NEW DITERPENOID ALKALOIDS FROM *DELPHINIUM STAPHISAGRIA* LINNE

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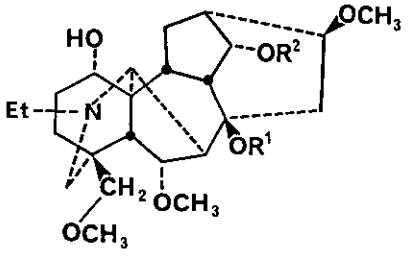
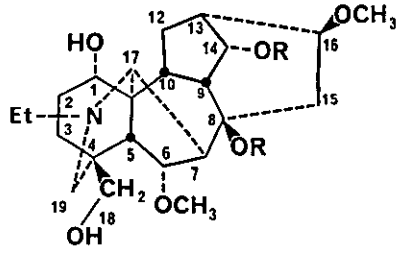
Abstract — Delstaphidine (7) and neolinine (8), two new C_{19}-diterpenoid alkaloids, and α-oxodelphinine (9) have been isolated from the seeds of Delphinium staphisagria. The structures of these alkaloids were determined on the basis of spectral data, correlation with alkaloids of established structures, preparation of certain derivatives and by synthesis. α-Oxodelphinine has not been isolated from a natural source previously. N-Deethyldelphisine (10) and N-deethyldelstaphidine (11), two new synthetic diterpenoid alkaloids, have been prepared by $KMnO_4$ oxidation of delphisine. Oxidative cyclization of delphisine with alkaline potassium ferricyanide gave delstaphinine (6)

The seeds of *Delphinium staphisagria* L., when extracted with ligroin yield an alkaloidal fraction of which delphinine[1] is the major component. The mother liquors accummulated during the isolation of a large quantity of delphinine furnished an amorphous fraction from which delphisine (1)[2], delphidine (2)[3], delphirine (1-epineoline)[4] and several novel *bis*-diterpenoid alkaloids have been isolated.[5-7]





1	$R^1 = R^2 = Ac$	Delphisine	4	R = Ac	Delstaphisagrine
2	$R^1 = Ac$; $R^2 = H$	Delphidine	8	R = H	Neolinine
5	$R^1 = H$; $R^2 = Ac$	Delstaphisagnine			
15	$R^1 = R^2 = H$	Neoline			

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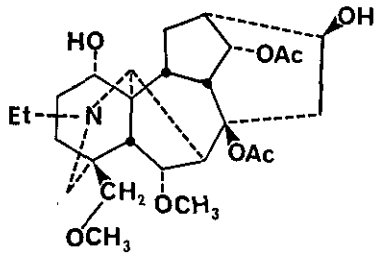
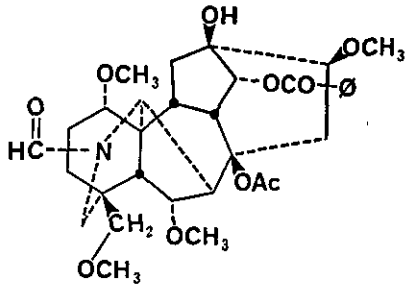
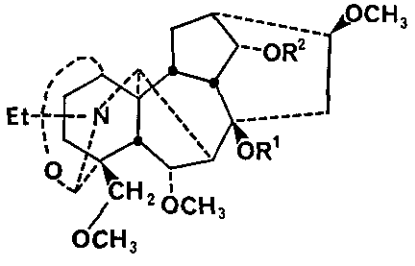
Recently the isolation of four new alkaloids, delstaphisine (3)[8], delstaphisagrine (4)[8], delstaphisagnine (5)[8], delstaphinine (6)[10] and the alkaloid 1-dehydrodelphisine[10] has been reported. In this paper we report separation of the amorphous fraction by a combination of gradient pH separation,[11] vacuum liquid chromatography (vlc)[12], preparative tlc and centrifugally accelerated, radial, thin-layer chromatography ("Chromatotron")[13,14] to give two new C_{19}-diterpenoid alkaloids, delstaphidine (7), neolinine (8) and the alkaloid, α-oxodelphinine (9). The latter has not been reported in nature before, but had been prepared by oxidation of delphinine.[15]

