BIOSYNTHESIS OF DAPHNILACTONE-B

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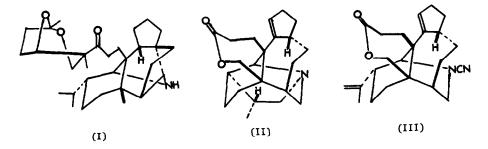
As reported in the previous paper,¹ daphniphylline, one of the major Daphniphyllum alkaloids, has been biosynthesized from six molecules of mevalonic acid (MVA) through a squalene-like intermediate, and the highest incorporation of DL- $[2-^{14}C]$ MVA into daphniphylline has also been recorded in June and July (total incorporation, 0 14%) However, DL- $[2-^{14}C]$ MVA has not been incorporated into yuzurimine-type alkaloids, the carbon skeleton of which consists of twenty-two carbon atoms. From a biogenetic point of view, many C_{22} -alkaloids represented by yuzurimine², methyl homodaphniphyllate³, methyl homosecodaphniphyllate⁴ and daphnilactone-B⁵ could not originate from four MVA molecules and an acetate,⁶ but must be derived from such a precursor as secodaphniphylline (I) or codaphniphylline by oxidative cleavage with loss of eight carbon atoms. In the present paper, the biosynthesis of daphnilactone-B (II), a main alkaloid of the fruits of Daphniphyllum teijsmanni Zollinger,⁵ is described

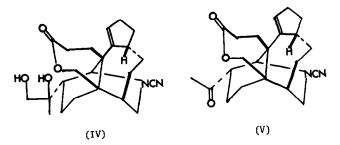
According to essentially the same procedure as reported in the previous paper,¹ the feeding experiments were carried out using a fresh spray of <u>Daphniphyllum teijsmanni</u> Zollinger with unripe fruits (ca 120 grains, ca. 56g)⁷, to which DL-[2-¹⁴C]MVA (100 μ Cl) was fed at room temperature for 14 days The alkaloidal components were extracted, and then carefully separated by preparative tlc [Kieselgel GF₂₅₄ nach Stahl in <u>n</u>-hexane- Et₂O-Et₂NH (20 20 1)] to give a radioactive fraction corresponding to daphnilactone-B (II),⁵ which was diluted with unlabelled daphnilactone-B (200mg) Recrystallization from benzene-

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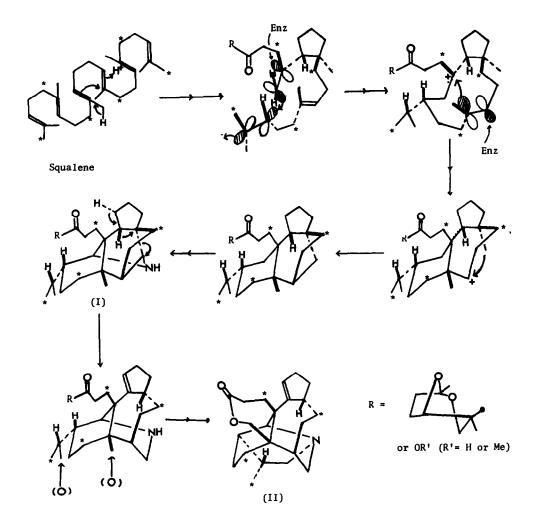
<u>n</u>-hexane afforded the pure daphnilactone-B with constant specific activity (4 27 x 10^4 dpm/mM) (total incorporation, 2.46 x 10^4 dpm, 0.01%) Degradation studies on the labelled daphnilactone-B revealed that four ¹⁴C atoms were included in II, and one-fourth of the. total radioactivity was located at the secondary methyl group, which was demonstrated by the following procedure Von Braun degradation of daphnilactone-B followed by dehydro-bromination with Ag - pyridine gave the known olefin (III), ⁵ which was oxidized with OsO₄ in pyridine - THF (room temp, overnight) to afford the corresponding glycol (IV)⁸ whose specific activity was 4.27 x 10^4 dpm/mM. Further oxidation with NaIO₄ in aq. dioxane

(room temp., overnight) gave a keto-cyanamide (V)⁹ (specific activity, 4.20 x 10^4 dpm/mM) and non-radioactive formaldehyde which was trapped by aq. solution of dimedone. Finally, the iodoform reaction of V afforded yellow needles of CHCI₃ (specific activity, 1.09 x 10^4 dpm/mM).





On the basis of the stereostructures of secodaphniphylline $(I)^{10}$ and daphnilactone-B $(II)^{11}$ coupled with the biogenetic consideration of the amine molety in I^1 , a plausible biosynthetic route to daphnilactone-B from a squalene-like intermediate can be demonstrated as follows.



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- 6 O. E. Edwards in "The Alkaloids Vol. 1", The Chemical Society, London, <u>1971</u>, p. 375, and references cited therein
- 7. Collected at Irako Cape (Aichi-ken, Japan) in the end of september
- 8. A colourless liquid (IV) C₂₃H₃₂O₄N₂ (m/e 400 2340), V_{max}(film) 3360, 2190 and 1725cm⁻¹;
 6 (CDCl₃) 1.28(3H, s), 3.03(1H, d, J= 14 0Hz), 3.50(2H, br s), 3.65(1H, d, J= 13.0Hz), 3.70(1H, brs), 4.14(1H, q, J= 14 0, 7.5Hz), 4 83(1H, d, J= 13.0Hz) and 5.83ppm (1H, br.s)
- 9. The ketocyanamide (V) \cdot m.p. 245 5-246°, $C_{22}H_{28}O_3N_2$, m/e 368(M⁺) and 325, \mathcal{Y}_{max} (KBr) 2190, 1730 and 1720cm⁻¹; $\mathcal{S}(CDC1_3)$ 2.32(3H, s), 3 01(1H, d, J= 14 0Hz), 3.69(1H, d, J= 13.0Hz), 3.75(1H, d, J= 3.0Hz), 4 08(1H, q, J= 14.0, 7 5Hz), 4.80(1H, d, J= 13.0Hz) and 5.90ppm (1H, br s)
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