

AMBIGUINE AND DIHYDROAJACONINE: TWO NEW
DITERPENOID ALKALOIDS FROM CONSOLIDIA AMBIGUA¹

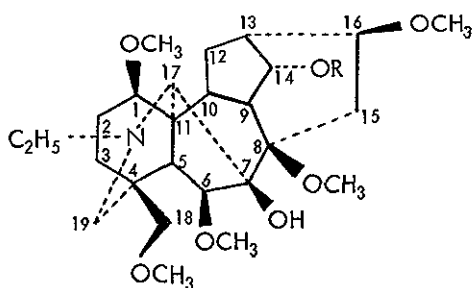
S. William Pelletier*, Rajinder S. Sawhney, and Naresh V. Mody
Institute for Natural Products Research and the Department of Chemistry,
University of Georgia, Athens, Georgia, 30602 U. S. A.

Chemical investigation of the minor alkaloids of Consolidia ambigua has led to the isolation and characterization of two new diterpenoid alkaloids, ambiguine (1) and dihydroajaconine (7). Ambiguine is the first lycotonine-type alkaloid in which a methoxy group is present at the C(8)-position, and dihydroajaconine is a reduced form of the known alkaloid, ajaconine.

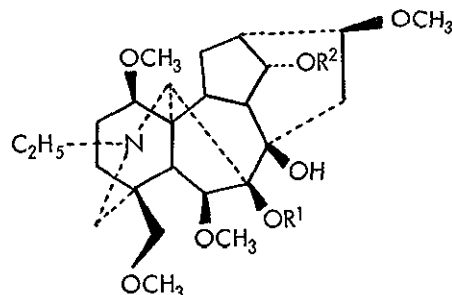
Consolidia ambigua¹ is an ornamental plant which is commonly known as garden larkspur. Some of the Consolidia (earlier reported as Delphinium) plants are known to possess insecticidal and growth inhibiting activities.² Recently, we have reported³ the structures of two new alkaloids, ajacusine and ajadine, isolated from the seeds of garden larkspur.¹ Further work on this plant has resulted into the isolation of two new minor alkaloids, ambiguine and dihydroajaconine. In this communication, we report the structures of ambiguine (1) and dihydroajaconine (7) by the aid of ¹³C nmr spectroscopy.

Ambiguine, C₂₈H₄₅NO₈ (element analysis and MS), mp 106-108° (corrected), $[\alpha]_D^{25} + 38^\circ$ (c 1.0, CHCl₃) was isolated from the 85% ethanol extract of the seeds by a combination of gradient pH separation, thick layer, and column chromatographic techniques. Infrared absorption at 3560, 1735 and 1090 cm⁻¹ indicated the presence of hydroxyl, acetate, and ether groups, respectively. The ¹H nmr spectrum of ambiguine in CDCl₃ exhibited the presence of an N-CH₂-CH₃ (3H, t, J 7.5 Hz) centered at δ 1.03, -OCOCH₃ group (3H, s) at δ 2.05 and 5 aliphatic methoxy groups (each 3H, s) at δ 3.28, 3.35, 3.38, 3.48 and 3.55. A doublet of doublets observed at δ 4.72 is typical

of a C(14) β -proton when an α -hydroxy group at C(14) is esterified. On the basis of these data and the nature of other C₁₉-diterpenoid alkaloids isolated from *C. ambigua*, we conclude that ambiguaine has a lycocotonine-type skeleton. Alkaline hydrolysis of ambiguaine (5% KOH in methanol) gave the aminoalcohol (2) which on acetylation (Ac₂O/pyridine) regenerated ambiguaine. Treatment of ambiguaine with Ac₂O/BF₃ or Ac₂O/*p*-toluenesulfonic acid did not acetylate the tertiary C(7)-hydroxy group, probably because of steric factors. The fragmentation pattern of ambiguaine in the mass spectrum also indicated the presence of a lycocotonine-type skeleton. The base peak at M⁺-31 in the mass spectrum revealed the presence of a methoxy group at C(1).⁴



1 R = Ac Ambiguaine
2 R = H



3 R¹ = R² = CH₃
4 R¹ = H; R² = CH₃ Delphatine
5 R¹ = R² = H Browniine
6 R¹ = H; R² = Ac

The ¹³C nmr spectrum of ambiguaine is consistent with structure 1. The chemical shifts of the 28-carbons in the molecule have been assigned (Table 1) on the basis of noise decoupled spectra, direct analysis of non-protonated carbons, single-frequency off-resonance decoupling (SFORD) techniques and by comparison with the ¹³C nmr spectra of closely related alkaloids.⁵ The signal at 75.0 ppm supports the presence of a C(14)-

