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NIRUROIDINE, A NORSECURININE-TYPE ALKALOID FROM *PHYLLANTHUS NIRUROIDES*

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Key Word Index—*Phyllanthus niruroides*; Euphorbiaceae; leaves; norsecurinine-type alkaloid; niruroidine.

Abstract—Leaves of *Phyllanthus niruroides* provided a new norsecurinine-type alkaloid niruroidine whose structure was elucidated on the basis of spectral measurements.

INTRODUCTION

Phyllanthus niruroides Muell. Arg. is a small plant widely used in Central African folk medicine, which has not been studied previously. The whole plant is used, along with *P. niruri* L., in the treatment of malaria. The present report deals with the isolation and structural determination of a new norsecurinine-type alkaloid **1**, which we have named niruroidine. Other members of the genus are reported to contain securinine- and norsecurinine-type alkaloids [*inter alia* 1–12], disaccharides of the polyfunctional bisabolene derivative, phyllanthocin [13–17], various triterpenoids [18–23], phytol [24], coumarins and flavonoids [25, 26], lignans and lignan glycosides [27–32].

RESULTS AND DISCUSSION

The CI mass spectrum ($[M + H]^+$ *m/z* 222) of the new alkaloid and the carbon and proton count from the ^{13}C NMR spectrum established the molecular formula as $C_{12}H_{15}NO_3$. Two of the oxygens were part of the characteristic α,β-unsaturated γ-lactone system represented in the IR spectrum by bands at 1745 and 1639 cm^{-1}, in the ^{13}C NMR spectrum by a singlet at δ174.8 (C-11), a doublet at δ110.8 (C-12) and a singlet at δ173.9 (C-13) and in the ^1H NMR spectrum (Table 1) by a signal at δ5.74 (H-12) [8–12, 33] allylically coupled to two mutually coupled (J = 19.5 Hz) signals at δ3.26 and 2.91 (H-14β and H-14α). H-14β was additionally coupled (2 Hz) to a one proton multiplet at δ3.15 (H-6), which was further coupled (J = 3 Hz) to a multiplet at δ4.45 (H-7).

The lactone oxygen was attached to a tertiary carbon (C-9 at δ83.9 in the range characteristic of this series [8, 9, 33]), whereas the third oxygen was that of a secondary hydroxyl group (strong IR band at 3370 cm^{-1}) obviously responsible for the H-7 multiplet at δ4.45 on a carbon atom responsible for one of two carbon doublets found at δ62.2 and 61.9. Further sequential decoupling showed that H-7 was also coupled (J = 9.5 Hz and J = 3 Hz) to two *gem*-coupled protons (J_{gem} = 14 Hz) giving rise to a *dd* at δ2.81 (H-8α) and a *ddd* at δ1.36 (H-8β). The δ1.36 signal, but not the δ2.81 signal, was coupled, apparently long-range (J = 1.5 Hz), to a *ddd* at δ3.02 (H-2) on a carbon atom responsible for the second of the two carbon doublets at δ62.2 and 61.9. Proton and carbon chemical shifts indicated that this carbon atom (C-2) was attached to nitrogen as were C-6 at δ55.7 and a third, C-5 at δ51.3. Completion of the carbon skeleton was achieved by carrying out further irradiations, which located the signals of H-3a, H-3b, H-4a, H-4b, H-5a and H-5b, the last two occurring at somewhat lower field, as expected for protons vicinal to nitrogen (Table 1).

In the relative configurations shown in **1**, the α-orientation of H-2 satisfies the requirement for W-coupling with H-8β. Furthermore, to satisfy the requirement for long-range (W) coupling between H-7 and H-14α, the hydroxyl group should be oriented as shown.

Niruroidine resembles one of the intermediate stages postulated for the biogenesis of the alkaloid, nirurine, in *P. niruri* [8]. However, the C-7 stereochemistry differs, thus preventing further elaboration to the pentacyclic ring system present in nirurine.

