# Costaricine, a New Antiplasmodial Bisbenzylisoquinoline Alkaloid from *Nectandra salicifolia* Trunk Bark

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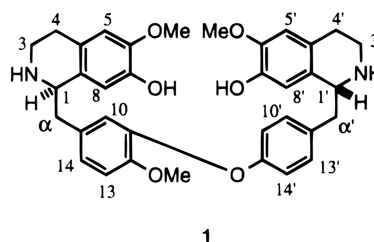
A MeOH extract of *Nectandra salicifolia* trunk bark, obtained during a diversity-based plant collection in a lower montane rainforest in Costa Rica, showed activity in an *in vitro* antiplasmodial assay measuring incorporation of [$^3$H]-labeled hypoxanthine by *Plasmodium falciparum*. In addition to 15 known alkaloids isolated from samples of trunk bark, roots, and leaves/twigs of this species, a new bisbenzylisoquinoline alkaloid (+)-costaricine [(+)-12-*O*-methyllindoldhamine] (**1**) was isolated from bark (0.038% yield) and from roots (0.001%). (+)-Costaricine was active in the antiplasmodial assay, with $IC_{50}$ values of 50 ng/mL vs. the chloroquine-sensitive D6 clone and 294 ng/mL vs. the chloroquine-resistant W2 clone of *P. falciparum*.

*Nectandra salicifolia* (H.B.K.) Nees (Lauraceae) was one of approximately 86 species in a diversity-based collection from two lower montane[1] rainforests in Costa Rica, with distinctly different species assemblages: site 1, the forested watershed of Quebrada Benjamin, north of the town of Palmar Norte, Puntarenas Province (8°96′ N, 83°28′ W), at an elevation of 300−350 m; and site 2, near the intersection of Quebrada Gonzalez and the Guapiles highway, in Braulio Carrillo National Park, Limon Province (10°10′ N, 83°48′ W), at 500−600 m. In a 0.1-ha plot in each forest, samples of trunk bark, roots, leaves/twigs, and fruits were collected where possible, from all tree species with individuals ≥5 cm in diameter at breast height (DBH) occurring in the plots. In a preliminary screening with the antiplasmodial assay of MeOH extracts of 238 samples, representing the various anatomical parts of the species collected, the extract of *N. salicifolia* bark showed this species to be one of the most active of those tested. A bioassay-guided fractionation of this sample was therefore initiated to isolate the active compound(s). To our knowledge, no previous phytochemical investigations have been conducted on this species.

Antiplasmodial assay of the fractions obtained during the initial chromatographic separation of a portion of the bark sample, followed by TLC of these fractions, indicated that the most active fractions contained alkaloids (determined by spraying the TLC plates with Dragendorff's reagent). The remainder of the samples were therefore subjected to acid/base alkaloid shake-outs,[2] and the alkaloids, purified by open column chromatography followed by preparative TLC, were tested individually in the antiplasmodial assay.

## Results and Discussion

A total of 16 alkaloids was isolated and characterized from the trunk bark, root, and leaf/twig samples collected from *N. salicifolia* (Table 1). The new bisbenzylisoquinoline (+)-costaricine (**1**) was first identified as a monobridged structure of the lindoldhamine type by the proton spectrum (Table 2).[3] This spectrum, obtained at 500.1 MHz, showed three aromatic OMe groups ($\delta$ 3.81−3.84) and no NMe groups (no peaks in the vicinity of $\delta$ 2.5). The spectrum was very similar to that of 7- or 7′-methoxylindoldhamine, with OMe groups at C-6 and C-6′, but it was at first unclear whether the third OMe in **1** occurred at C-7, C-7′ or C-12. A shift was noted in the position of the H-10 signal ($\delta$ 6.73), and there was a change in the appearance of the methoxy group region ($\delta$ 3.81−3.84), when the spectrum was recorded at different concentrations of the same sample. This is characteristic of the monobridged bisbenzylisoquinoline alkaloids and is perhaps due to differences in rotation of the monomeric units about the dimeric structure in solutions of different concentrations.




