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EXAMINATION OF THREE *SIPARUNA* SPECIES FOR ALKALOID CONTENT

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Key Word Index—*Siparuna dresslerana*; *S. nicaraguensis*; *S. patelliformis*; Monimiaceae; alkaloids; flavinantine; *O*-methylflavinantine; liriodenine.

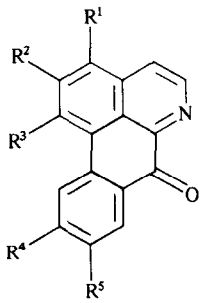
Abstract—Examination of the alkaloids of *S. dresslerana* has revealed the presence of flavinantine and *O*-methylflavinantine and two unidentified components. Traces of liriodenine were found in the twigs of *S. nicaraguensis* but no alkaloids were detected in the leaves. Traces of alkaloids were found in the methanolic extract of *S. patelliformis* but in insufficient quantity for characterization.

INTRODUCTION

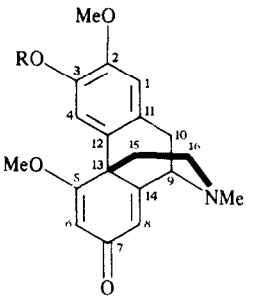
Plants of *Siparuna*, the largest genus of the Monimiaceae, are tree shrubs or climbers that inhabit tropical or subtropical regions. To our knowledge, only three species of *Siparuna* have been examined for alkaloid content. The fruit of *S. pauciflora* DC. was tested with reagents which indicated the presence of alkaloids but none were identified [1]. However, *S. guianensis* was found to contain the oxoaporphine alkaloids liriodenine (**1**) and cassamedine (**2**) [2], and *S. gilgiana* the oxoaporphine alkaloids, liriodenine (**1**) and oxonanteine (**3**) [3]. Here we report the examination of *S. dresslerana*, *S. nicaraguensis* and *S. patelliformis* for alkaloid content.

RESULTS AND DISCUSSION

We found that *S. dresslerana* contains the morphinandienone alkaloid *O*-methylflavinantine (**4**) identified by comparison of its physical and spectral properties with those reported in the literature [4–10; Lavie, D., personal communication]. An examination of the ^{13}CNMR spectrum of our sample run at 62.9 MHz in $CDCl_3$ was



1 Liriodenine $R^1 = R^4 = R^5 = H$; $R^2 + R^3 = OCH_2O$
2 Cassamedine $R^1 = OMe$; $R^2 + R^3 = R^4 + R^5 = OCH_2O$
3 Oxonantenine $R^1 = H$; $R^2 = R^3 = OMe$; $R^4 + R^5 = OCH_2O$



4 *O*-Methylflavinantine, R = Me
5 Flavinantine, R = H

also in agreement with the structural assignment. It has signals indicating the presence of 20 carbon atoms (eight quaternary, five methine, three methylene and four methyl) as determined by off-resonance and spin-sorting techniques. The ^{13}C signals that we report here can be tentatively assigned on the basis of chemical shifts and multiplicities as follows: δ180.9 (C-7), 161.7, 151.7, 148.8 and 148.5 (C-2, C-3, C-5 and C-14) 130.5 and 129.1 (C-11 and C-12), 122.4, 119.1, 111.0 and 109.5 (CH at C-1, C-4, C-6 and C-8), 61.2 (C-9), 55.7, 56.1 and 55.3 (3 OMe), 45.9 (C-16), 45.5 (C-13), 41.9 (NMe), 41.5 and 33.1 (C-10 and C-15). We believe that this is the first report of the ^{13}C spectrum of this alkaloid.

Flavinantine (**5**) was also isolated from *S. dresslerana* and identified on the basis of its mass and ^{1}HNMR spectra [11]. Other minor components were detected but not identified.

The twigs of *S. nicaraguensis* were found to contain liriodenine but no other alkaloids were detected; alkaloids were not detected in the leaves of this species. The leaves of *S. patelliformis* were extracted and examined for alkaloid content. Traces of alkaloids were indicated with test reagents but they were present in quantities insufficient

for characterization. The presence of morphinandienone alkaloids in the genus *Siparuna* is reported here for the first time.

EXPERIMENTAL

Plant material. All samples were collected in Panama by T. M. Antonio. *Siparuna dresslerana* (#5387–5389) and *S. patelliformis* (#5383–5386) were obtained in Cerro Jefe, 8.5 km from Goofy Lake toward the top of Cerro at an altitude of 900 m. *Siparuna nicaraguensis* (#5390) was collected in the vicinity of Cerro Campana, at an altitude of 700 m. Voucher specimens are deposited at the Herbarium in Field Museum (F) of Natural History, Chicago, IL under the numbers listed above.

