CHAPTER III

EXPERIMENTAL

3.1 General experimental procedures

All solvents were distilled prior to use. UV-Vis spectra were recorded on a Hewlet Packard 8452A diode array spectrophotometer in $CHCl_3$. IR spectra were obtained on a Perkin Elmer Model 1760x Fourier Transform Infrared Spectrophotometer. Spectra of solid samples were recorded as KBr pellets and liquid samples were recorded as thin films (NaCl cells). Low-resolution mass spectra was obtained with a Fisons Instruments Mass spectrometer model Trio 2000 at 70 eV. ^{1}H and ^{13}C NMR spectra were recorded at 200.13 and 50.32 MHz, respectively, on a Bruker Model AC-F200 spectrometer, and at 500.00 and 125.65 MHz on a JEOL JNM-A500 spectrometer in $CDCl_3$. Chemical shifts are given in parts per million using residual protonated solvents as reference. COSY, NOESY, HMQC and HMBC experiments were performed on the JEOL JNM-A500 spectrometer. Elemental Analyses were measured on a Perkin Elmer PE2400 SERIES II (CHN/O ANALYSER). Silica gel (Merck Kieselgel 60 and silica TLC plates (Si gels 60 F_{254}) were purchased from the Merck Company.

Plant specimens

Plant specimens have been collected from various locations in Thailand as shown in section 3.2. All of there were identified by comparison with specimen voucher specimen no. BKF 084729 deposited in the Royal Forest Department Herbarium in Bangkok, Thailand.

3.2 Preliminary screening of crude hexane from stem bark of *Croton oblongifolius* Roxb.

The stem bark of *Croton oblongifolius* Roxb. was obtained from various locations in Thailand as listed below :

Amphur Vicheinburi, Petchaboon province
Amphur Pakchong, Nakornratchasima province (Location 1 and 2)
Amphur Sai Yok, Kanchanaburi province (Location 1 and 2)
Amphur Panusnikom, Chonburi province
Amphur Sanamchaikate, Chachoengsao province
Amphur Dansai, Loei province
Amphur Nakornthai, Pitsanulok province
Amphur Hang Dong, Chiangmai province
Amphur Muang, Nakornpanum province
Amphur Muang, Uttaradit province
Amphur Muang, Sakonnakorn province
Amphur Muang, Udonthani province
Amphur Pranburi, Prachuabkirickhan province.

They were extracted with hexane and analyzed by ^{1}H-NMR and the NMR spectra were shown in Fig. 165-167.

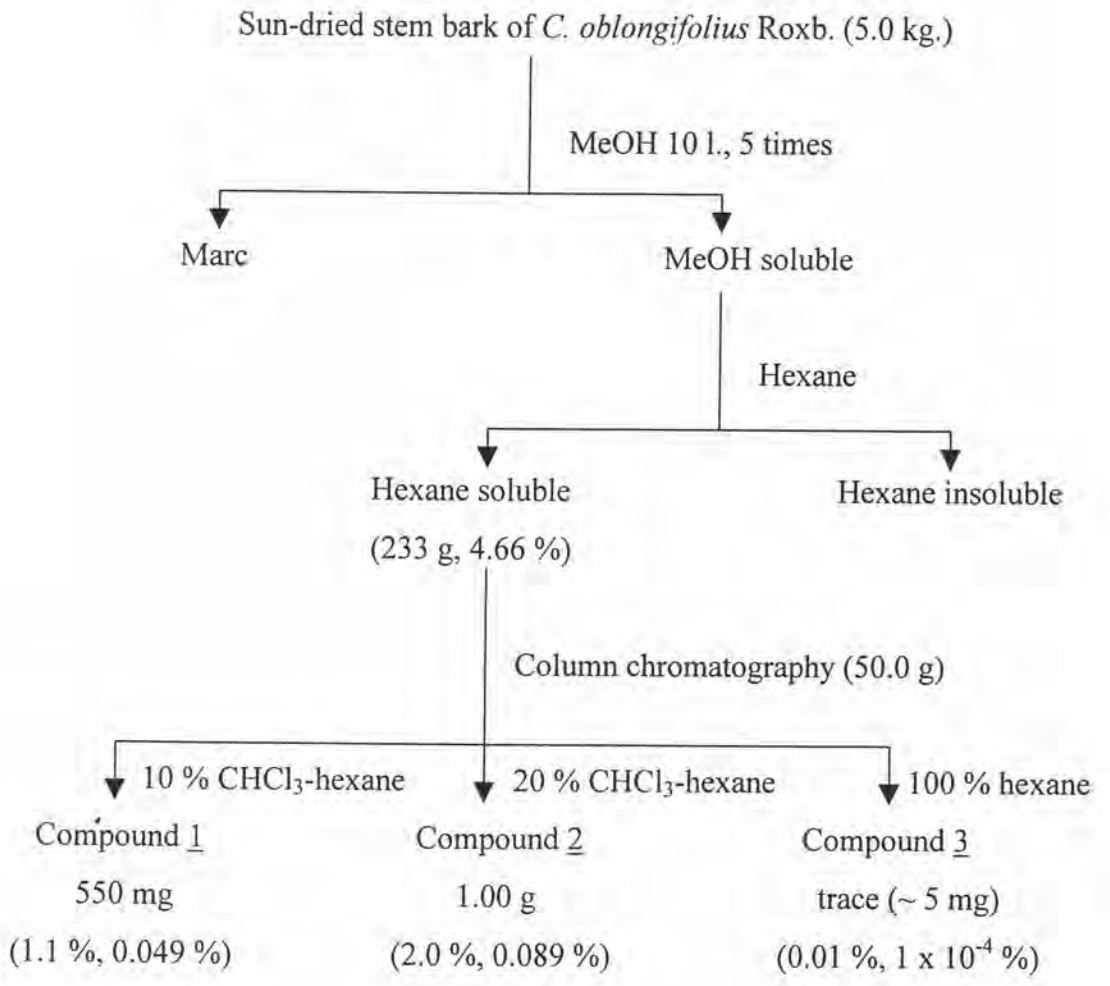
3.3 Extraction and isolation

3.3.1 Extraction and isolation of stem bark of *Croton oblongifolius* Roxb. from Amphur Vicheinburi, Petchaboon province

The powdered, sun-dried stem bark of *C. oblongifolius* Roxb. (5.0 kg.) from Amphur Vicheinburi, Petchaboon province was repeatedly extracted with methanol. The methanol extract was filtered and evaporated under reduced pressure to obtain a dark-red gummy residue that was repeatedly extracted with hexane. The hexane crude extract (233 g, 4.66 %) was obtained as yellowish green oil after evaporation. The

crude hexane extract (50.0 g) was fractionated by Si gel (400 g) column chromatography using Merck Si gel 60 (Art 7734.1000, 70-230 mesh ASTM) as adsorbent. The column was eluted with a hexane-chloroform-methanol gradient in a stepwise fashion. The extraction and isolation procedures are shown in Scheme 1.