**1**

The NOESY spectrum at 500.1 MHz showed NOE connectivity between the OMe at $\delta$ 3.84 and H-13 ($\delta$

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**Table 1.** Alkaloids of *Nectandra salicifolia* (H.B.K.) Nees

| alkaloid | trunk bark[a] | roots[a] | leaves/twigs[a] |
|---|---|---|---|
| (+)-boldine[b] | 159 mg (0.135%) | 24.7 mg (0.013%) | 1.9 mg (0.0007%) |
| (+)-isoboldine[b] | 3.6 mg (0.003%) | 1.0 mg (0.0005%) | |
| (+)-isocorydine[b] | 1.5 mg (0.001%) | | |
| (+)-laurolitsine [(+)-norboldine][b] | 1390 mg (1.18%) | 730 mg (0.384%) | |
| (+)-laurotetanine[b] | 63.9 mg (0.054%) | 36.6 mg (0.019%) | 1.0 mg (0.0004%) |
| (+)-*N*-methyllaurotetanine[b] | 35.0 mg (0.030%) | 5.2 mg (0.003%) | 1.7 mg (0.006%) |
| (+)-norisocorydine[b] | 3.3 mg (0.003%) | 4.0 mg (0.002%) | |
| (+)-norpurpureine[b] | 4.5 mg (0.004%) | 1.6 mg (0.0008%) | |
| (1*R*)-coclaurine[c] | 2.8 mg (0.002%) | | |
| (1*S*)-*N*-methylcoclaurine[c] | 4.1 mg (0.003%) | 1.0 mg (0.0005%) | |
| (1*S*)-juziphine[c] | 12.4 mg (0.011%) | 8.3 mg (0.004%) | |
| (1*S*)-norjuziphine[c] | 11.7 mg (0.010%) | 5.6 mg (0.003%) | |
| (1*S*)-reticuline[c] | 23.8 mg (0.020%) | 4.6 mg (0.002%) | |
| (+)-costaricine[d] | 45.0 mg (0.038%) | 1.6 mg (0.001%) | |
| (9*S*)-sebiferine [(9*S*)-*O*-methylflavinantine][e] | 6.0 mg (0.005%) | 2.4 mg (0.001%) | 2.3 mg (0.0008%) |
| (6a*S*)-glaziovine[f] | | 0.6 mg (0.0003%) | |

[a] Amount of alkaloid isolated from respective plant part and percentage of total sample weight (trunk bark, 117.5 g; roots, 190.0 g; and leaves/twigs, 274.3 g). [b] Aporphine alkaloid. [c] Benzylisoquinoline alkaloid. [d] Bisbenzylisoquinoline alkaloid. [e] Morphinandienone alkaloid. [f] Proaporphine alkaloid.

