solution of **7** (840 mg, 3.4 mmol) in dry THF (30 mL) was added BH_3•SMe_2 (10 M solution in THF, 1.7 mL, 17 mmol) at 0 °C under a nitrogen atmosphere; the mixture was stirred at room temperature for 3 h. After the addition of H_2O (20 mL), the resulting mixture was extracted with EtOAc (50 mL x 2), and the organic extracts were washed with sat. aq. $NaHCO_3$ and brine, dried (Na_2SO_4), then evaporated to dryness. The residue was dissolved in 60% aq. CH_3CN (40 mL), and CAN (4.0 g, 7.5 mmol) was added at 0 °C. After 5 min, the reaction mixture was diluted with $CHCl_3$, washed with H_2O and brine, dried (Na_2SO_4), then evaporated. Chromatographic purification ($CHCl_3$ / MeOH = 8:1) of a crude product afforded **8** (445 mg, 60%) as an

amorphous solid: IR (nujol) 1650, 1615, 1570, 1535, 1460 cm^{-1}; δ_H (CD_3OD) 2.68 (2H, t, J= 8 Hz), 3.77 (3H, s), 3.84 (3H, s), 3.98 (2H, t, J= 8 Hz), 5.90 (1H, s), 6.83 (1H, s); δ_C (CD_3OD) 18.9 (t), 35.7 (q), 50.9 (q), 57.0 (q), 105.5 (d), 116.2 (s), 117.8 (s), 129.8 (d), 143.0 (s), 158.1 (s), 160.0 (s), 172.1 (s). Found: *m/z* 216.0903. Calcd for $C_{12}H_{12}N_2O_2$: M, 216.0898.

Makaluvamine A (1).

A mixture of **8** (113 mg, 0.5 mmol) and NH_4Cl (270 mg) in MeOH (25 mL) was stirred at room temperature overnight. After evaporation, the residue was chromatographed on silica gel ($CHCl_3$ / MeOH / NH_4OH = 3:1:0.06) to give **1** (103 mg, 98%), which was treated with TFA in MeOH, leading to the corresponding TFA salt: IR (KBr) 1670, 1605 cm^{-1}; δ_H (DMSO-d_6) 2.84 (2H, t, J= 7.6 Hz), 3.76 (2H, t, J= 7.6 Hz), 3.89 (3H, s), 5.62 (1H, s), 7.31 (1H, s), 8.38 (1H, broad s), 9.15 (1H, broad s), 10.60 (1H, broad s); δ_C (DMSO-d_6) 18.0 (t), 35.8 (q), 42.0 (t), 86.5 (d), 117.8 (s), 122.3 (s), 123.0 (s), 131.0 (d), 155.9 (s), 156.7 (s), 168.2 (s). Found: *m/z* 202.0983. Calcd for $C_{11}H_{12}N_3O$: M+H, 202.0980.

Makaluvamine B (2).

A suspension of **1** (18.4 mg, 0.09 mmol) and 10% Pd-C (5 mg) in PhH (13 mL) was refluxed for 5 days. After filtration, the filtrate was evaporated to give a residue. Chromatographic purification on silica gel ($CHCl_3$ / MeOH = 5:1) afforded **2** (7.5 mg, 41%), which was characterized as a TFA salt: IR (KBr) 1680, 1600, 1495, 1330 cm^{-1}; δ_H (DMSO-d_6) 3.42 (2H, broad s), 4.28 (3H, s), 6.34 (1H, s), 7.61 (1H, d, J= 6.8 Hz), 7.95 (1H, d, J= 6.8 Hz), 8.17 (1H, broad s), 8.35 (1H, s); δ_C (DMSO-d_6) 38.0 (q), 88.4 (d), 111.3 (d), 120.6 (s), 122.5 (s), 129.9 (d), 133.4 (d), 144.0 (s), 155.7 (s), 166.3 (s). Found: *m/z* 199.0748. Calcd for $C_{11}H_9N_3O$: M, 199.0745.

1-Benzyl-3-ethoxycarbonylmethyl-6,7-dimethoxy-4-[N-methyl-(2-trimethylsilylethoxy)carbonyl-amino]indole (10).

