of 9-O-demethylhomolycorine (5 mg) which had been heated at 60 °C for 17 h in D_2O–CD_3OD (1:1) (0.5 mL).

Methylation of 9-O-Demethylhomolycorine. To a solution O-demethylhomolycorine (2 mg) in CH_3OH–Et_2O (1:1) (1 mL) was added 3 separate aliquots of 2 mL of ethereal CH_2N_2 solution over a period of 3 days. Removal of the exces CH_2N_2 and solvent left a solid residue (2 mg) which on crystallization from benzene gave homolycorine, mp 170–172 °C. The TLC and mass spectrum of this sample was identical in every respect with TLC and mass spectrum of authentic homolycorine.

Extraction of the Alkaloids from *C. scabrum*. The dry powdered plant (1.5 kg) was processed as described above for the extraction of *C. defixum*. After removal of the lycorine (0.52 g, 0.035%) by filtration, a crude alkaloid fraction (6.4 g) remained.

Chromatography of Crude Alkaloid Fraction from *C. scabrum*. The crude alkaloid extract (6.4 g) was preabsorbed on Al_2O_3 (neutral, grade II) in benzene. Fractions (150 mL) were collected by using the following linear solvent gradients. A: benzene–5% EtAc(1.5 L)–EtAc (1.5 L). B: EtOAc (1.5 L)–MeOH (8:2, v/v, 1.5 L). C: EtOAc–MeOH (1:1, 1 L). A total of 43 fractions were collected and combined as follows based upon analysis by TLC on silica gel with the solvent system ($CHCl_3$–EtOAc–MeOH (2:2:6). Fractions 1–12, unidentified alkaloid (s) (1.3 g); 13–24, crinamine (2.0 g); 25–26, mixture of lycorine and 6-hydroxycrinamine (1.1 g); 27–30, 6-hydroxycrinamine (0.3 g); 31–43 mixture of four unidentified alkaloids (1.4 g).

The alkaloids crinamine and 6-hydroxycrinamine were identified by IR, NMR, and MS spectral comparisons with the spectra of authentic samples and by the preparation of the following crystalline derivatives O-acetylcrinamine, mp 160–161 °C, 6-hydroxycrinamine methiodide, mp 170–172 °C, crinamine picrate, mp 271–271 °C, and 6-hydroxycrinamine methopicrate, mp 146 °C.

Extraction of the Alkaloids *C. latifolium*. Utilizing the same procedures as described for *C. defixium* and *C. scabrum*, the leaves (12.3 kg) and bulbs (700 g) of *C. latifolium* were extracted separately. The leaves afforded lycorine (1.8 g) and a crude alkaloid fraction (16.8 g) while the bulbs similarly gave lycorine (0.28 g) and a crude alkaloid residue (2.6 g). A TLC examination of the two crude alkaloid fractions indicated they contained the same alkaloids.

Chromatographic Separation of the Alkaloids of *C. latifolium*. A portion of the crude alkaloid fraction (10 g) was preabsorbed on Al_2O_3 (50 g) and placed on the top of a column containing Al_2O_3 (1 kg neutral, Activity II). The solvents used for linear gradient elution were the same as used for the chromatography of the *C. scabrum* alkaloids. Individual fractions (150 mL) were collected and combined as indicated below on the basis of their analysis by TLC on silica gel in $CHCl_3$–EtOAc–MeOH (2:2:6). Fraction 1–19, nonalkaloid material (0.9 g); 20–49, noncrystalline mixture of two bases (0.6 g); 50–59, crude hippeastrine and three other uncharacterized alkaloids (1.15 g). This fraction on standing gave crystalline material which on crystallization gave hippeastrine (50 mg), mp 215–217 °C, identified by suitable spectral comparisons with an authentic sample. Fraction 60–69, mixture of alkaloids (0.5 g), which onstanding deposited lycorine (12 mg); 70–75, unidentified alkaloids (0.3 g); 76–104, noncrystalline mixture of alkaloids (1.6 g).

Acknowledgment. We are indebted to the Egyptian Government for a fellowship to A.A. for a leave of absence from the Department of Pharmocognosy, Alexandria University, and to the National Institutes of Health for a predoctoral traineeship to D.C. We are most grateful to Dr. C. A. Evans, JEOL, for the ^{13}C-INEPT and ^1H NOE experiments on 9-O-demethylhomolycorine.

Registry No. 1, 13255-05-5; 3, 477-17-8; 4, 477-20-3; 7, 6879-81-8; lycorine, 476-28-8; crinamine, 639-41-8; 6-hydroxycrinamine, 545-66-4.

