# *CHAPTER 6*

# *EXPERIMENTAL*

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## 6.1 General

The optical rotations were recorded on Jasco (Japan) P1010 with tungsten lamp. Mass spectra were recorded on a Jeol JMS 700 spectrometer using NBA as the matrix for FAB analysis. The Automass Multi Thermofinnigan was used for HR ESI analysis and EIMS spectra were obtained on Shimadzu GC-MS-QP2000A Mass Spectrometer 70 eV.

Melting points were taken on a hot stage Gallen Kamp melting point apparatus and were uncorrected. The ultraviolet spectra were obtained in MeOH on Shimadzu UV-160A ultraviolet-visible spectrometer. The infrared spectra were taken on a Perkin Elmer 1600 Double-Beam recording Spectrometer, using chloroform or liquid film.

The $^{1}H$ NMR spectra were recorded in deuterated chloroform on a JEOL JNM-FX400 (unless stated otherwise); chemical shifts are reported in ppm on $\delta$ scale, and the coupling constants are given in Hz.

All solvents except those used for bulk extractions (distilled) are AR grade. Aluminium supported silica gel 60 $F_{254}$ plates were used for TLC. The plates were activated at $100^{0}C$ for one hour and stored in a dessicator until needed. TLC spots

were visualized under ultra-violet light (254 nm and 365 nm) followed by spraying with Dragendorff reagent.

Silica gel 60, 70-230 mesh ASTM (Merck 7734) and silica gel 60, 230-400 Mesh ASTM (Merck 9385) were used for column and flash chromatography respectively. Silica gel 604 $F_{254}$ were used for preparative TLC. Meyer reagent was used for alkaloid screening and alkaloid spotting (TLC).

Mayer's reagent : A solution of mercury (II) chloride (1.4 g) in distilled water (60 ml) was poured into a solution of potassium iodide (5.0 g) in distilled water (10 ml). The mixture was then made up to 150 ml.

Mayer's test : A positive result is indicated by the formation of white precipitate under acidic conditions.

**Dragendoff's reagent**

Solution A : Bismuth (III) nitrate (0.85 g) in a mixture of 10 ml glacial acid and 40 ml distilled water.

Solution B : Potassium iodide (8.0 g) in 20 ml distilled water.

Stock solution : A mixture of equal volumes of solution A and solution B.

Spray reagent : The stock solution (20 ml) was diluted in a mixture of 20 ml glacial acetic acid and 60 ml distilled water.

Dragendoff's test : A positive result is indicated by the formation of orange precipitates or spots.

## 6.2 Plant Material

All the species studied were identified with the assistance of the phytochemical survey of Malayan Herbarium, University Malaya and Forest Research Institute of Malaysia (FRIM), Kepong, Selangor. The locality and habitat of the plant species are shown in Table 6.1 below.

Table 6.1: The locality and habitat of the plant species.

| Species | Locality and Date Collected |
|---|---|
| *Phoebe grandis* (Nees) Merr. KL4224 | Gunung Stong permanent Forest Reserve, Terang Dam River, Kelantan, April 1993 |
| *Phoebe grandis* (Nees) Merr. KL4994 | Kuala Tahan Forest Reserve, Pahang, May 2001 |
| *Phoebe scortechinii* (Gamb.) Kochummen. KL4886 | Papulut Forest Reserve, Gerik, Perak, May 1997 |
| *Phoebe lanceolata* (Wall. Ex Nees) Nees. KL 4763 | Kepayang Forest Reserve, Pahang, Nov. 1997 |
| *Dehaasia longipedicellata* (Ridl.) Kosterm. KL4719 | Raub Forest Reserve, Pahang, July 1997 |
| *Dehaasia candolleana* (Meisn.) Kosterm. KL 4683 | Rimba Teloi Forest Reserve, Sik, Kedah, Mac 1997 |
| *Dehaasia incrassata* (Jack) Kosterm. KL 4640 | Gunong Basor Reserved Forest, Jeli, Kelantan. |

## 6.3 Extraction of Plant Material

Plant extractions were carried out by cold percolation or exhaustive extraction using the soxhlet extractor, following the general procedure described below.

Dried, grounded stems/leaves of the plant were first defatted with petroleum ether (40 - 60°C), after which they were air-dried for twenty four hours, then basified with 10% ammonia and left to soak overnight. They were then reextracted with dichloromethane and methanol, successively, checking for a Mayer's negative test after each extraction.

Dichloromethane extracts were concentrated under reduced pressure to a volume of about 500 ml and were examined for alkaloid contents using TLC and spotting with Dragendorff's reagent.