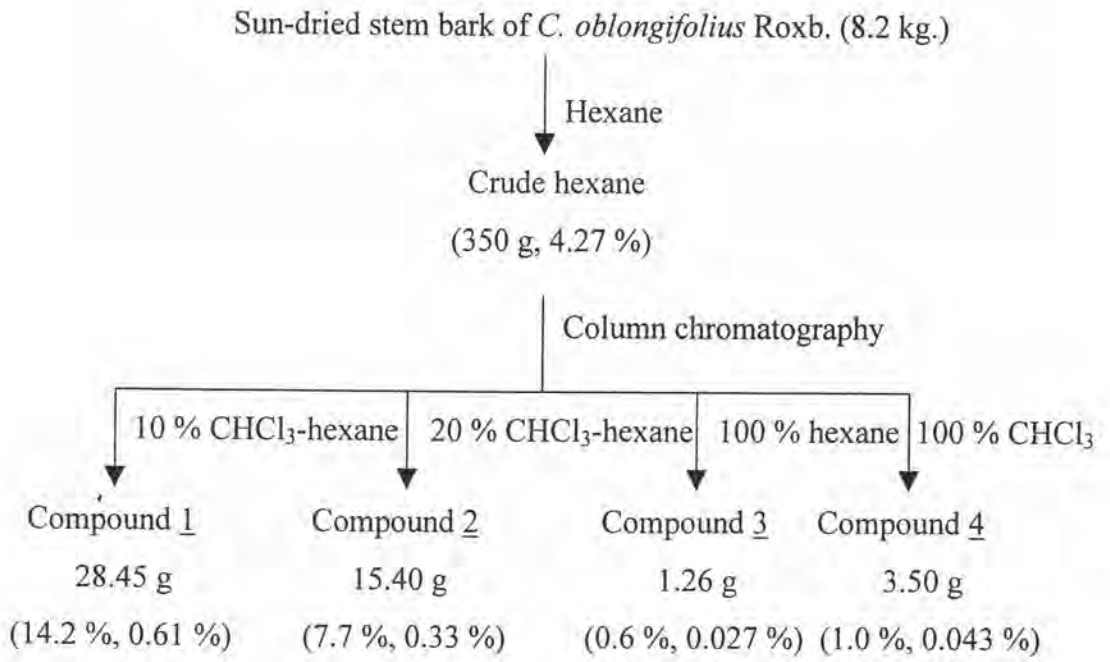
Scheme 1 Extraction and isolation of the stem bark of *C. oblongifolius* Roxb. from Amphur Vicheinburi, Petchaboon province



3.3.2 Extraction and isolation of stem bark of *Croton oblongifolius* Roxb. from Amphur Sai Yok, Kanchanaburi province

The powdered, sun-dried of stem bark of *C. oblongifolius* Roxb. (8.2 kg.) from Amphur Sai Yok, Kanchanaburi province was repeatedly extracted with hexane. The hexane extract was evaporated under reduced pressure to obtain yellowish green oil (350 g, 4.27 % from starting material). The crude hexane extract (200.0 g) was fractionated by Si gel (500 g) column chromatography. The column was eluted with a hexane-chloroform-methanol gradient in a stepwise fashion. The extraction and isolation procedures are shown in Scheme 2.

Scheme 2 Extraction and isolation of the stem bark of *C. oblongifolius* Roxb. from Amphur Sai Yok, Kanchanaburi province



3.3.3 Extraction and isolation of stem bark of *Croton oblongifolius* Roxb. from Amphur Muang, Prachuabkirikhan province

The powdered, sun-dried of stem bark of *C. oblongifolius* Roxb. (450 g) from Amphur Muang, Prachuabkirikhan province was repeatedly extracted with hexane. The hexane extract was evaporated under reduced pressure to obtain yellowish green oil (7.5 g, 1.67 % from starting material). The crude hexane extract was fractionated by Si gel (200 g) column chromatography. The column was eluted with a hexane-chloroform-methanol gradient in a stepwise fashion. The extraction and isolation procedures are shown in Scheme 2.

Scheme 3 Extraction and isolation of the stem bark of *C. oblongifolius* Roxb. from Amphur Muang, Prachuabkirikhan province

Sun-dried stem bark of *C. oblongifolius* Roxb. (450 g)

↓ Hexane

Crude hexane

(7.5 g, 1.67 %)

↓ Column chromatography

20 – 60 % $CHCl_3$ in hexane

Compound 5

4.25 g

(56.67 %, 0.94 %)

3.3.4 Extraction and isolation of stem bark of *Croton oblongifolius* Roxb. from Amphur Sai Yok, Kanchanaburi province (location 2)

The powdered, sun-dried of stem bark of *C. oblongifolius* Roxb. (3.25 kg) from Amphur Sai Yok, Kanchanaburi province (location 2) was repeatedly extracted with hexane. The hexane extract was evaporated under reduced pressure to obtain yellowish green oil (86.7 g, 2.67 % from starting material). The crude hexane extract was fractionated by Si gel (400 g) column chromatography. The column was eluted with a hexane-chloroform-methanol gradient in a stepwise fashion. The extraction and isolation procedures are shown in Scheme 4.

Scheme 4 Extraction and isolation of the stem bark of *C. oblongifolius* Roxb. from Amphur Sai Yok, Kanchanaburi province (location 2)

Sun-dried stem bark of *C. oblongifolius* Roxb. (3.25 kg.)

↓ Hexane

Crude hexane

(86.7 g, 2.67 %)

↓ Column chromatography

(20-60 % $CHCl_3$ in hexane)

Compound 6

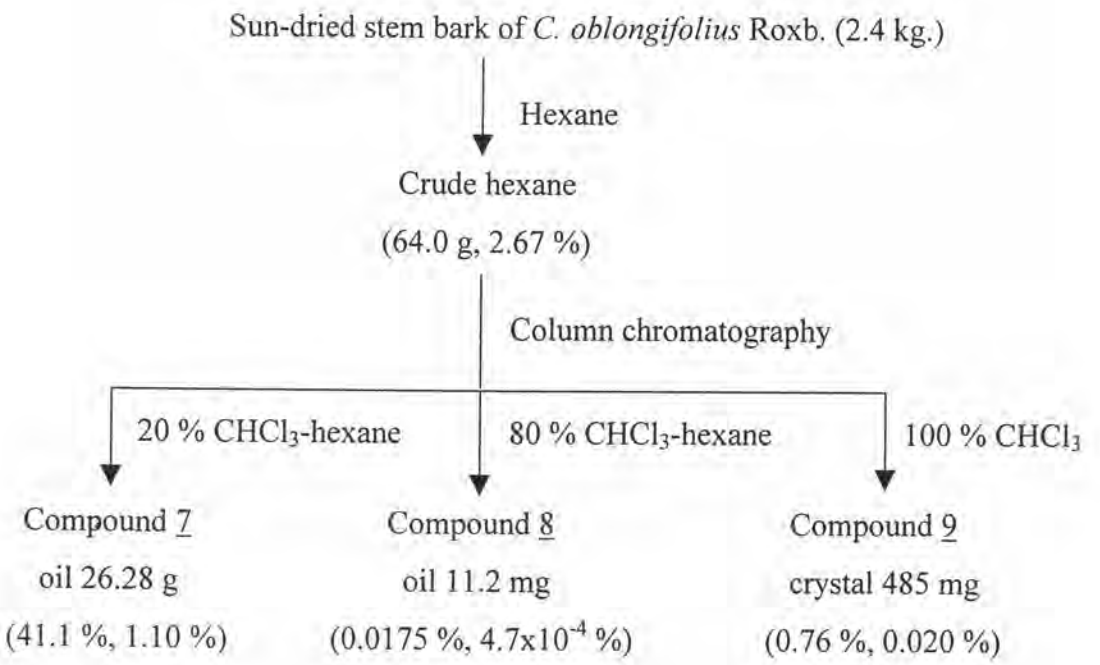
oil 42.78 g

(49.34 %, 1.32 %)

3.3.5 Extraction and isolation of stem bark of *Croton oblongifolius* Roxb. from Amphur Pakchong, Nakornratchasima province

The powdered, sun-dried of stem bark of *C. oblongifolius* Roxb. (2.4 kg) from Amphur Pakchong, Nakornratchasima province was repeatedly extracted with hexane. The hexane extract was evaporated under reduced pressure to obtain yellowish green oil (64.0 g, 2.67 % from starting material). The crude hexane extract was fractionated by Si gel (400 g) column chromatography. The column was eluted with a hexane-chloroform-methanol gradient in a stepwise fashion. The extraction and isolation procedures are shown in Scheme 5.