EXPERIMENTAL

Plant material. *P. niruroides* was collected in the vicinity of the University of Kinshasa campus and in Mayilu village (Kimwenza). Voucher specimens W. Robyn No. 4244 and Carlier No. 66 are on deposit in the INERA

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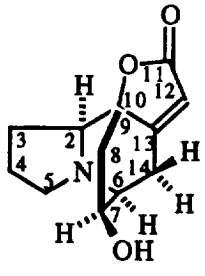
Table 1. ^{1}H NMR spectral data of compound **1** ($CDCl_3$, 500 MHz)

δ_H	Coupled to (H)	Assignment (H)
5.74 *t*	14α, 14β	12
4.45 *br ddd*	6, 8α, 8β, 14α	7
3.26 *ddd*	6, 12, 14α	14β
3.15 *m*	7, 14β	6
3.12 *m*	4a, 4b, 5b	5a
3.02 *ddd*	3a, 3b, 8β	2
2.91 *m*	7, 12, 14β	14α
2.89 *m*	4a, 4b, 5a	5b
2.81 *dd*	7, 8β	8α
1.97 *m*	2, 3b, 4a, 4b	3a
1.91 *m*	3a, 3b, 4b, 5a, 5b	4a
1.79 *m*	2, 3a, 4a, 5a, 5b	3b, 4b
1.36 *ddd*	2, 7, 8α	8β

J(Hz): 2,8β = 1.5; 2,3a = 2,3b = 7; 6,7 = 7,8β = 3; 6,14β = 2; 7,8α = 9.5; 7,14α = 1.5; 7,14β ~ O; 8α, 8β = 14; 12, 14α = 12; 14β = 2; 14α,14β = 19.5.



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herbarium, Biology Department, University of Kinshasa, Zaïre.

Extraction and isolation. Dried and powdered leaves (700 g) were defatted with hexane (3 × 21) at room temp. and subsequently extracted with MeOH (3 × 41), also at room temp. After removal of MeOH *in vacuo*, the residue was shaken with 2.5% aq. HCl and filtered. The filtrate was extracted with $CHCl_3$ to remove the neutral fr., brought to pH 9.5 with NH_4OH and extracted with $CHCl_3$. Evapn of the solvent gave crude alkaloids (140 mg) which were subjected to radial chromatography on silica gel, 5 ml frs being collected, using the following systems as eluents: hexane–Me_2CO 10:1 (frs 1–20), hexane–Me_2CO 5:2 (frs 21–44), hexane–Me_2CO 4:3 (frs 45–84), hexane–Me_2CO 1:1 (frs 85–100), hexane–Me_2CO–$CHCl_3$ 5:5:3 (frs 101–120), hexane–$CHCl_3$–MeOH–NH_4OH 10:17:3.8:0.25 (frs 121–201) and $CHCl_3$–MeOH–NH_4OH 17:3.8:0.25 (frs 202–300). Frs 151–163 yielded 20 mg of a mixt. (20 mg) which was subjected to TLC (silica gel, eluent hexane–$CHCl_3$–MeOH–NH_4OH, 10:17:3.8:0.25) to afford niruroidine (**1**), 8 mg (0.0015%) as an amorphous solid. MS PCI (*iso*butane) *m/z* (rel. int.) 222 ($[M + H]^+$, 100), 179 (5.2), 165 (5.2), 147 (4.9), 137 (8.6), 121 (5.3), 114 (9.3), 104 (9.4), 86 (7.4). IR ν^{KBr} cm^{-1}: 3370, 1745, 1639. ^{13}C NMR ($CDCl_3$, 50.3 MHz): δ174.8*s* (C-11), 173.9*s* (C-13), 110.8*d* (C-12), 83.9*s* (C-9), 62.1*d* and 61.5*d* (C-7 and C-2), 55.7*d* (C-6), 51.0*t* (C-5), 34.0 (C-8), 27.0, 26.9, 25.4 (C-3, C-4, C-14). ^{1}H NMR, Table 1.

