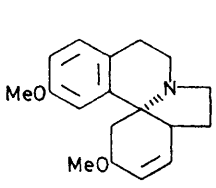
# Biosynthesis of Isococculidine

By DEWAN S. BHAKUNI,* AWADHESH N. SINGH, and RANDHIR S. KAPIL
(*Central Drug Research Institute, Lucknow-226001, India*)

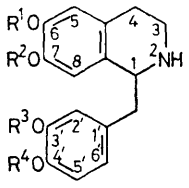
*Summary* Tracer experiments prove that the *Erythrina* alkaloid, isococculidine, is biosynthesised in *Cocculus laurifolius* from (+)-norprotosinomenine.

ISOCOCCULIDINE[1] (**1**), the neuromuscular blocking principle of *Cocculus laurifolius* DC (Menispermaceae) and a representative of 'abnormal' *Erythrina* alkaloids,[1,2] can be biosynthesised from norprotosinomenine (**2**), an established

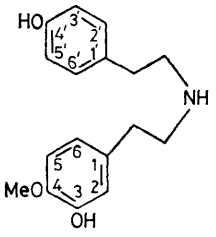
precursor of *Erythrina* alkaloids[3] *via* the intermediates[4] (**8**), (**9**), and (**10**). In the bioconversion, one of the oxygen functions of the precursor can be eliminated by a dienone–benzene rearrangement.[3] However, the possibility of the formation of (**1**) in Nature from nor-reticuline (**3**), nororientaline (**4**), and the amine (**6**) by oxidative coupling cannot be ruled out.[5]



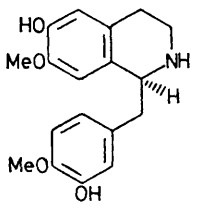
(1)



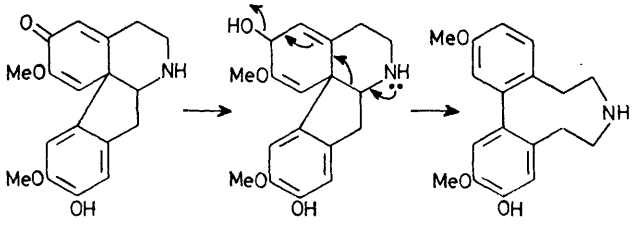
(2) $R^1 = R^3 = H$, $R^2 = R^4 = Me$
(3) $R^2 = R^3 = H$, $R^1 = R^4 = Me$
(4) $R^2 = R^4 = H$, $R^1 = R^3 = Me$
(5) $R^1 = R^2 = R^3 = R^4 = H$



(6)



(7)



(8) (9) (10)

TABLE. Tracer experiments on *C. laurifolius*.

| Expt. | Precursor fed | Incorporation into (**1**)/% |
|---|---|---|
| 1 | (+)-[*U*-$^{14}$C]Tyrosine | 0·11 |
| 2 | (±)-[1-$^3$H]-(**2**) | 0·18 |
| 3 | (±)-[5′,8-$^3H_2$]-(**4**) | 0·0014 |
| 4 | (±)-[2′,6′,8-$^3H_3$]-(**3**) | 0·007 |
| 5 | [2,3′,5′,6-$^3H_4$]-(**6**) | 0·001 |
| 6 | (±)-[1-$^3$H, 7-*OMe*-$^{14}$C]-(**2**) | 0·17 |
| 7 | (±)-[1-$^3$H, 4′-*OMe*-$^{14}$C]-(**2**) | 0·21 |
| 8 | (±)-[1-$^3$H]-(**5**) | 0·12 |
| 9 | (+)-[*Ar*-$^3$H]-(**7**) | 0·28 |
| 10 | (−)-[*Ar*-$^3$H]-(**7**) | 0·003 |
| 11 | (±)-[1-$^3$H]Protosinomenine | 0·007 |

Feeding of (±)-tyrosine (expt. 1) (Table) in parallel with (±)-nor-reticuline (**3**) (expt. 4), (±)-nororientaline (**4**) (expt. 3), and the amine (**6**) (expt. 5) established that *C. laurifolius* plants were actively biosynthesising (**1**), and that (**3**), (**4**), and (**6**) were being very poorly metabolised by the plants. Feeding with (±)-norprotosinomenine (**2**) (expts. 2, 6, and 7) showed that the compound is an efficient precursor of (**1**). (±)-Norlaudanosoline (**5**) (expt. 8) was also incorporated into (**1**). When (±)-protosinomenine (expt. 11) was fed to the plants it was poorly incorporated into (**1**). Feeding of doubly labelled (±)-norprotosinomenines (expts. 6 and 7) gave (**1**) labelled with both $^{14}$C and $^3$H. The $^{14}$C:$^3$H ratio in the precursor and the biosynthetic alkaloids in each experiment was practically unchanged. Cleavage of the methoxy groups of the isococculidine by the Zeisel method afforded radioactive methyl iodide trapped as triethylmethylammonium iodide which had, as expected, essentially half the molar activity of (**1**).

Parallel feeding with (+)-norprotosinomenine (**7**) (expt. 9) and (−)-norprotosinomenine (expt. 10) showed that the bioconversion of 1-benzylisoquinoline (**1**) was stereospecific.

Trapping experiments by feeding (±)-tyrosine (expt. 12) to *C. laurifolius* plants showed fairly high incorporation 0·42%) into norprotosinomenine. Thus (**2**) is a true precursor of (**1**).

(*Received, 21st December* 1976; *Com.* 1389.)

[1] D. S. Bhakuni, H. Uprety, and D. A. Widdowson, *Phytochemistry*, 1976, **15**, 739.
[2] R. Razakov, S. Yu. Yunusov, S-M. Nasyrov, A. N. Chekhlov, V. G. Andrianov, and Y. T. Struchkov, *J.C.S. Chem. Comm.*, 1974, 150.
[3] D. H. R. Barton, R. James, G. W. Kirby, D. W. Turner, and D. A. Widdowson, *J. Chem. Soc.* (*C*), 1968, 1529; D. H. R. Barton, R. B. Boar, and D. A. Widdowson, *ibid.*, 1970, 1213.
[4] H. Uprety and D. S. Bhakuni, *Tetrahedron Letters*, 1975, 1201.
[5] D. H. R. Barton, C. J. Potter, and D. A. Widdowson, *J.C.S. Perkin I*, 1974, 346.