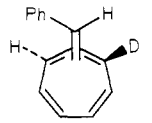
e.g., **4** (X = D) would give one enantiomer while elimination of HBr would give the other, the primary isotope effect should lead to an excess of one enantiomer over the other. Indeed, treatment of the mixture of bromocycloheptatrienes with potassium *tert*-butoxide in THF in the presence of the furan gave optically active adduct **6** (Table I). We take these results as unequivocal evidence for a chiral intermediate as the progenitor of adduct **6**.

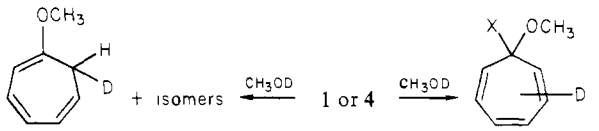
The effect of change in temperature[8] on the specific rotation of the adduct is also recorded in Table I. However, as described in footnote b of Table I, the endo adduct could not be separated from small (and varying) amounts of exo isomer. As a result, the only valid conclusion from these results is that up to 100 °C at least some of the activity persists. This requires that if the allene is competitively racemizing, at least some is bled off before complete racemization occurs.

In principle, chirality might also be used to assign the structure of the precursor to the spirononatriene. Thus, if styrene traps cycloheptatetraene in an allowed ($\pi^2$s + $\pi^8$s) reaction, the mode of attack on the double bond should be as pictured in **7**. Furthermore, solely on the basis of steric arguments, the phenyl ring would have a favored orientation; most likely the one shown. If this is the case, then optically active allene should give an excess of one enatiomeric transition state and the resulting spirononatriene (**5**) should be optically active. Of course, the achiral carbene would give racemic adduct.




7

The optically active bromide (**4**, X = D) was therefore treated with potassium *tert*-butoxide in the presence of styrene at −30 °C (to maximize asymmetric induction) for 5.5 h. The resulting spirononatriene (**5**) showed NO rotation ($\alpha_{obsd}$ 0.002 ± 0.002°; $[\alpha]^{25}_D$ 0.018°; limit of experimental method). This result must be taken as preliminary (a more sterically demanding system needs to be studied) and, admittedly, is negative evidence. Nonetheless, it is the result that would be expected if the carbene is the intermediate being trapped. At this point it should also be mentioned that Kirmse[10] has very recently reported that the intermediate from either **1** or **4** is trapped by alcohols to give **8**. This is the product expected of a carbene but not a strained cyclic allene, which should give **9**.[11]




+ isomers 9

8, X = H or D

**Acknowledgment.** We gratefully acknowledge financial support from the National Science Foundation. We are also indebted to Professor Ronald Breslow of Columbia University for a conversation that led to using chirality as a probe to distinguish between the structures discussed in this manuscript.

**Registry No. 1**, 83527-70-2; **2**, 17476-70-9; (+)-**3**, 83527-68-8; (−)-**3**, 83527-69-9; **4** (X = H), 32743-67-2; **4** (X = D), 83527-67-7; **5**, 50517-62-9; **6**, 83572-17-2; $PhCH{=}CH_2$, 100-42-5; 1,3-diphenylbenzo[*c*]furan, 5471-63-6; 2-bromocycloheptatriene, 3046-02-4; 3-bromocycloheptatriene, 3046-03-5; 7-deuterio-2-bromocycloheptatriene, 83527-71-3; 7-deuterio-3-bromocycloheptatriene, 83527-72-4; potassium methoxide, 865-33-8; potassium *tert*-butoxide, 865-47-4.

(9) Cervinka, O. *Collect. Czech. Chem. Commun.* **1965**, *30*, 1684, 2403.
(10) Kirmse, W.; Loosen, K.; Sluma, H. D. *J. Am. Chem. Soc.* **1981**, *103*, 5935.
(11) Mohanakrishnan, P.; Toyal, S. R.; Vaidyanathaswamy, R.; Devaprabhakara, D. *Tetrahedron Lett.* **1972**, 2871. Bottini, A. T.; Anderson, B. *Ibid.* **1973**, 3321.

