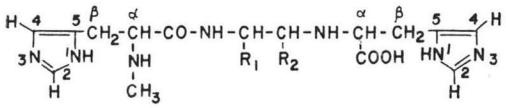
THE STRUCTURE OF FELDAMYCIN

Sir:

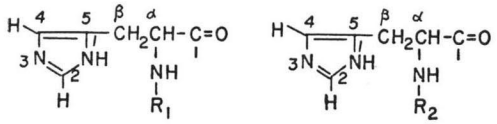
Feldamycin is an antibiotic isolated from culture filtrates of *Streptomyces ficellus*. The production, isolation, characterization and preliminary biological evaluation of feldamycin were described in a previous communication[1]. We now wish to report the structure of feldamycin which is represented by **Ia** (Fig. 1).

Fig. 1.



Ia: $R_1 = COOH$: $R_2 = CH_3$
Ib: $R_1 = CH_3$: $R_2 = COOH$

Fig. 2.



II **III**

Feldamycin, $C_{17}H_{25}N_7O_5$, mol. weight 407, is an amphoteric compound which is adsorbed on both anionic and cationic exchange resins and eluted by the use of appropriate solvents. The ir spectrum shows absorptions at 3130~3170 cm^{-1} (-OH; -NH) and at 1665 (shoulder), 1622 and 1570 (shoulder) cm^{-1} assigned[2] to the presence of amide, COO^- NH^+ zwitterions and the imidazole systems (Fig. 2) in feldamycin. The ir of feldamycin hydrochloride has a characteristic absorption at 1695 cm^{-1} assigned to free -COOH group(s).

The 100 MHz pmr spectrum* (Fig. 3) of a D_2O solution of feldamycin clearly indicated the presence of two histidine-like moieties (**II**, **III**, Fig. 2). The protons at C-2 and C-4 of the two imidazole rings appear at δ 7.95 (apparent s, 2H) and δ 7.15 (two apparent s, 2H) respectively. The complex absorptions at δ 4.00 (2H) and δ 3.20 (4H) are assigned to the α-CH and β-CH_2 of the two histidine-like moieties. The pmr assignments agree with cmr data (Table 1). The 25.16 MHz ^{13}C-nmr spectrum showed the presence of all 17 carbons of feldamycin. The peaks at δ 60.7 and 62.7, assigned to the α-CH carbons (Fig. 2) by single frequency proton decoupling at δ 4.0, are shifted downfield *ca.* 5~7 ppm compared to histidine[3]. This indicates that both amino groups of **II** and **III** (Fig. 2) are secondary (β-effect).[4,5]

Table 1. Chemical shifts observed in the C-13 nuclear magnetic resonance spectrum of feldamycin

Chemical shift, δ*	Multiplicity**	Assignment
60.7, 62.7	2d	**II**, **III** (Fig. 2) α-CH
28.7, 28.9	2t	" β-CH_2
136.6, 136.6	2d	" C-2
117.6, 118.0	2d	" C-4
131.3, 132.5	2s	" C-5
174.2, 174.9	2s	" carbonyl
13.1	q	CH_3
32.9	q	NCH_3
54.0	d	CH_3-CH
57.0	d	CH-COOH
171.2	s	O ‖ -C-OH

* Shifts given in ppm relative to TMS with dioxane used as an internal standard (δ, 67.4).
** Multiplicity in off-resonance decoupled spectrum: q=quartet; t=triplet; d=doublet; s=singlet.

A characteristic feature in the pmr spectrum of feldamycin is a singlet at δ 2.61 (3H) assigned to an N-CH_3 group. The presence of this absorption at too high field excludes attachment of the methyl group at N-1 or N-3 of the imidazole rings of either **II** or **III** (Fig. 2). The presence of an N-CH_3 in feldamycin is also indicated by absorption at δ 32.9 (q) in the cmr spectrum (Table 1).

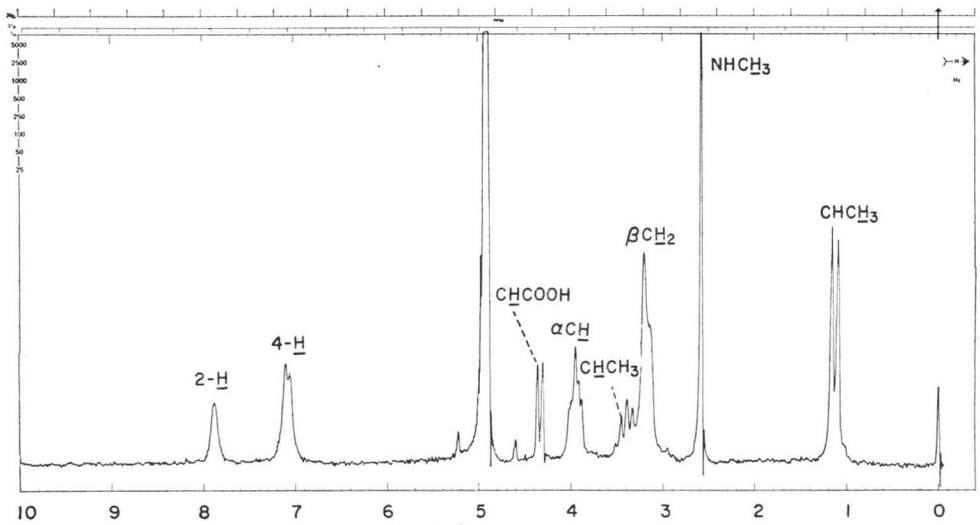
Furthermore, the pmr spectrum showed the presence of a CH_3 group at δ 1.15 (d, J=6.5, 3H). Spin decoupling indicated that the CH_3 is coupled to a -CH- at δ 3.45 (d of q, 1H) which in turn is coupled to a -CH- at δ 4.39 (d, J=5, 1H). The spin decoupling studies combined with the observed chemical shift of the methine protons indicate the presence of the fragment

$$CH_3\text{-}\underset{X}{CH}\underset{Y}{CH}\text{-}Z$$

(X, Y, Z=electronegative groups) in feldamycin. The cmr spectrum of feldamycin contains two carbonyl absorptions at δ 174.5 (s) and 174.9 (s)

