A Carbon-13 Nuclear Magnetic Resonance Spectroscopic Investigation of the Kiliani Reaction

Richard M. Blazer* and Thomas W. Whaley

Contribution from the Los Alamos Scientific Laboratory, University of California, Los Alamos, New Mexico 87545. *Received October 22, 1979*

Abstract: The Kiliani reaction of D-arabinose with sodium [^{13}C]cyanide (or [^{13}C,^{15}N]cyanide) was studied by ^{13}C NMR. The C-1 resonances of intermediates and products were observed as the reaction evolved. Intermediates were identified by addition of authentic samples to reaction solutions, interpretation of chemical shifts and coupling constants, and chemical experiments. Intermediates identified included cyanohydrins, amides, lactones, amidines, and an imidate. A discussion of the course of the reaction over the pH range 5.1–12.5 is presented. The final mannonate-to-gluconate ratio was shown to be a function of pH and not associated with the presence of certain metal ions.

The Kiliani–Fischer synthesis[1] is used commonly for the preparation of rare or unnatural sugars as well as sugars labeled with ^{13}C and ^{14}C. A 1-^{13}C-labeled aldose is prepared conveniently by chain extension of the next lower sugar via the Kiliani–Fischer cyanohydrin synthesis[2] employing sodium [^{13}C]cyanide. As a result of chirality of the starting aldose, unequal amounts of diastereomeric aldononitriles are produced in the cyanohydrin reaction. These initially produced epimeric aldononitriles are hydrolyzed in situ to the corresponding aldonates. Separation of the epimeric aldonates, lactonization, and reduction afford the desired aldoses. Since carbon–carbon bond formation occurs in the cyanohydrin reaction, it is this step that determines the relative amounts of epimeric aldoses obtained.

The paper by Isbell et al.[3] remains the definitive work on the Kiliani–Fischer synthesis of C-1-labeled glucose and mannose. Isbell was able to vary the ratio of mannonate to gluconate from 70:30 to 27:73 by altering reaction conditions; however, no explanation was proposed for the cause of the variation in epimer ratios. The identification of intermediates occurring in the Kiliani synthesis by paper chromatography and gas chromatography has been reported,[4] but we feel that the conclusions drawn were somewhat speculative. Species present in the crude chromatograms were often identified without the benefit of strong evidence such as comparison with authentic samples.

We wish to report a series of ^{13}C NMR studies on the evolving Kiliani cyanohydrin reaction that identify intermediate species and elucidate the course of the process (Scheme I). The use of ^{13}C NMR to monitor evolving reactions is a powerful technique, but has been reported for only a relatively few cases.[5]

Results and Discussion

Reactions between D-arabinose (**1**) and sodium [^{13}C]cyanide were conducted in NMR sample tubes, and consecutive spectra were obtained. Aqueous buffers were employed to eliminate variations of chemical shifts caused by pH changes during the reaction. The isotopic enrichment of the sodium [^{13}C]cyanide allowed the resonances from cyanide and C-1 of intermediates and products to be readily observable. As the reaction proceeded, the cyanide peak decreased and intermediate peaks were observed until finally only two product peaks were present in the spectrum. Dilute solutions and, where appropriate, subambient temperatures were employed to obtain reaction rates that were convenient for NMR studies.

Scheme I

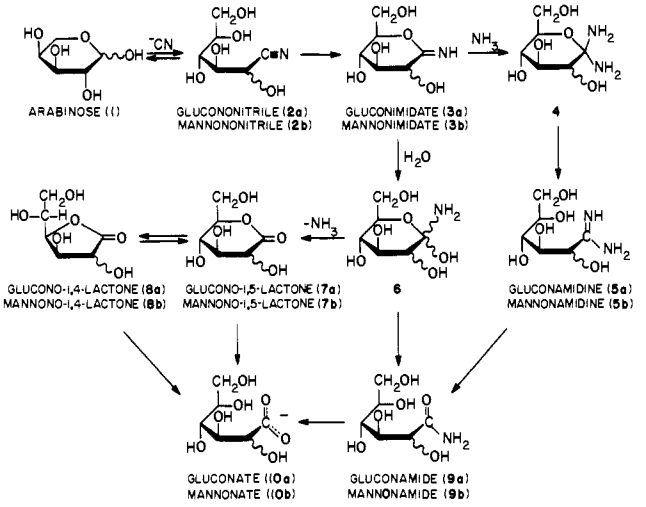

Table I. Chemical Shifts and Coupling Constants for ^{13}C-1 and ^{15}N Resonances Observed in Reactions of D-Arabinose with Isotopic Cyanide