The dichloromethane extracts were repeatly extracted with a solution of 5% hydrocloric acid until Mayer negative. The combined extracts were then basified with 10% ammonia solution to ca. pH 11 and then reextracted with dichloromethane. The crude alkaloid fraction was obtained as a dark gummy residue after washing the combined dichloromethane extracts with water, drying over anhydrous sodium sulphate and evaporation under reduced pressure.

The methanol extracts were first taken up to dryness and then acidified by the addition of 5% hydrocloric acid solution and left to stand overnight. The acid solution was then filtered and basified with 10% ammonia solution and reextracted with

dichloromethane. The dark residue obtained after washing, drying and evaporating to dryness were added to the crude alkaloid obtained from the dichloromethane extracts. Finally the yields of the crude alkaloid extracts from each plant material are presented in Table 6.2.

Table 6.2: Yields of alkaloid extracted from the species of *Phoebe* and *Dehaasia*.

| Species | Plant Part (Weight, g) | Weight of Crude (g) | % Yield |
|---|---|---|---|
| *Phoebe grandis* KL4224 | Bark (1000) | 7.500 | 0.750 |
| | Leaves (1500) | 7.228 | 0.480 |
| *Phoebe grandis* KL4994 | Bark (1000) | 4.020 | 0.402 |
| | Leaves (800 ) | 2.840 | 0.355 |
| *Phoebe scortechinii* KL 4886 | Bark (650) | 6.500 | 1.000 |
| | Leaves (800) | 10.500 | 1.313 |
| *Phoebe lanceolata* KL 4763 | Bark (1300) | 12.000 | 0.923 |
| | Leaves (2800) | 2.500 | 0.089 |
| *Dehaasia longipedicellata* KL 4719 | Leaves (1400) | 8.830 | 0.631 |
| *Dehaasia candolleana* KL 4683 | Leaves (1100) | 7.500 | 0.682 |
| *Dehaasia incrassata* KL 4640 | Bark (1000) | 5.940 | 0.594 |

## 6.4 Separation and purification of the alkaloids.

The basic work up on the crude alkaloid fraction of each plant follow the same general procedure described below.

The crude alkaloid fraction was subjected to column chromatography over silica gel; using the following solvents as stated below.

| Dichloromethane | : | Methanol |
|---|---|---|
| 100 | : | 0 |
| 99 | : | 1 |
| 95 | : | 5 |
| 90 | : | 10 |
| 80 | : | 20 |

100% Methanol + 1% $NH_4OH$

The fractions collected were grouped into a series of fractions monitoring with TLC. Each series were then treated separately to isolate and purify its alkaloid content by extensive column chromatography followed either by preparative TLC, as well as crystallization. Table 6.3 list the alkaloid isolated from the grouped fraction series of each plant. Purify of the alkaloids isolated were controlled by TLC (single spots), using several solvent systems.

Table 6.3: Alkaloids from the species of *Phoebe* and *Dehaasia*

| Species | Plant Part | Remarks (mg, $CH_2Cl_2$ : MeOH) |
|---|---|---|
| *Phoebe grandis* KL 4224 | Leaves | Phoebegrandine E **176** (3.9, 100: 0) |
| | | Tetrahydroglaziovine **178** (4.0, 96 : 4) |
| | | Phoebegrandine C **173** (12.4, 92 : 8) |
| | | Phoebegrandine A **28** (14, 92 : 8) |
| | | Phoebegrandine B **29** (12, 92 : 8) |
| | | Phoebegrandine D **174** (2.0, 90 : 10) |
| | Bark | Boldine **97** (7.5, 97 : 3) |
| | | Grandine A **179** (7.4, 95 : 5) |
| | | Grandine B **180** (3.4, 94 : 6) |
| | | Norboldine **98** (3.5, 94 : 6) |
| | | Lauformine **116** (5.3, 92 : 8) |
| *Phoebe grandis* KL 4994 | Leaves | Tetrahydropronuciferine **177** (9.42, 95 : 5) |
| | | Tetrahydroglaziovine **178** (1.46, 95 : 5) |
| | | Phoebescortechiniine A **175** (30.8, 93 : 7) |
| | | Phoebegrandine A **28** (24.6, 90 : 10) |
| | Bark | Grandine C **183** (4.9, 98 : 2) |
| | | Grandine D **184** (4.2, 98 : 2) |
| | | Lauformine **116** (0.8, 97 : 3) |
| | | Norhexahydromecambrine A **187** (4.3, 94 :6) |
| *Phoebe scortechinii* KL 4886 | Leaves | Tetrahydropronuciferine **177** (35, 95 : 5) |
| | | Phoebescortechiniine A **175** (30, 95 : 5) |
| | | Phoebegrandine A **28** (5.0, 92 : 8) |
| | | Phoebegrandine B **29** (2.0, 92 : 8, 90 : 10) |