Chemistry of Acronycine X. Oligomers of Noracronycine

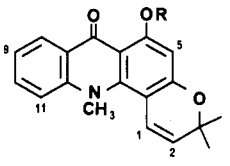
Shinji Funayama,† Geoffrey A. Cordell,*† Ronald D. Macfarlane,‡ and Catherine J. McNeal‡

Program for Collaborative Research in the Pharmaceutical Sciences, College of Pharmacy, University of Illinois at Chicago, Chicago, Illinois 60612, and Department of Chemistry, Texas A&M University, College Station, Texas 77843

Received July 25, 1984

Treatment of noracronycine (3) with methanolic hydrochloric acid as well as yielding the dimers AB-1 (6) and AB-2 (7) and the trimer AB-3 (8) also afforded AB-5. Plasma desorption mass spectrometry indicated this product to be a mixture of tetrameric and pentameric species which were separated and characterized. The pentamer AB-5A was deduced through spectroscopic interpretation to be the all-linear isomer 10 and the tetramer, AB-5B, was shown to have a partially rearranged linear–angular–angular–angular structure (11). The dihydro derivative of AB-5B was synthesized through the union of dihydro AB-1 (12) and AB-2 (7).

Acronycine (1), an alkaloid isolated from the bark of



	R
1	CH_3
3	H

Acronychia baueri Schott (Rutaceae),[1-4] possesses the broadest spectrum of in vivo antineoplastic activity of any alkaloid thus far tested.[5,6] In spite of this, very little is known of its chemistry or mode of action.[3,4]

Acronycine (1) is an acridone alkaloid with an additional hemiterpene unit attached at C-4 at the parent nucleus and cyclized to form a pyran ring. Initially there was a question as to whether acronycine had a linear or angular structure, and the presently accepted structure was de-

†University of Illinois.
‡Texas A&M University.

(1) Hughes, G. K.; Lahey, F. N.; Price, J. R.; Webb, L. J. *Nature (London)* **1948**, *162*, 223.
(2) Lahey, F. N.; Thomas, W. C. *Aust. J. Sci. Res.* **1949**, *2A*, 423.
(3) Brown, R. D.; Drummond, L. J.; Lahey, F. N.; Thomas, W. C. *Aust. J. Sci. Res.* **1949**, *2A*, 622.
(4) Drummond, L. J.; Lahey, F. N. *Aust. J. Sci. Res.* **1949**, *2A*, 630.
(5) Svoboda, G. H.; Poore, G. A.; Simpson, P. J.; Boder, G. B. *J. Pharm. Sci.* **1966**, *55*, 758.
(6) Svoboda, G. H. *Lloydia* **1966**, *29*, 206.

0022-3263/85/1950-1737$01.50/0

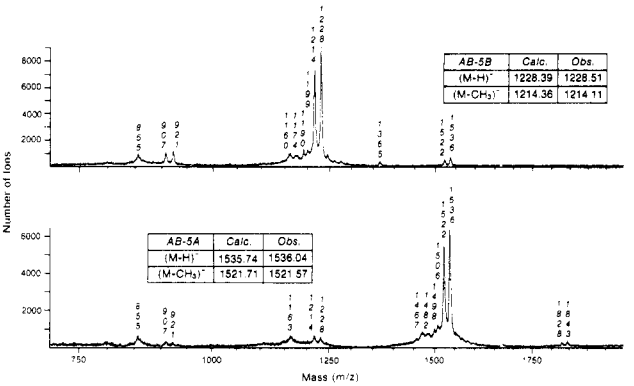
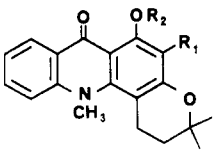


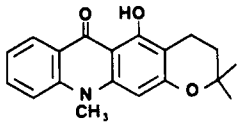
Figure 1.

duced from chemical[7] and spectroscopic evidence.[8] An X-ray crystallographic analysis of 5-bromo-1,2-dihydroacronycine (**2**) confirmed that the parent compound had the angular array.[9]



	R_1	R_2
2	Br	CH_3
4	H	H

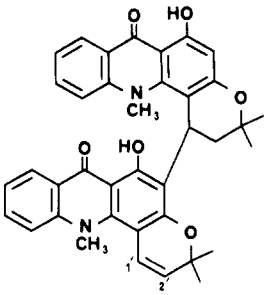
In the course of our work, we have reported on ^1H and ^{13}C NMR studies of acronycine (**1**) and simple derivatives,[10,11] on the dimerization and trimerization of noracronycine (**3**),[12,13] on the unexpected reactivity of dihydronoracronycine (**4**),[14] on the selective synthesis of dimers and trimers of noracronycine (**3**) and related compounds,[15] and on the facile conversion of dihydronoracronycine (**4**) to dihydroisonoracronycine (**5**).[16] We report



5

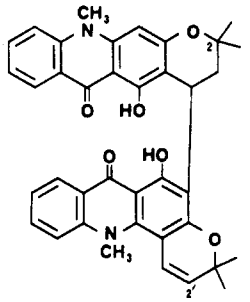
here on the minor products formed when noracronycine (**3**) is treated with acid.

