Tetrahedron Letters No.21, pp. 1961-1964, 1967. Pergamon Press Ltd. Printed in Great Britain.

STRUCTURE OF PIERICIDIN B AND STEREOCHEMISTRY OF PIERICIDINS

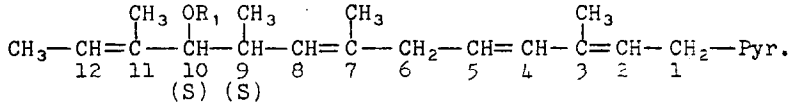
Nobutaka Takahashi, Akinori Suzuki, Yasuo Kimura, Satoshi Miyamoto
and Saburo Tamura
Department of Agricultural Chemistry, The University of Tokyo, Tokyo, Japan

(Received 27 February 1967)

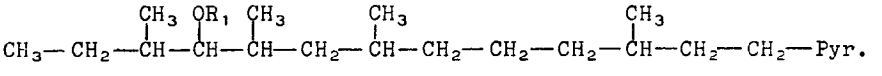
Piericidin A (PA) has been isolated (1) as an insecticide produced by Streptomyces mobaraensis and its structure (2) elucidated as Ia. Later, it was found to be the specific inhibitor (3) to coenzyme Q in the electron transport system of mitochondria. Besides PA, the presence of a homologous compound, piericidin B (PB), has been already suggested (1). In this communication, we wish to report the structure of PB as well as the stereochemistry of PA and PB.

We confirmed by feeding experiment of ^{14}C labelled PA that PA is converted into PB during a prolonged fermentation. Pure PB was separated from PA through the silicic acid adsorption chromatography. PB is rather unstable in the air and shows the same degree of insecticidal activity as PA. PB gives a monoacetate (Id) on the usual acetylation condition. On catalytic hydrogenation, Id absorbs four moles of hydrogen to give octahydropiericidin B (HPB)-monoacetate (IId), to which the molecular formula $C_{28}H_{49}NO_5$ was assigned from elemental analyses and mass spectrum (molecular ion peak at m/e 479). This suggests that PB should have the molecular formula $C_{26}H_{39}NO_4$, although it gave no satisfactory analyses because of its instability. The ultraviolet spectra of PB in neutral, basic and acidic solutions are quite similar to those of PA, suggesting that PB retains the same chromophore as PA. PA-monoacetate (Ib) and HPA-monoacetate (IIb) still contain an alcoholic hydroxyl group, while Id and IId contain no free hydroxyl group. The presence of an aliphatic methoxyl group in PB was shown by a band at 1100 cm^{-1} in its infrared spectrum as well as by a 3H singlet at δ 3.05 in its nmr spectrum (4). These evidences together with the comparison of the molecular formulae of PA and PB suggest that the hydroxyl group at C-10 in PA has been converted to a methoxyl group in PB as

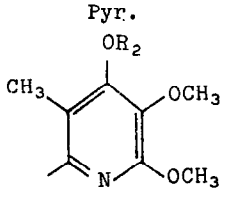
shown in Ic during fermentation. This was confirmed chemically as follows. Ib was methylated with diazomethane in the presence of boron trifluoride to give O-methyl-PA-monoacetate (Id), which was identical with PB-monoacetate in all respects, infrared, ultraviolet and nmr spectra and optical rotation. Thus, the structure Ic was assigned to PB.



I



II

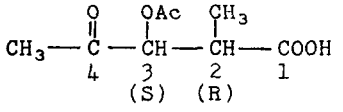


	R_1	R_2
a	H	H
b	H	$COCH_3$
c	CH_3	H
d	CH_3	$COCH_3$

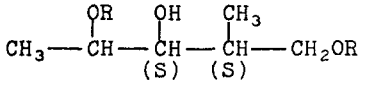
PA and PB contain only two asymmetric centers, C-9 and C-10. Since they are contained in Acid III (III) (2), the ozonolysis product of PA-diacetate, elucidation of the stereochemistry of III was attempted. This structure was further confirmed by following synthesis. Reformatsky reaction between methacrolein and methyl α-bromopropionate and successive acetylation gave methyl 3-acetoxy-2,4-dimethylpent-4-enoate, which was ozonolyzed to give the methyl ester of III. Although the synthetic III methyl ester was a racemic mixture of diastereomers, its infrared spectrum was essentially identical with that of the natural Acid III methyl ester. Gas-liquid chromatography of the synthetic ester on polyethylene glycol succinate column gave two peaks, while the natural ester gave one peak which corresponded to the second peak of the synthetic ester. This indicates that the natural Acid III has not been racemized in the process of the preparation from PA.