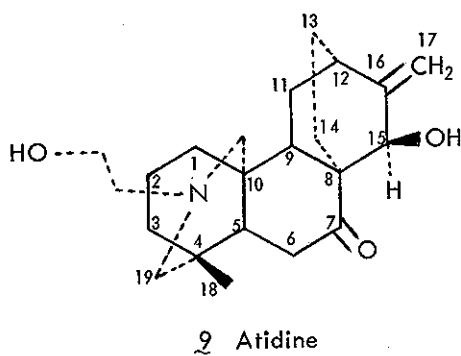
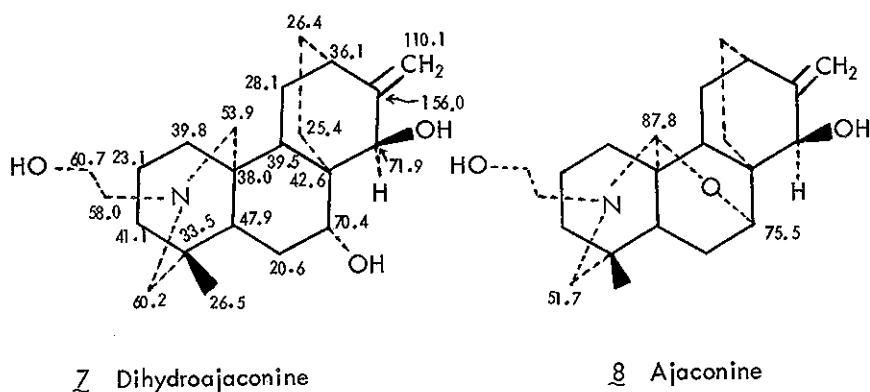
acetyl group. The chemical shift does not change (74.9 ppm) when the alkaloid is hydrolyzed to the aminoalcohol (2). We have observed⁵ the same behavior with several known C₁₉-diterpenoid alkaloids having an α -C(14)-acetate group. Comparison of the ¹³C nmr spectra of ambiguine with that of its hydrolyzed product (2) demonstrated the shielding (1.6 ppm) effect of the C(14)-acetate on the chemical shift of C(17) (Table 1). A similar effect has been observed in case of browniine (5) and its 14-acetate (6).

In order to exclude the possibility of a methoxy group at C(7) in ambiguine, the ¹³C nmr spectra of some model compounds were required. For this purpose, we have prepared the known compounds O,O dimethyllycoctonine (3) and delphatine (4) from lycoctonine by methylation with CH₃I and NaH at 110° for 26 hr. Correlation of compounds 3 through 6 with ambiguine and its hydrolyzed derivative was made through a study of their carbon-13 nmr spectra (Table 1). This comparison afforded evidence for the presence of a methoxyl group at C(8) as well as the basic lycoctonine-type skeleton in ambiguine. These ¹³C nmr studies indicate that ambiguine is similar to 14-acetylbrowniine except for the presence of a methoxyl group at C(8). The carbon-13 chemical shifts of ambiguine are in agreement with its structure (1). Although over twenty-five lycoctonine-type alkaloids are well known in nature, ambiguine is the first to bear a methoxyl group at C(8).

Dihydroajaconine, C₂₂H₃₅NO₃, mp 99-100° C, $[\alpha]_D^{24} - 35^\circ$ (c 1.0 Abs. EtOH), was isolated from the mother liquors accumulated during the isolation of ajaconine from garden larkspur. The ir spectrum in KBr exhibited peaks at 3350 cm⁻¹ (OH), 1660 and 920 (>C=CH₂). The ¹H nmr spectrum of dihydroajaconine in CDCl₃ revealed the presence of a singlet at δ 0.80 for the C(4)-methyl group and a multiplet centered at δ 5.12 for the exocyclic methylene group.

