Tetrahedron, 1963, Vol. 19, pp. 1437 to 1439. Pergamon Press Ltd. Printed in Northern Ireland

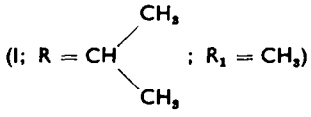
# THE STRUCTURE AND SYNTHESIS OF PIMPRININE

B. S. JOSHI and W. I. TAYLOR
Research Department, CIBA Pharmaceutical Company, Division of CIBA Corporation, Summit, New Jersey
and
D. S. BHATE and S. S. KARMARKAR
Hindustan Antibiotics Ltd., Pimpri (near Poona)

(*Received* 20 *April* 1963)

**Abstract**—The mould metabolite pimprinine obtained from *Streptomyces pimprina* has been shown by degradation and synthesis to be 5,3′-indolyl-2-methyloxazole (III).

*Streptomyces pimprina* is known to produce an antifungal antibiotic, hamycin, of the polyene type[1] which occurs in the mycelium, whereas the filtrates contain thiolutin (I; R, $R_1 = CH_3$), aureothricin (I; $R = CH_2CH_3$; $R_1 = CH_3$), isobutyropyrrothine




and a colourless crystalline compound, m.p. 205°.[2] The colourless crystalline compound has since been named pimprinine.

Pimprinine is a very weak base, and it analyses for the molecular formula $C_{12}H_{10}N_2O$ (molecular weight—Rast 198). Kuhn Roth determination indicates that it contains one C-methyl group, and N-acetyl determination yields a non-stoichiometric amount (15–20 per cent) of theoretical value of acetic acid for one such group. It does not contain methoxyl or N-methyl groups. The I.R. spectrum (nujol mull) shows peaks at 3150 $cm^{-1}$ (broad) OH or NH, 1640, 1630 and 1590 $cm^{-1}$, and none of the latter are strong enough to be carbonyls. The U.V. absorption spectrum has $\lambda_{max}^{EtOH}$ 224, 266, 284 (shld.) and 300 (shld.) m$\mu$; log $\epsilon$ 4·36, 4·17, 4·07 and 4·02 respectively. Pimprinine forms a methiodide and an N-acetyl derivative. These results indicate that pimprinine contains a basic tertiary nitrogen and a second nitrogen probably present as an —NH group. It also appears that the oxygen function, because of its unreactivity and the above physical data, is present, not as a carbonyl or a hydroxyl, but as an ether grouping. An important clue to the structure came from the products of the alkali fusion of pimprinine when indole, indole-3-acetic acid, acetic acid and ammonia were obtained. These fragments account for all the carbons, nitrogens and oxygen of the molecule and on this basis a partial structure (II) could be written.

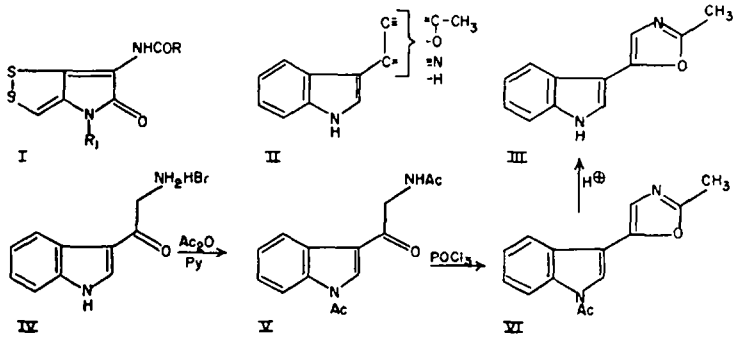
When pimprinine methiodide was warmed in alkali, a solution was obtained with U.V. absorption maxima at 334, 304 and 265 m$\mu$, reverting in acid to 296 and 265

[1] D. S. Bhate, G. R. Ambekar, K. K. Bhatnagar, R. K. Hulyalkar, *Hindustan Antibiot. Bull.* **4**, 139 (1961).
[2] D. S. Bhate, R. K. Hulyalkar and S. K. Menon, *Experientia* **16**, 504 (1960).

(shld.) mμ, behavior typical of 3-acetyl indole subjected to the same pH conditions.[3] The above results coupled with those of the P.M.R. spectrum* of pimprinine, unsplit methyl (2·40 p.p.m.) and six aromatic type protons (6·80–7·25 p.p.m.) suggested quite strongly that the structure of the mould metabolite was the biogenetically reasonable possibility III.†

3-Aminoacetylindole hydrobromide (IV)[4] was converted into its diacetyl derivative (V) and cyclized in refluxing phosphorous oxychloride. The resulting 5-1′-acetyl-3′-indolyl-2-methyloxazole (VI) upon hydrolysis in methanolic hydrochloride gave a product (III) indistinguishable from pimprinine. An alternative route involved the condensation of indole with acetylglycine in phosphorous oxychloride to give 3-acetamidoacetylindole, but the yield was too poor to be of practical use.

This is the first example of the isolation of an indole from *Streptomyces* species, and its biosynthesis in now under investigation.




## EXPERIMENTAL

*Pimprinine methiodide.* Pimprinine (0·5 g) in chloroform (50 ml) and methyl iodide (5 ml) was left 4 days. The precipitate was recrystallized from ethanol to m.p. 228° (dec).

If the methiodide was crystallized from either water or aqueous alcohol, pimprinine was regenerated.