Extraction of Siparuna dresslerana *T. Antonio.* The dried ground leaves (180 g) were extracted with MeOH in a Soxhlet for 48 hr. The MeOH was removed under N_2 and the residue extracted with 5% HCl (aq.). The aq. extract was filtered through Celite 503 and basified with conc NH_4OH. Extraction of the soln with $CHCl_3$ yielded a crude alkaloid extract which was partitioned into phenolic and nonphenolic fractions by extracting the crude extract with 5% NaOH (aq.). The nonphenolic bases remained in the $CHCl_3$ (1.3 g). The basic aq. layer was neutralized by bubbling CO_2 through the soln. Extraction of the soln with $CHCl_3$ yielded the phenolic bases (0.7 g).

Flash chromatography was performed on the nonphenolic fraction using silica gel 60 (0.04–0.063 mm) Merck, 50 cm × 2 cm. The column was developed in a stepwise manner with solns of $CHCl_3$ in MeOH at each of the following concns and in the order given: 98, 95, 90, 80, 70, 50, 30 and 0%; 250 ml portions of each soln were used.

Fractions (~18 ml) were collected and a sample of each was spotted onto a 0.2 mm silica gel TLC plate and developed with $CHCl_3$–MeOH (9:1). The plates were examined in UV light and similar fractions combined. Tests with Dragendorff's reagent and chloroplatinic acid indicated that alkaloids were present in fractions 23–33. These fractions were combined, evaporated and the residues submitted to HPLC on a preparative reverse-phase column (ODS-Magnum 9) using a MeOH–H_2O (17:3) mobile phase with a flow rate of 5 ml/min. A UV detector set at 254 nm was used. From this operation, *O*-methylflavinantine was isolated as a glass which failed to crystallize in a variety of solvents. EIMS (probe) 70 eV, *m/z* (rel. int.): 341 $[M]^+$ (100), 340 (19), 326 (32), 313 (18), 312 (12), 298 (38); high resolution mass spectrometry of $[M]^+$ (found 341.163, $C_{20}H_{23}NO_4$ requires 341.163) and $[M-Me]^+$ (found: 326.140; $C_{19}H_{20}NO_4$ requires 326.139); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1670, 1650 and 1625; UV λ_{max}^{EtOH} nm: (log ε) 231(3.4), 280 (3.0); ^1H NMR (400 MHz, $CDCl_3$): δ6.74, 6.25, 6.50, 6.56, (4 × *s*, aromatic Hs), 3.74, 3.79, 3.82, (3 × OMe) and 2.39 (*s*, NMe). The methiodide of *O*-methylflavinantine, prepared by refluxing the base with an excess of MeI in Me_2CO, was recrystallized from MeOH–Me_2CO, mp 245–250° (lit. 247–249° [7, 9], 252–254° [8]).

Fractions 63–80 also contained alkaloids. Chromatography of these fractions by HPLC (see above) yielded two nitrogen-containing components, one of which had an M_r of 325 with the base peak at *m/z* 282 and the other an M_r of 353 with the base peak at *m/z* 281. The M_rs of the components were verified by CIMS using CH_4 as the reagent gas. The compounds were not further characterized.

Flash chromatography was also performed on the phenolic fraction as described above. However, the solns used were 100, 98, 95, 90, 85, 80, 70, 50, 30 and 0% $CHCl_3$ in MeOH, each of 100-ml. Fractions were collected and examined as above. Fractions 22–28 contained a trace of *O*-methylflavinantine (**4**) while fractions 29–32 contained an alkaloid obtained as a glass which was identified as flavinantine (**5**). EIMS (probe) 70 eV, *m/z* (rel.int): 327 $[M]^+$ (100), 326 (20), 312 (35), 299 (17), 298 (13), 284 (34). ^1H NMR (80 MHz, $CDCl_3$): δ6.35, 6.40, 6.75 and 6.85 (4 × *s*, aromatic and vinylic Hs), 3.80, 3.90 (2 × *s*, 2 × OMe) and 2.45 (*s*, NMe).

Extraction of Siparuna nicaraguensis *Hemsl.* Twigs (125 g) were ground and extracted with MeOH as described above. The crude extract (0.4 g) was applied to a preparative layer silica gel 60 plate (2 mm × 20 cm × 2 cm) (Merck) and developed with $CHCl_3$. The bright yellow band $R_f \sim 0.5$ was removed from the plate and extracted into MeOH. Yellow needles were obtained on recrystallization from $CHCl_3$. The alkaloid was identified as liriodenine on the basis of its mass and ^1H NMR spectra [12, 13]. EIMS (probe) 70 eV, *m/z* (rel.int): 275 $[M]^+$ (100), 247 (17), 246 (12), 219 (10), 217 (15), 189 (22), 188 (30), 162 (21). High resolution mass spectrometry: $[M]^+$ (found: 275.060; $C_{17}H_9NO_3$ requires 275.058). ^1H NMR (250 MHz, DMSO-d_6): δ6.68 (2H, *s*, OCH_2O) and 7.60–8.98 (7H, *m*, aromatic Hs). Leaves of *S. nicarguensis* were extracted in the same manner as those of *S. dresslerana* but alkaloids were not detected.

Extraction of Siparuna patelliformis *Perk.* Leaves were extracted in the manner described for *S. dresslerana* but only traces of alkaloids were revealed. There was insufficient material for characterization.

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