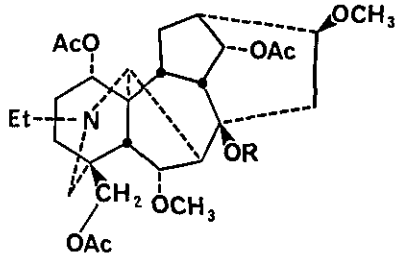


3 Delstaphisine



9 α-Oxodelphinine





6 $R^1 = R^2 = H$ Delstaphinine

7 $R^1 = R^2 = Ac$ Delstaphidine

12 $R^1 = H$; $R^2 = Ac$ 14-Acetyldelstaphinine

13 R = Ac Neolinine tetraacetate

14 R = H Neolinine 1,14,18,-triacetate

Delstaphidine (7) was obtained in a crystalline form, mp 192.5-194.5°C, $[\alpha]_D^{25}$ +27.4° (c, 0.27, $CHCl_3$), and its molecular formula $C_{28}H_{41}NO_8$ was deduced from the mass spectral (M^+, 519), and carbon-13 nmr data. The proton nmr spectrum exhibited the following signals: δ 1.08 (3H, t, J= 7 Hz, N-CH_2-CH_3), 1.92 and 1.96 (each 3H, s, OCOCH_3), 3.12, 3.22 and 3.26 (each 3H, s, OCH_3), 3.47 (1H, s, 19-H) and 4.72 (1H, dd, $J_1 = J_2 = 4.5$ Hz, C(14)-β-H).

The noise decoupled ^{13}C nmr spectrum of delstaphidine (7) exhibited 27 signals for the 28 carbon atoms of the molecule (Table 1). The mass spectrum showed a molecular ion M^+, m/z 519 (3%),

504 (M^+-CH_3, 7%), 489 (M^+-$2CH_3$, 13%), 488 (M^+-OCH_3, 32%), 474 (M^+-$3CH_3$, 2%), 463 (M^+-C_3H_4O, 34%), 433 ([M^+-$2CH_3$]-C_3H_4O, 6%), 432 ([M^+-OCH_3]-C_3H_4O, 17%), 234 (14%), 152 (60%), 45 (58%) and 43 (100%).

Structure **7** was deduced for delstaphidine from its spectral data and comparison of its data with those of the neoline group of aconitine-type C_{19}-diterpenoid alkaloids[16] and with those of delstaphinine[10], pentagynine[16], gadesine[17], 18-methoxygadesine[16] and nevadenine[17].

The DEPT spectra of delstaphidine exhibited 5 singlets at 46.5, 50.4, 84.1, 169.3 and 170.7 ppm. The two upfield signals (46.5 and 50.4 ppm) are assigned to the non-oxygenated quaternary C(4) and C(11), respectively. The signal at 84.1 ppm is assigned to the only oxygenated quaternary C(8). The two downfield signals (169.3 and 170.7 ppm) are assigned to two carbonyl groups of two acetates attached to C(8) and C(14), respectively. These two acetate groups were observed in the proton nmr spectrum at δ 1.92 and 1.96.

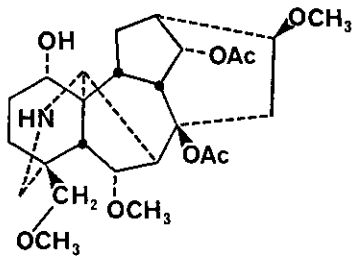
The ^{13}C nmr spectrum also showed the presence of 5 oxygenated CH doublets at 87.1, 84.9, 82.9, 75.2 and 69.2 ppm, and 3 methoxyl functions as quartets at 60.0, 57.9 and 56.5 ppm. The downfield doublets at 84.9 and 82.9 ppm are assigned to C(6) and C(16), respectively, which bear methoxyl groups. The corresponding methoxyl methyl quartets are those occuring at 57.9 and 56.5 ppm, respectively. The third quartet at 60.0 ppm is assigned to the methoxyl group attached to the only oxygenated methylene C(18), which is represented by the most downfield triplet at 75.2 ppm.

The last oxygen atom in the molecule is attached to C(1) and C(19) in a carbinolamine ether bridge. C(1) and C(19) are indicated by the two remaining doublets, the upperfield doublet at 69.2 ppm and the most downfield doublet at 87.1 ppm, respectively. The presence of the C(1)-O-C(19) ether bridge in delstaphidine was also confirmed by the existence of a peak at m/z 463 (34%) in its mass spectrum, caused by loss of an acrolein molecule from the molecular ion[18].

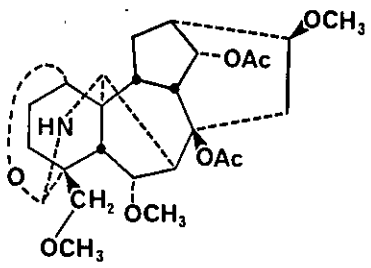
Alkaline hydrolysis of delstaphidine (**7**) with 5% methanolic KOH solution resulted in the production of a compound which was identical with delstaphinine (**6**) by tlc behavior, ir, proton and ^{13}C nmr spectra[10]. Reduction of delstaphidine with sodium borohydride afforded delphisine (**1**) in 50% yield, while reduction with sodium cyanoborohydride[19] gave (**1**) in almost quantitative yield. Delphisine was identified by the tlc behavior, mp, mmp, ir and proton nmr spectra.