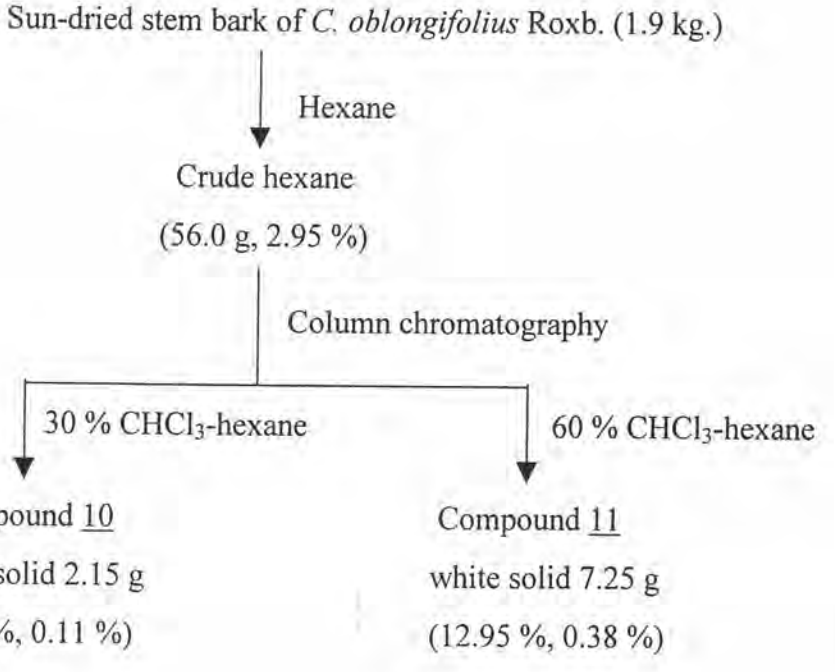
Scheme 5 Extraction and isolation of the stem bark of *C. oblongifolius* Roxb. from Amphur Pakchong, Nakornratchasima province



3.3.6 Extraction and isolation of stem bark of *Croton oblongifolius* Roxb. from Amphur Panusnikom, Chonburi province

The powdered, sun-dried of stem bark of *C. oblongifolius* Roxb. (1.9 kg) from Amphur Panusnikom, Chonburi province was repeatedly extracted with hexane. The hexane extract was evaporated under reduced pressure to obtain yellowish green oil (56.0 g, 2.95 % from starting material). The crude hexane extract was fractionated by Si gel (400 g) column chromatography. The column was eluted with a hexane-chloroform-methanol gradient in a stepwise fashion. The extraction and isolation procedures are shown in Scheme 6.

Scheme 6 Extraction and isolation of the stem bark of *C. oblongifolius* Roxb. from Amphur Panusnikom, Chonburi province



3.4 Purification and Properties of the Compounds from *Croton oblongifolius* Roxb.

3.4.1 Purification and properties of compound 1

Compound 1 was separated from stem bark of *C. oblongifolius* from Amphur Vicheinburi, Petchaboon province and Amphur Sai Yok, Kanchanaburi province (location 1, 8.2 kg). The crude hexane extract (350, 4.27 %) was fractionated with 20 % chloroform in hexane. Similar fractions were combined, and the solvents were removed by rotary evaporation. It was recrystallized from hexane for several times to obtain the white crystalline solid (28.45 g, 14.2 % yield from crude hexane and 0.61 % yield from starting material), m.p. 109-111^{O}C, $[\alpha]_D^{25}$ + 0.2^{O} (*c* 2.28, $CHCl_3$), UV ($CHCl_3$) λ_{max} 252 sh (log ε 4.29) ; HREIMS m/z found 302.2230, calcd for $C_{20}H_{30}O_2$, 302.2240; EA anal. C 79.47 %, H 9.66 % calcd. C 79.47 %, H 9.93 %. This compound is soluble in dichloromethane, chloroform, diethyl ether, hot ethyl acetate, methanol, ethanol, and slightly soluble in hexane. The R_f value was 0.15 in 100 % chloroform system (SiO_2).

FT-IR spectrum (KBr), ν_{max} (cm^{-1}): 2400-3500 (br), 2956 and 2960 (m), 1685 (s), 1644 (m), and 1445 (w). (Fig. 26)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 6.03 (1H, d), 6.01 (1H, t), 5.90 (1H, dd), 5.10 (1H, dt), 2.70 (2H, m), 2.41 (4H, m), 2.34 (1H, m), 2.20 (2H, m), 2.15 (4H, m) 1.73 (3H, s), 1.54 (3H, s), 1.04 (6H, d). (Fig. 27)

^{13}C-NMR, DEPT 90 and DEPT-135 spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 174.1 (s), 146.9 (s), 146.3 (d), 135.2 (s), 134.0 (s), 130.9 (s), 125.7 (d), 121.6 (d), 118.7 (d), 39.2 (t), 38.6 (t), 33.8 (d), 33.6 (t), 28.7 (t), 26.4 (t), 25.1 (t), 22.1 (qx2), 17.0 (q), 15.8 (q). (Fig. 28)

EI MS spectrum m/z: 302[M^+], 287, 256, 189, 152, 136, 121 and 93. (Fig. 32)

3.4.2 Purification and properties of compound 2

Compound 2 was separated from stem bark of *C. oblongifolius* from Amphur Vicheinburi, Petchaboon province and Amphur Sai Yok, Kanchanaburi province (location 1, 8.2 kg). The crude hexane extract (350, 4.27 %) was fractionated with 40 % chloroform in hexane. Similar fractions were combined, and the solvents were removed by rotary evaporation. It was recrystallized from hexane for several times to obtain the white crystalline solid (15.4 g, 7.7 % yield from crude hexane and 0.33 % yield from starting material), m.p. 128-130^{O}C, $[\alpha]_D^{25}$ – 0.3^{O} (*c* 2.55, $CHCl_3$), UV ($CHCl_3$) λ_{max} 247 sh (log ε 4.27) ; HREIMS m/z found 302.2230, calcd for $C_{20}H_{30}O_2$, 302.2240; EA anal. C 79.41 %, H 9.99 % calcd. C 79.47 %, H 9.93 %. This compound is soluble in dichloromethane, chloroform, diethyl ether, hot ethyl acetate, methanol, ethanol, and slightly soluble in hexane. The R_f value was 0.20 in 100 % chloroform system (SiO_2).

FT-IR spectrum (KBr), ν_{max} (cm^{-1}): 2400 – 3500 (br), 2955, 2930, and 2868 (m), 1690 (s), 1644 (m), and 1429 (m). (Fig. 33)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 6.89 (1H, t), 6.01 (1H, d), 5.91 (1H, d), 5.14 (1H, t), 2.39 (1H, m), 2.38 (2H, m), 2.36 (2H, m), 2.26 (2H, m), 2.23 (2H, m), 2.20 (2H, m), 2.15 (2H, m), 1.71 (3H, s), 1.68 (3H, s), 1.05 (6H, d). (Fig. 34)

^{13}C-NMR, DEPT-90 and DEPT-135 spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 173.5 (s), 146.5 (s), 145.7 (d), 135.6 (s), 134.8 (s), 132.1 (s), 127.8 (d), 120.0 (d), 118.6 (d), 38.5 (t), 37.7 (t), 34.6 (d), 30.5 (t), 29.1 (t), 26.7 (t), 24.7 (t), 22.1 (2xq), 18.0 (q), 17.4 (q). (Fig. 35)

EI MS spectrum m/z: 302[M^+], 287, 260, 189, 136, 121 and 93. (Fig. 40)

3.4.3 Purification and properties of compound 3

Compound 3 was separated from stem bark of *C. oblongifolius* from Amphur Vicheinburi, Petchaboon province and Amphur Sai Yok, Kanchanaburi province (location 1, 8.2 kg). The crude hexane extract (350 g, 4.27 %) was fractionated with 10 % chloroform in hexane, which was monitored by TLC using 2,4-dinitrophenylhydrazine solution as visualizing agent. Similar fractions were combined, and the solvents were removed by rotary evaporation to obtain a viscous transparent oil (1.26 g, 0.60 % yield from crude hexane and 0.027 % yield from starting material), UV (EtOH) λ_{max} 246 sh (log ε 4.21), EA; HREIMS m/z found 286.2290, calcd. For $C_{20}H_{30}O$, 286.2291. This compound is soluble in hexane, dichloromethane, chloroform, diethyl ether and ethyl acetate. The R_f value was 0.80 in 100 % chloroform system (SiO_2).