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REFERENCES

1. Parello, J., Melera, A. and Goutarel, R. (1963) *Bull Soc. Chim. Fr.*, 898.
2. Bevan, C. W. L., Patel, M. B., Rees, A. H. and Taylor, D. A. H. (1964) *Chem. Ind.*, 838.
3. Bevan, C. W. L., Patel, M. B. and Rees, A. H. (1964) *Chem. Ind.* 2054.
4. Parello, J. and Munavalli, S. (1965) *Compt. Rend. Acad. Sci.* **260**, 337.
5. Rouffiac, R. and Parello, J. (1969) *Plant. Med. Phytother.* **3**, 220.
6. Horii, Z., Imanishi, M., Yamauchi, M., Hanaoka, M., Parello, J. and Munavalli, S. (1972) *Tetrahedron Letters* 1877.
7. Mulchandani, N. B. and Hassaranjani, S. A. (1984) *Planta Med.* **50**, 104/
8. Petchnaree, P., Bunyapraphatsara, N., Cordell, G. A., Lowe, H. J., Cox, P. J., Howie, R. A. and Patt, S. L. (1986) *J. Chem. Soc. Perkin Trans.* **1**, 1551.
9. Joshi, B. S., Gawad, D. H., Pelletier, S. W. and Kartha, G. (1986) *J. Nat. Prod.* **49**, 614.
10. Mensah, J. L., Gleye, J., Moulis, C. and Fouraste, I. (1988) *J. Nat. Prod.* **51**, 1113.
11. Negi, R. S. and Fakhir, T. M. (1988) *Phytochemistry* **27**, 3027.
12. Hassaranjani, S. A. and Mulchandani, N. B. (1990) *Indian J. Chem.* **29B**, 801.
13. Kupchan, S. M., La Voie, E. J., Brautman, A. R., Tei, B. Y., Bright, W. M. and Bryan, R. F. (1977) *J. Am. Chem. Soc.* **99**, 3599.
14. Pettit, G. R., Cragg, G. M., Gust, D. and Brown, P. (1982) *Can. J. Chem.* **60**, 544.
15. Pettit, G. R., Cragg, G. M., Gust, D. and Brown, P. (1982) *Can. J. Chem.* **60**, 939.
16. Pettit, G. R., Cragg, G. M., Suffness, M. I., Gust, D., Boettner, F. E., Williams, M., Saenz-Renauld, J. A., Brown, P., Schmidt, J. M. and Ellis, P. P. (1984) *J. Org. Chem.* **49**, 4258.
17. Pettit, G. R., Schaufelberger, D. E., Nieman, R. A., Dufresne, C. and Saenz-Renauld, J. A. (1990) *J. Nat. Prod.* **53**, 1406.
18. Tanaka, R., Tabuse, M. and Matsunaga, S. (1988) *Phytochemistry* **27**, 3563.
19. Singh, B., Agrawal, P. K. and Thakur, R. S. (1989) *Indian J. Chem.* **28B**, 319.

20. Tanaka, R. and Matsunaga, S. (1989) *Phytochemistry* **28**, 1788.
21. Singh, B., Agrawal, P. K. and Thakur, R. S. (1989) *Phytochemistry* **28**, 1980.
22. Matsunaga, S., Tanaka, R., Takaoka, Y., In, Y., Ishida, T., Rahmani, M. and Ismail, H. B. M. (1993) *Phytochemistry* **32**, 165.
23. Tanaka, R., Masuda, K. and Matsunaga, S. (1993) *Phytochemistry* **32**, 472.
24. Singh, B., Agrawal, P. K. and Thakur, R. S. (1991) *Planta Med.* **57**, 98.
25. Hnatyszyn, O., Ferraro, G. and Coussio, J. D. (1993) *Fitoterapia* **64**, 556.
26. Hnatyszyn, O., Ferraro, G. and Coussio, J. D. (1987) *J. Nat. Prod.* **50**, 1156.
27. Satyanarayana, P., Subramanyam, P., Viswanatham, K. M. and Ward, R. S. (1988) *J. Nat. Prod.* **51**, 44.
28. Pettit, G. R. and Schaufelberger, D. E. (1988) *J. Nat. Prod.* **51**, 1104.
29. Singh, B., Agrawal, P. K. and Thakur, R. S. (1989) *J. Nat. Prod.* **52**, 48.
30. Satyanarayana, P. and Venkateswarlu, S. (1991) *Tetrahedron* **47**, 8931.
31. Huang, Y.-L., Chen, C. and Ou, J.-C. (1992) *Planta Med.* **58**, 473.
32. Samanabandhu, A., Nitayangkura, S., Mahidol, C., Ruchirawat, S., Likhitwitayawuid, K., Shieh, H.-L., Chai, H., Pezzuto, J. M. and Cordell, G. A. (1993) *J. Nat. Prod.* **56**, 233.
33. Livant, P. D. and Beutler, J. (1987) *Tetrahedron* **43**, 2915.