* Multiplicity in pmr or off resonance-cmr spectra: s=singlet; d=doublet, t=triplet; q=quartet; m=multiplet.

Fig. 3. 100 MHz Nuclear magnetic resonance spectrum of feldamycin*



* Refer to Structure **Ia**.

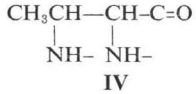
Table 2. High resolution mass spectral* data of feldamycin-TMS**

m/e (Found)	Theory	Elemental composition	Assignment
695.3494	695.3498	$C_{17}H_{21}N_7O_5 \cdot [Si(CH_3)_3]_4$	M^+
542.2643	542.2650	$C_{13}H_{17}N_5O_5 \cdot [Si(CH_3)_3]_3$	$M^+ - [CH_2\text{-}N\text{-}TMS]$
524.2555	524.2545	$C_{13}H_{15}N_5O_4 \cdot [Si(CH_3)_3]_3$	$M^+ - [CH_2\text{-}N\text{-}TMS] - H_2O$
511.2253	511.2228	$C_{12}H_{12}N_4O_5 \cdot [Si(CH_3)_3]_2$	$M^+ - [CH_2\text{-}N\text{-}TMS] - CH_3NH_2$
326.1704	326.1720	$C_8H_{10}N_3O_2 \cdot [Si(CH_3)_3]_2$	$CH_3CH{=}\overset{\oplus}{N}H\text{-}CH(COOTMS)\text{-}CH_2\text{-}N\text{-}TMS$
196.1265	196.1270	$C_6H_9N_3 \cdot Si(CH_3)_3$	$CH_3\overset{\oplus}{N}H{=}CHCH_2\text{-}N\text{-}TMS$
154.0915	154.0926	$C_4H_5N_2 \cdot Si(CH_3)_3$	$[CH_2\text{-}N\text{-}TMS + H]^+$

* High resolution mass spectra were recorded on photographic plate (Ionomet) using a CEC-21-110B mass spectrometer. Sample was introduced in the mass spectrometer using the direct probe. All spectra were recorded at 70 eV.

** TMS=trimethylsilyl.

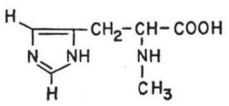
Fig. 4.



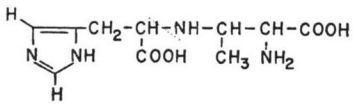
IV

assigned to the carbonyl groups of **II** and **III** (Fig. 2). A third carbonyl resonance at δ 171.2 was shown by single frequency decoupling[6] to be bonded to a methine group the proton of which appears at δ 4.39 in the pmr spectrum. In addition two methine carbons were found at δ 54.0 (d) and 57.0 (d) corresponding to the methine protons at δ 3.45 and 4.39, respectively. These carbon chemical shifts are at too high field for carbons bonded to oxygen, hence they must both be attached to nitrogen. We conclude therefore that in addition to the histidine-like

Fig. 5



V



VI

moieties **II** and **III** (Fig. 2) a fragment represented by **IV**, Fig. 4, is present in feldamycin. The data discussed indicate that **II**, **III**, **IV** and an N-CH_3 group account for all carbons of feldamycin. Cmr spectral data require that the secondary amino group of **II** and **III** (Fig. 2) are not involved in peptide bond formation. This requirement suggest structures **Ia** and **Ib** as the only possible structures for feldamycin.

High resolution mass spectral studies on both the trimethylsilyl and deuterated trimethylsilyl (TMS-d_9) derivatives of feldamycin allowed the assignment of the fragment ions reported in Table 2. All fragment ions with the exception of the ion at *m/e* 326 are in agreement with both structures **Ia** or **Ib**. The ion at *m/e* 326 permits differentiation between the two possible structures. The composition of this ion, $C_8H_{10}N_3O_2 \cdot [Si(CH_3)_3]_2$, indicates that this fragment can only originate from **Ia**. Cleavage of the structure **Ib** between the carbons substituted by R_1 and R_2 would give rise to ions occurring at *m/e* different from 326 with elemental composition containing N_3O_4 or N_4O_3 but not N_3O_2 as in the case for the ion at *m/e* 326. It is concluded therefore that feldamycin has structure **Ia** and is a dipeptide containing N-methylhistidine (**V**, Fig. 5) and a new aminoacid designated feldamycic acid (**VI**, Fig. 5).

It should be noted that the ion at *m/e* 326 is derived from the feldamycic acid part while the ion at *m/e* 196 (Table 2) results from the N-methylhistidine moiety of feldamycin. The stereochemistry of aminoacids **V** and **VI** is not known but we hope that studies, already underway, will resolve this question.

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