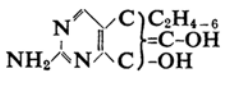
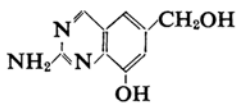
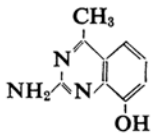


(I)



(II)



(III)

## *Structure of the $C_9$-Base, an Alkaline Degradation Product of Tetrodotoxin*

By Toshio GOTO, Yoshito KISHI and Yoshimasa HIRATA

(Received March 29, 1962)

By alkaline degradation of tetrodotoxin, Kawamura[1] obtained a yellow compound ($C_9$-base), to which he assigned a molecular formula $C_9H_{11}N_3O_2$ and a partial structure (I). 2-Aminopyrimidine moiety of the structure was rigorously established by isolation of 2-aminopyrimidine-4, 5-dicarboxylic acid from the oxidation products of the $C_9$-base. Tsuda et al.[2] further assumed that the pyrimidine nucleus would be fused with a five-membered ring containing an enol group. From the results reported, however, an alternate possibility that the $C_9$-base might be a phenol derivative, instead of the enol, can not be excluded. We wish to present a new structure, 2-amino-6-hydroxymethyl-8-hydroxyquinazoline (II), to the $C_9$-base from the following evidences.

The analytical value of the $C_9$-base hydrochloride is rather consistent with the formula of $C_9H_9N_3O_2 \cdot HCl$ (Found: C, 47.86; H, 4.47; N, 18.33, 18.09. Calcd.: C, 47.48; H, 4.42; N, 18.46%)[3]. Since one of the two hydroxyl groups in the $C_9$-base is alcoholic[1], only possible structure having a phenolic hydroxyl group is 2-amino-*x*-hydroxymethyl-*x'*-hydroxyquinazoline. For alkyl substituents would not influence much on the ultraviolet spectra of aromatic compounds, 2-amino-4-methyl-8-hydroxyquinazoline (III) was synthesized to compare its ultraviolet spectrum with that of the $C_9$-base. The quinazoline III, m. p. over 270°C (in sealed tube), was obtained from 2-nitro-3-hydroxyacetophenone by catalytic hydrogenation to 2-amino derivative, m. p. 165~168°C, and then by condensation with cyanamide. Ultraviolet spectra of the synthetic quinazoline III in different pH's almost superimposable, except for slightly hypsochromic shifts of 2~5 mμ, with those of the

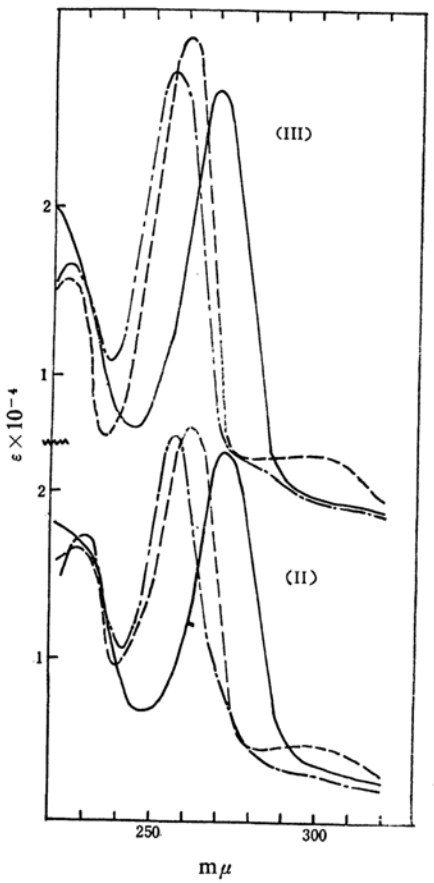



Fig. 1

——— 0.1 N NaOH —·—· $H_2O$
– – – 0.1 N HCl

1) M. Kawamura, *Chem. Pharm. Bull.* (*Japan*), **8**, 262 (1960).
2) K. Tsuda, M. Kawamura and R. Hayatsu, 3rd Symposium of Natural Products, Japan, October, 1959.
3) Analyses of the all derivatives of the $C_9$-base reported[1] are consistent with this formula.

$C_9$-base (Fig. 1). It is, therefore, quite reasonable to assume that the $C_9$-base has a 2-amino-8-hydroxyquinazoline skeleton[4].

NMR spectrum of the $C_9$-base shows four signals (all singlet) at +26, −48, −65, −113 c. p. s.[5] (area ratio ca. 2 : 1 : 1 : 1), which are assigned to two protons in $-CH_2O-$, two in benzene ring (two signals), and one attached at $C_4$-position, respectively. Since no coupling is observed between their signals, two aromatic protons are not in adjacent position. Thus the hydroxymethyl group must be attached to the $C_6$-position. NMR spectrum of the triacetate[1] of $C_9$-base possesses the following signals which are also consistent with the structure assigned: ($\tau$-values) 8.00 (NH); 7.80, 7.50, 7.30 (three acetyl groups); 4.70 ($-CH_2O-$); 2.40, 2.25 (arom.); 0.68 (proton at $C_4$); all signals are singlets[6].

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4) 2-Amino-4-methyl-6-hydroxyquinazoline was also synthesized, but its UV spectrum ($\lambda_{max}^{H_2O}$ 233 m$\mu$; $\lambda_{max}^{0.1N\ NaOH}$ 245 m$\mu$) was quite different from that of the $C_9$-base.

5) Spectra were taken on a Nihondensi JM (40 Mc) spectrometer using $D_2O$ containing NaOD as solvent, and a signal of DOH as an internal reference (0 c. p. s.).

6) Using $CDCl_3$ as solvent and tetramethylsilane as an internal reference ($\tau$ = 10).