To a suspension of NaH (60% dispersion in mineral oil, 63 mg, 1.6 mmol) in dry DMF (2 mL) was added **9**[4d] (449 mg, 0.88 mmol) in dry DMF (3.5 mL) at -50 °C followed by CH_3I (0.14 mL, 2.2 mmol); the resulting mixture was gradually warmed to room temperature for 3 h. After the addition of sat. aq. NH_4Cl (10 mL), the mixture was extracted with hexane - EtOAc (1:1, 20 mL x 3). The organic extracts were combined, washed with H_2O and brine, dried (Na_2SO_4), then evaporated to dryness. The residue was purified by a silica gel column (hexane / EtOAc = 4:1) to yield **10** (356 mg, 77%) as an oil: IR (film) 2950, 1735, 1700, 1520 cm^{-1}; δ_H -0.18 (9H, s), 0.83 (2H, t, J= 7.1 Hz), 1.27 (3H, t, J= 7.3 Hz), 3.23 (3H, s), 3.64 (3H, s, overlapped with 2H signal), 3.85 (3H, s), 4.10 ~ 4.20 (2h, complex), 4.15 (2H, q, J= 7.3 Hz), 5.52 (2H, s), 6.58 (1H, broad s), 6.96 (1H, s), 7.1 ~ 7.3 (5H, complex). Found: *m/z* 526.2480. Calcd for $C_{28}H_{38}N_2O_6Si$: M, 199.0745.

7,8-Dimethoxy-5-methyl-1,3,4,5-tetrahydropyrrolo[4,3,2-*de*]quinolin-4-one (11).

A solution of **10** (480 mg, 0.91 mmol) in AcOH (10 mL) and 60% $HClO_4$ (1 mL) containing Pd-black (700 mg) was stirred at room temperature for 2.5 h under a hydrogen atmosphere. After filtration, the filtrate was evaporated to give a residue, which was diluted with sat. aq. $NaHCO_3$, extracted with EtOAc (20 mL x 3). The organic extracts were dried (Na_2SO_4), and evaporated to give a residue, which was stirred at room temperature for 15 min in EtOH (6 mL) and 10M KOH (0.37 mL) under a nitrogen atmosphere. The reaction mixture was adjusted to pH ~5 by the addition of 4M HCl, and evaporated to dryness. The residue was dissolved in dry THF

(9 mL), and DCC (290 mg, 1.4 mmol) was added at -15 °C. The stirring mixture was kept at the same temperature for 2 h, then gradually warmed up to room temperature during overnight. The resulting mixture was filtered, and the filtrate was evaporated to give a residue, which on purification by silica gel column chromatography (hexane / EtOAc = 2:3) afforded **11** (91 mg, 41%) as an oil: IR (film) 3270, 1640, 1470 cm^{-1}; δ_H 3.41 (3H, s), 3.94 (3H, s), 3.95 (3H, s), 4.04 (2H, d, J= 1.3 Hz), 6.29 (1H, s), 6.78 (1H, broad s), 8.28 (1H, broad s); δ_C (DMSO-*d*$_6$) 28.2 (q), 33.5 (t), 58.6 (q), 60.5 (q), 93.2 (d), 106.1 (s), 111.8 (s), 112.5 (s), 117.9 (d), 126.8 (s), 129.2 (s), 148.0 (s), 167.5 (s). For an unstable property, this compound was used for the next reaction without further characterization.

Makaluvamine C (3).

To a solution of **11** (47 mg, 0.19 mmol) in dry THF (6 mL) was added BH_3•SMe_2 (2 M solution in THF, 0.5 mL, 1 mmol) at 0 °C. After being stirred at room temperature for 3.5 h, the resulting mixture was diluted with H_2O (20 mL) at 0 °C, then extracted with EtOAc (20 mL x 2). The organic extracts were washed with sat. aq. $NaHCO_3$ and brine, dried ($MgSO_4$), and evaporated. The residue was dissolved in 60% $HClO_4$ (4 mL), and CAN (360 mg, 0.6 mmol) was added at 0 °C. After being stirred for 5 min, the mixture was evaporated, and the residue was chromatographed on a silica gel column ($CHCl_3$ / MeOH / NH_4OH = 10:1:0.05) to yield **3** (9.9 mg, 26%), which was characterized as a TFA salt: IR (KBr) 1670, 1615 cm^{-1}; δ_H (DMSO-*d*6) 2.92 (2H, t, J= 7.6 Hz), 3.32 (3H, s), 3.90 (2H, t, J= 7.6 Hz), 5.69 (1H, s), 7.30 (1H, s), 8.68 (1H, broad s), 9.40 (1H, broad s); δ_C (DMSO-*d*6) 18.9 (t), 39.1 (q), 52.6 (t), 85.4 (d), 118.0 (s), 123.2 (s), 123.4 (s), 126.6 (d), 155.7 (s), 156.5 (s), 167.4 (s). Found: *m/z* 202.1002. Calcd for $C_{11}H_{12}N_3O$: M+H, 202.0980.

