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DITERPENOID ALKALOIDS FROM *ACONITUM CRASSICAULE*[1]

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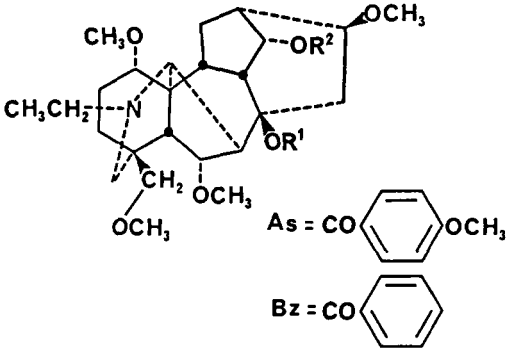
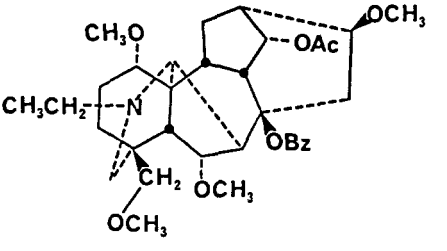
ABSTRACT.—From the roots of *Aconitum crassicaule* three new and five known diterpenoid alkaloids have been isolated. The new alkaloids are crassicaudine [**1**], crassicausine [**6**], and crassicautine [**11**]. The known alkaloids are chasmanine [**3**], crassicauline A [**5**], foresaconitine [**2**], forestine [**9**], and yunaconitine [**10**]. The isolation and determination of the structures of these alkaloids are described. The structures of the new alkaloids were established on the basis of spectral data and correlation with compounds of established structures. The structure of crassicaudine [**1**] was established by synthesis from chasmanine [**3**]. The structure of forestine [**9**] has been confirmed by correlation with crassicauline A [**5**].

The Chinese plant *Aconitum crassicaule* W.T. Wang (Ranunculaceae) has been shown to contain several diterpenoid alkaloids (1-3). In the course of studies of the total alkaloids of freshly collected roots of this plant, three new alkaloids—crassicaudine [**1**], crassicausine [**6**], crassicautine [**11**]—and five known alkaloids—chasmanine [**3**], crassicauline A [**5**], foresaconitine [**2**], forestine [**9**], and yunaconitine [**10**]—have been isolated. In this paper, we wish to describe the isolation and structure determination of these compounds. The total alkaloids obtained by an ion-exchange resin method (4) were separated by a combination of column chromatography, preparative tlc, and centrifugal tlc (Chromatotron®).

Crassicaudine [**1**] crystallized from Et_2O as colorless needles, mp 148-150°; $[\alpha]^{27}D$ +15° (*c* 0.32, $CHCl_3$); $C_{34}H_{47}NO_8$ (based on hrms); ^1H nmr δ1.06 (3H, t, J=7 Hz, NCH_2CH_3), 1.34 (3H, s, $OCOCH_3$), 3.16, 3.36 (each 3H, s, 2× OCH_3), 3.26 (6H, s, 2× OCH_3), 3.66 (2*H*, AB type, J=12 Hz, 18-Hz), 4.10 (1H, dd, J_1=6 Hz, J_2=1 Hz, 6 β-*H*, 5.10 (1H, t, J=4.5 Hz, 14 β-*H*, 7.36-8.20 (5H, m, aromatic protons); ms *m/z* 597(M^+), 566(M-OCH_3), 506(M-OCH_3-HAc), 105(100). The ^{13}C-nmr data for crassicaudine [**1**] are given in Table 1. The ^1H- and ^{13}C-nmr spectral data of crassicaudine are very similar to those of foresaconitine [**2**] (5,6) except for differences of



1 R^1=Ac, R^2=Bz (crassicaudine)
2 R^1=Ac, R^2=As (foresaconitine)
3 R^1=R^2=H (chasmanine)
3a R^1=H, R^2=Bz (14-benzoylchasmanine)



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[1]This article commemorates the 50th year of publication of the *Journal of Natural Products* (formerly *Lloydia*).

[2]Member of the Editorial Advisory Board of the *Journal of Natural Products* (*Lloydia*) since 1980.