	^{13}C NMR δ^{a-c}	^{15}N NMR δ^{b-d}	$^{1}J_{CN}$, Hz[e]	pH range
D-gluconononitrile	119.9		16.8	5.1–10.1
D-mannononitrile	121.2		16.9	5.1–10.1
D-gluconamidine	171.4	63.8	19.1	8.1–10.1
D-mannonamidine	172.2	64.7	19.0	8.1–11.6
D-glucono-1,5-lactone	175.1			8.1–9.3
D-mannono-1,5-lactone	175.6			8.1
D-mannonimidate	176.4	152.1	11.2	8.1–11.6
D-glucono-1,4-lactone	178.2			8.1
D-gluconamide	178.4	69.6	17.3	6.9–11.6
D-mannonamide	178.6	70.5	16.8	6.9–11.6
D-mannono-1,4-lactone	178.8			5.1–10.1
D-gluconate	179.4			5.1–12.5
D-mannonate	179.9			5.1–12.5

[a] Referenced to Me_4Si; see Experimental Section. [b] Chemical shifts varied slightly with pH. Values generally are ±0.2 ppm or better. [c] The ^{13}C NMR HCN/$^-$CN resonance varied from 113.1 ppm at pH 5.1 to 165.8 ppm at pH 12.5. This signal was not observed by ^{15}N NMR. [d] Referenced to Me_4N^+. [e] Values are ±0.2 Hz or better.

Identification of Intermediates and Products. Three methods were used to identify resonances of intermediates and products: (a) addition of authentic samples to reaction solutions; (b) interpretation of chemical shifts and coupling constants; (c) chemical experiments. The chemical shifts of the C-1 resonances for the species identified in the reaction are given in Table I along with the pH range over which the resonances were observable.

(1) Hudson, C. S. *Adv. Carbohydr. Chem.* **1945**, *1*, 1–36.
(2) For an alternative chain-extension method with nitromethane see: Sowden, J. C. *J. Biol. Chem.* **1949**, *180*, 55–58, and references cited therein.
(3) Isbell, H. S.; Karabinos, J. V.; Frush, H. L.; Holt, N. B.; Schwebel, A.; Galkowski, T. T. *J. Res. Natl. Bur. Stand.* **1952**, *48*, 163–171.
(4) Varma, R.; French, D. *Carbohydr. Res.* **1972**, *25*, 71–79.
(5) (a) Pines, S. H.; Douglas, A. W. *J. Am. Chem. Soc.* **1976**, *98*, 8119–8124. (b) *Tetrahedron Lett.* **1976**, 1955–1958. (c) *J. Org. Chem.* **1977**, *42*, 2786–2787. (d) Douglas, A. W. *J. Am. Chem. Soc.* **1978**, *100*, 6463–6469. (e) Norton, R. S.; Allerhand, A. *Biochemistry* **1976**, *15*, 3438–3445. (f) Ferris, J. P.; Edelson, E. H. *J. Org. Chem.* **1978**, *43*, 3989–3995.

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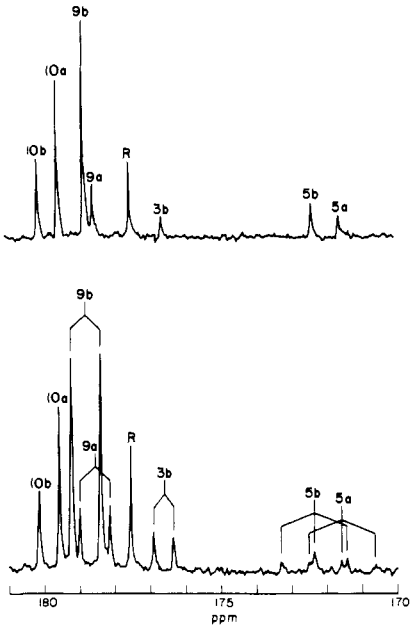


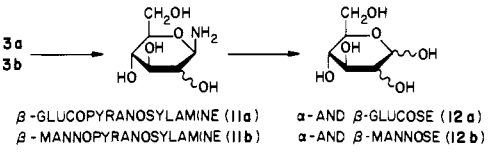
Figure 1. Typical ^{13}C NMR spectra of pH 10.1 reactions of D-arabinose with sodium [^{13}C]cyanide (top) and sodium [^{13}C,^{15}N]cyanide (bottom).