Heating a solution of acronycine (**1**) in methanolic 10 N hydrochloric acid (2.5:1, v/v) afforded an orange-yellow powder. The products other than noracronycine and acronycine were identified as AB-1 (**6**),[12] AB-2 (**7**),[12] AB-3



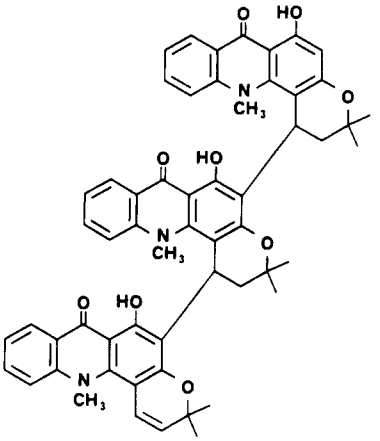
6

12 1′,2′-H_2



7

(**8**),[13] and so on, according to their R_f value. The diversity



8

of reaction products was formed in better yeild from noracronycine (**3**), and, subsequently, we have used noracronycine (**3**) as the starting material,[12] synthesized according to the method of Brown and co-workers.[3]

Noracronycine (**3**) possesses an angular arrangement of rings, and in one of the dimers, AB-1 (**6**), and the trimer AB-3 (**8**) this molecular array was maintained. However, in the dimer AB-2 (**7**) one of the chromene rings was rearranged to produce a linear four-ring system in the upper unit of the molecule. These two skeletons could be differentiated by establishing the chemical shift of the aromatic doublet peri to the *N*-methyl group. In AB-1 (**6**) or AB-3 (**8**) these signals appear about 0.5 ppm to higher field than those in the lower unit. In AB-2 (**7**) this signal appeared in the same region as those of the lower units.[15] During the course of this work essentially complete proton assignments were made for the dimers **6** and **7** and the trimer **8**.[15]

Discussion

AB-5 was obtained from the reaction mixture after AB-1

(7) McDonald, P. L.; Robertson, A. V. *Aust. J. Chem.* **1966**, *19*, 275.
(8) Govindachari, T. R.; Pai, B. R.; Subramaniam, P. S. *Tetrahedron* **1966**, *22*, 3245.
(9) Gougoutas, J. Z.; Kaski, B. A. *Acta Crystallogr., Sect. B* **1970**, *26B*, 853.
(10) Funayama, S.; Borris, R. P.; Cordell, G. A. *J. Nat. Prod.* **1983**, *46*, 391.
(11) Funayama, S.; Cordell, G. A. *Planta med.*, **1984**, *50*, 121.
(12) Funayama, S.; Cordell, G. A.; Wagner, H.; Lotter, H. L. *J. Nat. Prod.* **1984**, *47*, 143.
(13) Funayama, S.; Cordell, G. A. *Planta Med.* **1983**, *48*, 263.
(14) Funayama, S.; Cordell, G. A. *Heterocycles* **1983**, *20*, 2379.
(15) Funayama, S.; Cordell, G. A. *J. Nat. Prod.*, in press.
(16) Funayama, S.; Cordell, G. A. *J. Nat. Prod.*, submitted for publication.

(6), AB-2 (7), and AB-3 (8) had been isolated. ^{1}H NMR analysis of AB-5 indicated the presence of nine phenolic hydrogen-bonded protons and nine *N*-methyl signals. However, two aromatic singlets which could be assigned to H-5 of noracronycine units and two pairs of doublets (H-1 and H-2) were also observed. This suggested that AB-5 was a mixture, and the negative ion ^{252}Cf plasma desorption mass spectrum indicated that AB-5 was comprised of a pentamer and tetramer of noracronycine. Preparative TLC afforded two components, which were identified as AB-5A and AB-5B according to their R_f value.

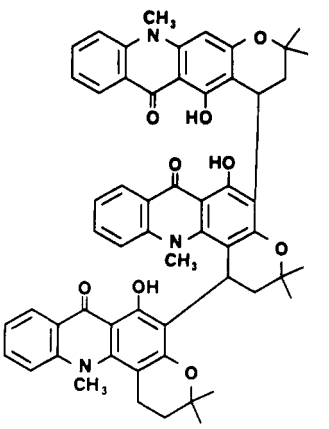
The negative ion spectra of AB-5A and AB-5B are shown in Figure 1. Both compounds produced prominent peaks corresponding to $(M - H)^-$ and $(M - CH_3)^-$. The spectrum of AB-5A showed that the sample is almost entirely comprised of the pentamer while the spectrum of AB-5B exhibited peaks due almost entirely to the tetramer moiety. The spectra of each compound also contained peaks of lower intensity corresponding to oligomers containing one more and one less noracronycine residue. In addition to loss of the methyl group, four unidentified fragment ions were observed in both spectra within m/z 100 of the molecular ion peak.