TABLE 1. Carbon-13 Chemical Shifts and Assignments for Crassicaudine [**1**], Foresacontine [**2**] (5), Crassicauline A [**5**] (20), Crassicausine [**6**], Aminoalcohol of Crassicausine [**7**], Bikhaconine [**8**], Forestine (**9**] (15), Yunaconitine [**10**] (11), Crassicautine [**11**], and Pseudaconine [**12**] (21, 22)

Carbon	Compounds									
	10	**5**	**1**	**2**	**6**	**7**	**8**	**9**	**11**	**12**
1	83.2	84.8	85.1	85.1	85.3	85.4	85.6	85.4	83.1	84.5
2	33.7	25.9	26.4	26.4	26.3	26.0	26.0	26.0	33.4	33.8 (35.9)[a]
3	71.3	35.4	35.0	34.9	34.8	34.9	34.9	34.9	71.8	72.2
4	43.2	39.1	39.1	39.1	39.1	39.2	39.3	39.3	43.2	43.5
5	47.4	48.9	49.3	49.2	49.0	49.1	49.9	49.6	47.5	49.3
6	82.3	83.3	82.3	82.6	82.9	82.8	82.3	82.5	82.6	82.4
7	44.8	50.4	44.9	44.9	48.4	48.2	52.4	48.3 (49.2)[a]	48.4	52.2
8	85.6	85.3	85.9	85.9	78.4	78.4	72.7	73.7	78.6	72.9
9	48.8	40.9	49.3	49.3	46.5	49.5	50.4	53.6	46.1	50.4
10	40.8	44.9	44.1	43.9	36.3	35.2	42.2	42.3	35.6	42.0
11	50.3	49.8	50.4	50.3	50.6	50.4	50.1	50.2	50.8	50.2
12	35.3	34.5	29.0	29.0	36.5	36.3	36.1	36.4	36.7	35.9 (33.8)[a]
13	74.8	75.1	39.1	39.1	85.4	76.4	76.7	76.1	75.3	76.9
14	78.6	78.9	75.6	75.4	80.3	80.2	80.6	80.1	79.1	79.6
15	39.6	38.9	37.9	37.9	41.6	41.7	39.8	41.9	41.5	40.1
16	83.6	83.9	83.5	83.5	84.0	84.0	84.5	83.3	83.9	83.2
17	61.6	61.6	61.6	61.7	61.4	62.0	62.5	62.2	61.1	62.4
18	76.8	80.4	80.5	80.4	80.3	80.2	79.5	80.6	79.1	77.5
19	48.8	53.8	53.9	53.8	53.9	53.6	53.9	53.6	47.6	48.7
N-CH_2	47.4	49.1	48.9	49.0	48.6	48.6	49.3	49.2 (48.3)[a]	48.6	47.4
CH_3	13.3	13.1	13.4	13.4	13.5	13.5	13.5	13.6	13.3	13.7
1′	56.8	56.1 (55.6)[a]	56.5	56.6	56.2	56.1	56.1	56.3	55.8	56.3
6′	58.8	57.8 (56.2)[a]	57.8	57.8	58.6	58.4	57.3	58.3	58.6	57.4
8′	—	—	—	—	58.8	58.8	—	—	58.8	—
16′	57.8	58.7 (59.3)[a]	55.9	56.0	58.8	58.8	57.7	57.5	58.8	57.9
18′	59.1	59.1 (57.8)[a]	59.0	59.1	59.0	59.0	59.1	59.2	59.1	59.4
O=C(8′)	169.9	169.8	169.6	169.8	—	—	—	—	—	—
CH_3	21.7	21.3	21.7	21.8	—	—	—	—	—	—

TABLE 1. *Continued*

Carbon	Compounds									
	10	**5**	**1**	**2**	**6**	**7**	**8**	**9**	**11**	**12**
O=C	—	—	166.3	—	—	—	—	—	—	—
1	—	—	130.7	—	—	—	—	—	—	—
2,6	—	—	128.4	—	—	—	—	—	—	—
3,5	—	—	129.7	—	—	—	—	—	—	—
4	—	—	132.8	—	—	—	—	—	—	—
O=C	166.1	166.3	—	166.2	166.5	—	—	166.6	166.3	—
1	122.6	123.1	—	123.0	123.3	—	—	122.4	123.3	—
2,6	131.7	131.9	—	131.8	131.8	—	—	131.8	131.8	—
3,5	113.8	114.1	—	113.7	113.5	—	—	113.8	113.5	—
4	163.5	163.8	—	163.5	163.5	—	—	163.4	163.3	—
OCH_3	55.4	55.4 (59.7)[a]	—	55.4	55.4	—	—	55.4	55.3	—