Table 6.3 [Continued]

| Species | Plant Part | Remarks (mg, $CH_2Cl_2$ : MeOH) |
|---|---|---|
| | Bark | Hexahydromecambrine A **186** (13, 98 : 2) |
| | | Grandine B **180** (12, 95 : 5) |
| | | Grandine C **183** (5, 98 : 2) |
| | | Norboldine **98** (12.5, 94 : 6) |
| | | Norhexahydromecambrine A **187** (23, 95 : 5, 90 : 10) |
| *Phoebe* | Leaves | Liriodenine **135** (15, 100 : 0) |
| *lanceolata* | | Roemerine **47** (5, 98 : 2) |
| KL 4763 | | Norboldine **98** (8, 95: 5) |
| | | Laurotetanine **99** (15, 92 : 8) |
| | Bark | Liriodenine**135** (4, 100 : 0) |
| | | Roemerine **47** (18.2, 98 : 2) |
| | | Sebiferine **71** (3.0, 98 : 2) |
| | | Norboldine **98** (50, 95: 5, 90 : 10) |
| | | Asimilobine **57** (6.3, 95 : 5) |
| | | Boldine **97** (34.2, 95 : 5 ) |
| *Dehaasia* | Leaves | (+)-Milonine **190** (70, 99 : 1) |
| *longipedicellata* | | (+)-Pallidinine **188** (45, 98 : 2) |
| KL 4719 | | (-)-Pallidine **189** (30, 95 : 5) |
| | | (-)-Sinoacutine **81** (25, 96 : 4) |
| | | (-)-8,14-dihydrosalutaridine **191** (2.3, 96 : 4) |
| *Dehaasia* | Leaves | Perakensol **193** (4, 99 : 1) |
| *candolleana* | | (-)-Sebiferine **71** (15, 95 : 5) |
| KL 4683 | | (+)-Sebiferine **192** (10, 95 : 5) |
| | | (-)-Pallidine **189** (2 mg, 90 : 10) |

Table 6.3 [Continued]

| Species | Plant Part | Remarks (mg, $CH_2Cl_2$ : MeOH) |
|---|---|---|
| *Dehaasia incrassata* KL 4640 | Bark | (-)-3', 4'-Dihydrostephasubine **198** (2.5, $CHCl_3$: MeOH; 96 : 4 saturated with $NH_4OH$,) (-)-Norstephasubine **201** (2.8, $CHCl_3$: MeOH; 96:4 saturated with $NH_4OH$) (-)-Gyrolidine **194** (4.2 mg, $CHCl_3$: MeOH; 97:3 saturated with $NH_4OH$ ). Stephasubine **201** (3.5, $CHCl_3$: MeOH; 95:5 saturated with $NH_4OH$) |

## 6.5 General spectral data

**Phoebegrandine C 173: $C_{28}H_{34}N_3O_3$**

Isolated as a brown amorphous

$[\alpha]_D^{23} = +3.82^o$ (c= 0.275, MeOH)

UV $\lambda_{max}$ nm : 238 , 301

IR $\upsilon_{max}$ $cm^{-1}$ : 3270, 2918

HRES (+ve mode) MS : 460.2616 (calc. 460.2600)

EIMS : 459(3), 184(12), 316(82)

$^1$H-NMR and $^{13}$C-NMR see Table 3.2.

DEPT : $8CH_2$, $2CH(sp^3)$, $5CH(sp^2$- aromatic), $2CH_3$, 2C quaternary, 9C-tertiary.

Principle NOE's (%) in $CDCl_3$: H5' to H4' (3.9), H2' to H3' (5), H3' to H2' (8), H4' to H-5' (5), H6a to H12eq (2), H12eq to H6a (2.3), H6a to H7$\alpha$ (1.3), H6a to 6NCH3 (1.3), H6a to H7$\beta$ (1), H4$\alpha$ to H4$\beta$ (16), H4$\alpha$ to H3 (3), H7$\alpha$ to H7$\beta$ (31), H7$\beta$ to H7$\alpha$ (7.6), H7$\alpha$ to H6a (5), H7$\alpha$ to H12eq (2.3), 2-Ome to H3 (2.7), H3 to 2-Ome (8.5), 1'NH to H2' (2.7), 1'NH to H11eq (1.9), 1'NH to H9eq (1.7) H9eq to H9 ax (10.6), H9ax to H9 eq (7.4), H-9 eq to 1'NH (3.7), H11ax to H11eq (7), H11ax to H12eq (6.5), H11ax to H12eq (4.5), H7$\beta$ to H8eq (4.6).