The positive ion spectra of AB-5A and AB-5B contained peaks due to $(M + H)^+$ and $(M + Na)^+$ of the pentamer and tetramer, respectively. There was also evidence for the presence of higher or lower order oligomers as had been observed in the negative ion spectra. The most intense peaks in the spectra were fragment ions formed by loss of one noracronycine residue. Since the mass of this fragment ion species is indistinguishable from an oligomer containing one less noracronycine unit, this complicated the interpretation of the AB-5A positive ion spectrum. However, the negative ion spectra clearly showed which of the oligomers was the major component of the sample. With the molecular weights of the products established, attention was focused on the structure determination.

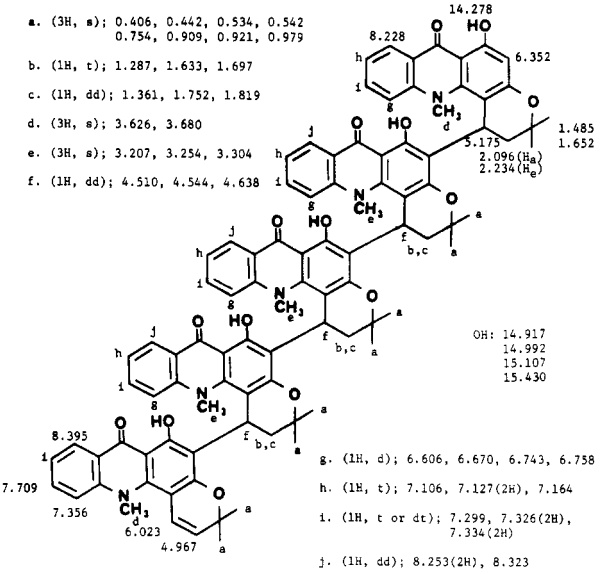
The UV and IR spectra of AB-5A were typical of those of acridone alkaloids.[17] In the ^{1}H NMR spectrum of AB-5A, five geminal methyl resonances, five *N*-methyl signals, five sets of aromatic signals, and five hydrogen-bonded phenolic OH protons were observed. Four aromatic doublets appeared about 0.5 ppm higher than those of AB-2 (7) and noracronycine (3), as had been observed in the ^{1}H NMR spectra of AB-1 (6) and AB-3 (8).[12,13,15] Four sets of geminal methyl resonances were also shifted by 0.5–1.0 ppm to higher field. Two sets of such signals had been observed in the ^{1}H NMR spectrum of AB-3 (8) and one set in the spectrum of AB-1 (6).[15]

On the other hand, three higher field shifted *N*-methyl signals were observed. Such signals had not been seen in the ^{1}H NMR spectrum of AB-1 (6), although one was noted in the ^{1}H NMR spectrum of AB-3 (8) and assigned to the *N*-methyl of the middle unit.[15] No such higher field shifted *N*-methyl signals were observed in the ^{1}H spectrum of the linear–angular–angular trimer 9. Consequently, it was considered that such signals would be diagnostic when a noracronycine (3) unit was inserted between two angular noracronycine (3) units.[15]

When noracronycine (3) was treated with methanolic hydrochloric acid at room temperature, no rearrangement of the upper unit was observed and AB-5A was formed together with AB-1 (6) and AB-3 (8), without forming AB-2 (7) or AB-5B. From these accumulated data, AB-5A was concluded to be constructed from five angular noracronycine units as shown in structure 10. ^{1}H NMR



9



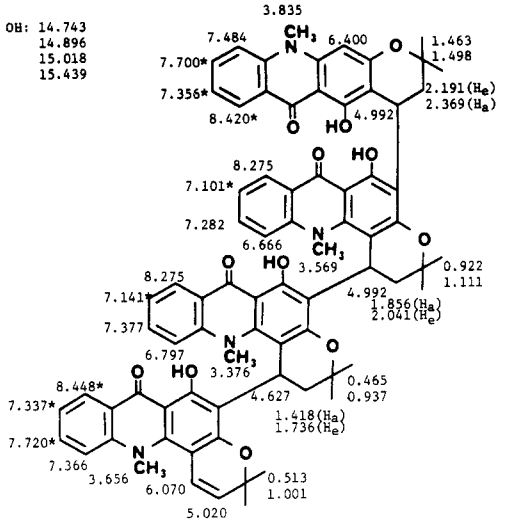
10

assignments for AB-5A were made principally by comparison with data for noracronycine (3), AB-1 (6), and AB-3 (8).[11,15]

The UV and IR specta of the tetramer AB-5B were typical of those of acridone alkaloids.[17] The ^{1}H NMR spectrum firmly established that it was indeed a tetramer, in that four sets of coupled aromatic signals and three sets of aliphatic pyran signals, as well as one aromatic singlet and a pair of doublets, were observed.