[a]Numbers in parentheses are old values.

signals in the aromatic region. Comparison of aromatic signals of foresaconitine with those of crassicaudine indicated that crassicaudine possesses a benzoyl group instead of an anisoyl group as in foresaconitine. Hydrolysis of crassicaudine with 10% NaOH in MeOH gave chasmanine [**3**]. The results lead to **1** or **4** as the structure for crassicaudine. Alkaloids without an oxygen substituent at C(13), but bearing a C(8)-OAc and a C(14)-OBz or C(14)-OAs, show a 3H singlet for the acetate methyl group between δ 1.34-1.46 and a 1H signal for the C(14)-H between δ 5.00-5.11. Examples are 8-acetyl-14-benzoylneoline (7), foresaconitine (5,6), isodelphinine (8), penduline (9,10), vilmorrianine A (6), and 3-acetylvilmorrianine A (6). However, alkaloids with the reverse arrangement, viz. C(8)-OBz or C(8)-anisoyl and C(14) OAc, such as ezochasmaconitine (11) and anisoylchasmaconitine (11,12), show a 3H singlet between δ 1.76-1.78 and a 1H signal for the C(14)-H between δ 4.80-4.82. Because crassicaudine shows a 3H singlet at δ 1.34 and a 1H signal at δ 5.10, clearly it has structure **1**. This structure was confirmed by synthesis from chasmanine [**3**]. Benzoylation of **3** with benzoyl chloride and pyridine gave 14-benzoylchasmanine [**3a**]. Acetylation of the latter with Ac_2O and *p*-toluenesulfonic acid gave 8-acetyl-14-benzoylchasmanine, mp 154-156°, that was identical in all respects with an authentic sample of crassicaudine [**1**]. Incidentally, both crassicaudine and foresaconitine can be separated by preparative tlc over alumina but not over silica gel GF254.

Crassicausine [**6**] was isolated as an amorphous compound showing one spot on tlc (alumina, Et_2O-1% MeOH); $[\alpha]^{23}D$ +32.7° (*c* 0.20, $CHCl_3$); $C_{34}H_{49}NO_9$ (based on hrms); ir 3500(OH), 1715(ester), 1609, 770(aromatic); ^1H nmr δ 1.10 (3H, t, J=7 Hz, NCH_2CH_3), 1.5-1.8 (2H, m, 3-H_2), 2.99, 3.26, 3.54 (each 3H, s, 3× OCH_3), 3.29 (6H, s, 2×OCH_3), 3.84 (3H, s, aromatic methoxyl), 4.00 (1H, dd, J_1=6 Hz, J_2=1 Hz, 6β-*H*), 4.87 (1H, d, J=4.5 Hz, 14β-*H*), 6.90, 8.04 (4H, A_2B_2 type, -OCO-C_6H_4-); ^{13}C nmr data for crassicausine are given in Table 1; ms *m/z* 615 (M^+), 600(M-CH_3), 584(M-OCH_3, 77.7%), 554(10.2), 466(23.9), 432(13.2), 149(12.8), 135(*p*-CO-C_6H_5-OCH_3, 100).

Of the aliphatic methoxyls, the signal at δ 2.99 is at a particularly high field. The one oxygen to be accounted for in addition to those of the anisoyl and five methoxyl groups may be present as a hydroxyl group based on ir absorption at 3500 cm^{-1}. The mass spectrum exhibited an intense characteristic fragment at *m/z* 584 (77.7%) corresponding to the loss of a methoxyl on C(1) (13,14).