Makaluvamine D (4).

A mixture of **12** (16 mg, 0.08 mmol) and tyramine hydrochloride (20 mg, 0.11 mmol) in MeOH (3 mL) was stirred at room temperature for 3 h in the presence of $NaHCO_3$ (45 mg). After filtration, the filtrate was evaporated to dryness, and the residue was chromatographed on silica gel ($CHCl_3$ / MeOH = 4:1) to yield **4** (22 mg, 92%), which was characterized as a TFA salt: IR (KBr) 1680, 1635, 1555 cm^{-1}; δ_H (DMSO-*d*6) 2.79 (2H, t, J= 7.0 Hz), 2.88 (2H, t, J= 7.6 Hz), 3.47 (2H, complex), 3.81 (2H, t, J= 7.6 Hz), 5.55 (1H, s), 6.71 (2H, t, J= 8.4 Hz), 7.06 (2H, d, J= 8.4 Hz), 7.33 (1H, broad s), 8.99 (1H, broad signal), 9.35 (1H, broad s), 10.78 (1H, broad s), 13.13 (1H, broad s); δ_C (DMSO-*d*6) 18.0 (t), 32.3 (t), 42.3 (t), 45.0 (t), 84.1 (d), 115.2 (d), 118.6 (s), 122.5 (s), 123.7 (s), 126.8 (d), 128.1 (s), 129.5 (d), 152.9 (s), 155.9 (s), 157.0 (s), 167.4 (s). Found: *m/z* 307.1290. Calcd for $C_{18}H_{17}N_3O_2$: M, 307.1319.

4-Acetoxy-1-[(2-benzyloxycarbonylamino-1-hydroxy)ethyl]benzene (15).

To a stirring solution of **14** (1.10 g, 6.0 mmol as a hydrochloride) in dioxane (12 mL) and H_2O (12 mL) was added ZCl (1.7 mL, 12 mmol). The pH value of the reaction mixture was maintained basic by the addition of sat. aq. $NaHCO_3$. After 1 h, the resulting mixture was extracted with EtOAc (100 mL x 2), and the organic extracts were dried ($MgSO_4$), then evaporated. A mixture of the residue and K_2CO_3 (0.3 g) in MeOH (70 mL) was heated at 40 °C for 1.5 h. After evaporation, the residue was partitioned between EtOAc and H_2O, and the organic layer was dried ($MgSO_4$), then evaporated. A solution of the residue in pyridine (5 mL) and Ac_2O (2 mL) was kept at room temperature for 2 h, and evaporated. The residue was dissolved in EtOAc, washed with H_2O, 0.1M HCl, sat. aq. $NaHCO_3$ and brine, dried ($MgSO_4$), then evaporated to give the corresponding acetate (1.82 g, 98%)

as an amorphous solid: IR (nujol) 3380, 1758, 1735, 1682 cm^{-1}; δ_H 2.33 (3H, s), 4.70 (2H, d, J= 4.6 Hz), 5.16 (2H, s), 5.81 (1H, broad t, J= 4.6 Hz), 7.23 (2H, d, J= 8.7 Hz), 7.30 ~ 7.42 (5H, complex), 8.00 (2H, d, J= 8.7 Hz); δ_C 21.1 (q), 47.8 (t), 67.0 (t), 122.2 (d), 128.1 (d), 128.2 (d), 128.6 (d), 129.5 (d), 131.9 (s), 136.4 (s), 155.0 (s), 156.3 (s), 168.7 (s), 192.9 (s).

