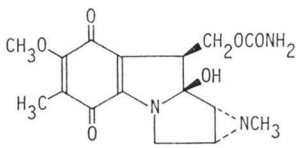
NEW MITOMYCIN, 10-DECARBAMOYL-OXY-9-DEHYDROMITOMYCIN B FROM *STREPTOMYCES CAESPITOSUS*

Sir:

In 1956, HATA and his co-workers reported that *Streptomyces caespitosus* produced mitomycins A and B[1]. Thereafter, WAKAKI and his co-workers found mitomycin C from the same strain[2], HERR and his co-workers found porfiromycin from *Streptomyces ardus*[3], and LEFEMINE and his co-workers found mitiromycin from *Streptomyces verticillatus*[4]. These compounds are generally called mitomycin antibiotics.

In this communication, we described a new mitomycin which was recovered from a fractionation segment rich in mitomycin B. The rich-cut was obtained as a chloroform eluate from alumina chromatography performed upon chloroform extracts of the broth filtrate from a *Streptomyces caespitosus* fermentation[2]. One hundred milliliters of the rich-cut was evaporated under reduced pressure. To the residue, a small amount of chloroform was added and the insoluble materials were filtered off. The filtrate

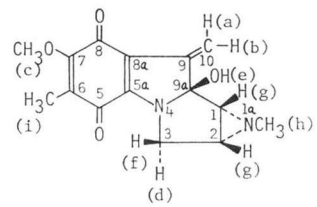


Mitomycin B[5]

was chromatographed on silica gel with acetone - chloroform (0: 1~1: 1) in a gradient manner. Less polar blue colored fractions eluting prior to mitomycin B were collected and evaporated under reduced pressure. The residue was subjected to chromatography over silica gel with acetone - chloroform (1: 1). The main eluate was evaporated under reduced pressure. By crystallizing the residue from acetone, 40 mg of bluish-purple needles (**1**) were obtained. Properties of the compound are shown in Table 1. The color of **1** indicates that it is probably a mitomycin antibiotic. A 60 MHz p.m.r. spectrum showed that 7-methoxy protons (δ 4.06), la-methyl protons (δ 2.18) and 6-methyl protons (δ 1.74) in the mitosane structure are present. The signals of δ 6.13 and δ 5.33 indicated the existence of olefinic protons (>C=CH_2). The

Table 1. Properties and structure of **1**.

Color	Bluish-purple
Rf*1	0.70 (mitomycin B; 0.30)
M^+*2	*m/z* 288.1135 (288.1110 calcd. for $C_{15}H_{16}N_2O_4$)
i.r. ν_{max}(KBr)*3	3,400, 2,950, 1,660, 1,625, 1,560
p.m.r. $\delta(CDCl_3)$*4	(a) 6.13 (1H, s), (b) 5.53 (1H, s), (c) 4.06 (3H, s), (d) 3.98 (1H, d, J=13 Hz), (e) 3.73 (1H, bs), (f) 3.45 (1H, d, J=13 Hz), (g) 2.27 (2H, s), (h) 2.18 (3H, s), (i) 1.74 (3H, s)
^{13}C n.m.r. $\delta(CDCl_3)$*5	182.58; (5) } or 177.59; (8) } 47.92; 1a-CH_3 157.53; 5a 43.95; 1 151.06; 7 43.83; 2 143.38; 9 8.17; 6-CH_3 124.43; (8a) } or 113.39; (6) } or 112.23; 10 } or 100.04; (9a) } 61.27; 7-OCH_3 48.77; 3



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*1 Silica gel TLC plate; Merck Art. 5719 (actone - chloroform=1 : 1).

*2 JEOL JMS-OISG-2 mass spectrometer.

*3 Hitachi 215 spectrophotometer.

*4 Varian T-60 spectrometer (TMS as internal standard).

*5 JEOL PFT-100 NMR spectrometer (25 MHz, TMS as internal standard).

signal of δ 3.73 disappeared on addition of deuterium oxide. Amino protons (*ca.* δ 5~7) of the urethane group in mitomycins were absent. In the i.r. spectrum of **1**, the carbonyl absorption (usually *ca.* 1720 cm^{-1}) of the urethane group was not observed. The molecular ion peak in the mass spectrum of **1** indicated that the molecular formula was $C_{15}H_{16}N_2O_4$. Since the molecular formula of mitomycin B is $C_{16}H_{19}N_3O_6$, the molecule of **1**, accordingly, is smaller by CH_3NO_2 (H_2NCOOH) as compared with mitomycin B.

From these facts, it was presumed that **1** must be 10-decarbamoyloxy-9-dehydromitomycin B. The structure is shown in Table 1. The n.m.r. spectra, i.r. spectrum and mass spectrum of **1** were well explained by this structure. The p.m.r. spectrum and ^{13}C n.m.r. spectrum were assigned as shown in Table 1 in reference to our accumulated data and the report[6] by LOWN and his coworker on mitomycin chemistry. The configuration of **1** was determined to be as shown in Table 1, since **1** has been derived chemically from mitomycin B[7]. The compound **1** is the first mitomycin having an exo-cyclic double bond in its structure.* It shows antibacterial activity and cytotoxic activity. Details of **1** will be published elsewhere[7].

Acknowlegements

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* Independently of our work, two new mitomycins similar to **1** were found from *Streptomyces caespitosus* by an other group at our laboratory. These compounds were presented, with other new naturally occurring mitomycins, at a recent symposium—SHIRAHATA, K.; M. KONO, I. MATSUBARA & M. KASAI: New mitomycins, structure determination, derivation and their anticancer activity. 23rd Symposium on the Chemistry of Natural Products (Nagoya, Japan, October 22~25, 1980).

CHIKAHIRO URAKAWA
HARUKO TSUCHIYA
KIN-ICHI NAKANO

Tokyo Research Laboratory
of Kyowa Hakko Kogyo Co., Ltd.
3-6-6, Asahimachi, Machidashi, Tokyo, Japan

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