# Revision of Assignment of Structure to the Pyrrolodiazepinone Antitumor Antibiotic Sibiromycin

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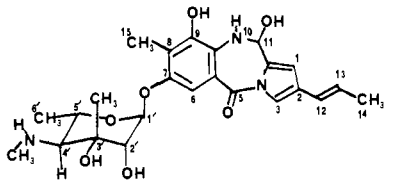
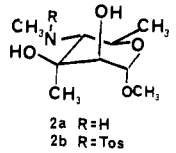
*Received August 9, 1982*

Studies directed toward the synthesis of sibiromycin recently led us to revise the structure of the carbohydrate portion of this antitumor antibiotic.[1] Further comparison of the synthetic material derived from the natural product and examination of the spectroscopic properties of anhydrosibiromycin now allow us to reassign the structure of sibiromycin as the α-L-glycoside **1**.



1




Sibiromycin was obtained from a culture of *Streptosporangium sibiricum* (American Type Culture Collection, original specimen contributed by Gause)[2] grown as outlined by Hurley.[3] Degradation[4] followed by tosylation[1] and isolation by flash chromatography gave material that proved to be levorotatory,[5] whereas compound **2b** (synthetic, from D-glucose) was dextrorotatory. We conclude therefore that natural sibirosamine is L-4,6-dideoxy-3-*C*-methyl-4-(methylamino)mannose. The large negative rotational shift observed by Mesentsev when tetraminecopper(II) sulfate was added to methyl sibirosaminide[4,6] is consistent with this assignment.

We were able to address the question of anomeric configuration (previously assigned β[7]) by comparison of the nuclear Overhauser effects of the axial C-3′ methyl group on the axial C-5′ proton and the anomeric C-1′ proton in anhydrosibiromycin (N-10, C-11 anhydro).[8,9] NOE difference spectroscopy[10,11] (irradiation of C-3′ methyl resonance at δ 1.35) indicated enhancement of the signals for H-2′ (δ 3.87) and H-5′ (δ 3.74) but not for H-1′ (δ 5.74). Therefore we conclude that sibiromycin is an α-glycoside.

Finally, we sought to verify the original designation of the natural product as a C-7 (rather than C-9) glycoside by spectroscopic means.[12] The selectively decoupled $^{13}C$ NMR spectrum

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(1) Parker, K. A.; Babine, R. E. *Tetrahedron Lett.* **1982**, *23*, 1763 and references therein.
(2) The American Type Culture Collection, *Catalog of Strains I*, 13th ed.; Rockville, MD, 1978; p 175, catalog no. ATCC 29053.
(3) Hurley, L. H.; Lasswell, W. L.; Malhotra, R. K.; Parry, R. *Biochemistry* **1979**, *18*, 4225.
(4) Mesentsev, A. S.; Kuljaeva, V. V. *Tetrahedron Lett.* **1973**, 2225.
(5) Optical rotation data: methyl *N*-tosyl α-sibirosaminopyranoside. Synthetic **2b** from D-glucose[1] (*c* 1.56, $CHCl_3$): +62° (589 nm), +73° (578 nm), +81° (546 nm), +138° (436 nm), +211° (365 nm). Degradation product, from sibiromycin (*c* 0.08, $CHCl_3$): −54° (589 nm), −65° (578 nm), −78° (546 nm), −162° (436 nm), −202° (365 nm).
(6) Umezawa, S.; Tsuchija, T.; Tatsuta, K. *Bull. Chem. Soc. Jpn.* **1966**, *39*, 1235.
(7) Mesentsev, A. S.; Kuljaeva, V. V.; Rubasheva, L. M. *J. Antibiot.* **1974**, *27*, 866.
(8) The studies described below were carried out on anhydrosibiromycin, which may be obtained as very clean (but noncrystalline solid) material; sibiromycin itself is difficult to purify. H-4′ and H-5′ must be axial because of the large coupling (*J* = 9.6 Hz) between them. Also the C-3′ methyl must be trans to the C-4′ proton because methyl *N*-tosyl-α-D-sibirosaminide is prepared by 3,4-epoxide opening with the sodium salt of *N*-methyltosylamide.[1] Therefore the C-3′ methyl group is necessarily axial in anhydrosibiromycin.
(9) Refer to Table I for complete $^1H$ NMR data.
(10) Hall, L. D.; Sanders, J. K. M. *J. Am. Chem. Soc.* **1980**, *102*, 5703.
(11) All spectra discussed in this paper were measured in a Bruker WM 250 NMR spectrometer.
(12) We felt it was necessary to check the previous assignment, which was based on the failure to observe reaction of the phenolic hydroxyl of anhydrosibiromycin with diazomethane.