**(-) Phoebegrandine D 174: $C_{28}H_{34}N_3O_3$.**

Isolated as a brown amorphous

$[\alpha]_D$= -22.16° (c= 0.167, MeOH)

UV $\lambda_{max}$ nm : 233.0, 300.0 nm

IR $\upsilon_{max}$ $cm^{-1}$ : 3270, 2918 $cm^{-1}$

HRES(+ve mode)-MS : $[M+H]^+$ m/z 460.2596 (cacd 460.2600)

EIMS m/z(rel. int) : 459(3), 184(12), 316(82)

$^1$H-NMR and $^{13}$C-NMR see Table 3.3.

**Phoebescortechiniine A 175: $C_{30}H_{38}N_3O_3$**

Isolated as a brown amorphous

$[\alpha]_D^{23}$ = -3.26° (*c* 1.0, MeOH)

UV $\lambda_{max}$ nm (log $\epsilon$) : 215.0 (4.55) and 272.0 (3.97).

HRMS (m/z) : $[M+H]^+$ 488.2913 (calcd.) found 488.2924.

$^1$H and $^{13}$C NMR, see Table 3.4

**(-)-Phoebegrandine A 28: $C_{29}H_{35}N_3O_3$**

Isolated as a brown amorphous

$[\alpha]_D$= -10.56° (c= 0.125, MeOH)

UV $\lambda_{max}$ nm (log $\epsilon$) : 224 (4.55), 277 (3.97), 308 sh (3.51)

EIMS m/z (rel. int.) : 473$[M]^+$ (23), 230(88), 229 (100), 227 (87),214 (81).

$^1$H and $^{13}$C-NMR see Table 3.5

**(-) Phoebegrandine B 29: ($C_{13}H_{14}N_2O$)**

Isolated as a brown amorphous

$[\alpha]_D$= -8.9° (c= 0.167, MeOH)

UV $\lambda_{max}$ nm (log $\epsilon$) : 225 (4.55), 278 (3.97), 308 sh (3.51)

EIMS m/z (rel. int.) : 473$[M]^+$ (42), 230 (42), 227 (40), 214 (100).

$^1$H and $^{13}$C-NMR see Table 3.5

**(-) Phoebegrandine E 167: $C_{16}H_{18}N_3$**

Isolated as a brown amorphous

$[\alpha]_D$= -38.44 (c=0.167, $CHCl_3$)

UV $\lambda_{max}$ nm (log $\epsilon$) : 233 (3.95), 300 (3.60) and 339 (3.87)

IR $\upsilon_{max}$ $cm^{-1}$ : 2260 (CN)

APCI (+ve mode) m/z : 252.0

EIMS m/z(rel. int) : 225 (100), 197 (30), 169 (43).

$^1$H and $^{13}$C-NMR see Table 3.6

**Tetrahydropronuciferine 177: $C_{19}H_{25}NO_3$.**

Isolated as a brown amorphous

$[\alpha]_D$ -50.0° ($CHCl_3$, *c* 1.0), $[\alpha]_D$ -1.66° (MeOH, *c* 0.391)

UV $\lambda_{max}$ (log $\epsilon$) nm : 249 (2.30), 285 (1.93), 306 (3.51),

IR $\nu_{max}$ ($CHCl_3$) : 1713 (CO),

HRES (+ve mode) m/z : 316.1957 (calc. 316.1913).

EIMS m/z (rel. int. %) : 315, 314 (100), 300, 284, 272.

$^1$H and $^{13}$C NMR, see Table 3.7.

Principle NOE's (%) in $CDCl_3$: H6a to H12eq (3.9), 12eq to H6a (6.03), H6a to H7$\alpha$ (5.2), H6a to H7$\beta$ (2.0), H4$\alpha$ to H4$\beta$ (8.3), H4$\alpha$ to H3 (1.4), H7$\beta$ to H7$\alpha$ (18.9), H7$\beta$ to H12eq (4.6), H12eq to H7$\beta$ (5.0), 2-OMe to H3 (3.9), H3 to 2-OMe (6.2), H12ax to H12eq (24.7), H8ax to H8eq (23.4), H8eq to H8ax (17.4), H8ax to 1-OMe (1.2).