FT-IR spectrum (NaCl), υ_{max} (cm^{-1}): 2960, 2925, and 2873 (m), 2710 (w), 1690 (s), 1639 (w), and 1445 (m). (Fig. 41)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 9.28 (1H, s), 6.39 (1H, t), 5.96 (1H, d), 5.84 (1H, dd), 5.13 (1H, t), 2.50 (2H, m), 2.40 (1H, heptet), 2.32 (2H, m), 2.24 (2H, m), 2.22 (2H, m), 2.20 (2H, m), 2.12 (2H, m), 1.68 (3H, s), 1.68 (3H, s), 1.67 (3H, s), and 1.04 (6H, d). (Fig. 42)

^{13}C-NMR spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 194.7 (s), 155.0 (d), 146.2 (s), 143.9 (s), 135.4 (s), 134.0 (s), 128.0 (d), 119.8 (d), 118.4 (d), 38.5 (t), 37.4 (t), 34.0 (d), 30.0 (t), 28.7 (t), 24.8 (t), 24.1 (t), 22.0 (2xq), 18.0 (q), and 17.2 (q). (Fig. 43)

EI MS spectrum m/z: 286[M^+], 243, 189, 175, 137, 121 and 93. (Fig. 48)

3.4.4 Purification and properties of compound 4

Compound 4 was separated from stem bark of *C. oblongifolius* (8.2 kg) from Amphur Sai Yok, Kanchanaburi province. It separated from concentrated hexane solution. Compound 4 was recrystallized from methanol for several times to obtain the white crystalline solid (3.50 g, 1.0 % yield from crude hexane and 0.043 % yield from starting material), m.p 165-167^{O}C This compound is soluble in dichloromethane, chloroform, diethyl ether, hot ethyl acetate, methanol and ethanol. The R_f value was 0.30 in 5 % methanol in chloroform system (SiO_2).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) : 2940, 2909, and 2865 (m), 1731 (s), 1506 and 1440 (m), 1204 (m), and 1025 (s). (Fig. 49)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 7.37 (1H, d), 7.35 (1H, t), 6.37 (1H, d), 5.31 (1H, dd), 5.26 (1H, s), 5.11 (1H, d), 4.48 (1H, ddd), 3.69 (3H, s), 2.83 (1H, dd), 2.28-2.40 (2H, m), 2.23 (1H, dd), 2.13 (1H, dd), 2.02 (1H, dt), 1.95 (1H, m), 1.84 (1H, m) 1.70 (2H, m), 1.57 (2H, m), 1.43 (1H, ddd), 1.34 (1H, m), 0.93 (3H, d). (Fig. 50)

^{13}C-NMR spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 170.2 (s), 143.4 (d), 139.3 (d), 127.2 (s), 108.6 (d), 104.4 (d), 100.7 (d), 75.7 (d), 74.9 (d), 54.0 (d), 51.6 (q), 53.0 (s), 44.3 (s), 38.9 (d), 38.6 (t), 37.4 (d), 31.5 (t), 25.6 (t), 20.2 (t), 16.9 (q). (Fig. 51)

EI MS spectrum m/z: 374[M^+] 328, 248, 234, 176, 163 and 94. (Fig. 56)

3.4.5 Purification and properties of compound 5

Compound 5 was separated from stem bark of *Croton oblongifolius* from Amphur Muang, Prachubkirikhan province (450 g). The crude hexane extract (7.5 g, 1.67 %) was fractionated hexane-chloroform solvent system. Compound 5 was obtained in with 20-60 % chloroform in hexane. Similar fractions were combined, and the solvents were removed by rotary evaporation to obtain a transparent oil (4.25 g, 56.67 % yield from crude hexane and 0.94 % yield from starting material), $[\alpha]_D^{25}$ – 128.9^{O} (*c* 0.30, $CHCl_3$), UV ($CHCl_3$) λ_{max} 244 sh (log ε 4.05). This compound is

almost soluble in all solvents, for example, dichloromethane, chloroform, diethyl ether, ethyl acetate, methanol, ethanol, and slightly soluble in hexane. The R_f value was 0.20 in 100 % chloroform system (SiO_2).

FT-IR spectrum (KBr), ν_{max} (cm^{-1}): 2400 – 3500 (br), 2955, 2930, and 2868 (m), 1682 (s), 1620 (m), and 1429 (m). (Fig. 57)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 6.05 (2H, m), 5.56 (1H, t-like), 5.21 (1H, d), 5.15 (1H, d), 3.00 (2H, m), 1.30-2.50 (11H, m), 1.80 (3H, s), 1.65 (3H, s), 0.82 (3H, d), 0.80 (3H, d). (Fig. 58)

^{13}C-NMR, DEPT-90 and DEPT-135 spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 173.7 (s), 147.8 (d), 135.1 (s), 131.2 (d), 130.9 (s), 130.5 (d), 128.9 (d), 128.0 (s), 125.7 (d), 47.9 (d), 38.6 (t), 32.8 (d), 32.1 (t), 29.5 (t), 26.2 (t), 25.9 (t), 20.9 (q), 20.0 (q), 19.3 (q), 14.4 (q). (Fig. 59)

EI MS spectrum m/z: 302[M^+], 287, 260, 189, 136, 121 and 93. (Fig. 60)

3.4.6 Purification and properties of compound 6

Compound 6 was separated from stem bark of *C. oblongifolius* (3.25 kg) from Amphur Sai Yok, Kanchanaburi province (location 2). The crude hexane extract (86.7 g, 2.67 %) was fractionated with 20-60 % chloroform in hexane. Similar fractions were combined, and the solvents were removed by rotary evaporation to obtain a viscous transparent oil (42.78 g, 49.34 % yield from crude hexane and 1.32 % yield from starting material). This compound is soluble in hexane, dichloromethane, chloroform, diethyl ether, ethyl acetate, methanol and ethanol. The R_f value was 0.55 in 100 % chloroform system (SiO_2).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) : 3350 (br), 2935, 2879, and 2858 (s), 1670 (s), 1455 and 1020 (m). (Fig. 61)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 5.38 (1H, t-like), 5.24 (1H, br), 4.11 (2H, d), 2.09 (1H, m), 1.99 (1H, m), 1.95 (2H, m), 1.85 (2H, dd), 1.78 (1H, m),

1.66 (3H, s), 1.65 (3H, dd), 1.56 (1H, m), 1.40 (1H, m), 1.33 (1H, m), 1.30 (1H, ddd), 1.22 (1H, m) 1.15 (1H, m), 1.04 (1H, dt), 1.01 (3H, s), 0.78 (3H, s), 0.74 (3H, d). (Fig. 62)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 140.9 (s), 139.8 (s), 123.1 (d), 122.8 (d), 59.4 (t), 44.6 (d), 40.0 (s), 37.7 (t), 37.3 (d), 36.8 (s), 36.4 (t), 33.0 (q), 32.7 (t), 28.7 (t), 19.7 (q), 17.7 (t), 17.2 (q), 16.5 (q), 15.9 (q). (Fig. 63)

EI MS spectrum m/z : 290[M^+] 272, 257, 190, 109 and 95. (Fig. 68)

3.4.7 Purification and properties of compound 7

Compound 7 was separated from stem bark of *C. oblongifolius* (2.4 kg) from Amphur Pakchong, Nakornratchasima province with 20 % chloroform in hexane. Similar fractions were combined, and the solvents were removed by rotary evaporation to obtain a viscous transparent oil (26.28 g, 41.1 % yield from crude hexane and 1.10 % yield from starting material). This compound is soluble in hexane, dichloromethane, chloroform, diethyl ether, ethyl acetate, methanol and ethanol. The R_f value was 0.30 in 100 % chloroform system (SiO_2).