**Table 2.** $^1$H- and $^{13}$C-NMR Assignments of (+)-Costaricine (**1**)[a]

| position | $^1$H | $^{13}$C | position | $^1$H | $^{13}$C |
|---|---|---|---|---|---|
| 1 | 4.05 (dd, 9.0, 3.5) | 56.4 | 1′ | 4.08 (dd, 10.0, 3.7) | 56.7 |
| 3 | 2.64 (m), 3.16 (m) | 40.9 | 3′ | 2.72 (m), 3.23 (m) | 40.6 |
| 4 | 2.63 (m), 2.88 (m) | 29.3 | 4′ | 2.76 (m), 2.92 (m) | 29.2 |
| 4a | | 126.3 | 4a′ | | 126.1 |
| 5 | 6.51 (s) | 111.2 | 5′ | 6.56 (s) | 111.2 |
| 6 | | 145.4 | 6′ | | 145.5 |
| 7 | | 144.0 | 7′ | | 143.9 |
| 8 | 6.69 (s) | 112.6 | 8′ | 6.73 (s) | 112.5 |
| 8a | | 130.3 | 8a′ | | 130.5 |
| α | 2.85 (m), 3.04 (dd, 14.0, 3.5) | 41.1 | α′ | 2.80 (dd, 13.5, 3.7), 3.14 (m) | 41.7 |
| 9 | | 131.4 | 9′ | | 133.1 |
| 10 | 6.73 (d, 2.0) | 120.9 | 10′ | 7.13 (br d, 8.5) | 130.4 |
| 11 | | 145.4 | 11′ | 6.86 (br d, 8.5) | 117.9 |
| 12 | | 149.6 | 12′ | | 155.9 |
| 13 | 6.92 (d, 8.5) | 112.6 | 13′ | 6.86 (br d, 8.5) | 117.9 |
| 14 | 6.96 (dd, 8.5, 2.0) | 125.1 | 14′ | 7.13 (br d, 8.5) | 130.4 |
| 6-MeO | 3.81 (s) | 55.7 | 6′-MeO | 3.82 (s) | 55.7 |
| 12-MeO | 3.84 (s) | 56.0 | | | |

[a] Chemical shifts are reported in ppm from TMS in $CDCl_3$; multiplicity and coupling constants in Hz are in parentheses.

6.92, d, $J = 8.5$ Hz), which first alerted us to the position of the third OMe at C-12, rather than at C-7 or C-7′. Further evidence was provided by a 1D NOE experiment, which showed enhancement of the proton signal at $\delta$ 3.84 by irradiation of the resonance assigned to H-13 ($\delta$ 6.92). A contour of the NOESY spectrum showed clearly that the three-proton singlet at $\delta$ 3.81 is due to 6-OMe (connectivity with H-5, $\delta$ 6.51), and the three-proton singlet at $\delta$ 3.82 can be assigned to 6′-OMe (connectivity with H-5′, $\delta$ 6.56).

The COSY spectrum at 500.1 MHz made possible the assignment of groups of aliphatic protons at positions 3 and 4 and at 3′ and 4′. NOESY connectivities between $\delta$ 2.63 (H-4a) and $\delta$ 6.51 (H-5) and between $\delta$ 2.76 (H-4′a) and $\delta$ 6.56 (H-5′) allowed the assignment of these groups of protons with COSY connectivities to rings B and B′. In addition, the COSY spectrum showed the correlation between the proton resonating at $\delta$ 4.05 with those at $\delta$ 2.85 and $\delta$ 3.04, and between the proton at $\delta$ 4.08 with those occurring with chemical shifts of $\delta$ 2.80 and $\delta$ 3.14.

Returning to the NOESY spectrum, connectivities between the signal at $\delta$ 4.05 and the signals at $\delta$ 2.64 (H-3a) and $\delta$ 6.69 (H-8) established $\delta$ 4.05 as the resonance of H-1. NOESY connectivities of H-$\alpha_a$ ($\delta$ 2.85) and H-$\alpha_b$ ($\delta$ 3.04) with H-10 ($\delta$ 6.73) completed the evidence for the assignments of these protons. Similarly, in the NOESY spectrum, the proton resonating at $\delta$ 4.08 showed connectivity with H-8′ ($\delta$ 6.73), and connectivities of H-$\alpha'_a$ ($\delta$ 2.80) and H-$\alpha'_b$ ($\delta$ 3.14) with a proton resonating at $\delta$ 7.13 (H-10′ or H-14′) completed the evidence for these assignments.

A HETCOR spectrum at 300/75.4 MHz allowed the assignment of most of the direct C-H connectivities but was ambiguous with respect to the assignments of carbons α, α′, 3, and 3′. A HETCOR spectrum at 500.1/125.8 MHz allowed the unambiguous assignments of the resonances of these four carbons.