Closer examination of the ^{1}H NMR spectrum, and the observation that this compound was not formed by treating noracronycine (3) with methanolic hydrochloric acid at room temperature, suggested that AB-5B was not comprised only of angular units like AB-1 (6), AB-3 (8), and AB-5A (10). For if this were the case, three aromatic doublets should be evident at about 0.5 ppm higher than those of AB-2 (7) and noracronycine (3),[15] however, only two such signals were observed. Similarly, two higher field shifted *N*-methyl signals would be expected from the all-angular tetrameric structure,[15] but only one such signal was evident. From all these observations, AB-5B was envisaged as a tetramer with the linear–angular–angular–angular system 11 and an attempt to confirm this was made through the synthesis of the corresponding dihydroderivative.

(17) Reisch, J.; Szendrei, K.; Minker, E.; Novak, I. *Die Pharmazie* **1972**, *27*, 208.



11

13 $1''', 2'''\text{-}H_2$

After it was established that AB-2 (7) would not undergo self-condensation, AB-2 (7) and dihydro-AB-1 (12) were mixed in the ratio 1:5 and stirred in methanolic hydrochloric acid under N_2 at room temperature. By mixing these compounds in this ratio, it was anticipated that a molecule of AB-2 (7) would preferentially react with a molecule of 12. In the event, dihydro-AB-5B (13) was obtained and its identification was achieved by direct comparison with dihydro-AB-5B (13) produced through the catalytic (Pd/C) hydrogenation of AB-5B (11). Preliminary ^1H NMR assignments for AB-5B are shown.

Some of the NMR assignments of AB-5B were confirmed by NOE enhancement experiments. When the singlet at δ 6.400 (C_{12}–H) was irradiated, a 15% NOE was observed in the three-proton singlet at δ 3.835 (N_{11}–CH_3) and when the δ 3.835 signal was irradiated, a 6% NOE at δ 7.484 (C_{10}–H) and a 12% NOE at δ 6.400 were observed. Irradiation at δ 4.627 caused a 4% NOE at δ 3.376. Because the shielded *N*-methyl signal could be assigned to the unit between two angular type noracronycine (3) moieties, these signals were assigned to H-1″ and $N_{12''}$–CH_3, respectively. In addition, when the signal at δ 3.376 was irradiated, an 11% NOE was evident at δ 6.797. This indicated that a set of four aromatic protons (δ 6.797, 7.377, 7.141 (or 7.101), and 8.275) could be assigned to the third unit. Finally, through irradiation at δ 4.992 (C_4– and C_1–H) which caused a 3% NOE at δ 3.569, the latter signal could be assigned to $N_{12'}$–CH_3.

There are very few examples of tetrameric and pentameric alkaloids; the quadrigemines from *Hodgkinsonia frutescens*[18] and psychotridine from *Psychotria beccarioides*[19] appear to be the only previous examples, respectively. We therefore believe the compounds described here to be the only other tetramers or pentamers of alkaloids to be obtained synthetically or from natural sources.

Experimental Section

Preparation of Acronycine (1), Noracronycine (3), and Dihydronoracronycine (4). The preparation and properties of these compounds were described previously.[10]

Formation and Isolation of AB-1 (6), AB-2 (7), AB-3 (8), AB-5A (10), and AB-5B (11). The formation, isolation, and properties of AB-1 (6), AB-2 (7), and AB-3 (8) have been described previously.[12,13] Repeated preparative TLC of the polar products from the reaction of noracronycine with methanolic hydrochloric acid afforded AB-5A (10, 3.0 mg) and AB-5B (11, 4.1 mg).