The structure of delstaphidine (**7**) was also confirmed by synthesis from delphisine (**1**). In principle, this objective should be attainable by one-step oxidation of delphisine in such a manner that C(1)-hydroxyl group is converted to a C(1)-O-C(19) carbinolamine ether. However, oxidation of delphisine by treatment with potassium permanganate in acetone-H_2O solution resulted in the formation of four compounds. The two main products were isolated and identified as *N*-deethyldelphisine (**10**) and *N*-deethyldelstaphidine (**11**). Compounds (**10**) and (**11**) are new synthetic C_{19}-diterpenoid alkaloids.

Oxidative cyclization of delphisine with alkaline potassium ferricyanide[20] afforded delstaphinine (**6**). Delstaphinine by acetylation with acetic anhydride and pyridine gave the new synthetic alkaloid 14-acetyldelstaphinine (**12**), whereas acetylation with acetyl chloride afforded delstaphidine (**7**). Identity of the natural and synthetic delstaphidine was confirmed by tlc behav-



10 N-Deethyldelphisine



11 N-Deethyldelstaphidine

ior, mp, mmp, ir, proton and ^{13}C nmr spectra.

Neolinine (8) was isolated in crystalline form, mp 226.5-228.5°C, $[\alpha]_D^{26}$ +29.3 (c, 0.19, $CHCl_3$), and its molecular formula $C_{23}H_{37}NO_6$ was deduced from the mass spectral (M^+, 423), and carbon-13 nmr data. The proton nmr spectrum exhibited the following signals: δ 1.10 (3H, t, J = 7 Hz, N-CH_2-CH_3), 3.30 and 3.32 (each 3H, s, OCH_3), 4.15 (1H, dd, J_1 = 1 Hz, J_2 = 7 Hz, C(6)-β-H).

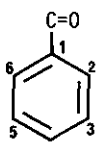
The noise decoupled ^{13}C nmr spectrum of neolinine (8) exhibited 20 signals for the 23 carbon atoms of the molecule (Table 1). The mass spectrum showed a molecular ion M^+, m/z 423 (6%), 408 (M^+-CH_3, 15%), 406 (M^+-OH, 70%), 390 ([M^+-H_2O]-CH_3, 8%), 374 ([M^+-H_2O]-OCH_3, 6%), 367 (6%), 319 (6%), 96 (32%), 91 (27%), 85 (33%), 71 (45%), 67 (24%), 58 (100%), 44 (81%), 43 (99%).

Structure 8 was deduced for neolinine from its spectral data and comparison of its data with neoline (15)[16] and delstaphisagrine (4)[8]. The ^{13}C nmr spectrum exhibited 3 singlets at 39.0, 49.6 and 74.1 ppm. The two upfield signals (39.0 and 49.6 ppm) are assigned to the non-oxygenated quaternary carbons 4 and 11, respectively. The signal at 74.1 ppm is assigned to the only oxygenated quaternary C(8). The two methoxyl signals at δ 3.30 and 3.32 in the proton nmr spectrum correspond to C(6) and C(16), as indicated by signals at 82.7 and 82.0 ppm for C(6) and C(16) and signals at 57.9 and 56.3 ppm for C(6)-OCH_3 and C(16)-OCH_3, respectively.

The presence of C(1)-α-OH is shown by the appearance of a signal at 72.1 ppm and by the occurrence of C(2) and C(3) signals at 29.6 ppm in the ^{13}C nmr spectrum. The signal at 72.1 ppm is analogous to that found for neoline (15) at 72.3 ppm. The location of an OH group on C(18) is deduced by the change of the chemical shift of the only oxygenated C(18) methylene from ~ 80.0 to 70.9 ppm and by the absence of the OCH_3 signal at ~ 59.0 ppm in the ^{13}C nmr spectrum.

Alkaline hydrolysis of delstaphisagrine (4) afforded a compound which was identical with neolinine (8) by tlc behavior, mp, mmp, ir and proton spectra. Acetylation of neolinine (8) with acetic anhydride and pyridine afforded neolinine 1,14,18-triacetate (14), and acetylation of 8 with acetyl chloride yielded neolinine tetraacetate (13).

