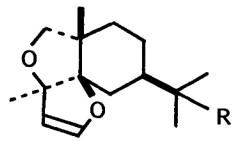
CHEMISTRY LETTERS, pp. 1377-1378, 1987.

Biosynthesis of Phytuberin[1)]

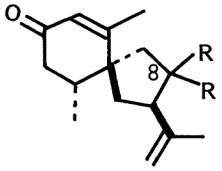
Akio MURAI,* Yuko YOSHIZAWA, Hiroshi MIYAZAKI,
Tadashi MASAMUNE, and Norio SATO†
Department of Chemistry, Faculty of Science,
Hokkaido University, Sapporo 060
†Hokkaido National Agricultural Experiment Station,
Sapporo 004

Feeding experiments with (±)-[8,8-$^2H_2$]solavetivone in diseased potato tuber tissues indicated that phytuberin, one of the most representative phytoalexins in the Solanaceae, was biosynthesized from solavetivone.

Phytuberin (1)[2)] is a representative phytoalexin in the genus Solanum and has been an attractive biosynthetic target because of the novel structure characterized by the presence of two hydrofuran rings and the biological importance. Stoessl[3)] proposed the biogenetic pathway of 1 and its hydrolyzed product, phytuberol (2), on the basis of the experiments using doubly ^{13}C-labelled method, and indicated that formation of the hydrofuran rings would be initiated by oxidative cleavage of the C-1 and C-2 bond of an eudesmane precursor. On the other hand, in view of the co-existence of 1 and 2 with solavetivone (3) in diseased potato tissues,[4)] we assumed that 3 would be a biosynthetic precursor of 1 and 2. We disclose herein that this is indeed the case.



1, R=OAc
2, R=OH



3, R=H
3-D, R=D

Aged tissue plugs of potatoes (Rishiri, Solanum tuberosum × S. demissum) were inoculated with Fusarium roseum Lk. cultivatied on Irish Cobbler at 23 °C for 21 h, when 1 and 2 were detected on TLC. These plugs were then halved and the cut surface was incubated with (±)-[8,8-$^2H_2$]solavetivone [(±)-3-D, $^2H_2$-content, ∿100%][5)] at 23 °C for 22 h. The disks were blended with chloroform-methanol (1:1), and the chloroform extracts were fractionated as usual to give pure samples of (-)-1-D, (-)-2-D ($[\alpha]_D^{33}$ -40° (c 0.02, EtOH); natural sample,[2)] -36.9°), and the recovered (+)-3-D ($[\alpha]_D^{31}$ +39° (c 0.10, EtOH); natural sample,[6)] -119°) in 6×10^{-5}%, 6×10^{-5}%, and 5.4×10^{-4}% yields, respectively. Compound 1-D was further saponified to give (-)-2-D, $[\alpha]_D^{33}$ -15° (c 0.04, EtOH). The EI-MS spectra of these phytuberols revealed that they contained at least 0.6-0.7% of the $^2H_2$ derivatives. The ^2H NMR spectrum of 2-D derived from 1-D is given in Fig. 1. The broad singlets at 1.95 and 1.73 ppm were reasonably identified as D(8)eq and D(8)ax, respectively.

These facts established that phytuberin (1) and phytuberol (2) originate from solavetivone (3) in diseased potato tissues. Although it has not completely been demonstrated yet how 3 is metabolized to 1 and 2, a plausible biogenetic route is illustrated in Scheme 1, which is not contradictory to the Stoessl proposal. Interestingly, 2 seems to be a hydrolyzed product of 1 in vivo, because inoculation of natural (-)-1 in healthy thin slices of potato tubers (Rishiri) at 23 °C for 48 h led to isolation of (-)-2 in 13.2% yield. It is to be noted that phytuberin skeleton would be constructed from a spirovetivane via an eudesmane. We emphasize that the presumable hydroperoxide would participate in this biogenetic route and will report the details in the near future.