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Table I. Assignment of Resonances in the $^1H$ NMR Spectrum of Anhydrosibiromycin (250 MHz, $CDCl_3$ containing $D_2O$)

| chem shift, ppm | multiplicity (coupl const) | integral | assignment | basis for assignment |
|---|---|---|---|---|
| 1.35 | d ($J$ = 6.0 Hz) | 3 H | H-6′ | *a* |
| 1.46 | s | 3 H | C-3′$CH_3$ | *a* |
| 1.91 | d ($J$ = 5.8 Hz) | 3 H | H-14 | *a* |
| 2.27 | s | 3 H | C-8$CH_3$ | *a* |
| 2.56 | d ($J$ = 9.9 Hz) | 1 H | H-4′ | irradiation at δ 3.74 results in collapse to singlet |
| 2.62 | s | 3 H | N-$CH_3$ | *a* |
| 3.74 | dq ($J$ = 9.9, 6.0 Hz) | 1 H | H-5′ | irradiation at δ 1.35 results in collapse to doublet ($J$ = 9.9 Hz) |
| 3.87 | d ($J$ = 1.1 Hz) | 1 H | H-2′ | *a* |
| 5.74 | d ($J$ = 1.1 Hz) | 1 H | H-1′ | *a* |
| 6.21 | dq ($J$ = 16.5, 5.8 Hz) | 1 H | H-13 | irradiation at δ 1.91 results in collapse to doublet ($J$ = 16.5 Hz) |
| 6.37 | d ($J$ = 16.5 Hz) | 1 H | H-12 | coupling to H-13 |
| 7.14 | d ($J$ = 2.0 Hz) | 1 H | H-1 | *a* |
| 7.94 | s | 1 H | H-6 | *a* |
| 8.18 | d ($J$ = 2.0 Hz) | 1 H | H-3 | *a* |
| 8.28 | s | 1 H | H-11 | *a* |

[a] Multiplicity and chemical shift.

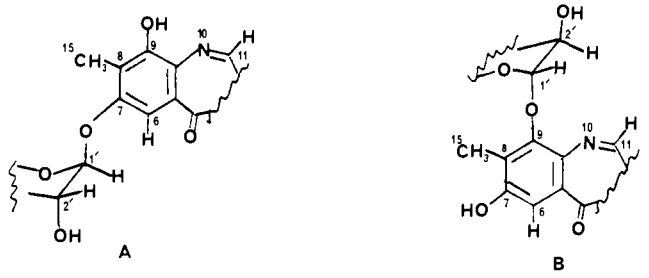

**Figure 1.** Partial structures of the C-7 (A) and C-9 (B) glycosides of anhydrosibiromycinone.

(C-8 methyl protons irradiated) of anhydrosibiromycin (see Figure 1) would be expected to exhibit a doublet of doublets for the C-7 absorption (two-bond coupling to the C-6 proton and three-bond coupling to the C-1′ proton) and a singlet for the C-9 absorption (when exchange of the phenolic proton is fast). On the other hand, the single frequency decoupled $^{13}C$ NMR spectrum of the C-9 glycoside (see Figure 1) should contain a doublet for the C-7 absorption (two-bond coupling to the C-6 proton, no coupling to the phenolic proton when exchange is fast) and a doublet for the C-9 absorption (three-bond coupling to the C-1′ proton).

Examination of the off-resonance decoupled $^{13}C$ NMR spectrum[11] of anhydrosibiromycin (Table II) allowed us to assign the singlets at 154.87 and 154.03 ppm to the two aromatic carbons bearing oxygen.[13] Measurement of the proton noise decoupled spectrum of a sample to which a small amount of a 1:1 mixture of $H_2O$ and $D_2O$ had been added also showed only two singlets (154.87 and 153.95 ppm),[14] thereby indicating that exchange of the phenolic proton was rapid.