AB-5A (10) crystallized from $CHCl_3$ as yellow rhombic cubes; mp 303 °C dec; IR (KBr) ν_{max} 1627, 1588, 1559, 1498, 1484, 1450, 1438, 1326, 1265, 1145 cm^{-1}; UV ($CHCl_3$) λ_{max} 257, 283, 307, 347, 417 nm; ^1H NMR (360 MHz, $CDCl_3$) δ 0.406, 0.442, 0.534, 0.542, 0.754, 0.909, 0.921 and 0.979 (s, each 3 H, 13′-, 13″-, 13‴-, 13⁗-, 14′-, 14″-, 14‴-, 14⁗-CH_3), 1.287 (t, J = 12.6 Hz, 1 H), 1.361 (dd, J = 6.9, 12.4 Hz, 1 H), 1.485 (s, 3 H, 13- or 14-CH_3), 1.633 (t, J = 14.3 Hz, 1 H) and 1.697 (t, J = 11.8 Hz, 1 H) ($C_{2'}$–, $C_{2''}$–, and $C_{2'''}$–H_a), 1.652 (s, 3 H, 14- or 13-CH_3), 1.752 (dd, J = 7.0, 12.9 Hz, 1 H) and 1.819 (dd, J = 6.7, 13.0 Hz, 1 H) ($C_{2'}$–, $C_{2''}$–, and $C_{2'''}H_e$), 2.096 (t, J = 12.5 Hz, 1 H, C_2–H_a), 2.234 (dd, J = 7.0, 13.0 Hz, 1 H, C_2–H_e), 3.207, 3.254, and 3.304 (s, each 3 H, $N_{12'}$–, $N_{12''}$–, and $N_{12'''}$–CH_3), 3.626 and 3.680 (s, each 3 H, N_{12}– and $N_{12'''}$–CH_3), 4.510 (dd, J = 6.7, 12.1 Hz, 1 H), 4.544 (dd, J = 6.6, 12.0 Hz, 1 H) and 4.638 (dd, J = 6.7, 11.8 Hz, 1 H, $C_{1'}$–, $C_{1''}$–, and $C_{1'''}$–H), 4.967 (d, J = 9.6 Hz, 1 H, $C_{2''''}$–H), 5.175 (dd, J = 6.9, 11.6 Hz, C_1–H), 6.023 (d, J = 9.6 Hz, 1 H, $C_{1''''}$–H), 6.352 (s, 1 H, C_5–H), 6.606 (d, J = 8.4 Hz, 1 H), 6.670 (d, J = 8.5 Hz, 1 H), 6.743 (d, J = 8.5 Hz, 1 H), and 6.758 (d, J = 8.4 Hz, 1 H) (C_{11}–, $C_{11'}$–, $C_{11''}$–, $C_{11'''}$–H), 7.106 (t, J = 7.7 Hz, 1 H), 7.127 (t, J = 7.4 Hz, 2 H) and 7.164 (t, J = 7.8 Hz, 1 H) (C_9–, $C_{9'}$–, $C_{9''}$–, and $C_{9'''}$–H), 7.299 (dt, J = 1.4, 8.4 Hz, 1 H), 7.326 (t, J = 8.7 Hz, 2 H) and 7.334 (t, J = 8.3 Hz, 2 H) (C_{10}–, $C_{10'}$–, $C_{10''}$–, $C_{10'''}$–, and $C_{9''''}$–H), 7.356 (d, J = 7.9 Hz, 1 H, $C_{11''''}$–H), 7.709 (dt, J = 1.1, 7.7 Hz, 1 H, $C_{10''''}$–H), 8.228 (dd, J = 1.3, 8.4 Hz, 1 H, C_8–H), 8.253 (dd, J = 1.5, 8.5 Hz, 2 H) and 8.323 (dd, J = 1.0, 8.2 Hz, 1 H) ($C_{8'}$–, $C_{8''}$–, and $C_{8'''}$–H), 8.395 (dd, J = 1.0, 8.2 Hz, 1 H, $C_{8''''}$–H), 14.278 (s, C_6–OH, 1 H), 14.917, 14.992, 15.107 and 15.430 (s, each 1 H) ($C_{6'}$, $C_{6''}$, $C_{6'''}$, and $C_{6''''}$–OH); MS, see Figure 1.

AB-5B (11) crystallized from $CHCl_3$ as fine yellow rhomboids; mp 262–267 °C; IR (KBr) ν_{max} 1627, 1588, 1559, 1498, 1448, 1327, 1264, 1186, 1146, 1122 cm^{-1}; UV ($CHCl_3$) λ_{max} 225, 285, 304, 347 (sh), 413 nm; ^1H NMR (360 MHz, $CDCl_3$) δ 0.465 (s, 3 H, 13″- or 14″-CH_3), 0.513 (s, 3 H, 13‴- or 14‴-CH_3), 0.922 (s, 3 H, 13′- or 14′-CH_3), 0.937 (s, 3 H, 14″- or 13″-CH_3), 1.001 (s, 3 H, 14‴- or 13‴-CH_3), 1.111 (s, 3 H, 14′- or 13′-CH_3), 1.418 (t, J = 11.9 Hz, 1 H, $C_{2''}$-Ha), 1.463 (s, 3 H, 13- or 14-CH_3), 1.498 (s, 3 H, 14- or 13-CH_3), 1.736 (dd, J = 6.9, 12.4 Hz, 1 H, $C_{2''}$–H_e), 1.856 (t, J = 12.2 Hz, 1 H, $C_{2'}$–H_a), 2.041 (dd, J = 6.9, 12.8 Hz, 1 H, C_2–H_e), 2.191 (dd, J = 7.6, 13.2 Hz, 1H, C_2–H_e), 2.369 (t, J = 12.5 Hz, 1 H, C_2–H_a), 3.376 (s, 3 H, $N_{12''}$–CH_3), 3.569 (s, 3 H, $N_{12'}$–CH_3), 3.656 (s, 3 H, $N_{12'''}$–CH_3), 3.835 (s, 3 H, N_{11}-CH_3), 4.627 (dd, J = 6.7, 11.8 Hz, 1 H, $C_{1''}$–H), 4.992 (dd, J = 7.4, 11.6 Hz, 2 H, C_4– and $C_{1'}$–H), 5.020 (d, J = 9.5 Hz, 1 H, $C_{2'''}$–H), 6.070 (d, J = 9.5 Hz, 1 H, $C_{1'''}$–H), 6.400 (s, 1 H, C_{12}–H), 6.666 (d, J = 8.4 Hz, 1 H, $C_{11''}$–H), 6.797 (d, J = 8.6 Hz, 1 H, $C_{11''}$–H), 7.101 (t, J = 7.4 Hz, 1 H, $C_{9''}$– or $C_{9'''}$–H), 7.141 (dd, J = 6.2, 7.7 Hz, 1 H, $C_{9'''}$– or $C_{9''}$–H), 7.282 (dt, J = 1.2, 8.3 Hz, 1 H, $C_{10''}$–H), 7.337 (t, J = 7.1 Hz, 1 H, C_8– or $C_{9''''}$–H), 7.356 (t, J = 7.0 Hz, 1 H, $C_{9''''}$– or C_8–H), 7.377 (d, J = 1.2, 7.5 Hz, $C_{10''}$–H), 7.366 (d, J = 7.8 Hz, 1 H, $C_{11'''}$–H), 7.484 (d, J = 8.8 Hz, 1 H, C_{10}–H), 7.700 (dt, J = 1.4, 8.1 Hz, 1 H, C_9– or $C_{10'''}$–H), 7.720 (ddd, J = 1.3, 7.9, 8.0 Hz, 1 H, $C_{10'''}$– or C_9–H), 8.275 (dd, J = 1.0, 7.6 Hz, 2 H, $C_{8'}$– and $C_{8''}$–H), 8.448 (dd, J = 1.4, 8.5 Hz, 1 H, $C_{8'''}$–H), 14.743, 14.896, 15.018, and 15.439 (s, each 1 H) ($C_{5'}$-, $C_{6'}$-, $C_{6''}$-, and $C_{6'''}$-OH). MS, see Figure 1.