FT-IR spectrum (NaCl), λ_{max} (cm^{-1}): 2800-3600 (br), 2966, 2940, and 2873 (s), 1701 (s), and 1511 and 1460 (m). (Fig. 69)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 7.32 (1H, t), 7.18 (1H, t), 6.24 (1H, m), 1.37-2.38 (15H, m), 1.27 (3H, s), 0.88 (3H, d), 0.87 (3H, s). (Fig. 70)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 184.0 (s), 142.7 (d), 138.4 (d), 136.3 (s), 131.3 (s), 125.7 (s), 111.0 (d), 47.5 (s), 41.1 (s), 36.5 (t), 33.6 (t), 27.7 (d), 26.9 (t), 25.1 (t), 24.3 (q), 20.8 (q), 20.0 (t), 19.5 (t), 16.1 (q). (Fig. 71)

EI MS spectrum m/z: 316[M^+] 221, 175 and 119. (Fig. 76)

3.4.8 Purification and properties of compound 8

Compound 8 was separated from stem bark of *C. oblongifolius* (2.4 kg) from Amphur Pakchong, Nakornratchasima province with 80 % chloroform in hexane. Similar fractions were combined, and the solvents were removed by rotary evaporation to obtain a viscous transparent oil (11.2 mg g, 0.0175 yield from crude hexane and $4.7x10^{-4}$ % yield from starting material). This compound is soluble in hexane, dichloromethane, chloroform, diethyl ether, ethyl acetate, methanol and ethanol. The R_f value was 0.40 in 5 % methanol in chloroform system (SiO_2).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) : 2850-3600 (br), 2966, 2935, and 2873 (m), 1721 and 1696 (s), 1455 (m), and 1270 (s). (Fig. 77)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 8.00 (1H, d), 7.96 (1H, d), 7.33 – 7.56 (5H, m) , 6.43 (1H, d), 6.19 (1H, dd), 2.40 (1H, dd), 2.11 (1H, br), 0.95-1.94 (10H, m), 0.92 (3H, s), 0.88 (3H, d), 0.84 (3H, s). (Fig. 78)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm); 184.0 (s), 165.5 (s), 143.2 (d), 139.8 (d), 136.2 (s), 133.0 (d), 130.4 (s), 130.2 (s), 129.6 (2xd), 128.4 (2xd), 126.5 (s), 108.9 (d), 65.8 (d), 47.2 (s), 40.7 (s), 40.6 (t), 35.8 (t), 33.8 (d), 27.5 (d), 26.8 (d), 25.7 (d), 23.3 (q), 21.6 (q), 20.0 (t), 16.1 (q). (Fig. 79)

EI MS spectrum m/z: 376[M^+] 315, 330, 268, 220, 176 and 105. (Fig. 84)

3.4.9 Purification and properties of compound 9

Compound 9 was separated from stem bark of *C. oblongifolius* (2.4 kg) from Amphur Pakchong, Nakornratchasima province with 100 % chloroform. Similar fractions were combined, and the solvents were removed by rotary evaporation. It was recrystallized from methanol for several times to obtain the white crystalline solid (485 mg, 0.076 % yield from crude hexane and 0.020 % yield from starting material), 168-170 ^{O}C; $[\alpha]_D^{25}$ + 86.5^{O} (*c* 1.0, $CHCl_3$), UV ($CHCl_3$) λ_{max} 254 sh (log ε 3.57). This compound is soluble in dichloromethane, chloroform, diethyl ether, methanol and ethanol. The R_f value was 0.25 in 5 % methanol in chloroform system (SiO_2).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}): 2850-3600 (br), 2966, 2935, and 2884 (m), 1726 and 1690 (s), 1562 and 1506 (m), and 1163 (m). (Fig. 85)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 7.33 (1H, d), 7.22 (1H, d), 6.26 (1H, s), 1.37-2.38 (15H, m), 1.29 (3H, s), 0.88 (3H, d), 0.86 (3H, s). (Fig. 86)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 184.8 (s), 142.6 (d), 138.4 (d), 135.9 (s), 131.3 (s), 125.8 (s), 111.1 (d), 47.5 (s), 40.9 (s), 36.5 (t), 35.4 (t), 33.2 (d), 26.8 (t), 25.9 (t), 25.1 (t), 22.8 (q), 20.8 (q), 19.5 (d x 2), 16.0 (q). (Fig. 87)

EI MS spectrum m/z: 330[M^+] 312, 268, 220, 175, 159 and 119. (Fig. 92)

3.4.10 Purification and properties of compound 10

Compound 10 was separated from stem bark of *C. oblongifolius* from Amphur Panusnikom, Chonburi province (1.9 kg). The crude hexane extract (56 g, 2.95 %) was fractionated with hexane-chloroform solvent system. Compound 10 was obtained in 30 % chloroform in hexane. It was recrystallized from 80% chloroform in hexane for several times to obtain the white crystalline solid (2.15 g, 3.84 % yield from crude hexane and 0.11 % yield from starting material), m.p. 100-102 °C. This compound is soluble in dichloromethane, chloroform, diethyl ether, hot ethyl acetate, methanol and ethanol. The R_f value was 0.30 in 100 % chloroform (SiO_2).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}): 2400-3600 (br), 2971, 2930, and 2873 (m), 1685(s), 1634(m), and 1460 and 1396(m) and 1381(m) and 1280(m). (Fig. 93)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 7.33 (1H, s), 7.19 (1H, s), 6.85 (1H, s), 6.26 (1H, s), 2.16–2.50 (6H, m), 1.40-1.73 (8H, m) 1.26 (3H, s), 0.84 (3H, d), 0.76 (3H, s). (Fig. 94)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 172.8(s), 142.6 (d), 141.5 (s), 140.2 (d), 138.3 (d), 125.4 (s), 110.9 (d), 46.7 (d), 38.8 (s), 38.6 (t), 37.6 (s), 36.3 (d), 35.8 (t), 27.5 (t), 27.3 (t), 20.5 (q), 18.4 (q), 18.2 (t), 17.5 (t), 16.0 (q). (Fig. 95)

EI MS spectrum m/z: 316[M^+] 299, 283, 221, 203, 105 and 96. (Fig. 96)