The FLOCK spectrum at 500.1/125.8 MHz clearly showed three-bond connectivities between H-5 ($\delta$ 6.51) and C-8a ($\delta$ 130.3) and between H-5′ ($\delta$ 6.56) and C-8a′ ($\delta$ 130.5). This spectrum showed similar connectivities between H-8 ($\delta$ 6.69) and C-4a ($\delta$ 126.3) and between H-8′ ($\delta$ 6.73) and C-4a′ ($\delta$ 126.1), allowing the unambiguous assignments of these four quaternary carbons, none of which could be assigned with selective INEPT experiments. This finding was presumably due to the irradiation in each selective INEPT experiment overlapping the resonance frequencies of several protons in close proximity, giving rise to more enhancements than would be expected from irradiation of the resonance frequencies of single protons.

The chemical shifts of several other quaternary carbons with resonances occurring close together in the

**Table 3.** Antiplasmodial Activities of *N. salicifolia* Alkaloids

| compound | *P. falciparum* clone ($IC_{50}$, ng/mL) D6 | W2 |
|---|---|---|
| (+)-costaricine | 50 | 294 |
| (+)-isoboldine | 668 | 904 |
| (+)-laurolitsine [(+)-norboldine] | 1240 | 1680 |
| (+)-norpurpureine | 1510 | 1750 |
| (+)-boldine | 2130 | 1470 |
| (+)-norisocorydine | 1990 | 1900 |
| (+)-isocorydine | 1900 | 2830 |
| (+)-laurotetanine | 3900 | 2530 |
| (1*S*)-*N*-methylcoclaurine | 2730 | 3810 |
| (1*S*)-norjuziphine | 4100 | 3100 |
| (1*S*)-juziphine | 4090 | 4480 |
| (+)-reticuline | 5800 | 3650 |
| (+)-*N*-methyllaurotetanine | 7150 | 2700 |
| (9*S*)-sebiferine [(9*S*)-*O*-methylflavinantine] | 7100 | 9020 |
| standards[a] | | |
| chloroquine[b] | 3.4 ± 0.6 | 51.8 ± 36.5 |
| quinine[b] | 15.7 ± 9.2 | 38.8 ± 19.6 |
| mefloquine[c] | 4.3 ± 0.6 | 1.0 ± 0.6 |
| artemisinin[c] | 4.1 ± 2.2 | 2.2 ± 0.7 |

[a] n = 3. [b] Sigma, St. Louis, MO. [c] Walter Reed Army Institute of Research, Silver Springs, MD.

$^{13}$C-NMR spectrum were likewise assigned by this FLOCK experiment. C-6 ($\delta$ 145.4) was assigned from its connectivity with H-8 ($\delta$ 6.69), as were C-6′ ($\delta$ 145.5) with H-8′ ($\delta$ 6.73), C-7 ($\delta$ 144.0) with H-5 ($\delta$ 6.51), C-7′ ($\delta$ 143.9) with H-5′ ($\delta$ 6.56), and C-11 ($\delta$ 145.4) with H-13 ($\delta$ 6.92).

The CD spectrum of (+)-costaricine was compared with that of an authentic sample of thalibrine (1-*S*, 1′-*S*)[4] taken at the same time. The CD spectrum of (+)-costaricine was the inverse of that of thalibrine; thus, the stereochemistry of (+)-costaricine was assigned as (1*R*,1′*R*).

The results of antiplasmodial testing of the 16 alkaloids isolated from *N. salicifolia* are summarized in Table 3. (+)-Costaricine clearly has the strongest activity of the individual alkaloids isolated from this plant and tested for this activity. In the course of investigation of the antiplasmodial activity of bisbenzylisoquinoline alkaloids (to be published), this compound can be considered as one of the more interesting leads.

(+)-Costaricine was isolated from trunk bark in an appreciable concentration (0.038%), which could account for much of the antiplasmodial activity of the crude MeOH extract. Another compound that likely contributed to the antiplasmodial activity of the whole MeOH extract is (+)-laurolitsine [(+)-norboldine], obtained from bark at a much higher concentration of 1.2%. The question of whether there is an enhancement of the antiplasmodial activity of the whole extract due to an interaction of the activities of these two compounds was tested using combinations of them in varying ratios (costaricine−laurolitsine 3:1, 1:1, and 1:3). The resulting data, together with the $IC_{50}$ values for the pure compounds, were used to construct the isobologram.[5] The isobole (data not shown) was linear, indicating an additive effect of the antiplasmodial activities of these two compounds coexisting in *N. salicifolia*.

