THE STRUCTURES OF COMPONENT A₁ (=LL-AB664) AND COMPONENT A₂ (=LL-AC541),
STREPTOTHRICIN-LIKE ANTIBIOTICS

Sir:

Two streptothricin-like antibiotics effective against *Serratia marcescens*, tentatively designated as components A₁ and A₂, were isolated from broth filtrate of *Streptoverticillium olivoreticuli*¹²). Component A₁ was identified to be LL-AB664 (=BD-12)³⁶) by Avicel and silica gel TLC²) and component A₂ as the LL-AC541 (=BY-81, citromycin, E-749-C)⁵⁹), respectively. Streptothricin group antibiotics give streptolidine, D-gulosamine, L-β-lysine, ammonia and carbon dioxide by acid hydrolysis, while streptolidine or N-methylstreptolidine, N-methyl-D-gulosamine, glycine with or without formic acid, ammonia and carbon dioxide were found in the acid hydrolysate of streptothricin-like antibiotics. Though, structures have been proposed for several streptothricin-like antibiotics, the location of the carbamoyl group has not yet been fully determined, it is either on the C₃- or C₄-OH group of the N-methyl-D-gulosamine moiety. Thus, ¹H-NMR and ¹³C-NMR spectra of component A₂ (I-b) and its partial hydrolysis product, N-guan-streptolidyl N'-methyl-β-D-gulosaminide (II), were studied to determine the location of the carbamoyl group on the aminosugar. The partial hydrolysis product II was obtained from I-b according to the method reported by BORDERS et al.⁴).

¹H-NMR spectra of I-b and II were taken in D₂O. The complete assignment of the proton resonances of II has already been achieved by BORDERS et al.⁴¹. They have also partly assigned the proton resonances of I-b, however the protons of the aminosugar moiety have been left unidentified except C₁-H and C₆-H.

The ¹³C-NMR spectra of both compounds in D₂O were assigned by the help of proton selective decoupling experiments. For the spectrum of II every proton has been properly assigned. For the spectrum of I-b, carbon signals of the streptolidine moiety were assigned by irradiation of known proton resonance position*. By the same method, C₁ and C₆ signals of the aminosugar moiety of I-b were assigned to δ 77.8 and 61.1, respectively. ¹³C-Chemical shifts for the N-methylgulosamine moiety of I-b (see below) and II are shown in Table 1.

NAGANAWA et al. reported the ¹³C-NMR of bleomycins, in which the effects of substitution of a carbamoyl group on sugar hydroxyl groups were precisely studied¹⁰). They found that the α-carbon signal was shifted downfield by 1.9~5.0 ppm on substitution, while the β-carbon signal was shifted upfield by 0.3~2.7 ppm. The chemical shift of C₂'-H (δ 5.2) reported by BORDERS et al. should be corrected to δ 4.8 according to our decoupling experiment.

![Fig. 1.](image)

**Component A₁ (I-a):** R = CH₃

**Component A₂ (I-b):** R = H

![Fig. 2.](image)

**N-guan-Streptolidyl N'-methyl-β-D-gulosaminide (II).**

![Fig. 3.](image)

**Table 1.** ¹³C-Chemical shifts for N-methyl-D-gulosamine moiety of component A₂ (I-b) and N-guan-streptolidyl N'-methyl-β-D-gulosaminide (II).

<table>
<thead>
<tr>
<th></th>
<th>C₁</th>
<th>C₂</th>
<th>C₃</th>
<th>C₄</th>
<th>C₅</th>
<th>C₆</th>
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<tbody>
<tr>
<td>I-b</td>
<td>77.8</td>
<td>61.7</td>
<td>61.7</td>
<td>71.5</td>
<td>74.7</td>
<td>61.1</td>
</tr>
<tr>
<td>II</td>
<td>78.4</td>
<td>57.5</td>
<td>65.7</td>
<td>68.2</td>
<td>75.4</td>
<td>61.6</td>
</tr>
<tr>
<td>Δδ</td>
<td>-0.6</td>
<td>+4.2</td>
<td>-4.0</td>
<td>+3.3</td>
<td>-0.7</td>
<td>-0.5</td>
</tr>
</tbody>
</table>

Measured at 25.15 MHz using D₂O as a solvent. Chemical shifts were calculated from internal dioxane (δ 67.4).
resonance was shifted upfield by 0.3~2.7 ppm (Fig. 3).

The resonance of C₅ of the aminosugar moiety of I-b was assigned to δ 74.7 by consideration of shift trend, which would be shifted at most 3 ppm on substitution of the carbamoyl group either on the C₃- or C₄-OH group. Among remaining three doublet signals (actually two, due to overlapping), the C₄, C₃ and C₂ signals were tentatively assigned to δ 71.5, 61.7 and 61.7 respectively, based on the assumed C₁ substitution. By this assignment, C₄ (α-carbon) was shifted downfield by 3.3 ppm, while C₅ and C₃ (β-carbons) were shifted upfield by 0.7 and 0.4 ppm, respectively. The large shift of C₃ of I-b was explained by the additive β-effect of the amido formation at C₂-N. An additional β-effect of C₂-N substitution was observed at C₁ (−0.6 ppm). C₂ Carbon (α-position) was probably shifted downfield by 4.2 ppm. Alternative assignments of C₁, C₂, and C₃ could not reasonably explain the shift induced by substitution. This corroborated our tentative assignment. The carbon resonances of the aminosugar moiety of I-a in D₂O were also identical to those of I-b.

In conclusion, the position of the carbamoyl group was determined to be on the C₄-OH group. Thus, the structures of components A₁ and A₂ were determined to be I-a and I-b as shown in Fig. 1.

Acknowledgement

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References