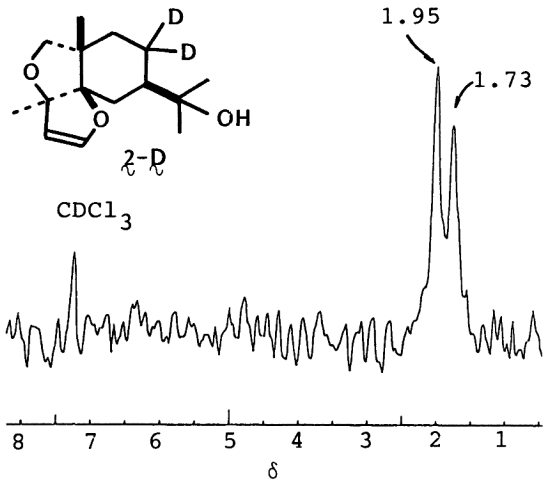
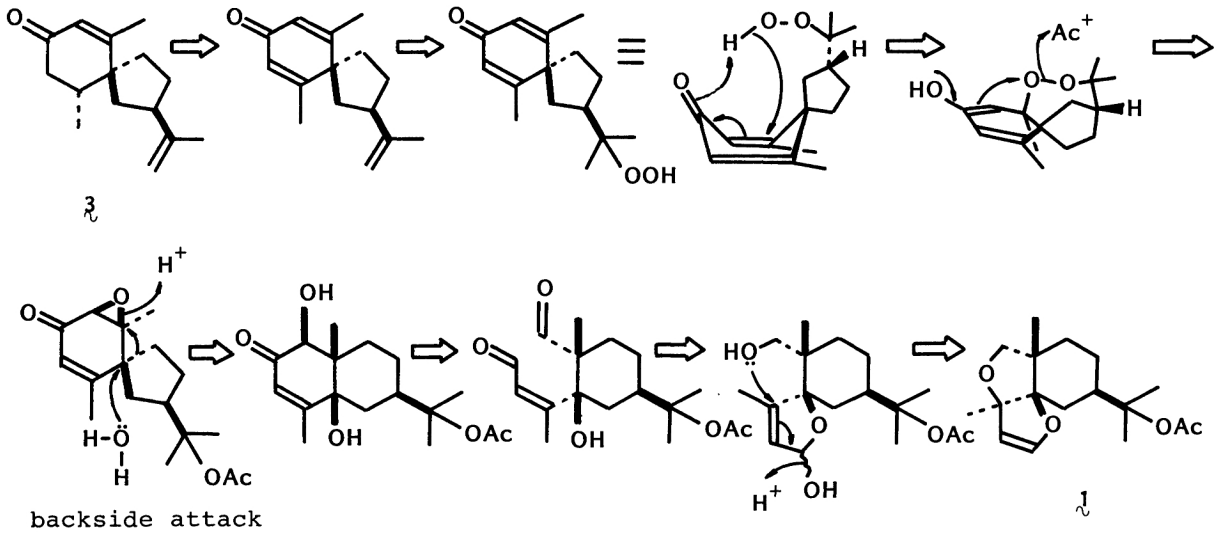


Fig. 1. Fourier-transform ^2H NMR spectrum (61.44 MHz, 10 mm tube, 4200 transients) of 2-D as solution in CCl_4-$CHCl_3$ (10:1).



Scheme 1.

References

1) Part XLIV of "Studies on the Phytoalexins." Part XLIII, A. Murai, H. Miyazaki, K. Watanabe, and T. Masamune, Chem. Lett., 1987, 651.
2) A. Murai, M. Ono, and T. Masamune, Bull. Chem. Soc. Jpn., 55, 1202 (1982) and references cited therein.
3) A. Stoessl, J. B. Stothers, and E. W. B. Ward, Can. J. Chem., 56, 645 (1978).
4) N. Katsui, Y. Takahashi, N. Sato, A. Murai, and T. Masamune, Nippon Kagaku Kaishi, 1981, 659.
5) A. Murai, S. Sato, and T. Masamune, Bull. Chem. Soc. Jpn., 57, 2282 (1984).
6) D. T. Coxon, K. R. Price, B. Howard, S. F. Osman, E. B. Kalan, and R. M. Zacharius, Tetrahedron Lett., 1974, 2921.

(Received April 24, 1987)