Table 1. ^{13}C nmr Chemical Shifts and assignments for delphisine (1), *N*-deethyldelphisine (10), delstaphinine (6), delstaphidine (7), *N*-deethyldelstaphidine (11), 14-acetyldelstaphinine (12), neolinine (8), neolinine tetraacetate (13), neolinine 1,14, *18*-triacetate (14), *neoline* (15), *delstaphisagrine* (4) *and α-oxodelphinine* (9).

Carbon	1	10	6	7	11	12	8	13	14	15	4	9
C(1)	72.1	71.9	69.2	69.2d	69.3	69.2	72.1	77.1	77.6	72.3	71.9d	83.3d
C(2)	29.5	29.4[a]	26.7	26.6t	27.1	26.6	29.6	27.5	27.4	29.5	29.5t	24.9t
C(3)	30.1	29.8[a]	22.7	22.7t	22.3	22.7	29.6	33.7	34.0	29.9	29.8t	33.0t
C(4)	38.1	38.2s	46.2s	46.5s	46.7s	46.1s	39.0s	38.0s	38.2s	38.2	39.0s	37.8s
C(5)	44.1	44.1	36.8	36.6d	36.5	36.4	46.7	49.8	50.4	44.9	45.2d	48.9d
C(6)	84.2	83.7	84.2	84.9d	84.5	84.3	82.7	83.0	82.1	83.3	83.7d	81.7d
C(7)	48.3	49.1	58.9	58.9d	58.1	58.8	51.7	49.3	53.4	52.3	47.6d	48.5d
C(8)	85.8	85.6s	71.5s	84.1s	83.7s	72.1s	74.1s	85.3s	73.2s	74.3	85.8s	84.3s
C(9)	43.3	43.1	49.9	49.7d	49.3	49.4	48.3	43.9	46.8	48.3	43.1d	40.7d
C(10)	38.5	38.5	39.1	38.8d	38.9	36.6	40.5	38.3	38.2	40.7	38.4d	43.7d
C(11)	49.8	49.4s	50.2s	50.4s	49.5s	50.3s	49.6s	49.2s	49.1s	49.6	49.8s	49.6s
C(12)	29.2	28.9	29.6	30.0t	29.8	29.6	29.6	29.2	29.0	29.8	29.4t	34.1t
C(13)	43.3	42.8	46.8	41.7d	41.5	44.7	44.2	43.9	44.0	44.3	43.1d	74.8s
C(14)	75.5	75.5	75.5	75.2d	74.9	75.2	75.9	74.9	76.5	75.9	75.6d	78.4d
C(15)	38.5	38.0	40.3	37.9t	37.5	41.8	42.8	37.5	41.1	42.7	38.4t	38.9t
C(16)	82.7	82.2	81.8	82.9d	82.8	81.9	82.0	83.0	81.9	82.3	82.6d	82.6d
C(17)	62.7	59.1	60.8	60.0d	58.8	61.6	63.7	60.5	60.9	63.6	63.8d	59.2d
C(18)	79.8	79.7	75.3	75.2t	74.9	76.7	70.9	71.3	71.6	80.3	70.2t	79.5t
C(19)	56.8	54.9	87.8	87.1d	82.6	87.6	56.9	53.8	54.0	57.2	56.6t	55.4t
N-CH_2	48.0	-	48.0	47.5t	-	47.8	48.3	48.4	48.6	48.2	48.4t	162.4*d
CH_3	12.9	-	14.2	14.0q	-	14.1	12.9	13.3	13.4	13.0	12.9q	-
C(1)'	-	-	-	-	-	-	-	-	-	-	-	54.8q
C(6)'	58.0	57.8	57.8	57.9q	58.0	57.8	57.9	58.0	57.6	57.8	58.2q	57.6q
C(16)'	56.5	56.5	56.4	56.5q	56.5	56.0	56.3	56.4	56.1	56.3	56.6q	58.9q
C(18)'	59.0	58.1	60.5	58.9q	58.0	60.0	-	-	-	59.1	-	59.0q
C(1)-C=O	-	-	-	-	-	-	-	170.6[a]s	170.5[a]s	-	-	-
CH_3	-	-	-	-	-	-	-	21.1[b]	21.2[b]	-	-	-
C(8)-C=O	169.3	169.5s	-	169.3s	169.2s	-	-	169.4s	169.3s	-	169.7s	169.6s
CH_3	22.2	22.3	-	22.4q	22.3	-	-	22.4	22.4	-	22.4q	21.4q
C(14)-C=O	170.4	170.6s	-	170.7s	170.6s	170.3s	-	170.1[a]s	170.1[a]s	-	170.7s	-
CH_3	21.1	21.1	-	21.9q	21.1	21.2	-	21.8[b]	22.0[b]	-	21.3q	-
C(18)-C=O	-	-	-	-	-	-	-	170.9[a]s	-	-	-	-
CH_3	-	-	-	-	-	-	-	20.8[b]	-	-	-	-
 C=O	-	-	-	-	-	-	-	-	-	-	-	166.2s
1	-	-	-	-	-	-	-	-	-	-	-	130.0s
2,6	-	-	-	-	-	-	-	-	-	-	-	129.7d
3,5	-	-	-	-	-	-	-	-	-	-	-	128.5d
4	-	-	-	-	-	-	-	-	-	-	-	133.2d