3.4.11 Purification and properties of compound 11

Compound 11 was separated from stem bark of *C. oblongifolius* from Amphur Panusnikom, Chonburi province (1.9 kg). The crude hexane extract (56.0 g, 2.95 %) was fractionated with hexane-chloroform solvent system. Compound 11 was obtained in 60 % chloroform in hexane fraction. Similar fractions were combined, and the solvents were removed by rotary evaporation. It was recrystallized from 50% chloroform in hexane for several times to obtain the white solid (7.25 g, 12.95 % yield from crude hexane and 0.38 % yield from starting material), m.p. 146-149 °C. This compound is soluble in dichloromethane, chloroform, diethyl ether, hot ethyl acetate, methanol and ethanol. The R_f value was 0.20 in 5 % methanol in chloroform system (SiO_2).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) : 3421 (br), 2950, 2925, and 2879 (s), 1465 (m), and 1090 (m). (Fig. 97)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 6.29 (1H, dd), 5.52 (1H, t), 5.02 (1H, d), 4.87 (1H, d), 3.48 (1H, dd), 2.82 (2H, br), 2.35 (1H, dt), 2.15 (1H, dt), 1.81 (1H, ddd), 1.73 (3H, s), 1.54 (2H, m), 1.37 (2H, m), 1.26 (1H, dd), 1.23 (1H, t), 1,10 (3H, s), 1.00 (1H, dd), 0.85 (3H, s), 0.77 (3H, s). (Fig. 98)

^{13}C-NMR spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 141.5(d), 135.7 (d), 132.5 (s), 110.4 (t), 80.2 (d), 78.1 (s), 60.2 (d), 53.5 (d), 41.6 (t), 39.7 (t), 39.2 (s), 33.4 (t), 33.1 (s), 27.8 (t). 23.5 (t), 21.5 (q), 18.4 (t), 17.8 (q), 15.5 (q), 11.8 (q). (Fig. 99)

EI MS spectrum m/z: 306[M^+] 289, 252, 207, 177, 124 and 109. (Fig. 104)

3.5 Modification of compound from *Croton oblongifolius* Roxb.

3.5.1 Modification of compound 1

3.5.1.1 Methylation of compound 1

The compound 1 (510.8 mg, 1.69 mmol) was methylated with diazomethane in diethyl ether under the conditions described earlier [59,60] and to give compound 1a as a viscous transparent oil (450.4 mg, 84.3 % yield), UV ($CHCl_3$) λ_{max} 250 sh (log ε 4.19); R_f = 0.75 (100% chloroform).

FT-IR spectrum (KBr) υ_{max} (cm^{-1}) 2960, 2925, and 2873 (m), 1721 (s), 1644 (w), 1445 (m) and 1204 (m). (Fig. 105)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 5.76-5.99 (3H, m), 5.05 (1H, dd), 3.67 (3H, s), 2.57 (2H, m), 2.38 (5H, m), 2.11-2.34 (8H, m), 1.69 (3H, s), 1.50 (3H, s), 0.98 (6H, d). (Fig. 106)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 168.6 (s), 146.0 (s), 143.2 (d), 134.8 (s), 133.9 (s), 131.4 (s), 125.4 (d), 121.8 (d), 118.7 (d), 51.0 (q), 39.2 (d), 38.6 (d), 33.4 (t), 28.6 (t), 26.2 (t), 25.1 (t), 22.1 (q), 16.9 (q), 15.7 (q). (Fig. 107)

EI MS spectrum m/z: 316[M^+], 257, 166, 136, 121, and 93. (Fig. 108)

3.5.1.2 Reduction of compound 1a

Methyl ester of compound 1a (202.5 mg, 0.64 mmol) in 15 ml of anhydrous diethyl ether was added slowly from a dropping funnel into a stirred solution of lithium aluminium hydride [61] (200 mg, 5.13 mmol) in 20 ml of anhydrous diethyl ether in a 50 ml round bottom flask. After the addition was completed, the reaction mixture was stirred for 5 hours at room temperature. The reaction was stopped and worked up by dropping some water into the solution. The organic layer was concentrated by rotary evaporator and purified by silica gel column chromatography and eluting with 10 % chloroform in hexane to give compound 1b as a viscous

transparent oil (138.5 mg, 75.1 % yield), UV ($CHCl_3$) λ_{max} 255 sh (log ε 4.28); R_f = 0.50 (100% chloroform).

FT-IR spectrum (KBr) υ_{max} (cm^{-1}) 3365 (s), 2960, 2920, and 2868 (s), 1650 and 1614 (w), 1440 (m) and 1009 (m). (Fig. 109)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 5.86-6.05 (2H, m), 5.22 (1H, t), 4.97 (1H, t), 4.00 (2H, s), 2.40-2.14 (10, m), 2.11 (3H, s), 1.71 (3H, s), 1.48 (3H, s), 1.03 (6H, d). (Fig. 110)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 146.4 (s), 138.3 (s), 134.9 (s), 133.8 (s), 129.4 (d), 125.1 (d), 122.0 (d), 118.5 (d), 61.5 (t), 39.3 (t), 38.8 (t), 33.5 (t), 33.0 (d), 28.6 (t), 25.4 (t), 24.3 (t), 22.2 (2 x q), 16.8 (q), 15.7 (q). (Fig. 111)

EI MS spectrum m/z: 288[M^+], 257, 136, 121, and 93. (Fig. 112)

3.5.1.3 Oxidation of compound 1b

The compound 1b (75.6 mg, 0.26 mmol) was oxidized with manganese dioxide (760 mg) in acetone 10 ml. [62] The organic solution layer was concentrated by rotary evaporator and purified by silica gel column chromatography and eluted with 10 % chloroform in hexane to give compound 1c as a viscous transparent oil (60.5 mg, 81.4 % yield), UV ($CHCl_3$) λ_{max} 245 sh (log ε 4.13); R_f = 0.75 (100 % chloroform).

FT-IR spectrum (KBr) υ_{max} (cm^{-1}) 3053 (w), 2966, 2920, and 2873 (m), 2756 (w), 1680 (s), 1629 (w) and 1450 (m). (Fig. 113)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 10.06 (1H, s), 6.36 (1H, t), 5.97 (1H, d), 5.81, (1H, d), 5.05 (1H, dd), 2.70 (2H, m), 2.35-2.01 (10H, m), 1.70 (3H, d), 1.51 (3H, s), 0.98 (6H, d). (Fig. 114)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 191.2 (d), 149.5 (d), 145.8 (s), 140.0 (s), 134.5 (s), 133.1 (s), 126.0 (d), 122.2 (d), 118.5 (d), 39.4 (t), 38.4 (t), 32.7 (d), 28.1 (t), 27.7 (t), 25.3 (t), 23.4 (t), 22.1 (2 x q), 16.6 (q), 15.7 (q). (Fig. 115)

EI MS spectrum m/z: 287[$M+H^+$], 243, 136, 121, and 93. (Fig. 116)

3.5.2 Modification of compound 2

3.5.2.1 Methylation of compound 2

The compound 2 (508.4 mg, 1.68 mmol) was methylated with diazomethane in diethyl ether under the conditions described earlier [59, 60] and to give compound 2a as a viscous transparent oil (422.9 mg, 79.7 % yield), UV ($CHCl_3$) λ_{max} 248 sh (log ε 4.22); R_f = 0.75 (100% chloroform).

FT-IR spectrum (KBr) υ_{max} (cm^{-1}) 2966, 2925, and 2879 (m), 1711 (s), 1639 (w), 1434 (m) and 1199(m). (Fig. 117)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 6.72 (1H, t), 5.96 (1H, t), 5.91 (1H, dd), 5.12 (1H, t), 3.68 (3H, s), 2.11-2.40 (10, m), 1.70 (3H, s), 1.66 (3H, s), 1.04 (6H, d). (Fig. 118)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 168.4 (s), 146.5 (s), 143.0 (d), 135.4 (s), 134.9 (s), 132.5 (s), 127.6 (d), 120.1 (d), 118.6 (d), 51.5 (q), 38.6 (d), 37.8 (t), 34.8 (d), 30.4 (d), 29.0 (t), 27.2 (t), 24.7 (t), 24.7 (t), 22.1 (q x 2), 17.9 (q), 17.4 (q). (Fig. 119)

EI MS spectrum m/z: 316[M^+], 256, 166, 136, 121, and 93. (Fig. 120)

3.5.2.2 Reduction of compound 2a

Methyl ester of compound 2a (212.4 mg, 0.67 mmol) was reduced with $LiAlH_4$ (200 mg, 5.13 mmol, in 20 ml of anhydrous diethyl ether) under the conditions described earlier [61] and to give compound 2b as a viscous transparent oil (140.7 mg, 72.9 % yield), UV ($CHCl_3$) λ_{max} 255 sh (log ε 4.26); R_f = 0.50 (100% chloroform).