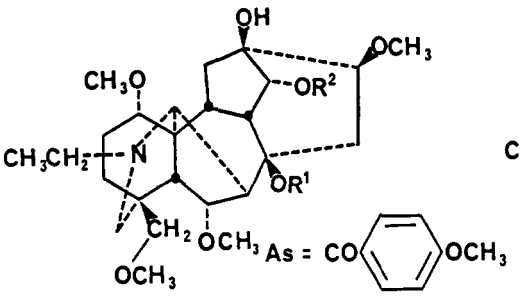
The ^{13}C-nmr data (Table 1) of crassicausine are similar to those of forestine [**9**] (15), which suggested the presence of methoxyls at C(1), C(6), C(16), and C(18). The ^{13}C-nmr spectrum of crassicausine showed the presence of two quaternary carbon atoms bearing an oxygenated group; one signal at 78.4 ppm has been assigned to C(8) and the other at 85.4 ppm must be either C(9) or C(13) because the C(14)β-proton is a doublet. The presence of a hydroxyl group at C(13) is suggested by the fact that crassicausine does not manifest any signal downfield of 28 ppm apart from the one at 26.3 ppm attributed to the C(2) methylene. If there were no OH group at C(13), one would expect the C(12) methylene triplet to appear around 26-30 ppm. Therefore, structure **6** may be assigned to crassicausine. This assignment was established by correlation of crassicausine with crassicauline A [**5**]. Because the 8-acetyl group of C_{19}-diterpenoid alkaloids is readily replaceable by an alkoxyl group by treatment with the corresponding alcohol (16), crassicauline A was heated with MeOH at 135-145° for 9 h. The product was shown to be identical in all respects with crassicausine, confirming structure **6** for this alkaloid.

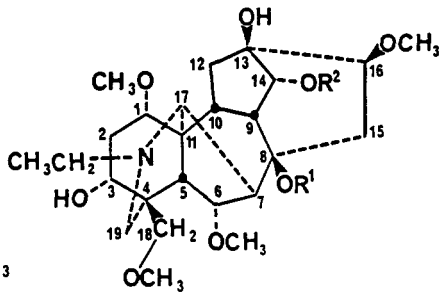
The structure of forestine was confirmed by correlation with crassicauline A [**5**]. Heating a mixture of crassicauline A perchlorate and H_2O in a sealed glass tube at 140-145° for 8 h afforded a compound identical in all respects with forestine [**9**]. The ^{13}C-

nmr assignments of C(7) and NCH_2 in forestine are corrected based on its ^{13}C DEPT spectrum.

Crassicautine [**11**] is an amorphous compound having one spot on tlc; $[\alpha]^{24}D$ +26.1° (*c* 0.18, $CHCl_3$); $C_{34}H_{49}NO_{10}$ (based on hrms); ir 3510(OH), 1715(ester), 1609, 1512, 770 (aromatic); ^1H nmr δ 1.10 (3H, t, J=7 Hz, NCH_2CH_3), 3.00, 3.27, 3.54 (each 3H, s, 3×OCH_3), 3.30 (6H, s, 2×OCH_3), 3.87 (3H, s, aromatic methoxyl), 4.05 (1H, dd, J_1=6 Hz, J_2=1 Hz, 6β-*H*), 4.89 (1H, d, J=4.5 Hz, 14β-*H*), 6.94, 8.07 (4H, A_2B_2 type, OCO-C_6H_4-); ms *m/z* 631(M^+), 616(M-CH_3), 600(M-OCH_3, 63.1%), 135(*p*-CO-C_6H_5-OCH_3, 100). ^{13}C-nmr data for crassicautine are given in Table 1.

The ir, ^1H- and ^{13}C-nmr spectra of crassicautine are very similar to those of crassicausine [**6**]; the difference of 16 mass units indicated that crassicautine possesses one more hydroxyl group. Comparison of its ^1H-nmr spectrum with that of crassicausine revealed that the two-proton multiplet at δ 1.5-1.8 due to the methylene at C(3) for crassicautine is not present. Also many features of the ^{13}C-nmr spectrum resemble those of yunaconitine [**10**] and pseudaconine [**12**]. In particular, the signal at 71.8 ppm compared with the value of 34.8 ppm for crassicausine [**6**] indicates a hydroxyl at C(3) in crassicautine.





5 R^1=Ac, R^2=As (crassicauline A)
6 R^1=CH_3, R^2=As (crassicausine)
7 R^1=CH_3, R^2=H
8 R^1=R^2=H (bikhaconine)
9 R^1=H, R^2=As (forestine)

10 R^1=Ac, R^2=As (yunaconitine)
11 R^1=CH_3, R^2=As (crassicautine)
12 R^1=R^2=H (pseudaconine)

In order to prove the above deduction, the correlation of crassicautine A with yunaconitine [**10**] was carried out. Heating **10** with MeOH at 140-145° for 9 h gave an amorphous compound [**11**] that proved to be identical in all respects with crassicautine. Therefore, crassicautine must have structure **11**.