Table II. Assignment of Resonances in the $^{13}C$ NMR Spectrum ($CD_3SOCD_3$ Solvent) of Anhydrosibiromycin

| chemical shift[a] | multiplicity | assignment |
|---|---|---|
| 160.42 | s | C-5[c] |
| 154.87 | s | C-7[d] |
| 154.03 | s | C-9[d] |
| 143.15 | d | C-11[e] |
| 128.78 | s | na[f] |
| 128.15 | s | na |
| 128.11 | s | na |
| 127.72 | d | C-12 or C-13[e] |
| 122.31 | d | C-3[e] |
| 121.99 | d | C-12 or C-13[e] |
| 120.03 | d | C-1[e] |
| 119.06 | s | na |
| 118.29 | s | na |
| 106.46 | d | C-6[e] |
| 99.09 | d | C-1′[e] |
| 73.54 | d | C-5′[g] |
| 71.71 | s | C-3′[g] |
| 69.92 | d | C-2′[g] |
| 66.15 | d | C-4′[c] |
| 37.96 | q | N-$CH_3$[c] |
| 19.84 | q | na |
| 19.15 | q | na |
| 18.16 | q | na |
| 9.70 | q | na |

[a] ppm relative to $Me_4Si$ (0 ppm). [b] This multiplicity arises from one bond coupling as determined in a single frequency off-resonance decoupled spectrum. [c] Assigned on chemical shift considerations. [d] Assigned by the selective frequency decoupling experiment; see text. [e] Assigned by a Birdsall "crossover" experiment (see: Birdsall, B.; Birdsall, N. J. M.; Feeney, J. *J. Chem. Soc., Chem. Commun.* 1972, 316. [f] na = not assigned. [g] Assigned on the basis of SFORD multiplicity.

Selective decoupling by low-power irradiation[15] at a frequency corresponding to a proton frequency of δ 2.27 (aromatic methyl) resulted in a spectrum in which the carbon absorption at 154.87 ppm was observed as an apparent triplet ($J$ = 3 Hz) and the resonance at 154.03 ppm was observed as a singlet. If the apparent triplet is in fact a doublet of doublets, this pattern is consistent with anhydro **1** (A, Figure 1) but not with a structure that is a C-9 glycoside (B, Figure 1). Single frequency decoupling at higher power[15] resulted in a spectrum in which the 154.87 ppm signal appeared as a doublet; under these conditions, one of the two protons coupled to this carbon (the C-1′ proton) is being decoupled along with the aromatic methyl protons. Therefore, the apparent triplet observed at lower decoupling power is indeed a doublet of doublets and corresponds to C-7 in anhydrosibiromycin (anhydro **1**); the singlet corresponds to C-9.

With the exception of the configuration of the C-11 carbon, then, the structure of sibiromycin can be completely assigned. Superposition of a CPK molecular model of this revised structure (**1**) on a model of DNA[16] (alkylation of a guanine N-2 by the sibiromycin carbinolamine) revealed no destabilizing interactions.

**Acknowledgment.** This work was supported by the American Cancer Society (CH222), to whom we are most grateful. K.A.P. also acknowledges assistance in the form of a Teacher–Scholar Award from the Camille and Henry Dreyfus Foundation. The NMR studies were accomplished on Brown University's WM 250 spectrometer, which was purchased with funds from the National Science Foundation and from Montedison, S.P.A. (Milan, Italy). We thank Professor David E. Cane, Professor Ronald G. Lawler, Dr. James Van Epp, and Tzyy-Chyau Liang for consultations that were most helpful in the design and execution of the $^{13}C$ NMR experiment.

(13) Hurley[3] assigned some of the signals in the $^{13}C$ NMR spectrum of diacetyl sibiromycinone. We have been able to assign most but not all of the signals in the $^{13}C$ NMR spectrum of anhydrosibiromycin.

(14) Measured relative to the 160.42 ppm peak. We believe that the upfield shift, although small, is significant. Upfield shifts when $^{13}COH$ is completely exchanged to $^{13}COD$ are approximately 0.2 ppm; see: Gorst-Allman, C. P.; Pachler, K. G. R.; Steyn, P. S.; Wessels, P. L.; Scott, D. B. *J. Chem. Soc., Perkin Trans. 1* **1977**, 2181.

(15) Low decoupling power, 0.013 W (DP = 12 L); high decoupling power, 0.13 W (DP = 2 L).

(16) Petrusek, R. L.; Anderson, G. L.; Garner, T. F.; Fannin, Q. L.; Kaplan, D. J.; Zimmer, S. G.; Hurley, L. H. *Biochemistry* **1981**, *20*, 1111 and references therein.