Supplementary materials

Absolute configuration of plumeridoid C from the Amazonian traditional medicinal plant *Himatanthus sucuuba*

Birgit Waltenberger, Judith M. Rollinger, Ulrich J. Griesser, Hermann Stuppner, Thomas Gelbrich*

Institute of Pharmacy, University of Innsbruck, Innrain 52c, 6020 Innsbruck, Austria.

* Correspondence email: Thomas.Gelbrich@uibk.ac.at

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SI 1. General experimental conditions used for the isolation and identification of (I) to (XI).

All chemicals used were of analytical grade. Solvents used were either analytical grade or puriss. grade and distilled before use.

Column chromatography was performed under TLC monitoring using Merck silica gel 60, 40-63 µm or Pharmacia Sephadex® LH-20, 20-100 µm.

TLC was carried out using hexane, ethyl acetate and formic acid in varying concentrations as mobile phase and Merck silica gel 60 PF_{254} as stationary phase. The spots were detected at UV 254 and UV 366 nm as well as with staining reagents vanillin/H_2SO_4 (1% w/v and 5% v/v solutions in methanol, respectively) at VIS.

Preparative HPLC was performed on a Dionex preparative HPLC system with a UVD 170U detector.

Analytical HPLC was conducted on a Hewlett-Packard HP-1050 or a Hewlett-Packard HP-1090 system with a photodiode array detector (DAD), respectively.

HPLC-MS analysis was accomplished on a Hewlett-Packard HP-1100 system with a DAD, coupled with an Esquire 3000 plus ion-trap mass spectrometer (Bruker Daltonics) equipped with electrospray ionization (ESI) in positive and negative mode (spray voltage: 4.5 kV; sheath gas: N_2, 30 psi; dry gas: N_2, 6 L min^{-1}, 350 °C; scanning range: 50-1000 *m/z*).

NMR experiments were carried out on a Bruker TXI600 or a Bruker DRX300.

Optical rotation was measured on a Perkin-Elmer 341 polarimeter.

The FT-IR spectrum was recorded on a Bruker IFS 25 FT-IR spectrometer in transmission mode within the range of 4000 to 600 cm^{-1}. The sample was rolled on a ZnSe disk of 2 mm thickness.

SI 2. Detailed isolation protocols of compounds (I) to (XI).

Powdered bark material of *Himatanthus sucuuba* (batch number BEL0207) was purchased from Raintree Nutrition, Inc. (Carson City, Nevada, U.S.A.) in July, 2008. The identity was specified by the provider including a certificate of analysis. The sample was further examined with light microscopy. Based on the anatomic information provided by Amaral *et al.* it was confirmed that the material consists of the bark of *Himatanthus sucuuba*. A voucher specimen (No. JR-20080730-A1) has been deposited at the Institute of Pharmacy, Department of Pharmacognosy, University of Innsbruck, Austria.

The plant material (1.9 kg) was extracted with petroleum ether (RT, 2 x 2 L). The defatted and dried bark powder was then extracted with methanol (RT, 5 x 3 L) yielding 225 g of extract. The methanol extract was suspended in water and partitioned consecutively with ethyl acetate and n-butanol (4 x, respectively). The ethyl acetate extract (32 g) was subjected to column chromatography (silica gel, 9 x 25 cm) using a step gradient of dichloromethane, ethyl acetate and methanol (225:25:0, 200:5:0,

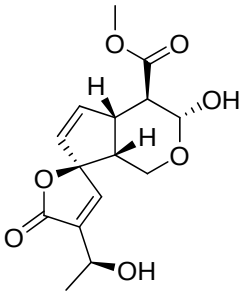
175:75:0, 150:100:0, 125:125:0, 100:150:0, 75:175:0, 150:600:0, 25:225:0, 0:1100:0, 0:450:50, 0:200:50, 0:175:75, 0:150:100, 0:125:125, 0:100:150, 0:75:175, 0:50:200, 0:25:225, 0:0:3350 mL) to give 15 fractions (S1F1-15).

Fraction S1F6 (1.28 g) was subjected to silica gel column chromatography (Merck silica gel 60 PF_{254}, 213g; 65 x 4 cm) using a gradient system of dichloromethane, ethyl acetate and methanol (500:0:0, 450:50:0, 200:50:0, 280:120:0, 120:80:0, 100:100:0, 80:120:0, 50:150:0, 0:200:0, 0:160:40, 0:140:60, 0:120:80, 0:100:100, 0:40:60, 0:60:140, 0:20:80, 0:10:90, 0:0:1750 mL) which yielded 9 fractions (S3F1-9). Fraction S3F4 (299 mg) was chromatographed by Sephadex® LH-20 [mobile phase: DCM/acetone (85:15)] to give 13 fractions (S4F1-13). Plumericin (II) was recrystallized in methanol from fraction S4F3 as colorless crystals, whereas dihydrobiochanin A (VI) was recrystallized in dichloromethane from S4F9 as beige amorphous solid. Fraction S3F5 (185 mg) was subjected to Sephadex® LH-20 column chromatography [mobile phase: DCM/acetone (85:15)] yielding 5 fractions (S5F1-5). S5F5 (42 mg) was purified using Sephadex® LH-20 column chromatography (mobile phase: acetone) to give 8 fractions (S6F1-8). S6F4 was identified as biochanin A [(V), beige microcrystalline solid].