D-[1-^{13}C]Gluconononitrile (**2a**), D-[1-^{13}C]gluconamide (**9a**), and D-[1-^{13}C]mannonamide (**9b**) were identified by addition of the corresponding authentic ^{13}C-labeled compounds at appropriate times. Additions of D-[1-^{13}C]glucono-1,5-lactone (**7a**) and D-[1-^{13}C]mannono-1,4-lactone (**8b**) to pH 10.1 reaction solutions were used to identify [1-^{13}C]gluconate (**10a**) and [1-^{13}C]-mannonate (**10b**) since aldonolactones are hydrolyzed rapidly to aldonates at this pH. It was possible to identify D-[1-^{13}C]-mannono-1,4-lactone (**8b**) in a pH 5.1 reaction by addition of an authentic sample because at this lower pH the hydrolysis of mannono-1,4-lactone is slow. D-[1-^{13}C]Mannono-1,5-lactone (**7b**), D-[1-^{13}C]glucono-1,5-lactone (**7a**), and D-[1-^{13}C]glucono-1,4-lactone (**8a**) were identified by comparison of chemical shifts of resonances observed in a pH 8.1 reaction solution with pH 8.1 solutions of D-[1-^{13}C]glucono-1,5-lactone (**7a**) and D-[1-^{13}C]-mannono-1,4-lactone (**8b**). At this pH, steady-state mixtures of aldonate, aldono-1,4-lactone, and aldono-1,5-lactone are seen. The two C-1 gluconolactone peaks were differentiated by a ^{13}C NMR time study of the equilibration of D-[1-^{13}C]glucono-1,5-lactone (**7a**) with its 1,4-lactone (**8a**) and aldonate (**10a**).

The use of sodium [^{13}C,^{15}N]cyanide in the Kiliani reaction afforded additional information about the nitrogen-containing intermediates.[6] The multiplicity of a carbon resonance indicated the number of nitrogen atoms (99 mol % ^{15}N) bonded to C-1 of the intermediate. Furthermore, values of $^1J_{CN}$ provided some information on the hybridization of the carbon and nitrogen atoms involved. Figure 1 shows typical spectra obtained during the reaction of D-arabinose (**1**) with [^{13}C]- and [^{13}C,^{15}N]cyanide. D-[1-^{13}C,^{15}N]Gluconamide (**9a**), D-[1-^{13}C,^{15}N]mannonamide (**9b**), and D-[1-^{13}C,^{15}N]gluconononitrile (**2a**) resonances appeared as doublets with appropriate $^1J_{CN}$ coupling constants[7] (Table I). The resonance at 121.2 ppm appeared as a doublet ($^1J_{CN}$ = 16.9 Hz) and was assigned to D-[1-^{13}C,^{15}N]mannononitrile (**2b**) on the basis of chemical shift, coupling constant, and its position relative to gluconononitrile. As can be seen from Table I, the downfield C-1 resonance of each epimeric pair is from the mannono isomer.[8]

The resonances at 172.2 and 171.4 ppm appeared as 1:2:1 triplets in experiments employing sodium [^{13}C,^{15}N]cyanide, suggesting a carbon coupled to two equivalent ^{15}N atoms. Possible intermediates containing this structural feature are D-[1-^{13}C,$^{15}N_2$]gluconamidine (**5a**) and D-[1-^{13}C,$^{15}N_2$]mannonamidine (**5b**), which would exist primarily as amidinium ions over the pH range studied in this work. Amidine formation can be rationalized quite easily by the addition of [^{15}N]ammonia (which is released in the reaction sequence) to a cyclic imidate (**3**, Scheme I) followed by ring opening. Given this reasoning, it follows that amidine formation would be promoted by a high ammonia concentration. Indeed, the reaction of D-arabinose (**1**) and sodium [^{13}C]cyanide in pH 9.7 ammonium bicarbonate buffer showed almost exclusively the two amidine peaks in a ^{13}C NMR spectrum recorded at 2 h reaction time. An analogous experiment with sodium [^{13}C,^{15}N]cyanide gave doublets for the two amidine peaks indicating one ^{15}N from cyanide and one ^{15}N from the buffer. Singlet amidine peaks appeared with time as the result of nitrogen exchange with the buffer. The amide resonances displayed the same phenomenon of initial doublets giving way to singlets. Since the amidines may be viewed as precursors of the amides in ammonium bicarbonate buffer, it was possible to identify the upfield amidine as D-gluconamidine (**5a**) by comparison of the extent of exchange with buffer for the amides and amidines. It was shown that mannonate (**10b**) was produced at the expense of the 172.2-ppm resonance. Therefore, the downfield amidine was identified as D-mannonamidine (**5b**).