a and b The assignments may be interchanged in any vertical column.
*N-CHO

α-Oxodelphinine was isolated in a crystalline form, mp 217.5-219.5°C, $[\alpha]_D^{22}$ -62.8 (c, 0.28, 95% ethanol). This compound proved to be identical with a synthetic sample of α-oxodelphinine by tlc behavior, mp, mmp, ir, mass, proton and ^{13}C-nmr spectra. α-Oxodelphinine has not previously been reported in nature.

Table 1 gives the carbon-13 chemical shifts and assignments for delphisine (1), *N*-deethyldelphisine (10), delstaphinine (6), delstaphidine (7), *N*-deethyldelstaphidine (11), 14-acetyldelstaphinine (12), neolinine (8), neolinine tetraacetate (13), neolinine 1,14,18-triacetate (14), neoline (15), delstaphisagrine (4) and α-oxodelphinine (9).

EXPERIMENTAL

Melting points are corrected and were taken on a Thomas-Kofler hot stage equipped with a microscope and a polarizer. Optical rotations were measured on a Perkin-Elmer model 141 polarimeter. Infrared spectra were taken on a Perkin-Elmer model 1420 spectrophotometer. ^1H and ^{13}C nmr spectra were recorded on JEOL FT models FX-60 and FX-90 Q spectrometers in $CDCl_3$. Mass spectra were determined on a Finnegan Quadrupole 4023 instrument. For chromatographic separations on a Chromatotron[13,14] rotors were coated with a 1 or 2 mm thick layer of silica gel PF_{254} + 365 (EM Art. #7741), for vlc[12], silica gel HR (EM Art. #7744), and for ptlc, silica gel PF-254 (EM Art #7747).

Fractionation of Mother Liquors of Delphinium staphisagria — 106 g of the amorphous powder of mother liquors of *D. staphisagria* was fractionated into 6 groups by adopting gradient pH extraction techniques.[11] These groups were: group 1 (neutral fraction A, 36,17 g), group 2 (neutral fraction B, 24.11 g), group 3 (pH 4.5, 20.61 g), group 4 (pH 8, 20.02 g), group 5 (pH 10, 1.32 g), group 6 (pH 12, 0.21 g).

Isolation of Delstaphidine (7) — A fraction of 17.64 g of group 3 (pH 4.5) was chromatographed (vlc) on silica. Elution was performed with hexane-$CHCl_3$ in a manner of increasing polarity. Fractions eluted with hexane-$CHCl_3$ (60:40) and hexane-$CHCl_3$ (40:60) were similar on tlc plates, and were combined (10.62 g). The combined fraction was crystallized from acetone-hexane mixture several times to give 7.345 g of delphisine (1), mp 122-123°C. The mother liquors were purified twice on a silica rotor of a Chromatotron to give 186 mg of delstaphidine (7), mp 192.5-194.5°C, $[\alpha]_D^{25}$ +27.4 (c 0.27, $CHCl_3$). For ^{13}C nmr data see Table 1.

Hydrolysis of Delstaphidine (7) — To 30 mg of delstaphidine (7) in 5 ml CH_3OH was added 5 ml of 5% methanolic KOH solution. The mixture was stirred at room temperature for 18 h. Methanol was distilled and 10 ml of H_2O was added. The solution was extracted with 4 x 15 ml of $CHCl_3$. The $CHCl_3$ extract was dried over anhydrous Na_2SO_4 and the solvent was removed under reduced pressure to give 26 mg of residue. This was chromatographed on one plate of silica using 5% CH_3OH in $CHCl_3$ as an eluent. The major zone was cut and extracted to give 20 mg of a residue which was identical with delstaphinine (6) by tlc behavior, and ir, proton and ^{13}C nmr spectra[10].