## Experimental Section

**General Experimental Procedures.** Optical rotations were measured with a Perkin-Elmer 241 polarimeter. CD spectra were obtained with a JASCO J-710 spectropolarimeter, UV spectra, in MeOH with a Beckman DU-7 spectrophotometer and IR spectra, with a Nicolet MX-1 FT-IR spectrophotometer. NMR spectra were recorded with General Electric GE Omega 500, Nicolet NMC-360, and Varian XL-300 spectrometers in $CDCl_3$ solution (with the exception of $CD_3OD$ for [+]-laurolitsine) with TMS as internal standard. The $^1$H-NMR spectrum of (+)-costaricine was obtained on the GE Omega instrument operating at 500.1 MHz. The $^{13}$C-NMR and APT spectra were obtained on the Varian XL-300 instrument, operating at 75.4 MHz. Selective INEPT experiments were conducted on the Nicolet NMC-360 spectrometer operating at 90.8 MHz. HETCOR spectra were obtained at 300/75.4 MHz and 500.1/125.8 MHz using standard pulse sequences from the Varian and GE libraries. COSY and NOESY spectra were obtained by Dr. Edward J. Kennelly at 500.1 MHz using standard programs from the GE library. The FLOCK spectrum was obtained at 500.1/125.8 MHz on the GE Omega 500 spectrometer with $^nJ_{CH}$ = 6.3 Hz. EIMS, CIMS, and HRMS(+FAB) were recorded on a Finnegan MAT-90 instrument.

**Plant Material.** Trunk bark (117.5 g), root (190.0 g), and leaf/twig (274.3 g) samples were collected from a tree of 20-m height and 22-cm diameter DBH, located on a 23% slope at an elevation of approximately 330 m, in the lower montane rainforest north of the town of Palmar Norte (8°96′ N, 83°28′ W), on 8 July, 1992. No flowers or fruit were found. A specimen was taxonomically identified by L. J. P. at the Museo Nacional de Costa Rica, and vouchers *(Böhlke 48)* were deposited at this institution and at the John G. Searle Herbarium at the Field Museum of Natural History, Chicago, IL.

**Extraction and Isolation.** A portion (50.0 g) of the air-dried trunk-bark sample was extracted with 0.5 L MeOH, and the solution was evaporated *in vacuo*. In the antiplasmodial assay, this extract produced $IC_{50}$ values of 1100 ng/mL vs. clone D6 and 9890 ng/mL vs. clone W2 of *Plasmodium falciparum*. The dried MeOH extract was resuspended in 10% $H_2O$ in MeOH and partitioned with hexane. The aqueous MeOH layer was dried, redissolved in $H_2O$, and partitioned successively with $CHCl_3$ and EtOAc. These two partitions yielded three fractions each, which included material insoluble in either layer, but soluble in MeOH.

The residues of the hexane and EtOAc fractions were inactive against *P. falciparum* ($IC_{50}$ for both clones >10 000 ng/mL), the $CHCl_3$ residue was slightly active against clone D6 ($IC_{50}$ 2040 ng/mL), while the two MeOH-soluble and the $H_2O$-soluble residues were more active ($IC_{50}$ values vs. clone D6, clone W2: 787, >10 000; 489, >10 000; and 582, 6350 ng/mL, respectively). The latter three residues were combined and subjected to vacuum-liquid chromatography Si gel 60 H (E. Merck, Darmstadt, Germany). The column was conditioned with $CHCl_3$, and fractions were eluted with increasing concentrations of MeOH in $CHCl_3$. Alkaloid-containing fractions were eluted by mixtures of $CHCl_3$/MeOH (90:10 to 80:20). The two most active fractions in this separation had $IC_{50}$ values of 191 ng/mL and 262 ng/mL vs. clone D6 and 1210 ng/mL and 1300 ng/mL vs. clone W2 of *P. falciparum*. Further purification of alkaloids was achieved by open column chromatography [Si gel 60 H with $CHCl_3$/MeOH/$NH_4OH$ (28%) 98:2:1 to