The tlc examination of the crude extract of *A. crassicaule* (not treated with MeOH) shows the existence of crassicausine and crassicautine in the total alkaloid mixture. This result excludes the possibility that crassicausine and crassicautine are artifacts.

The ^{13}C-nmr chemical shifts and assignments for new alkaloids **1, 6,** and **11** and related compounds **2-5, 7-10,** and **12** are summarized in Table 1.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Melting points were determined on the Thomas-Kofler block (corrected), ir spectra on a Perkin-Elmer model 1420 spectrophotometer; ^1H-nmr spectra on a Varian EM-390, 90 MHz spectrometer; ^{13}C-nmr spectra on a JEOL model FX-60 spectrometer in $CDCl_3$ with TMS as an internal reference. Mass spectra were measured on a ZAB VG analytical mass spectrometer. A polyvinyl sulfonic acid ion exchange resin (H^+-form, cross linking 1×1.1, from Huabei Pharmaceutical works, China) was used in the separation of total alkaloids from the percolate.

PLANT MATERIAL.—Roots of *A. crassicaule* were collected in Yunlong (Yunnan), China, and the

plant was identified by Professor W.T. Wang, Institute of Botany, Academia Sinica, Beijing, China. A voucher specimen is deposited in that institute.

EXTRACTION AND SEPARATION.—According to the published method (4), 3,200 g of powdered roots of *A. crassicaule* was percolated with 0.05 N HCl until about 3.5 liters was collected. A column of 6,400 g of wet resin (dry weight 640 g) was used to treat the percolate. After exchange, the resin was washed repeatedly on a suction filter with deionized H_2O, spread out, and air-dried overnight. The resin was mixed well with 3,800 ml of 10% NH_4OH, placed in a Soxhlet appartus and then extracted with two portions of Et_2O (500 ml ×2) for 1 h and 3 h, respectively. Evaporation of the Et_2O extracts gave 79.7 g of total alkaloids as a white foam. This mixture (28 g) was chromatographed on a column containing 360 g of Merck alumina 60 PF254 +365 (Type E) and eluted successively with hexane, hexane-2% EtOAc, hexane-8% EtOAc, EtOAc, EtOAc-2% MeOH, MeOH (400-500 ml fractions) to afford fractions A (0.047 g), B (14.360 g), C (2.257 g), D (1.136 g), E (0.571 g), F (2.947 g), G (0.811 g), and H (1.322 g).

ISOLATION OF CHASMANINE [**3**].—Crystallization of fraction D from hexane containing a few drops of MeOH gave colorless needles (107 mg) showing one spot on tlc (silica gel GF254, $CHCl_3$-10% MeOH), mp 85-87° [lit., (17), mp 90-91°]; $[\alpha]^{29}D$ +35.8° (*c* 0.46, EtOH), which were identified as chasmanine [**3**] by comparison of the ^{13}C-nmr data with those reported (18).

ISOLATION OF CRASSICAUDINE [**1**] AND FORESACONITINE [**2**].—Fraction B (14.36 g) was chromatographed on a column containing 120 g of silica gel 60 and eluted successively with hexane, hexane-10% EtOAc, hexane-20% EtOAc, hexane-40% EtOAc, hexane-60% EtOAc, EtOAc, CH_2Cl_2-10% MeOH, MeOH (400-500 ml fractions) to give fractions B1 (0.551 g, no alkaloidal material), B2 (0.168 g), B3 (0.864 g), B4 (2.582 g), B5 (4.735 g), B6 (0.814 g), B7 (0.820 g), and B8 (0.990 g). Fraction B3 was separated on a Chromatotron (19) (silica gel GF254, 1 mm) and successively eluted with $CHCl_3$ containing increasing amounts of EtOAc (10-100%) and CH_2Cl_2 containing increasing amounts of MeOH (0.2%-10%). Combination of similar fractions gave fractions B3-1 (223 mg), B3-2 (104 mg, crude crassicauline A), B3-3 (29 mg), and B3-4 (588 mg). Fraction B3-4 (274 mg) was separated on seven preparative tlc plates (silica gel GF254, 3 mm, (20×20 cm^2), $CHCl_3$-5% MeOH-25% EtOAc, developing each plate 3 or 4 times to give the fractions B3-4A (290 mg), B3-4B (84 mg), B3-4C (73 mg), and B3-4D (11 mg). Finally, fraction B3-4C was chromatographed on a Chromatotron (19) over alumina GF254 with hexane containing increasing amounts of Et_2O (5-30%) to give crude foresaconitine (33 mg) and crassicaudine [**1**] (20 mg). The crude foresaconitine was crystallized from Et_2O to give about 20 mg of colorless needles, mp 148-152°; $[\alpha]^{27}D$+26.1° (*c* 0.41, $CHCl_3$), shown to be identical with foresaconitine [**2**] (5) by comparison of its ^{13}C-nmr spectra, tlc, mmp, and ir spectra with those of an authentic sample. The crude crassicaudine was crystallized from Et_2O to give 15 mg of colorless needles [**1**], mp 148-150°. Hrms 597.32871. $C_{34}H_{47}NO_8$ requires 597.33017.