Reduction of Delstaphidine (7) to Delphisine (1) — A. Reduction with sodium borohydride: To 25 mg of delstaphidine in 50 ml of CH_3OH was added 50 mg of $NaBH_4$. The mixture was stirred at room temperature for 10 min, then another 50 mg of $NaBH_4$ was added and the mixture was stirred for 10 min. Methanol was distilled and 25 ml of water was added. The solution was extracted with 5 x 25 ml of $CHCl_3$. The $CHCl_3$ extract was dried over anhydrous sodium sulphate and was distilled under reduced pressure to give 21 mg of residue which was chromatographed over one plate of silica using 4% CH_3OH in $CHCl_3$ as an eluent. The major zone was cut and extracted to give 10 mg of delphisine (1), crystalline from acetone-hexane mixture, mp 122-123°C. Delphisine was identified by tlc behavior, mp, mixture mp, and ir and proton nmr spectra.

B. Reduction with sodium cyanoborohydride — To a solution of 6 mg of delstaphidine and 6 mg of sodium cyanoborohydride[19] in 2 ml CH_3OH was added a few drops of 10% aqueous HCl to adjust the pH of the reaction mixture between 6 and 7. The resulting solution was stirred continuously at 24°C for 3 h and the pH of the reaction mixture was maintained between 6 and 7. The solvent was removed *in vacuo*, the residue was treated with 1.5 ml of Na_2CO_3 solution and the mixture was extracted with 3 x 5 ml of $CHCl_3$. The $CHCl_3$ extracts were dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 5.8 mg, which when crystallized afforded delphisine, mp 122-123°C, identified by tlc behavior, ir and proton nmr spectra.

Oxidation of Delphisine (1) to *N*-Deethyldelphisine (10) and *N*-Deethyldelstaphidine (11) — To a solution of delphisine (250 mg) in acetone (150 ml) was added $KMnO_4$ (250 mg) in 300 ml of H_2O-acetone (1:5). After one h at room temperature with occasional shaking an additional amount of $KMnO_4$ (250 mg) in 300 ml of water-acetone (1:5) was added and the mixture was warmed by immersing in a water bath heated to (80-90°C) for 10 min. Acetone was evaporated under reduced pressure, water (250 ml) was added and excess $KMnO_4$ was decomposed by addition of about 100 mg of $NaHSO_3$ and 2 drops of H_2SO_4. The mixture was cooled in an ice bath, made basic with aqueous Na_2CO_3 and extracted with 8 x 250 ml of $CHCl_3$. The $CHCl_3$ extract was dried over anhydrous Na_2SO_4 and distilled under vacuum to give 218 mg of residue. This residue was fractionated twice on silica rotors of a Chromatotron to give 40 mg of *N*-deethyldelphisine (**10**) and 65 mg of *N*-deethyldelstaphidine (**11**) with characteristics as listed below:

N-Deethyldelphisine (10) — Amorphous; $[\alpha]_D^{25}$ +27.7 (c, 0.25, $CHCl_3$); ^1H nmr: δ 1.95 (3H, s, $OCOCH_3$), 2.01 (3H, s, $OCOCH_3$), 3.24 (3H, s, OCH_3), 3.30 (6H, s, $2XOCH_3$), 4.77 (1H, dd, $J_1 = J_2$ = 4.5 Hz, C(14)-β-H). For ^{13}C nmr data see table 1. The mass spectrum showed a molecular ion M^+, m/z 493 (0.14%), 476 (M^+-OH, 4%), 475 (M^+-H_2O, 2%), 460 ([M^+-H_2O]-CH_3, 1%), 444 (2%), 91 (13%), 85 (10%), 71 (24%), 45 (53%), 43 (100%).

N-Deethyldelstaphidine (11) — Crystalline from ethanol-hexane mixture; mp 267.5-269.5°C; $[\alpha]_D^{24}$ +46.0 (c 0.27, $CHCl_3$); ^1H nmr: δ 1.90 (3H, s, $OCOCH_3$), 1.97 (3H, s, $OCOCH_3$), 3.16, 3.21, 3.24 (3H each, s, OCH_3), 3.47 (1H, s, C(19)-H), 4.74 (1H, dd, $J_1 = J_2$ = 4.5 Hz, C(14)-β-H). For ^{13}C nmr data see table 1. The mass spectrum showed a molecular ion M^+, m/z 491 (1%), 490 (M^+-H, 2%), 476 (M^+-CH_3, 1%), 414 (1%), 400 (1%), 384 (1%), 205 (5%), 155 (7%), 129 (12%), 109 (24%), 91 (25%), 85 (44%), 71 (38%), 56 (6%), 55 (28%), 45 (70%), 43 (100%), 41 (33%).