The acetate (1.00 g, 3.3 mmol) was dissolved in MeOH (10 mL) and CH_2Cl_2 (2 mL), and $NaBH_4$ (70 mg) was added at 0 °C. After 15 min, the mixture was diluted with H_2O (50 mL), then extracted with $CHCl_3$ (100 mL). The organic layer was washed with H_2O and brine, dried ($MgSO_4$), and evaporated to give a crude product, which on chromatographic purification (hexane / EtOAc = 1:1) provided **15** (0.84 g, 81%) as needles: mp 113 - 114 °C (hexane - EtOAc); IR (nujol) 3300, 1758, 1698 cm^{-1}; δ_H 2.29 (3H, s), 3.07 (1H, broad s), 3.25 (1H, ddd, J= 5.2, 8.1, 14.1 Hz), 3.52 (1H, ddd, J= 3.7, 7.0, 14.1 Hz), 4.78 (1H, m), 5.10 (2H, s), 5.25 (1H, m), 7.04 (2H, d, J= 8.5 Hz), 7.30 ~ 7.37 (7H, complex); δ_C 21.0 (q), 48.4 (t), 66.9 (t), 72.8 (d), 121.5 (d), 126.9 (d), 128.0 (d), 128.1 (d), 128.4 (d), 136.3 (s), 139.2 (s), 150.1 (s), 157.1 (s), 169.5 (s). Found: *m/z* 329.1264. Calcd for $C_{18}H_{19}NO_5$: M, 329.1262.

7-{[2-Hydroxy-2-(4-hydroxyphenyl)ethyl]amino}-1,3,4,8-tetrahydropyrrolo[4,3,2-*de*]quinolin-8-one (16).

A solution of **15** (371 mg, 0.1 mmol) in MeOH (20 mL) containing 10% Pd-C (15 mg) was stirred at room temperature for 3 h under a hydrogen atmosphere. After filtration, the filtrate was evaporated to give a crude amine (230 mg), which was added to a solution of **8** (125 mg, 0.59 mmol) in MeOH (5 mL). After being stirred at room temperature overnight, the mixture was evaporated, and purified by silica gel chromatography ($CHCl_3$ / MeOH / NH_4OH = 3:1:0.06) to give **16** (194 mg, 97%) as a brown solid: IR (film) 1670, 1610, 1560 cm^{-1}; δ_H (DMSO-d_6) 2.68 (2H, t, J= 7.6 Hz), 3.2 ~ 3.3 (2H, complex), 3.87 (2H, t, J= 7.6 Hz), 3.88 (3H, s), 4.75 (1H, complex), 5.47 (1H, s), 5.56 (1H, broad signal), 6.73 (2H, complex), 7.15 (1H, s), 7.18 (2H, complex), 9.34 (1H, broad s). For an unstable property, this compound was used for the next reaction without further characterization.

Makaluvamine E (5).

A mixture of **16** (95 mg, 0.28 mmol) and BF_3•OEt_2 (0.33 mL, 2.8 mmol) in DMF (10 mL) was stirred at 100 °C overnight, then cooled to room temperature. After the addition of pyridine (0.24 mL), the resulting mixture was evaporated, and the residue was purified successively by silica gel chromatography ($CHCl_3$ / MeOH = 3:1) and Sephadex LH 20 (MeOH) to yield **5** (54 mg, 60%), which was characterized as a TFA salt: IR (nujol) 3460, 1680, 1620, 1600, 1545 cm^{-1}; δ_H (CD_3OD) 2.93 (2H, t, J= 7.5 Hz), 3.85 (2H, t, J= 7.5 Hz), 3.95 (3H, s), 5.87 (1H, s), 6.74 (2H, d, J= 8.8 Hz), 6.77 (1H, d, J= 13.7 Hz), 7.09 (1H, s), 7.18 (1H, d, J= 13.7 Hz), 7.32 (2H, d, J= 8.8 Hz); δ_C (CD_3OD) 19.5 (t), 36.5 (q), 44.1 (t), 87.1 (d), 116.8 (d), 119.6 (s), 121.3 (d), 123.8 (s), 125.1 (d), 127.9 (s), 129.2 (d), 131.9 (d), 148.6 (s), 158.2 (s), 159.4 (s), 169.1 (s). Found: *m/z* 320.1408. Calcd for $C_{19}H_{18}N_3O_2$: M+H, 320.1398.

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