HYDROLYSIS OF CRASSICAUDINE [**1**].—Crassicaudine (33 mg) was dissolved in 5 ml of 10% methanolic NaOH and allowed to stand at room temperature overnight. Removal of solvent under reduced pressure gave a residue which was mixed with a small amount of H_2O and extracted with CH_2Cl_2 (15 ml ×7). The extracts were dried over Na_2SO_4 and evaporated to give a yellowish residue (33 mg) which was purified by preparative tlc over alumina (20 ×20 cm^2, 3 mm, Et_2O) to give a compound (25 mg) showing a single spot on tlc. This compound was identical with chasmanine [**3**] (18) by comparison of its ^{13}C nmr, ir spectra, and tlc behavior with those of an authentic sample.

PREPARATION OF 14-BENZOYLCHASMANINE [**3a**].—To a solution of chasmanine (140 mg) in 1 ml of pyridine was added 7 drops of benzoyl chloride. The solution was allowed to stand at room temperature for 18 h. To the residue obtained on evaporation of solvent was added 15 ml of H_2O; the mixture was basified with NH_4OH to pH 11 and then extracted with CH_2Cl_2 (30 ml ×5). The combined extracts were dried over Na_2SO_4. The residue (199 mg) obtained on evaporation of solvent was separated on a Chromatotron (silica gel GF-254, 1 mm) eluting with $CHCl_3$ containing increasing amounts of MeOH (6-10.5%) to give 14-benzoylchasmanine (54 mg) [**3a**] in 20% yield. ^1H nmr δ 1.20 (3H, t, J=7 Hz, NCH_2CH_3), 3.15, 3.27 (each 3H, s, 2×OCH_3), 3.24 (6H, s, 2×OCH_3), 4.15 (1H, dd, J_1=6 Hz, J_2=1 Hz, 6β-H), 5.09 (1H, t, J=4.5 Hz, 14β-H), 7.25-8.10 (5H, m, aromatic protons). Structure **3a** was assigned on the basis of ^1H- and ^{13}C-nmr spectra.

SYNTHESIS OF CRASSICAUDINE (8-ACETYL-14-BENZOYLCHASMANINE) [**1**].—A solution of chasmanine (230 mg) in 3 ml of a mixture of dry pyridine and CH_2Cl_2 (1:1) was allowed to stand in a refrigerator for 12 h. To the residue obtained on evaporation of solvent was added 15 ml of H_2O; the mixture was basified with NH_4OH to pH 11 and then extracted with CH_2Cl_2 (30 ml ×5). The combined extracts were dried over Na_2SO_4, evaporated to give a brownish foam (254 mg) containing 14-benzoylchasmanine [**5**] as the major component in about 90% yield. This residue, dissolved in 1 ml of acetyl chloride plus a little *p*-toluenesulfonic acid, was sealed in a glass tube, and heated on an oil bath at 44° with stirring for 20 h.

Work up gave a pale brown residue which was separated on a Chromatotron of silica gel GF-254 (thick 1 mm) eluting with $CHCl_3$ containing increasing amounts of MeOH (33%) to give a foamy residue (256 mg). Separation of this residue on preparative tlc of alumina (20 ×20 cm^{-2}×3), eluting with a solvent system of hexane-30% Et_2O, gave a crude 8-acetyl-14-benzoylchasmanine (186 mg) in 69.1% yield. Crystallization of the crude 8-acetyl-14-benzoylchasmanine from Et_2O plus a little MeOH gave 102 mg of colorless needles having one spot on tlc. This compound was identical with natural crassicaudine [**1**] by direct comparison of their tlc, mmp, specific rotation, ir, and ^{1}H- and ^{13}C-nmr spectra.