On the hydrogenation of Acid III in acetic acid over Adams' catalyst, a

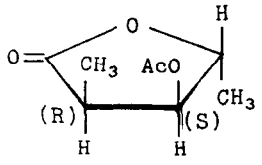
1:1 mixture of a lactone and hydroxy acid was obtained. Nmr spectrum of the lactone (3H, d, δ 1.09; 3H, d, δ 1.28; 3H, s, δ 2.10; 1H, d-q, δ 2.82; 1H, d-q, δ 4.53; 1H, d-d, δ 5.43) and the double resonance experiment supported the proposed structure IV. The fact that $J_{H-2,H-3}$ and $J_{H-3,\ H-4}$ are 6 and 3 cps, respectively, suggests that in IV H-2 and H-3 exist in cis-configuration, and H-3 and H-4 in trans, on the application of Karplus equation on qualitative base (5, 6). The following chemical evidences support the above assumption. The formation of the hydroxy acid in 50% yield on hydrogenation in acidic medium indicates that this acid probably has the opposite configuration to IV at C-4, and can not form a γ-lactone ring because of large steric hindrance from all cis-substituents. Furthermore, catalytic hydrogenation of the synthetic Acid III methyl ester in methanol gave a complex mixture of the hydrogenation products, in which the presence of a γ-lactone was indicated by the band at 1782 cm^{-1} in its infrared spectrum, while the natural Acid III gave only a trace of a γ-lactone. The simultaneous lactonization on catalytic reduction in a neutral solvent may be expected in the case of the diastereomer of the natural Acid III to form a γ-lactone having all trans-substituents. These evidences allow to assign the relative configuration of C-2 and C-3 in the natural Acid III as S-R or R-S and consequently C-9 and C-10 in piericidins as R-R or S-S.



III



V R=H, VI R=SO_2CH_3



IV

Lithium aluminum hydride reduction of Acid III methyl ester gave a triol (V), which was converted into dimesylate (VI). Reduction of VI with lithium aluminum hydride gave 2-methyl-3-pentanol, which was characterized as a 3,5-dinitrobenzoate, mp. 98-99°; $[\alpha]_D^{17}$ =+7.67° (C=2.9 in $CHCl_3$). Since the (-)-ester

or (-)-alcohol has been correlated to L-glyceraldehyde (7), the absolute configuration of C-3 in (+)-dinitrobenzoate of 2-methyl-3-pentanol must be R. It is now evident that both C-9 and C-10 in piericidins have the S-configuration from the stereochemical correlation between (+)-2-methyl-3-pentanol and piericidins.

Acknowledgement. The authors are indebted to Kaken Chemical Co. for the preparation of crude piericidin, Mr. H. Sato, the Naka Works, Hitachi Ltd., for the measurement of mass spectrum and Mr. K. Aizawa, Department of Agricultural Chemistry, The University of Tokyo, for measurement of infrared, ultraviolet and nmr spectra. This research was supported in part by the research fund from Waksman Foundation of Japan Inc., to which authors wish to express their thanks.

REFERENCES

1. S. Tamura, N. Takahashi, S. Miyamoto, R. Mori, S. Suzuki and J. Nagatsu, Agr. Biol. Chem., 27, 576 (1963).
2. N. Takahashi, A. Suzuki and S. Tamura, J. Am. Chem. Soc., 87, 2066 (1965); N. Takahashi, A. Suzuki and S. Tamura, Agr. Biol. Chem., 30, 1 (1966).
3. C. Hall. M. Wu, F. L. Crane, N. Takahashi, S. Tamura and K. Folkers, Biochem. Biophy. Res. Communs., 25, 373 (1966).
4. Chemical shifts are expressed in δ-value (ppm) from tetramethylsilane as internal standard.
5. G. W. Perold and K. G. R. Pachler, J. Chem. Soc., C, 1966, 1918.
6. Although recent investigations indicate that Karplus equation should not be applied on five membered ring on quantitative base, it has been suggested (5) that qualitative conclusions may be drawn from experimentally determined parameters.
7. G. Büchi, L. Crombie, P. J. Godin, J. S. Katenbronn, K. S. Siddalingaiah and A. A. Whiting, J. Chem. Soc., 1961, 2844.