Conversion of Delphisine (1) to Delstaphinine (6) — A solution of potassium ferricyanide[20] (2.5 g) in 20 ml of 8% aqueous NaOH was added to 495 mg of delphisine in 100 ml of 50% ethanol. The reaction mixture was stirred at room temperature while the progress of the reaction was monitored by tlc on silica gel. After 2 h the reaction mixture was concentrated *in vacuo*. Water was added (200 ml) and the mixture was extracted with 4 x 200 ml of $CHCl_3$. The combined extracts were dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 426 mg of residue. This residue was purified twice on a silica rotor of a Chromatotron[13,14] to give 330 mg of pure delstaphinine (6), amorphous. For ^{13}C nmr data: see table 1. Delstaphinine (6) was identified by tlc behavior, and by comparison of ir, mass, proton and ^{13}C nmr spectral data[10] with those of an authentic sample.

Conversion of Delstaphinine (6) to Delstaphidine (7) — Twenty ml of acetyl chloride was added to 55 mg of delstaphinine (6) and the resulting solution was stirred continuously at 25°C for 4 days. The resulting mixture was evaporated to dryness *in vacuo*, the residue was treated with 10 ml of Na_2CO_3 solution and the mixture was extracted with 3 x 20 ml of $CHCl_3$. The combined extracts were dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 52 mg of residue which was purified on a silica rotor of a Chromatotron[13,14] to give 41 mg of pure delstaphidine (7), mp 192.5-194.5°C. The synthetic and natural samples of delstaphidine (7) were identical by tlc behavior, mp, mixture mp, ir, proton and ^{13}C nmr spectra

Conversion of Delstaphinine (6) to 14-Acetyldelstaphinine (12) — Fifteen ml of a mixture of acetic anhydride and pyridine (1:1) was added to 95 mg of delstaphinine (6) and left overnight. Thirty ml of iced water was added and the reaction mixture was rendered alkaline with $NaHCO_3$. The mixture was extracted with 3 x 60 ml of $CHCl_3$. The combined extracts were dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 97 mg of residue. This residue was purified on a silica rotor of a Chromatotron[13,14] to give 83 mg of pure 14-acetyldelstaphinine (12). ^{13}C nmr: see table 1; ^1H nmr: δ 1.08 (3H, t, J = 7 Hz, N-CH_2-CH_3), 2.00 (3H, s, OCOCH_3), 3.25 (3H, s, OCH_3), 3.29 (6H, s, 2XOCH_3), 3.51 (1H, d, J = 1.5 Hz, 19-H), 4.80 (1H, dd, $J_1 \approx J_2$ = 4.5 Hz, C(14)-β-H). The mass spectrum showed a molecular ion M^+, m/z 477 (0.5%), 462 (M^+-CH_3, 1%), 447 (M^+-$2CH_3$, 2%), 446 (M^+-OCH_3, 6%), 421 (M^+-C_3H_4O, 12%), 390 ([M^+-C_3H_4O]-OCH_3, 7%), 152 (9%), 85 (13%), 71 (13%), 58 (8%), 57 (6%), 56 (12%), 55 (12%), 45 (38%), 43 (100%).

Isolation of Neolinine (8) — A fraction of 19.96 g of group 4 (pH 8) was chromatographed (vlc)[12] on silica gel. Elution was performed with hexane, hexane:$CHCl_3$, then $CHCl_3$:CH_3OH mixtures in a manner of increasing polarities. Fractions eluted with $CHCl_3$ and $CHCl_3$:CH_3OH (99:1) were similar on tlc plates, and were combined (5.76 g). This fraction was rechromatographed (vlc) on silica. Elution was carried out with hexane, hexane:$CHCl_3$ and $CHCl_3$:CH_3OH mixtures. Fractions, eluted with $CHCl_3$:CH_3OH (97:3) (100 mg), were combined and chromatographed (ptlc) over 2 plates of silica, using $CHCl_3$:CH_3OH (90:10) as an eluent. The major zone was extracted to give 49 mg of neolinine (8), crystalline from acetone, mp 226.5-228.5°C, $[\alpha]_D^{26}$ +29.3 (c, 0.19, $CHCl_3$). For ^{13}C nmr data see table 1.