ISOLATION OF CRASSICAULINE A [**5**] AND YUNACONITINE [**10**].—Fraction B5 (4.735 g) was crystallized from Me_2CO/MeOH to give granular crystals (4.069 g) that were recrystallized from the same solvent mixture to afford granular crystals (3.792 g) having one spot on tlc, mp 160-164°; $[\alpha]^{25.5}$D +33.4° (*c* 0.625, $CHCl_3$). This compound was identified as crassicauline A (20) by its ^{13}C-nmr spectral data.

Fraction B7 (0.820 g) was a yellowish, amorphous substance, which was crystallized from Et_2O containing a little Me_2CO to give granular crystals, mp 140-143°; $[\alpha]^{27}$D +30.6° (*c* 0.47, $CHCl_3$). This compound was identified as yunaconitine [**10**] (11) by its ^{13}C-nmr spectrum.

ISOLATION OF CRASSICAUSINE [**6**] AND CRASSICAUTINE [**11**].—Fraction B8 (0.990 g) was chromatographed on preparative tlc (silica gel GF254, 20×20 cm^2×7, 3 mm, $CHCl_3$-10% MeOH, 8-10 times development) to afford the fractions B8-1 (3 mg), B8-2 (52 mg), B8-3 (157 mg), B8-4 (170 mg), B8-5 (205 mg), B8-6 (149 mg). Fraction B8-4 was separated over silica gel GF254 on a chromatotron with hexane-50% $CHCl_3$, hexane-90% $CHCl_3$, and $CHCl_3$ containing increasing amounts of MeOH (2-20%) to give the fractions B8-4A (52 mg), B8-4B (81 mg), and B8-4C (35 mg). Fraction B8-4B was separated again on a Chromatotron over alumina 60 GF254 with Et_2O containing increasing amounts of MeOH (0,0.5, 5%) to give the fractions B8-4B-1 (6 mg), B8-4B-2 (crassicausine, **6**, 24 mg), B8-4B-3 (30 mg), and B8-4B-4 (crassicautine, **11**, 23 mg). Hrpms of crassicausine: 615.34044. $C_{34}H_{49}NO_9$ requires 615.34073. Hrpms of crassicautine: 631.33807. $C_{34}H_{49}NO_{10}$ requires 631.33565.

ISOLATION OF FORESTINE [**9**].—Fractions B8-4A, B8-4B, B8-4B-1, B8-4B-3 with similar fractions obtained from B7-5 were combined and separated by preparative tlc over alumina GF254 (20×20 cm^2×7, 3 mm, Et_2O) to give amorphous forestine (15) (99 mg), having one spot on tlc. It was identified by direct comparison of its ^{13}C nmr, ir spectra, and tlc with those of an authentic sample.

CONVERSION OF CRASSICAULINE A [**5**] TO FORESTINE [**9**].—Crassicauline A (1 g) was dissolved in 10 ml of 70% EtOH with warming, and perchloric acid was added to pH 4 (Congo Red). Evaporation of the solution gave a white residue (1.25 g). The residue (500 mg) was added to 4 ml of distilled H_2O in a sealed glass tube, and the mixture was heated at 140-145° for 8 h. When cooled, colorless needles of crassicauline A perchlorate (11 mg) separated. The mother liquor was basified with NH_4OH to pH 10, extracted with CH_2Cl_2, dried (Na_2SO_4), and evaporated to give a pale yellow residue (260 mg) which was separated by preparative tlc [alumina, (20×20 cm^2)×5, Et_2O-20% hexane] to give crude material corresponding to forestine (121 mg). This material was separated again by preparative tlc as well as on a small column of silica gel (72-325 mesh), eluting with CH_2Cl_2 and MeOH to give an amorphous compound (42 mg), $[\alpha]^{28}$D +55.7° (*c* 0.42, $CHCl_3$) that proved to be identical with forestine [**9**] (15) by comparison of its ^{13}C nmr, ir spectra, and tlc with those of an authentic sample.