**Tetrahydroglaziovine 178: $C_{18}H_{23}NO_3$**

Isolated as a brown amorphous

UV $\lambda_{max}$ nm : 226, 283, 302

IR $\nu_{max}$ ($CHCl_3$) : 3345, 1710 (CO), 856

EIMS m/z (rel. int) : $M^+$ 301, 300, 286, 270, 258.

$^1$H-NMR ppm : 3.75 (s, 3H, 2-$OCH_3$), 6.45 (s, 1H, H-3), 2.37 (3H, 6-$NCH_3$), 1.79-3.27 (aliphatic protons).

$^{13}$C-NMR see Table 3.7.

**Grandine A 179: $C_{18}H_{23}NO_3$**

Isolated as a white amorphous

$[\alpha]_D^{23}$ −50.0 (*c* =0.1, MeOH)

UV $\lambda_{max}$ nm : 226, 283 and 302 nm

IR $\nu_{max}$ (liquid film) : 1672, 3345, 945.4($OCH_2O$) $cm^{-1}$

HRES(+) : $[M+H]^+$ 302.1707 (calc. 302.1756).

EIMS: m/z (rel. int. %): 301, 300 (100).

$^1H$ and $^{13}C$ NMR see Table 3.8

**Grandine B 180: $C_{18}H_{23}NO_3$**

Isolated as a white amorphous

$[\alpha]_D^{23}$ +19.29° (*c* =0.225, MeOH)

UV: $\lambda_{max}$ (log $\epsilon$) nm : 298 (2.30), 240 (1.93)

IR: $\nu_{max}$ (liquid film) : 3285.6(OH), 908.3 (-$OCH_2O$)

EIMS: m/z (rel. int. %): 285(57), 284(79.1), 268(46), 256(46), 202(100), 189(69.7).

HR FAB MS : $[M+H]^+$ m/z 286.1439 (calc. 286.1431)

$^1H$ and $^{13}C$ NMR see Table 3.10 .

**Grandine C 183: $C_{17}H_{15}NO_4$**

Isolated as a yellow amorphous solid

$[\alpha]_D^{23}$ +100° (*c* =0. 5, $CHCl_3$)

UV: $\lambda_{max}$ (log $\epsilon$) nm : 330, 350, 360.5, 401 (3.101)

IR: $\nu_{max}$ (liquid film) : 3360, 2926,2857, 1725, 1596-1428, 1367-1212, 1132-961 (-$OCH_2O$), 858-664

HR-FAB $[M+H]^+$ : 298.1079 (calc 298.1078)

EIMS: m/z (rel. int. %): 297, 279

$^1H$ and $^{13}C$ NMR see Table 3.11

**Grandine D 184: $C_{17}H_{13}NO_4$**

Isolated as a brown amorphous

$[\alpha]_D^{23}$ +55° (*c* =1.0, $CHCl_3$)

UV: $\lambda_{max}$ nm : 250, 320

IR: $\nu_{max}$ (liquid film) : 3376, 1927, 2922

HR-EIMS : $[M+Na]^+$ 318.0768 (Calc 318.0742)

EIMS: m/z (rel. int. %): 295, 277, 249, 268, 237, 267

$^1H$ and $^{13}C$ NMR see Table 3.12

**Lauformine 116**

Isolated as a white amorphous

$[\alpha]_D$ +3.28° (MeOH, *c* 0.225)

UV: $\lambda_{max}$ (log $\epsilon$ ) nm : 300 (2.30), 247 (1.93), 267 (1.60)

IR: $\nu_{max}$ ($CHCl_3$) : 3605.5 (NH), 3420.9 (OH), 2930.8, 1125.2, 1462.4, 1251.0, 945.4 ($OCH_2O$)

HRES (+) mass spectrum: m/z 288.1632 (calc. 288.1600)

EIMS: m/z (rel. int. %): 287 (37.5), 286 100), 258 (47.5), 189 (50), 214 (22.5)

**(-)- Hexahydromecambrine A 186: $C_{18}H_{25}NO_3$**

Isolated as a white amorphous

$[\alpha]_D$ - 7.2° (MeOH, *c* 0.15)

UV: $\lambda_{max}$ (log $\epsilon$ ) nm : 300 (2.30), 247.2 (1.93), 267.0 (1.60)

IR: $\nu_{max}$ ($CHCl_3$) : 3420.9 (OH), 934 (-$OCH_2O$)

EIMS: m/z (rel. int. %): 301 (33.3), 300 (100), 258 (46.67), 203 (72),185 (20)