Conversion of Neolinine (8) to Neolinine Tetraacetate (13) — Five ml of acetyl chloride was added to 11 mg of neolinine (8) and the resulting solution was stirred continuously at room temperature for 48 h. The resulting mixture was evaporated to dryness *in vacuo*, the residue was treated with 10 ml of Na_2CO_3 solution and the mixture was extracted with 4 x 25 ml of $CHCl_3$. The combined extracts were dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 14 mg of residue which was purified by tlc to give 11.2 mg of neolinine tetraacetate (13), $[\alpha]_D^{24}$ -3.1 (c, 0.25, $CHCl_3$). For ^{13}C nmr data see table 1; ^1H nmr: δ 1.11 (3H, t, J = 7 Hz, N-CH_2-CH_3), 1.97 and 2.04 (each 3H, s, $OCOCH_3$), 2.05 (6H, s, 2 $XOCOCH_3$), 3.20 and 3.32 (each 3H, s, OCH_3), 4.16 (1H, dd, J_1 = 1 Hz, J_2 = 7 Hz, C(6)-β-H) 4.75 (1H, dd, J_1 = J_2 = 4.5 Hz, C(14)-β-H); mass spectrum: 533 ([M^+-$COCH_3$]-CH_3, 3%), 532 (M^+-59, 11%), 58 (20%), 57 (5%), 56 (9%), 45 (15%), 43 (100%).

Conversion of Neolinine (8) to Neolinine 1,14,18-Triacetate (14) — Two ml of a mixture of acetic anhydride and pyridine (1:1) was added to 9 mg of neolinine, warmed at 50°C for 1 h, then left overnight at room temperature. Ice water (15 ml) was added and the reaction mixture was rendered alkaline with $NaHCO_3$. The mixture was extracted with 4 x 25 ml of $CHCl_3$. The combined extracts were dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 11 mg residue which was purified by tlc to give 7.8 mg of neolinine 1,14,18-triacetate (14). For ^{13}C nmr data see table 1; ^1H nmr: δ 1.12 (3H, t, J = 7 Hz, N-CH_2-CH_3), 2.05 (9H, s, 3 x $OCOCH_3$), 3.24 and 3.31 (each 3H, s, OCH_3), 4.15 (1H, dd, J_1 = 1 Hz, J_2 = 7 Hz, C(6)-β-H), 4.75 (1H, dd, J_1 = J_2 = 4.5 Hz, C(14)-β-H); mass spectrum: 549 (M^+, 0.04%), 534 (M^+-CH_3, 0.22%), 492 (M^+-57, 2.4%), 491 ([M^+-$COCH_3$]-CH_3, 14%), 490 (M^+-59, 45%), 58 (41%), 57 (6%), 56 (21%), 45 (16%), 43 (100%).

Conversion of Delstaphisagrine (4) to Neolinine (8) — To 21.5 mg of delstaphisagrine in 5 ml CH_3OH was added 5 ml of 5% methanolic KOH solution and kept over night. Methanol was distilled and 10 ml of water was added. The solution was extracted with 4 x 15 ml $CHCl_3$. The chloroform extract was dried over anhydrous Na_2SO_4 and was distilled under reduced pressure to give 18 mg of residue which was crystallized from acetone to give 11 mg of neolinine (8), mp 227.5-229.5°C. The natural and synthetic neolinine were identical by tlc behavior, mp, mmp and ir spectra.

Isolation of α-Oxodelphinine (9) — A fraction of 15 g of group 1 (neutral fraction A) was chromatographed (vlc) on silica. Elution was carried out with hexane, hexane:$CHCl_3$ mixtures in a manner of increasing polarities. Fractions eluted with hexane:$CHCl_3$ (50:50) and hexane:$CHCl_3$ (40:60) were similar on tlc plates, and were combined (2.11 g). This fraction was crystallized from $CHCl_3$: hexane mixture to give 208 mg of α-oxodelphinine (9), mp 217.5-219.5°C, $[\alpha]_D^{22}$ -62.8 (c, 0.28, 95% ethanol); ir (nujol): 3540 cm^{-1} (OH), 1712, 1648 cm^{-1} (C=O); ^1H nmr: δ 1.26 (3H, s, $OCOCH_3$), 3.13, 3.18, 3.27, 3.56 (3H each, s, OCH_3), 4.08 (1H, dd, J_1 = 1 Hz, J_2 = 7 Hz, C(6)-β-H), 4.90 (1H, d, J = 4.5 Hz, C(14)-β-H), multiplets between δ 7.3-8.1 (5H, aromatic protons of benzoyl radicle); mass: M^+ 613 (0.1%), 553 (M^+-60, 0.1%), 522 (0.2%), 521 (0.2%), 416 (0.4%), 400 (0.7%), 399 (2%), 296 (2%), 222 (4%), 218 (3%), 211 (3%), 149 (8%), 109 (7%), 105 (100%), 85 (17%), 77 (39%), 71 (49%), 57 (11%), 55 (22%), 45 (63%), 43 (57%), 41 (28%). For ^{13}C nmr data see table 1. This compound proved to be identical with a synthetic sample of α-oxodelphinine by tlc behavior, mp, mixture mp, ir, mass, proton and ^{13}C nmr spectra.

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