Scheme II



The resonance at 176.4 ppm was suspected to be from C-1 of one of the imidates, **3a** or **3b**, since it appeared early in the reaction and existed as a doublet with an appropriate[7] coupling constant, $^1J_{CN}$ = 11.2 Hz, in sodium [^{13}C,^{15}N]cyanide experiments. Chemical evidence supporting this hypothesis was obtained by adding sodium borohydride to a reaction solution at a time when the 176.4-ppm peak was at a maximum. There was a dramatic decrease in the relative height of the 176.4-ppm peak with the appearance of two new peaks at 83.4 ($^1J_{CN}$ = 5.1 Hz) and 85.9 ppm ($^1J_{CN}$ = 6.0 Hz). With time these peaks disappeared and resonances from α- and β-D-[1-^{13}C]glucose (**12a**) and α- and β-D-[1-^{13}C]mannose (**12b**) appeared. The 83.2- and 85.9-ppm peaks corresponded to the C-1 resonances of authentic samples of β-D-mannopyranosylamine and β-D-glucopyranosylamine, respectively. These results were interpreted in terms of reduction of the cyclic imidates,[9] **3a** and **3b**, to the corresponding aldosylamines (**11a** and **11b**) with subsequent hydrolysis to the aldoses (Scheme II). When it was assumed that the observed imidate had the mannono configuration, the ratio of the sums of the peak heights for the two epimeric series remained nearly constant throughout the reaction, while this ratio varied greatly with time if the glucono configuration was assumed for the imidate. Furthermore, in a sodium [^{13}C,^{15}N]cyanide experiment conducted in ammonium bicarbonate buffer, comparison of singlet-to-doublet ratios for imidate and amidine resonances, which reflected the degree of exchange with buffer for the two epimeric series, indicated that the 176.4-ppm peak was from D-[1-^{13}C]-mannonimidate (**3b**).

In a pH 9.3 reaction between D-arabinose and [^{13}C,^{15}N]cyanide where the reactant concentrations were four times normal, new resonances were observed at 84.5 (dd, $^1J_{CN}$ = 7.8 and 2.2 Hz) and 87.8 ppm (dd, $^1J_{CN}$ = 7.8 and 2.1 Hz). These signals are likely to arise from C-1 of the epimeric *gem*-diamino ethers **4**. When a similiar high-concentration reaction was conducted at pH 8.1, resonances were also seen at 104.4 (d, $^1J_{CN}$ = 9.0 Hz)

(6) Other examples include: (a) Nadzan, A. M.; Rinehart, K. L., Jr. *J. Am. Chem. Soc.* **1977**, *99*, 4647–4654. (b) Yamada, H.; Hirobe, M.; Higashiyama, K.; Takahashi, H.; Suzuki, K. T. *Tetrahedron Lett.* **1978**, 4039–4042.

(7) (a) Axenrod, T. In "Nitrogen NMR"; Witanowski, M., Webb, G. A., Eds.; Plenum Press: New York, 1973; pp 286–287. (b) Witanowski, M.; Stefaniak, L.; Webb, G. A. *Annu. Rep. NMR Spectrosc.* **1977**, *7*, 117–244.

(8) Exceptions we have observed are the β-D-aldoses and the β-D-aldopyranosylamines.

(9) Neilson, D. G. In "The Chemistry of Amidines and Imidates"; Patai, S., Ed.; Wiley: New York, 1975; pp 460–461.

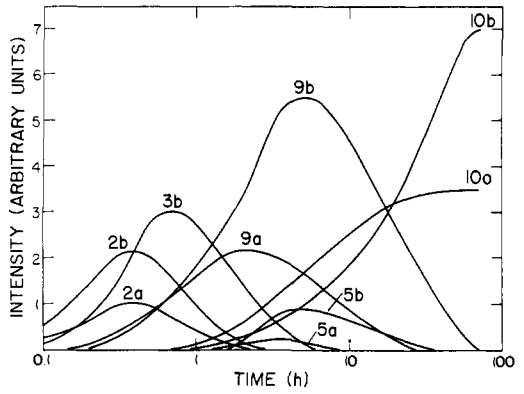


Figure 2. Graphic representation of the appearance and disappearance of the principal species present in the reaction of D-arabinose (**1**) with sodium [^{13}C]cyanide in pH 9.3 buffer.

and 105.0 ppm (d, $^1J_{CN}$ = ca. 7 Hz). These doublets could well be from the epimeric *gem*-hydroxyamino ethers **6**.