Hydrogenation of AB-5B (11). AB-5B (11, 0.5 mg) was dissolved in EtOAc (3.0 mL), 10% Pd/C (0.5 mg) was added, H_2 gas was introduced, and the mixture was stirred at room temperature for 24 h. The yellow powder obtained after filtration and evaporation and was purified by preparative TLC on silica gel eluting with benzene–EtOAc (9:1) to afford dihydro AB-5B (13, 0.4 mg).

Reaction of AB-2 (7) with Methanolic Hydrochloric Acid at Room Temperature. AB-2 (7, 1.8 mg) was dissolved in methanolic 10 N aqueous hydrochloric acid (2.5:1, v/v, 3.0 mL) and the mixture sitrred under N_2 at room temperature for 24 h. The reaction mixture was worked up in the usual way to afford unreacted AB-2 (7) as the only detectable product.

Preparation of Dihydro-AB-1 (12). The preparation and physical and spectroscopic properties of dihydro-AB-1 (12) were described previously.[12,15]

(18) Parry, K. P.; Smith, G. F. *J. Chem. Soc., Perkin Trans. 1* **1978**, 1671.

(19) Hart, N. K.; Johns, S. R.; Lamberton, L. A.; Summons, R. E. *Aust. J. Chem.* **1974**, *27*, 639.

Coupling of AB-2 (7) and Dihydro-AB-1 (12). AB-2 (7, 1.2 mg) and dihydro-AB-1 (12, 2.7 mg) were dissolved in methanolic 10 N aqueous hydrochloric acid (2.5:1, v/v, 2.0 mL) and the mixture was stirred under N_2 at room temperature. After 3 days the reaction mixture was worked up in the usual way to afford on orange-yellow powder (3.6 mg). TLC analysis on silica revealed the presence of dihydro-AB-5B (13), unreacted AB-2 (7), dihydro-AB-1 (12), and other minor products.

Reaction of Noracronycine (3) with Methanolic Hydrochloric Acid at Room Temperature. The procedure for this experiment has been described previously.[12] Through TLC analysis, AB-1 (6), AB-3 (8), and AB-5A (10) were detected, as well as unreacted noracronycine (3) and several other minor products. AB-2 (7) and AB-5B (11) were not detected in the reaction mixture.

^{252}Cf Plasma Desorption Mass Spectrometry (^{252}Cf PDMS). A description of the basic operational principles of the ^{252}Cf PD mass spectrometer and the method of mass calibration have been described.[20] The m/z of the ions is measured by the time-of-flight method utilizing a 45-cm path length. The acceleration voltage was ±10 kV: no post acceleration was used. The mass resolution was approximately 450 M/ΔM at full-width half-maximum. At this low resolution, the measured masses closely approximate the chemically averaged masses.[21] The fission fragment flux through the sample was approximately 1500^{-1} cm^{-2}.

Thin solid films of the noracronycine oligomers were prepared by first dissolving the sample in chloroform then diluting with a 50/50 v/v solution of methanol–2-propanol (Burdick and Jackson distilled in glass). The concentrations were estimated to be ≃1 μg/mL. A volume of 25 μL was electrosprayed onto a 1.5-μm-thick aluminized mylar foil (Steiner Film Co.) producing a uniform film 250 nm thick.