FT-IR spectrum (KBr) υ_{max} (cm^{-1}) 3385 (br), 2960, 2925, and 2868 (s), 1650 (w), 1460 (m), and 1015 (m). (Fig. 121)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 5.93-6.08 (2H, m), 5.42 (1H, t), 5.08 (1H, t), 3.98 (2H, s), 2.37-2.05 (12H, m), 17.2 (3H, s), 1.68 (3H, d), 1.05 (6H, d). (Fig. 122)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 146.7 (s), 139.3 (s), 135.9 (s), 135.1 (s), 128.1 (d), 127.2 (d), 119.8 (d), 118.7 (d), 67.3 (t), 39.2 (t), 37.5 (t), 34.8 (d), 28.9 (t), 28.8 (t), 28.3 (t), 24.8 (t), 22.2 (2 x q), 18.2 (q), 17.8 (q). (Fig. 123)

EI MS spectrum m/z: 288[M^+], 270, 245, 136, 121, and 93. (Fig. 124)

3.5.2.3 Oxidation of compound 2b

The compound 2b (80.6 mg, 0.28 mmol) was oxidized with manganese dioxide (800 mg) in acetone 10 ml. [62] The organic layer was concentrated by rotary evaporator and purified by silica gel column chromatography and eluted with 10 % chloroform in hexane to give compound 2c as a viscous transparent oil (50.5 mg, 63.1 % yield), UV ($CHCl_3$) λ_{max} 245 sh (log ε 4.16); R_f = 0.75 (100% chloroform).

FT-IR spectrum (KBr) υ_{max} (cm^{-1}) 3058 (w), 2966, 2930, and 2873 (m), 2720 (w), 1696 (s), 1639 (w) and 1455 (m). (Fig. 125)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 9.29 (1H, s), 6.40 (1H, t), 5.90 (1H, d), 5.84 (1H, d), 5.12 (1H, d), 1.68 (6H, s), 1.05 (6H, d). (Fig. 126)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 194.8 (d), 155.2 (d), 146.3 (s), 143.9 (s), 135.5 (s), 134.1 (d), 128.1 (d), 119.8 (d), 118.4 (d), 38.5 (t), 37.4 (t), 34.1 (d), 30.0 (t), 28.7 (t), 24.8 (t), 24.1 (t), 22.1 (2 x q), 18.1 (q), 17.3 (q). (Fig. 127)

EI MS spectrum m/z: 286[M^+], 243, 218, 175, 136, 121, and 93. (Fig. 128)

3.5.3 Modification of compound 5

3.5.3.1 Methylation of compound 5

The compound 5 (700.0 mg, 2.32 mmol) was methylated with diazomethane in diethyl ether under the conditions described earlier [59, 60] and to give compound 5a as a viscous transparent oil (650 mg, 89.3 %), UV ($CHCl_3$) λ_{max} 246 sh (log ε 3.99). This compound is usually soluble in all solvents. The R_f value was 0.80 in 100 % chloroform system (SiO_2).

FT-IR spectrum (KBr), υ_{max} (cm^{-1}) 2966, 2925, and 2879 (m), 1711 (s), 1639 (w), 1434 (m) and 1199(m). (Fig. 129)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 6.04 (1H, d), 5.80 (1H, t-like), 5.52 (1H, t-like), 5.18 (1H, d), 5.12 (1H, d), 3.74 (3H, s), 2.95 (2H, m), 1.25-2.47 (11H, m), 1.77 (3H, s), 1.61 (3H, s), 0.82 (3H, d), 0.76 (3H, d). (Fig. 130)

^{13}C-NMR, DEPT-90 and DEPT-135 spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 168.3 (s), 144.0 (d), 135.1 (s), 131.2 (d), 131.1 (s), 130.4 (d), 129.5 (s), 127.6 (d), 125.7 (d), 50.7 (q), 47.9 (d), 38.6 (t), 32.7 (d), 32.3 (t), 29.3 (t), 26.2 (t), 25.4 (t), 20.8 (q), 19.9 (q), 19.5 (q), 14.4 (q). (Fig. 131)

EI MS spectrum m/z: 316[M^+], 256, 166, 136, 121, and 93. (Fig. 132)

3.5.3.2 Reduction of compound 5a

Methyl ester of compound 5a (400.0 mg, 1.27 mmol) in 15 ml of anhydrous diethyl ether was reduced with $LiAlH_4$ (500 mg, 12.82 mmol, in 20 ml of anhydrous diethyl ether) under the conditions described earlier [61] and to give compound 5b as a white amorphous (324.2 mg, 88.37 %), R_f = 0.50 (100% chloroform).

FT-IR spectrum (KBr), υ_{max} (cm^{-1}) 3385 (br), 2960, 2925, and 2868 (s), 1650 (w), 1460 (m), and 1015 (m). (Fig. 133)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 6.04 (1H, d), 5.52 (1H, t-like), 5.20 (1H, m), 5.13 (2H, m), 4.24 (1H, d), 3.97 (1H, d), 3.00 (2H, m), 1.25-2.47 (11H, m), 1.80 (3H, s), 1.60 (3H, s), 0.84 (3H, d), 0.80 (3H, d). (Fig. 134)

^{13}C-NMR, DEPT-90 and DEPT-135 spectrum ($CDCl_3$, 125.65 MHz) δ (ppm), 136.0 (s), 135.1 (s), 130.9 (d), 130.8 (s), 130.3 (d), 129.7 (d), 127.1 (d), 125.5 (d), 59.4 (t), 48.5 (d), 38.7 (t), 32.8 (d), 32.4 (t), 27.9 (t), 26.2 (t), 22.8 (t), 20.8 (q), 19.9 (q), 19.8 (q), 14.4 (q). (Fig. 135)

EI MS spectrum m/z: 288[M^+], 270, 245, 136, 121, and 93. (Fig. 136)

3.5.3.3 Oxidation of compound 5b

The compound 5b (200.6 mg, 0.70 mmol) was oxidized with manganese dioxide (2.0 g) in acetone 20 ml. [62] The organic layer was concentrated by rotary evaporator and purified by silica gel column chromatography and eluted with 10% chloroform in hexane to give compound 5c as a viscous transparent oil (128.5 mg, 64.5 %), R_f 0.80 (100% chloroform).