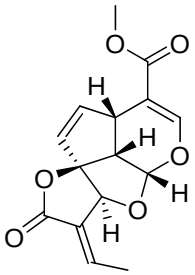
Fraction S1F8 (1.68 g) was chromatographed by Sephadex® LH-20 [mobile phase: DCM/acetone (85:15)] yielding 10 fractions (S10F1-10). Since a part of fraction S1F8 could not be eluted from the column using DCM/acetone (85:15), the Sephadex® LH-20 material was washed with methanol. This washing fraction (202 mg) was re-chromatographed using Sephadex® LH-20 (mobile phase: MeOH) to give 10 fractions (S11F1-10). S11F6 (18 mg) and S11F7 (10 mg) were separated via semi-preparative HPLC and after purification dalbergioidin [(VII), beige amorphous solid], naringenin [(VIII), yellow needles], ferreirin [(IX), light yellow amorphous solid] and dihydrocajanin [(X), beige microcrystalline solid] were yielded. S10F4 (788 mg) was suspended in methanol and filtered. The filtrate was separated by Sephadex® LH-20 column chromatography (mobile phase: MeOH) to obtain 8 fractions (S13F1-8). S13F5 (32 mg) was further separated using semi-preparative HPLC. After purification using Sephadex® LH-20 column chromatography (mobile phase: methanol) the compounds allamandicin [(IV), yellowish amorphous solid] and pinoresinol [(XI), yellowish amorphous solid] were gained, respectively. S13F6 was identical to compound (XI).

Fraction S1F10 was separated using Sephadex® LH-20 column chromatography [mobile phase: DCM/acetone (85:15)] to yield 14 fractions (S7F1-14). Compound (I) was obtained as colorless prisms by recrystallization of S7F7 from methanol. Fraction S7F8 (81 mg) was subjected to Sephadex® LH-20 column chromatography (mobile phase: MeOH) yielding 7 fractions (S8F1-7). S8F3 (43 mg) was further purified via Sephadex® LH-20 column chromatography [mobile phase: DCM/acetone (85:15)] to give 10 fractions (S9F1-10). S9F5 was identified as plumieridin [(III), light yellow amorphous solid].

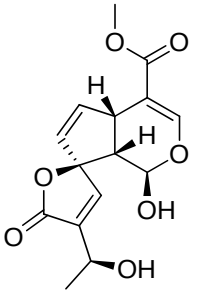
SI 3. Chemical structures of compounds (I) to (XI).



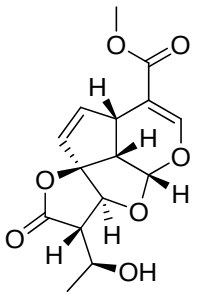
(I) Plumeridoid C



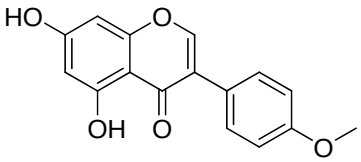
(II) Plumericin



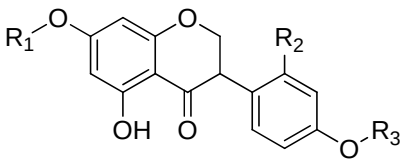
(III) Plumieridin



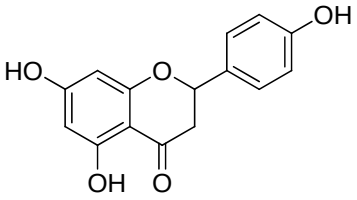
(IV) Allamandicin



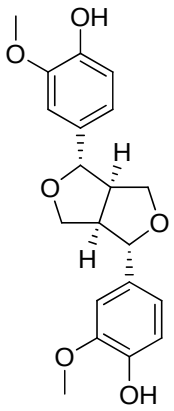
(V) Biochanin A



(VI) R_1 = H, R_2 = H, R_3 = CH_3 Dihydrobiochanin A
(VII) R_1 = H, R_2 = OH, R_3 = H Dalbergioidin
(IX) R_1 = H, R_2 = OH, R_3 = CH_3 Ferreirin
(X) R_1 = CH_3, R_2 = OH, R_3 = H Dihydrocajanin



(VIII) Naringenin



(XI) Pinoresinol

SI 4. Additionally determined parameters of compound (I).

Plumeridoid C (I):

colorless prisms; m.p.: the melting process starts at 181°C under decomposition; UV/Vis (MeOH): λ_{max} nm = 195 (sh, 1336), 214 (1785); $[\alpha]_D^{20}$ +70.54 (c = 0.97, methanol); FT-IR: V_{max} = 3329, 3190, 2953, 2891, 1758, 1732, 1465, 1440, 1379, 1318 cm^{-1}; ESI-MS: *m/z* = 309 $[M - H]^-$, 333 $[M + Na]^+$, 643 $[2M + Na]^+$.

SI 5. NMR spectral data of compounds (I) and (Ia).