ESI (+) : m/z $[M+H]^+$ : 302.13

$^1H$ and $^{13}C$-NMR see Table 3.14

**(-)- Norhexahydromecambrine A 187: $C_{17}H_{21}NO_3$**

Isolated as a white amorphous

$[\alpha]_D^{23}$ –2.15 (c= 0.325, MeOH)

UV: $\lambda_{max}$ (log $\epsilon$ ) nm : 300 (2.30), 247 (1.93), 267 (1.60)

IR: $\nu_{max}$ ($CHCl_3$) : 3605.5 (NH), 3420.9 (OH), 2930.8, 1125.2, 1462.4, 1251.0, 945.4($OCH_2O$)

HRESI(+) : (m/z) $[M+H]^+$ at 288.1634 (calc. 288.1600)

EIMS: m/z (rel. int. %): 287 (33.3), 286 (100).

$^1H$ and $^{13}C$-NMR see Table 3.15

**(-)-Pallidine 189: $C_{19}H_{21}NO_4$**

Isolated as a yellow amorphous

$[\alpha]_D$ = -9.6° (c= 0.08, $CHCl_3$)

UV: $\lambda_{max}$ (MeOH) nm : 208, 245, 295

IR: $\upsilon_{max}$ ($CHCl_3$) $cm^{-1}$ : 3346, 3001, 2937, 2842, 1666, 1642, 1620, 1514, 1463, 1266, 1220, 1176, 1122, 1016, 889, 861

MS: m/z(%) : 327 (100), 312 (44.1), 299 (36.7), 284 (76.5), 268 (40.3), 256 (22.8), 242(33.8), 227(27.9)

$^1$H and $^{13}$C-NMR see Table 3.19

**(+)-Pallidinine 188: $C_{19}H_{23}NO_4$**

Isolated as a yellow amorphous

$[\alpha]_D^{23}$ = +45.0° (c= 1.0, $CHCl_3$)

UV: $\lambda_{max}$ (log $\epsilon$), MeOH nm: 205 (4.45), 228, 260 (3.99)

IR: $\upsilon_{max}$ ($CHCl_3$) $cm^{-1}$ : 1688, 1685, 1620

MS: m/z(rel. abundance,%): $M^+$ 329 (100), 314 (44.1), 286 (50), 243(15), 218(15), 192(60).

FAB(+)-MS, $[M+H]^+$ 330.1700 (calc 330.1688)

$^1$H and $^{13}$C-NMR see Table 3.18

**(+)- Milonine 190: $C_{19}H_{23}NO_4$**

Isolated as a pale yellow amorphous

$[\alpha]_D$ +40.0° (c= 1.0, $CHCl_3$).

UV: $\lambda_{max}$ (log $\epsilon$), MeOH nm: 210 (4.52), 264 (4.01),

IR: $\upsilon_{max}$ ($CHCl_3$) $cm^{-1}$ : 3500, 1680, 1620

MS: m/z(%) : $M^+$ 329 (55), 314 (100), 286 (24), 192(25).

$^1$H and $^{13}$C-NMR see Table 3.20

**(-)-Sinoacutine 81: $C_{19}H_{21}NO_4$**

Isolated as a yellow amorphous

$[\alpha]_D^{23}$ –5.75° (c= 0.12, $CHCl_3$)

UV: $\lambda_{max}$ (MeOH) nm : 241 (4.31), 278 (3.82)

IR: $\upsilon_{max}$ ($CHCl_3$) $cm^{-1}$ : 3410, 1676, 1646, 1487, 1286

HRES(+)-MS : $[M+H]^+$ 328.1574 (calc. 328.1549)

MS: m/z(%) : 327 (100), 312 (44.1), 299 (36.7), 284 (76.5), 268 (40.4), 256 (22.8), 242 (33.8), 227 (27.9)

$^1H$ and $^{13}C$-NMR see Table 3.21

**(+)-Sebiferine 192: $C_{20}H_{23}NO_4$**

Isolated as a pale yellow amorphous

$[\alpha]_D$ = +1.38° (c= 0.08, $CHCl_3$)

UV: $\lambda_{max}$ (MeOH) nm: 210, 240, 296

IR: $\upsilon_{max}$ ($CHCl_3$) $cm^{-1}$ : 2924, 2853, 1667, 1642, 1615, 1517, 1463, 1351, 1246, 1221, 1167, 1102, 1006, 888, 753, 664, 616, 532

MS: m/z(%) : 341 (100), 326 (39.8), 313 (29.1), 298 (72.8), 282 (43.0), 270 (20.3), 256 (27.2), 240 (13.9)