FT-IR spectrum (KBr), υ_{max} (cm^{-1}) 3058 (w), 2966, 2930, and 2873 (m), 2720 (w), 1696 (s), 1639 (w) and 1455 (m). (Fig. 137)

^{1}H-NMR spectrum ($CDCl_3$, 500 MHz) δ (ppm) 10.5 (1H, s), 6.32 (1H, dd), 6.00 (1H, d), 5.55 (1H, t-like), 5.18 (2H, m), 3.00 (2H, m), 1.25-2.56 (11H, m), 1.77 (3H, s), 1.65 (3H, s), 0.80 (3H, d), 0.75 (3H, d). (Fig. 138)

^{13}C-NMR, DEPT-90 and DEPT-135 spectrum ($CDCl_3$, 125.65 MHz) δ (ppm) 191.8 (d), 150.2 (d), 137.8 (s), 135.8 (s), 131.2 (d), 130.2 (d), 129.8 (s), 128.7 (d), 125.4 (d), 48.2 (d), 38.3 (t), 32.7 (d), 29.6 (t), 28.7 (t), 26.2 (t), 22.4 (t), 20.8 (q), 19.9 (q), 19.4 (q), 14.4 (q). (Fig. 139)

EI MS spectrum m/z: 286[M^+], 243, 218, 175, 136, 121, and 93. (Fig. 140)

3.5.4 Modification of compound 6

3.5.4.1 Oxidation of compound 6

The compound 6 (505.6 mg, 1.74 mmol) was oxidized with manganese dioxide on activated carbon 500 mg in acetone 10 ml. [62] The organic layer was concentrated by rotary evaporator and purified by silica gel column chromatography and eluted with 10 % chloroform in hexane to give compound 6a as a viscous transparent oil (408.5 mg, 81.5 %), R_f = 0.75 (100% chloroform).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}): 2940, 2884, and 2863 (m), 2766 (w), 1680 (s), 1634 (w) and 1450 (w). (Fig. 141)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 9.95 (1H, d), 5.89 (1H, dd), 5.25 (1H, br), 2.16(3H, d), 2.12-1.92 (6H, m), 1.65 (3H, d), 1.55 – 1.75 (2H, m), 1.44-1.19 (6H, m), 1.02 (1H, s), 0.81 (3H, s), 0.75 (3H, d). (Fig. 142)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 191.2 (d), 165.5 (s), 139.7 (s), 127.1 (d), 123.1 (d), 44.7 (d), 40.2 (s), 37.7 (t), 37.4 (d), 36.9 (s), 35.8 (t), 34.2 (t), 33.1 (q), 28.7 (t), 23.9 (t), 19.7 (q), 17.8 (t), 17.7 (t), 17.2 (q), 15.9 (q). (Fig. 143)

EI MS spectrum m/z: 288[M^+] 273, 255, 189, 121, 107 and 95. (Fig. 144)

3.5.4.2 Oxidation of compound 6a

The compound 6a (204.0 mg, 0.71 mmol) was oxidized with silver nitrate 250 mg in sodium hydroxide (Tollen's solution) in aqueous ethanol. [63] The organic layer was concentrated by rotary evaporator and purified by silica gel column chromatography and eluted with 20 % chloroform in hexane to give compound 6b as a white solid (145.9 mg, 67.6 %), R_f = 0.30 (100% chloroform).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}): 2400-3600 (br), 2935, 2879, and 2858 (0s), 1690 (s), 1639 (s), and 1440 (m). (Fig. 145)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 5.69 (1H, br), 5.26 (1H, br), 2.17 (3H, d), 2.05-1.91 (4H, m), 1.66 (3H, s), 1.58 – 1.09 (6H, m), 1.02 (3H, s), 0.80 (3H, s), 0.76 (3H, d). (Fig. 146)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 172.5 (s), 164.6 (s), 139.7 (s), 123.1 (d), 114.9 (d), 44.6 (d), 40.2 (s), 37.7 (s), 37.4 (t), 36.9 (d), 36.1 (t), 34.8 (t), 33.1 (q), 28.7 (t), 24.0 (t), 19.8 (q), 19.5 (q), 17.7 (t), 17.2 (q), 15.9 (q). (Fig. 147)

EI MS spectrum m/z: 304[M^+] 289, 261, 190, 123, 107 and 95. (Fig. 148)

3.5.4.3 Methylation of compound 6b

The compound 6b (60.7 mg, 0.20 mmol) was methylated with diazomethane in diethyl ether under the conditions described earlier [59, 60] and to give compound 6c as a viscous transparent oil (55.5 mg, 87.3 %), R_f = 0.75 (100% chloroform).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) 2940, 2879, 2858 (s), 1729 (s), 1650 (m), 1440 (m) and 1153 (s). (Fig. 149)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 5.65 (1H, d), 5.24 (1H, br), 3.65 (3H, s), 2.15 (3H, d), 2.10-1.86 (6H, m), 1.64 (3H, d), 1.60 – 1.03 (4H, m), 1.00 (3H, s), 0.79 (3H, s) and 0.74 (3H, d). (Fig. 150)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 167.3 (s), 161.9 (s), 139.7 (s), 123.1 (d), 114.7 (d), 50.7 (q), 44.6 (d), 40.2 (s), 37.7 (t), 37.4 (d), 36.8 (s), 36.1 (t), 34.5 (t), 33.1 (q), 28.7 (t), 24.0 (t), 19.7 (q), 19.2 (q), 17.7 (t), 17.2 (q) and 15.9 (q). (Fig. 151)

EI MS spectrum m/z: 318[M^+] 303, 243, 159, 123, 107 and 95. (Fig. 152)

3.5.5 Methylation of compound 7

The compound 7 (202.2 mg, 0.64 mmol) was methylated with diazomethane in diethyl ether under the conditions described earlier [59, 60] and to give compound 7a as a viscous transparent oil (160.5 mg, 76.0 %), R_f = 0.85 (100% chloroform).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) 2940, 2879, and 2843 (m), 1737 (s), 1460 and 1434 (W) and 1168 (m). (Fig. 153)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 7.31 (1H, t), 7.20 (1H, d), 6.23 (1H, d), 3.62 (3H, s), 2.40 (1H, m), 1.31-2.24 (12H, m), 1.25 (3H, s), 0.88 (3H, s), 0.86 (3H, d). (Fig. 154)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 178.2 (s), 142.7 (d), 138.4 (d), 135.6 (s), 131.7 (s), 125.7 (s), 111.0 (d), 51.8 (q), 47.7(d), 41.0 (t), 36.6 (t), 36.4

(d), 33.6 (t), 27.6 (t), 26.9 (t), 25.1 (t), 24.4 (q), 20.8 (q), 20.1 (q), 19.5 (q), 16.1 (q). (Fig. 155)

EI MS spectrum m/z: 330[M^+] 371, 236, 175, 119 and 81. (Fig. 160)

3.5.6 Methylation of compound 9

The compound 9 (102.5 mg, 0.31 mmol) was methylated with diazomethane in diethyl ether under the conditions described earlier [59, 60] and gave compound 9a as a viscous transparent oil (91.5 mg, 85.6 %), R_f = 0.75 (100% chloroform).

FT-IR spectrum (KBr), λ_{max} (cm^{-1}) 2940, 2879, and 2843 (m), 1737 (s), 1460 and 1434 (W) and 1168 (m). (Fig. 161)

^{1}H-NMR spectrum ($CDCl_3$, 200 MHz) δ (ppm) 7.95 (1H, s), 7.33 (1H, s), 6.68 (1H, s), 3.55 (3H, s), 2.85 (1H, d), 2.68 (1H, d), 1.30-2.10 (10H, m), 1.22 (3H, s), 0.90 (3H, s), 0.78 (3H, d). (Fig. 162)

^{13}C-NMR spectrum ($CDCl_3$, 50.25 MHz) δ (ppm) 193.9 (s), 178.3 (s), 146.8 (d), 144.0 (d), 134.7 (s), 130.0 (s), 129.2 (s), 108.6 (d), 51.6 (q), 47.5 (s), 46.8 (t), 41.3 (s), 35.8 (t), 34.6 (d), 26.7 (t), 25.9 (t), 25.8 (t), 23.7 (q), 21.1 (q), 19.8 (t), 16.2 (q). (Fig. 163)

EI MS spectrum m/z: 330[M^+] 371, 236, 175, 119 and 81. (Fig. 164)