80:20:1 and $C_6H_6/CHCl_3/MeOH/NH_4OH$ 25:55:20:1] and by preparative TLC (E. Merck Si gel 60 F254, 0.25-mm thick, with $CHCl_3/MeOH/NH_4OH$ 80:20:1 or $C_6H_6/CHCl_3/MeOH/NH_4OH$ 25:55:20:1).

The remaining trunk bark (67.5 g), root (190.0 g), and leaf/twig (274.3 g) samples were extracted with MeOH (10 mL MeOH/g sample), and the solutions were dried *in vacuo*. The dried residues were treated with 1% HCl, and the acidic solutions were filtered, made basic with 28% $NH_4OH$ to pH 8−9, and repeatedly extracted with $CHCl_3$ until the aqueous layers gave negative tests with Mayer's reagent. The alkaloid mixtures were separated by vacuum-liquid chromatography (Si gel 60 H with $CHCl_3/MeOH$ 98:2 to 80:20). Further purification of alkaloids was achieved by open column chromatography (Si gel 60 H with $CHCl_3/MeOH/NH_4OH$ 80:20:1 or $C_6H_6/CHCl_3/MeOH/NH_4OH$ 25:60:15:1) and preparative TLC (Si gel 60 F254, 0.25-mm thick, with $CHCl_3/MeOH/NH_4OH$ 80:20:1 or $C_6H_6/CHCl_3/NH_4OH$ 25:60:15:1 to 25:55:20:1).

**(+)-Costaricine (1):** yellow amorphous powder ($CHCl_3/MeOH/NH_4OH$), $[\alpha]_D$ +46.4° (*c* 0.248, $CHCl_3$); CD (MeOH) $\Delta\epsilon$ (nm): 0 (300), −2.6 (289), 0 (271), +0.2 (266), 0 (262), +0.6 (248), 0 (242), −5.6 (226), −4.3 (216), −5.0 (213), 0 (206); UV (MeOH) $\lambda$ max (log $\epsilon$) 210 (4.73), 224 (sh, 4.53), 284 (4.12) nm; IR (film) $\nu$ max 2926, 2843, 1609, 1591, 1507, 1449, 1272, 1223, 1127, 1028, 802 $cm^{-1}$; HRMS (+FAB) *m*/*z* calcd for $C_{35}H_{39}N_2O_6$ $[M + H]^+$ 583.2808; found 583.2817 (20), 178 (100); CIMS *m*/*z* $[M + H]^+$ (100); EIMS *m*/*z* $[M - H]^+$ 581 (0.4), 405 (9), 403 (9), 192 (55), 179 (10), 178 (100); $^1H$ NMR and $^{13}C$ NMR (Table 2).

**Other Compounds.** The identification of the known alkaloids and determination of stereochemistry[6,7] was accomplished by comparisons of CD, UV, MS, $^1H$ NMR, and $^{13}C$ NMR with data reported previously for these alkaloids.[8−18] Additional data obtained in the current work for these compounds follow.

**(1*R*)-Coclaurine:** CD (MeOH) $\Delta\epsilon$ (nm); 0 (326), −0.2 (300), −0.9 (288), −0.1 (276), 0 (249), +0.2 (240), 0 (238), −1.8 (227), −0.4 (219), −1.1 (212), 0 (205).

**(6a*S*)-Glaziovine:** CD (MeOH) $\Delta\epsilon$ (nm), 0 (343), −4.2 (275), 0 (253), +4.3 (242), 0 (225), −4.1 (213), −3.5 (211), −4.3 (208), 0 (203).