$^1H$ and $^{13}C$-NMR see Table 3.22

**(-)-Sebiferine 71: $C_{20}H_{23}NO_4$**

Isolated as a pale yellow amorphous

$[\alpha]_D^{23}$ = -8.47° (c= 0.275, MeOH)

UV: $\lambda_{max}$ (MeOH) nm: 209, 238, 292

IR: $\upsilon_{max}$ ($CHCl_3$) $cm^{-1}$ : 2923, 2853, 1662, 1639, 1519, 1462, 1377, 1222, 1136, 1080, 1016, 914, 888, 722

MS: m/z(%) : 341 (100), 326 (45.8), 313 (36.9), 298 (83.1), 282 (52), 270 (23.1), 256 (33.8), 240 (18.5)

$^1H$ and $^{13}C$-NMR see Table 3.22

**Asimilobine 57: $C_{17}H_{17}O_2N$**

Isolated as white amorphous solids.

$[\alpha]_D^{23}$ = -36.13° (c= 0.075, MeOH)

UV $\lambda_{max}$ (log ε) nm : 273 (4.21), 308 (3.51).

IR $\upsilon_{max}$ $cm^{-1}$ : 3255, 3550, 1035.

Mass spectrum m/e (%) : 267 (70), 266 (100), 238, 223 252, 236, 194, 177.

$^1H$ NMR (400 MHz) ppm : 3.59 (3H, s, 1-$OCH_3$), 6.70 (1H, s, H-3), 8.3 (1H, m, H-11), 2.8 3.2 (3H, m, H - 8, H - 9, H - 10).

$^{13}C$ NMR ($CDCl_3$) : 48.3 (C-1), 142 - 9 (C -2), 125 (C - 1a), 132 (C - 1b), 114.6 (C - 3), 127 (C - 3a), 28.9 (C - 4), 43.2 (C - 5), 53.6 (C - 6a), 36.1 (C - 7), 136.1 (C - 7a), 129.8 (C - 8), 128.0 (C - 9), 128.3 (C -10), 127.2 (C - 11), 131 (C - 11a), 60.4 ($OCH_3$ - C - 1).

**Roemerine 47: $C_{18}H_{17}O_2N$**

Isolated as orange yellow amorphous.

$[\alpha]_D^{23}$ = -21.92° (c= 0.075, MeOH)

UV $\lambda_{max}$ (log ε) nm : 273 (4.21), 295, 318

IR $\upsilon_{max}$ $cm^{-1}$ : 940, 1048, 1454.

Mass spectrum m/e (%) : 279, 278, 236.

$^1$H NMR (400 MHz) ppm : 6.54 (s, H-3), 2.58-2.78 (m, H-4, 7), 3.09-3.26 (m, H-5, H-6a), 7.21 (m, H-8), 7.29 (m, H-9, 10), 8.03 (d, H-11), 6.07 br s and 6.19 br s (-$OCH_2O$-), 2.58 (N-Me).

**Boldine 97: $C_{19}H_{21}NO_4$**

White amorphous solid.

UV $\lambda_{max}$ (MeOH) nm : 283 (4.21), 304 (4.23).

IR $\upsilon_{max}$ $cm^{-1}$ : 3533.4 (OH).

Mass spectrum m/e (%) : 327 (70%), 326 (100), 312, 310, 296, 284 253.

$^1$H NMR ($CDCl_3$) ppm : 87.89 (s, 1H, H - 11), 6.83 (s, 1H, H - 8), 6.63 (s, 1H, H - 3), 2.52 (3H, s, N - $CH_3$), 2.58 – 3.20 (aliphatic protons).

$^{13}$C NMR : 142 (C - 1), 148 (C - 2), 113.3 (C - 3), 130 (C - 3a), 126 (C - 1a), 125 (C - 1b), 28.9 (C - 4), 53.4 (C - 5), 62.6 (C - 6a), 34.2 (C - 7), 130.2 (C - 7a), 114.2 (C - 8), 145.1 (C - 9), 145 (C - 10), 110 (C - 11), 123 (C - 119), 56.1 (OMe - C - 10), 60.2 (OMe - C - 1).

**Norboldine 98: $C_{18}H_{19}NO_4$**

White amorphous solid.

$[\alpha]_D^{23}$ = -86.77° (c= 0.167, MeOH)

UV $\lambda_{max}$ nm (log ε) : 284 (4.13), 304 (3.17).

IR $\upsilon_{max}$ $cm^{-1}$ : 3500, 2936.