The ¹³C nmr spectrum of dihydroajaconine (7) showed the presence of one methyl group, eleven methylene carbons, five methine carbons, four quaternary carbon atoms, and two olefinic carbons. Comparison of the ¹³C nmr spectrum of dihydroajaconine with that of ajaconine (8)⁶ indicated that the only difference between these two alkaloids was the presence of a C(7)-hydroxyl group in the α -configuration in 7 instead of a C(7)-C(20) carbinolamine ether linkage. Unambiguous carbon signal assignment in dihydroajaconine (7) was achieved by using conventional techniques, additivity relationships, a direct

analysis of non-protonated carbon centers and by comparison with the ^{13}C nmr spectra of closely related alkaloids⁷, e.g. atidine, ajaconine.



Reduction of ajaconine with sodium borohydride in aq. methanol afforded, in a quantitative yield, a product which was identical with natural dihydroajaconine. This correlation established the complete structure of the new alkaloid as **7**. To our knowledge, dihydroajaconine has not previously been isolated as a natural product.⁸

Biogenetically, dihydroajaconine may be an important precursor between ajaconine (**8**) and atidine (**9**)⁹.

Table 1. Carbon-13 Chemical Shifts and Assignments for Ambiguine and Related Alkaloids

Carbon	1	2	3	4	5	6
1	83.4	83.9	84.3 ^c	83.9 ^c	85.2	84.2
2	25.3	24.9	26.2	26.2	25.5	26.2
3	31.5	31.7	32.5	32.4	32.5	32.4
4	38.3	38.6	38.1	38.1	38.4	38.1
5	40.9 ^e	42.7 ^e	44.6 ^e	43.3 ^e	45.1 ^e	42.6 ^e
6	91.1	91.0	85.1	90.6	90.1	90.3
7	90.0	89.9	92.8	88.4	89.1	88.3
8	80.4	79.6	80.0	77.5	76.3	77.1
9	52.0	52.6	50.5	49.8	49.6	51.2
10	35.9	36.6	39.0	38.1	36.4	38.1
11	47.2	46.4	49.0	48.9	48.2	49.5
12	27.4	27.6	29.0	28.7	27.5	28.2
13	46.1 ^e	46.7 ^e	45.7 ^e	46.1 ^e	46.1 ^e	45.7 ^e
14	75.0	74.9	84.7 ^c	84.3 ^c	75.3	76.0
15	28.5	26.8	33.5	33.5	33.1	33.7
16	81.9	82.3	82.9	82.6	81.7	82.4
17	66.7	68.3	66.5	64.8	65.4	64.8
18	79.3	79.1	78.2	78.1	78.0	78.0
19	53.6	53.9	52.3	52.8	52.7	52.7
N-CH ₂	52.6	52.9	51.5	51.1	51.3	48.8
 CH ₃	14.9	15.3	14.0	14.2	14.3	14.2
1'	55.6	55.7	55.8 ^d	55.7	56.0	55.8
6'	59.4	59.2	56.0 ^d	57.3	57.5	57.3
7'	-	-	55.5	-	-	-
8'	53.7	54.0	-	-	-	-
14'	-	-	57.8	57.8	-	-
16'	56.3	56.5	56.3	56.3	56.5	56.2
18'	59.4	59.5	59.1	59.0	59.1	59.0

Carbon	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>	<u>5</u>	<u>6</u>
-C=O	171.3	-	-	-	-	171.9
 CH ₃	21.4	-	-	-	-	21.5

^a Chemical shifts in ppm downfield from TMS. Solvent deuteriochloroform.

^b Spectra were taken at 15.03 MHz in the Fourier mode using a FX-60 spectrometer in conjunction with a JEC-980 computer.

^{c,d} These assignments may be interchanged in any vertical column.

^e These assignments may be interchanged in any vertical column. At the present we do not have model compounds with a C(13)-hydroxyl or methoxyl group to confirm the assignments.

ACKNOWLEDGMENT We thank Dr. L. H. Zalkow for providing a sample of mother liquors accumulated during the isolation of ajaconine from garden larkspur.

REFERENCES

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- 8 Sastry and Waller (Chem. and Ind., 1972, 381) reported the presence of dihydro-ajaconine in GC-MS studies of ajaconine isolated from Delphinium ajacis. A sample of what had been earlier identified as pure ajaconine furnished a mixture of five components when the deuterated trimethylsilyl derivative was analysed on the GC-MS. The temperature on the GC column (215°C) and the time required for elution (12.7-27.8 min.) may have been responsible for the rearrangement products. However, the authors believed their evidence demonstrated that the ajaconine sample was a mixture of five components.
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