Plumeridoid C (I):

^{1}H-NMR (CD_3OD, 600 MHz): 3.83 (1H, dd, *J* = 4.4, 13.1, H-1a), 3.74 (1H, dd, *J* = 1.3, 12.9, H-1b), 4.77 (1H, d, *J* = 8.5, H-3), 2.24 (1H, dd, *J* = 8.6, 11.2, H-4), 3.28 (1H, m, H-5), 6.26 (1H, dd, *J* = 2.8, 5.7, H-6), 5.64 (1H, d, *J* = 5.7, H-7), 2.68 (1H, ddd, *J* = 1.5, 4.4, 7.6, H-9), 7.35 (1H, d, *J* = 1.3, H-10), 4.56 (1H, dq, *J* = 1.4, 6.6, 6.5, 6.5, H-13), 1.42 (3H, d, *J* = 6.6, H-14), 3.75 (3H, s, H-16), δ_C values obtained from HSQC and HMBC experiments: 61.4 (C-1a), 61.4 (C-1b), 95.6 (C-3), 54.0 (C-4), 44.8 (C-5), 138.7 (C-6), 130.8 (C-7), 96.8 (C-8), 44.1 (C-9), 148.5 (C-10), 137.8 (C-11), 171.4 (C-12), 62.4 (C-13), 21.1 (C-14), 172.7 (C-15), 51.1 (C-16);

^{1}H-NMR (C_5D_5N, 300 MHz): 3.99 (1H, dd, *J* = 1.5, 12.9, H-1a), 3.84 (1H, m, H-1b), 5.29 (1H, d, *J* = 8.4, H-3), 2.74 (1H, dd, *J* = 8.3, 11.4, H-4), 3.45 (1H, ddd, *J* = 2.6, 7.5, 10.6, H-5), 6.25 (1H, dd, *J* = 2.8, 5.7, H-6), 5.60 (1H, d, *J* = 5.7, H-7), 2.81 (1H, m, H-9), 7.75 (1H, d, *J* = 1.5, H-10), 5.03 (1H, d, *J* = 6.8, H-13), 1.68 (3H, d, *J* = 6.6, H-14), 3.75 (3H, s, H-16), δ_C values obtained from HSQC and HMBC experiments: 61.9 (C-1a), 61.9 (C-1b), 96.6 (C-3), 54.8 (C-4), 45.4 (C-5), 139.1 (C-6), 131.7 (C-7), 97.0 (C-8), 44.8 (C-9), 148.2 (C-10), 139.7 (C-11), 171.7 (C-12), 62.8 (C-13), 23.1 (C-14), 173.2 (C-15), 51.7 (C-16).

Epiplumeridoid C (Ia):

^{1}H-NMR (CD_3OD, 600 MHz): 4.16 (1H, *J* = dd, 4.7, 12.7, H-1a), 3.30 (1H, m, H-1b), 5.46 (1H, d, *J* = 3.5, H-3), 2.50 (1H, dd, *J* = 3.5, 11.5, H-4), 3.48 (1H, m, H-5), 6.38 (1H, dd, *J* = 2.9, 5.7, H-6), 5.60 (1H, d, *J* = 5.7, H-7), 2.68 (1H, m, H-9), 7.32 (1H, d, *J* = 1.3, H-10), 4.56 (1H, m, H-13), 1.42 (3H, d, *J* = 6.6, H-14), 3.74 (3H, s, H-16), δ_C values obtained from HSQC and HMBC experiments: 53.8 (C-1a), 53.8 (C-1b), 89.6 (C-3), 50.1 (C-4), 37.6 (C-5), 141.2 (C-6), 130.7 (C-7), 97.7 (C-8), 44.1 (C-9), 148.6 (C-10), 137.8 (C-11), 171.6 (C-12), 62.4 (C-13), 21.1 (C-14), 171.0 (C-15), 51.1 (C-16);

^{1}H-NMR (C_5D_5N, 300 MHz): 4.45 (1H, dd, *J* = 4.6, 12.6, H-1a), 3.58 (1H, d, *J* = 12.5, H-1b), 5.92 (1H, t, *J* = 3.3, H-3), 2.76 (1H, dd, *J* = 3.4, 11.6, H-4), 3.87 (1H, m, H-5), 6.46 (1H, dd, *J* = 2.9, 5.7, H-6), 5.65 (1H, d, *J* = 5.7, H-7), 2.89 (1H, dd, *J* = 4.5, 7.7, H-9), 7.82 (1H, d, *J* = 1.5, H-10), 5.09 (1H, d, *J* = 6.7, H-13), 1.73 (3H, d, *J* = 6.6, H-14), 3.70 (3H, s, H-16), δ_C values obtained from HSQC and HMBC experiments: 54.5 (C-1a), 54.5 (C-1b), 90.2 (C-3), 51.0 (C-4), 38.2 (C-5), 141.6 (C-6), 131.6 (C-7), 97.6 (C-8), 44.5 (C-9), 148.6 (C-10), 139.7 (C-11), 171.9 (C-12), 62.9 (C-13), 23.2 (C-14), 171.5 (C-15), 51.6 (C-16).