Mass spectrum m/e (%) : 313 (70), 312 (100), 282, 298, 269, 284, 253.

$^1H$ NMR ($CDCl_3$) ppm : 7.91 (s, 1H, H - 11), 6.67 (s, 1H, H - 8), 6.65 (s, 1H, H - 3), 3.60 (3H, s, OMe), 3.80 (3H, s, OMe) 2.8 - 3.20 (aliphatic protons).

**Laurotetanine 99: $C_{19}H_{21}NO_4$**

White amorphous solid.

$[\alpha]_D^{23}$ = 0 (c= 0.275, MeOH)

UV $\lambda_{max}$ nm : 278 (3.83), 221 (4.31), and 305 (4.17).

IR $\upsilon_{max}$ $cm^{-1}$ : 3350 (OH).

Mass spectrum m/e (%) : 327 (100), 326, 312, 269, 298, 253.

$^1H$ and $^{13}C$ NMR ppm : See Table 3.17

**Liriodenine 135: $C_{17}H_9O_3N$**

Crystallized as yellow needles from chloroform mp 278 - 280°C (dec) lit[146] 275 - 276°C).

UV $\lambda_{max}$ (log ε) nm : 248 (3.91), 275 (4.1), 310 (3.18) sh, 415 (3.10).

IR $\upsilon_{max}$ $cm^{-1}$ : 1660, 960, 860.

Mass spectrum m/e (%) : 275 (100), 247, 219, 189, 188, 162.

$^1$H NMR ($CDCl_3$) ppm : 6.34 (2H, s, -$OCH_2O$-), 7.17 (1H, s, H - 3), 7.62 - 7.80 (2H, m, H - 9, 10), 8.63 (1H, dd, $J$ = 8 Hz, $J'$ = 1 Hz, H - 8), 8.71 (1H, dd, $J''$ = 8 Hz, $J'''$ = Hz, H - 11), 7.77 (1H, d, $J$ = 5.4 Hz, H - 4), 8.96 (1H, d, $J$ = 5.4 Hz, H - 5).

$^{13}$C NMR ppm : See Table 3.16

**(-)-Gyrolidine 194: $C_{38}H_{42}N_2O_6$**

Isolated as a brownish amorphous state

$[\alpha]_D^{23}$ –53.0 ($c$ =0.02, MeOH)

UV $\lambda_{max}$ nm : 208, 261 and 284 nm (MeOH)

IR $\upsilon_{max}$ $cm^{-1}$ : 1635, 1508, 1356.

EI Mass spectrum m/e (%) : 622(100), 516(10), 515(25), 395(97), 381(65), 198(75).

$^1$H NMR ($CDCl_3$) ppm : See Table 3.24

**(-)-3', 4'-Dihydrostephasubine 198: $C_{36}H_{36}N_2O_6$**

Isolated as a brownish amorphous state

$[\alpha]_D^{23}$ –196.0 ($c$ =0.01, MeOH).

UV $\lambda_{max}$ nm : 215, 287 and 337 nm (MeOH)

IR $\upsilon_{max}$ $cm^{-1}$ : 3410 (OH), 1605 (C=N), 1510, 1464.

EI Mass spectrum m/z (%) : 592.1(90), 591.1(100)

APCI mass spectrum m/z : $[M+H]^+$ 593.30, $[M-H]^-$ 591.15

$^1$H and $^{13}$C-NMR : See Table 3.25.

**(-)-Norstephasubine 201: $C_{35}H_{32}N_2O_6$**

Isolated as a brownish amorphous state

$[\alpha]_D^{23}$ –46.0 (*c* =0.1, MeOH)

UV $\lambda_{max}$ nm : 219, 260 and 337 nm (MeOH)

IR $\upsilon_{max}$ $cm^{-1}$ : 3421 (OH), 1637 (C=N), 1508, 1452.

EI Mass spectrum m/e (%) : 576 (90), 575 (100)

ESI mass spectrum m/z : $[M+H]^+$ 577.2

HR ESI mass spectrum m/z : $[M+H]^+$ 577.2328

$^1H$ NMR ($CDCl_3$) ppm : See Table 3.26

**Stephasubine 202: $C_{36}H_{34}N_2O_6$**

Isolated as a brownish amorphous state

UV $\lambda_{max}$ nm : 219, 239 and 337 nm (MeOH)

APCI mass spectrum m/z : $[M+H]^+$ 591.13, $[M-H]^-$ 589.14

$^1H$ NMR ($CDCl_3$) ppm : See Table 3.26