Acknowledgment. This work was supported at the University of Illinois at Chicago, in part, by Grant CA-20164 from the Division of Cancer Treatment, National Cancer Institute, Bethesda, MD. High-field nuclear magnetic resonance spectra were obtained at the Midwest Regional NMR Facility, University of Illinois at Urbana, Urbana-Champaign. This facility is funded, in part, by the National Science Foundation, Grant CHE 79-16100. At Texas A&M University this work was supported by grants to R.D.M. from the National Institutes of Health (GM-26096) and the Robert A. Welch Foundation (Grant A258). We are indebted to Vanessa Mancill for her unfailing assistance.

Registry No. 3, 13161-79-0; 6, 88151-49-9; 7, 88151-50-2; 8, 88151-51-3; 10, 95784-52-4; 11, 95797-83-4; 12, 88188-93-6; 13, 95784-53-5.

(20) Macfarlane, R. D. *Anal. Chem.*, in press.
(21) McNeal, C. J.; Ogilvie, K. K.; Theriault, N. Y.; Nemer, J. J. *J. Am. Chem. Soc.* **1982**, *104*, 976.

Synthesis of 4-Substituted 5-Amino-2-(β-D-ribofuranosyl)thiazoles and 4-Substituted 5-Amino-2-(β-D-ribofuranosyl)selenazoles and Their Respective Conversion into 2-(β-D-Ribofuranosyl)thiazolo[5,4-*d*]pyrimidines and 2-(β-D-Ribofuranosyl)selenazolo[5,4-*d*]pyrimidines. A New Synthesis of Tiazofurin and Selenazofurin

William J. Hennen,* Barbara C. Hinshaw, Timothy A. Riley, Steven G. Wood, and Roland K. Robins

Cancer Research Center, Department of Chemistry, Brigham Young University, Provo, Utah 84602

Received July 30, 1984

A novel ring closure has been devised to produce fully substituted thiazoles and selenazoles from the condensation of thio- and selenoates with various 2-aminoacetonitrile derivatives. The syntheses of methyl 2,5-anhydroallonothioate (3) and methyl 2,5-anhydroallonoselenoate (4) from methyl 2,5-anhydroallonimidate are described. The condensations of these carboxylates with the appropriate 2-aminoacetonitrile derivatives to give the corresponding 5-amino-2-(β-D-ribofuranosyl)thiazole and -selenazole nucleosides bearing the carboxamide (10 and 13), ethyl carboxylate (11 and 14), and cyano (12) functions at the four positions are reported. Further manipulation of these functionalized thiazoles and selenazoles yielded the corresponding thiazolo- and selenazolo[5,4-*d*]pyrimidine nucleosides (20, 21, and 22), as well as a thiazolo[5,4-*d*][1,2,3]triazine nucleoside (23). New syntheses of tiazofurin (1) and selenazofurin (2) via the reductive dediazotization products of ethyl 5-amino-2-(β-D-ribofuranosyl)-thiazole-4-carboxylate (11) and ethyl 5-amino-2-(β-D-ribofuranosyl)selenazole-4-carboxylate (14) are also reported.

Tiazofurin,[1] 2-(β-D-ribofuranosyl)thiazole-4-carboxamide[2] (1), and selenazofurin,[3] 2-(β-D-ribofuranosyl)selenazole-4-carboxamide[4] (2), are promising antitumor agents currently under study by the National Cancer Institute (NCI). Tiazofurin (1) has been shown to be an effective antitumor agent in animals.[5] Selenazofurin (2) was similarly demonstrated to possess both significant antitumor properties in animals[4] and has been shown to possess broad spectrum antiviral activity[6] in cell culture experiments.

Several modifications of the parent structure of tiazofurin, 1, have been reported. The carboxamido function has been transformed to the thiocarboxamide,[2] the amidine,[7] and the 2-thiazole-4-carboxamide[7] functions. Of

(1) Generic name given to compound 1.
(2) Srivastava, P. C.; Pickering, M. V.; Allen, L. B.; Streeter, D. G.; Campbell, M. T.; Witkowski, J. T.; Sidwell, R. W.; Robins, R. K. *J. Med. Chem.* **1977**, *20*, 256.
(3) Generic name given to compound 2.
(4) Srivastava, P. C.; Robins, R. K. *J. Med. Chem.* **1983**, *26*, 445.
(5) Robins, R. K.; Srivastava, P. C.; Narayanan, V. L.; Plowman, J.; Paull, K. D. *J. Med. Chem.* **1982**, *25*, 107.
(6) Kirsi, J. J.; North, J. A.; McKernan, P. A.; Murry, B. K.; Canonico, P. G.; Huggins, J. W.; Srivastava, P. C.; Robins, R. K. *Antimicrob. Agents Chemother.* **1983**, *24*, 353.

0022-3263/85/1950-1741$01.50/0