*The alkali fusion of pimprinine.* Pimprinine (8 g) and sodium hydroxide (40 g) were heated at 250–260° for 1 hr in an atmosphere of nitrogen, and the volatile products were absorbed in N hydrochloric acid (basic fraction). The fused mass was dissolved in water and extracted with ether (neutral fraction), and the aqueous solution was acidified with 50% sulphuric acid, thoroughly extracted with ether, which in turn was shaken out with sodium bicarbonate (acidic fraction).

The hydrochloric acid solution (basic fraction) was concentrated and chloroplatinic acid added to furnish a yellow precipitate. This was recrystallized from water to yield pure ammonium chloroplatinate (Found: N, 6·4. $(NH_4)_2PtCl_6$ requires: N, 6·3%).

The neutral fraction was crystallized from pet. ether (b.p. 40–60°) and gave indole, m.p. 49–50° (Found: C, 81·8; H, 5·6; N, 12·0. $C_8H_7N$ requires: C, 82·1; H, 6·0; N, 12·0%).

The acidic fraction was crystallized from benzene and from water to give indole-3-acetic acid, m.p. 168° (dec.) undepressed by an authentic sample.

*Run by J. Siragusa in trifluoroacetic acid on a Varian A60 using tetramethylsilane as internal standard.

†This structure is in complete agreement with the observation that hydrogenation of pimprinine gave acetyltryptamine (Dr. A. Hofmann, personal communication.)

[3] J. A. Ballantine, C. B. Barrett, R. J. S. Beer, B. G. Boggiano, S. Eardley, B. E. Jennings and A. Robertson, *J. Chem. Soc.*, 2227 (1957); M. F. Bartlett, D. F. Dickel and W. I. Taylor, *J. Amer. Chem. Soc.* **80**, 126 (1958).

[4] K. Bodendorf and A. Walk, *Arch. Pharm.* **294**, 484 (1961).

If the crude alkali fusion mixture of pimprinine was steam distilled at a low pH, acetic acid (0·88 mole equivalent) could be isolated and characterized as its 5-benzylthiouronium salt, m.p. 139–140°.

1-*Acetyl*-3-*acetamidoindole* (V). 3-Aminoacetylindole hydrobromide (5 g) and acetic anhydride (5 ml) were stirred in pyridine (15 ml) until a clear solution was obtained. The excess solvent was removed *in vacuo* and the residue triturated with water. The product crystallized from methanol-water or benzene and had m.p. 179°, $\lambda_{max}$ m$\mu$ ($\varepsilon \times 10^{-3}$) 217 (30·18), 258 shld. (6·56) and 300 (12·62); $\lambda_{min}$ 268 (4·96); $\nu_{C=O}$ 1723, 1682 and 1650 cm$^{-1}$. (Found: C, 65·0; H, 5·5. $C_{14}H_{14}N_2O_3$ requires; C, 65·1; H, 5·5%).

3-*Acetamidoacetylinsole*. The above acetyl compound (100 mg) was heated with methanol (9 ml) and 6N hydrochloric acid (5 ml) at 60° for 30 min and poured into water. It was basified with ammonia and extracted into methylene chloride. The product crystallized from methanol, m.p. 225°, $\lambda_{max}$ m$\mu$ ($\varepsilon \times 10^{-3}$) 240 (12·9), 256 (9·15) and 297 (12·3); $\lambda_{min}$ 226 (4·88), 253 (9·1) and 271 (6·72); $\nu_{C=O}$ 1660 and 1638 cm$^{-1}$ (Found: C, 66·9; H, 5·6; N, 13·4. $C_{12}H_{12}N_2O_2$ requires: C, 66·7; H, 5·6; N, 13·0%).

5-1′-*Acetyl*-3′-*indolyl*-2-*methyloxazole* (VI). 1-Acetyl-3-acetamidoindole (200 mg) was heated under reflux with phosphorous oxychloride (5 ml) for 4 hr. The solution was poured into water, basified with ammonia and the precipitate (190 mg) was crystallized from methanol. It had m.p. 160°, $\lambda_{max}$ m$\mu$ ($\varepsilon \times 10^{-3}$) 236 shld. (21·4), 243–247 (23·0), 268–274 plateau (9·84), 283 (8·52), 306 (14·9) and 311 shld. (13·8); $\lambda_{min}$ 220 (15·00), 279 (7·95) and 288 (7·8); $\nu_{C=O}$ 1650 and 1720 cm$^{-1}$. (Found: C, 70·4; H, 5·0; N, 11·8. $C_{14}H_{12}N_2O_2$ requires: C, 70·0; H, 5·0; N, 11·7%).

*Pimprinine* (III). The above acetyl compound (50 mg) in methanol (6 ml), 6N hydrochloric acid (2 ml) was heated at 60° for 15 min. The solution was basified and extracted with methylene chloride. From the organic layer pimprinine (42 mg) was obtained, m.p. 205° (undepressed mixed m.p. with the natural material) from benzene, $\lambda_{max}$ m$\mu$ ($\varepsilon \times 10^{-3}$) 224 (24·46), 264–266 (14·74), 238 shld. (12·15) and 299 shld. (10·59); $\lambda_{min}$ 243 (8·34). (Found: C, 72·5; H, 5·3. $C_{12}H_{10}N_2O$ requires: C, 72·7; H, 5·1%).