**(+)-Isoboldine:** CD (MeOH) $\Delta\epsilon$ (nm); 0 (331), −6.9 (308), −4.8 (289), −6.4 (280), 0 (258), +34.0 (241), 0 (226), −18.9 (218), −12.2 (210), −11.5 (203).

**(1*S*)-Juziphine:** CD (MeOH) $\Delta\epsilon$ (nm), −0.5 (298), −1.2 (278), −0.6 (248), −2.8 (237), −1.3 (226), −5.1 (213), 0 (207); $^{13}C$ NMR ($CDCl_3$, 90.8 MHz) $\delta$ 60.5 (C-1), 43.9 (C-3), 21.9 (C-4), 126.2 (C-4a), 119.4 (C-5), 109.2 (C-6), 144.2 (C-7), 142.5 (C-8), 123.3 (C-8a), 38.6 (C-$\alpha$), 130.8 (C-9), 129.8 (C-10), 115.6 (C-11), 154.8 (C-12), 115.6 (C-13), 129.8 (C-14), 41.7 (NMe), 56.1 (OMe) ppm.

**(+)-Laurolitsine [(+)-norboldine]:** CD (MeOH) $\Delta\epsilon$ (nm); 0 (340), −1.5 (328), −5.9 (316), −4.6 (295), −7.4 (281), 0 (257), +59.2 (242), 0 (228), −42.7 (215).

**(1*S*)-*N*-Methylcoclaurine:** CD (MeOH) $\Delta\epsilon$ (nm), 0 (275), +0.7 (258), +4.8 (243), 0 (228), −2.2 (217), −1.6 (209).

**(+)-Norisocorydine:** CD (MeOH) $\Delta\epsilon$ (nm); 0 (326), −3.2 (304), −3.2 (292), −7.8 (274), 0 (255), +41.6 (235), 0 (221), −10.6 (214), −10.3 (212), −12.0 (206).

**(1*S*)-Norjuziphine:** CD (MeOH) $\Delta\epsilon$ (nm); −0.3 (292), −0.7 (286), 0 (276), +0.1 (273), 0 (268), −0.2 (257), 0 (248), +1.0 (238), 0 (232), −0.5 (227), 0 (219), +0.3 (217), 0 (214).

**(+)-Norpurpureine:** CD (MeOH) $\Delta\epsilon$ (nm), 0 (328), −4.1 (313), −5.1 (280), 0 (259), +32.8 (243), 0 (229), −14.8 (218); EIMS *m*/*z* $[M]^+$ 371 (100), 370 (78), 356 (29), 340 (32), 327 (13), 324 (16), 311 (14).

**(+)-Reticuline:** CD (MeOH) $\Delta\epsilon$ (nm), 0 (298), +2.0 (291), 0 (279), 0 (258), +11.7 (239), +1.4 (221), +12.5 (209), 0 (205).

**Alkaloid Yields.** The total alkaloid yields of the samples were: trunk bark, 1.5% (1.76 g/117.5 g); roots, 0.44% (0.827 g/190.0 g); and leaves/twigs, 0.0025% (0.0069 g/274.3 g).

**Antiplasmodial Assay Procedure.** The protocol used for the antiplasmodial assay, measuring incorporation of $[^3H]$-labeled hypoxanthine by *P. falciparum*, was that previously described,[19] with the following changes: (a) the complete RPMI 1640 medium contains 0.5% AlbuMAXII lipid-rich bovine serum albumin (GIBCO Laboratories, Grand Island, NY), replacing the 10% heat-inactivated type-$A^+$ human plasma; and (b) the contents of the microtiter plates were harvested on glass fiber filters (90 × 120 mm, Wallac, Turku, Finland) using a TOMTEC Harvester 96 (TOMTEC Inc., Orange, CT). The glass fiber filters were dried, and radioactivity retained on the filters was counted with a Wallac Microbeta liquid scintillation counter.

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