CONVERSION OF CRASSICAULINE A [**5**] TO CRASSICAUSINE [**6**].—A solution of crassicauline A (100 mg) in 5 ml of MeOH was sealed in a glass tube and heated in an oil bath at 135-145° for 9 h. Evaporation of the mixture gave a white residue. To this a small amount of H_2O was added; the mixture was basified with NH_4OH to pH 11, extracted with $CHCl_3$ (15 ml×4), dried over Na_2SO_4, and evaporated to dryness to afford a white residue (99 mg). The latter was purified on a Chromatotron [silica gel GF254, 1 mm, eluting with $CHCl_3$ containing increasing amounts of MeOH (1 to 50%)], followed by passing through a column (6 mm diameter) of 1 g silica gel covered with 1 g of alumina to give an amorphous compound (32 mg) having one spot on tlc (alumina, Et_2O-10% hexane), $[\alpha]^{23}$D +32.0° (*c* 0.19, $CHCl_3$) [crassicausine: $[\alpha]^{23}$D +32.7° (*c* 0.20, $CHCl_3$)]; ir 3500 (OH), 1715 (ester), 1609, 770 (cm^{-1}) (aromatic). This compound is identical with crassicausine [**6**] based on ir, ^{1}H- and ^{13}C-nmr spectral data.

PREPARATON OF COMPOUND [**7**].—A solution of crassicauline A (200 mg) in 5 ml of MeOH was sealed in a glass tube, heated in an oil bath at 135-145° for 9 h, and then evaporated to dryness under the reduced pressure to give a white foam. To this 5 ml of 10% NaOH in MeOH was added, and the mixture was allowed to stand at room temperature overnight. After completion of reaction (by monitoring with tlc), the reaction mixture was evaporated; to this a small amount of H_2O was added, the mixture was extracted with $CHCl_3$ (20 ml×5), and evaporated to give a white residue (117 mg). This residue was separated on a Chromatotron (alumina, 1 mm, eluting subsequently with Et_2O, $CHCl_3$, $CHCl_3$-5% MeOH and $CHCl_3$-50% MeOH) to give an amorphous compound [**7**] (44 mg) having one spot on tlc. ^{1}H nmr δ 1.03 (3H, t, J=7 Hz, NCH_2CH_3), 1.44-1.72 (2H, m, 3-H_2), 3.20, 3.21, 3.30, 3.32, 3.45 (each 3H, s, 5× OCH_3), 4.13 (1H, dd, J_1=6 Hz, J_2=1 Hz, 6β-*H* . For ^{13}C-nmr data see Table 1.

PREPARATION OF BIKHACONINE [**8**].—To 99 mg of crassicauline A [**5**] was added 4 ml of 10% NaOH in MeOH. This mixture was allowed to stand at room temperature for 3 days and then was evaporated to give a residue (83 mg) that was separated on a Chromatotron [silica gel GF254, 1 mm, eluting with $CHCl_3$ containing increasing amounts of MeOH (10-50%)]. The amorphous product (53 mg) was purified again on a Chromatotron [alumina, 1 mm, eluting with $CHCl_3$ containing increasing amounts of MeOH (10-50%)] to obtain a white amorphous compound (32 mg) that proved to be bikhaconine [**8**] on the basis of ^{1}H- and ^{13}C-nmr (Table 1) analysis.

CONVERSION OF YUNACONITINE [**10**] TO CRASSICAUTINE [**11**].—A mixture of yunaconitine (70 mg) and 5 ml of MeOH was sealed in a glass tube, heated in an oil bath at 140-145° for 9 h, and evaporated to give a residue. To this a little H_2O was added, the mixture was basified with NH_4OH to pH 11, extracted with $CHCl_3$ (15 ml×4), dried over Na_2SO_4, and evaporated to dryness to afford a white residue (67 mg). This residue was separated on a Chromatotron [silica gel GF254, 1 mm, eluting with $CHCl_3$ containing increasing amounts of MeOH (5-50%)] to give an amorphous compound (57 mg) having one spot on tlc, $[\alpha]^{24}D$ +21.3° (*c* 0.15, $CHCl_3$, less than natural crassicautine possibly due to impurities) [crassicautine: $[\alpha]^{24}D$ +26.1° (*c* 0.18, $CHCl_3$)]; ir 3510 (OH), 1715 (ester), 1609, 1512, 770 (aromatic). This compound was identified as crassicautine [**11**] by direct comparison of ir, ^{1}H- and ^{13}C-nmr spectra with those of natural crassicautine.

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