In order to corroborate evidence obtained from ^{13}C NMR experiments, a ^{15}N NMR study was made of the reaction between D-arabinose (**1**) and ^{15}N-enriched cyanide at pH 9.3. Resonances for D-[$^{15}N_2$]gluconamidine (**5a**), D-[$^{15}N_2$]mannonamidine (**5b**), D-[^{15}N]mannonimidate (**3b**), D-[^{15}N]gluconamide (**9a**), and D-[^{15}N]mannonamide (**9b**) were identified in experiments that employed potassium [^{15}N]cyanide (Table I). Assignments were based on chemical shifts,[10] relative peak areas, order of appearance, and values of $^1J_{CN}$ obtained in reactions using sodium [^{13}C,^{15}N]cyanide. A small ammonia–ammonium ion peak was identified, but neither cyanide nor the aldononitriles were detected, presumably owing to long relaxation times. Two small peaks at 9.8 and 11.1 ppm were observed early in the reaction. These chemical shifts imply [^{15}N]amino groups, and well could be from diamino ethers **4**, whose ^{13}C NMR peaks are present over the same time interval. This ^{15}N NMR study confirmed that only one imidate achieves a substantial concentration—it is highly unlikely that the epimers would have identical ^{13}C and ^{15}N NMR chemical shifts and coupling constants.

Course of the Reaction. The appearance and disappearance of intermediates and the appearance of products with time are illustrated in Figure 2 for the reaction between D-arabinose (**1**) and sodium [^{13}C]cyanide at pH 9.3. All identified intermediates are seen at this pH except D-[1-^{13}C]glucono-1,4-lactone (**8a**). The data support the course of reaction outlined in Scheme I. Cyanide–hydrogen cyanide decreases from zero time on, and the cyanohydrins, **2a** and **2b**, are clearly the initial intermediates to form. D-[1-^{13}C]Mannonimidate (**3b**) reaches its maximum concentration after D-[1-^{13}C]mannonitrile (**2b**) does. The epimeric aldonamides, **9a** and **9b**, and aldonamidines, **5a** and **5b**, form later in the reaction. The amidines appear soon after D-[1-^{13}C]-mannonimidate (**3b**) and before the amides when ammonia is present from the beginning of the reaction (vide supra). The observed aldonolactones, **7a**, **7b**, and **8b**, are detectable only relatively late in the reaction. Finally, only aldonates remain. The ^{15}N NMR studies confirmed the information obtained from ^{13}C data, and also revealed early appearing peaks that were probably from tetrahedral intermediates, **4**, as well as, ultimately, an ammonia–ammonium ion peak.

Table I gives the pH ranges over which various species were observed by ^{13}C NMR. The greatest number of intermediates was observable in the pH 8.1–10.1 range. All intermediates were seen at pH 8.1, but none at pH 12.0 or 12.5.

Isbell et al.[3] obtained their highest yield of barium gluconate by allowing D-arabinose and sodium cyanide to react in unbuffered aqueous calcium chloride solution. They suggest 3 days' reaction time at room temperature to form the cyanohydrins followed by heating in vacuo to effect hydrolysis to the aldonates. We repeated this reaction employing sodium [^{13}C]cyanide, and ^{13}C NMR showed only aldonates remaining after approximately 7 h at ambient temperature.

In the pH range 5.1–10.1 the rate of the overall Kiliani sequence yielding D-[1-^{13}C]gluconate (**10a**) and D-[1-^{13}C]mannonate (**10b**) increased with increasing pH. At pH 11.6 the rate of the overall reaction was the fastest observed, although the conversion of the aldonamides to aldonates was slower than at pH 10.1. The reaction between D-arabinose and sodium [^{13}C]cyanide at pH 12.0 was slower than at pH 11.6, and a further decrease in the overall reaction rate was seen at pH 12.5.

Reaction rates differed between the glucono and mannono series. The glucono epimer of a given type of intermediate generally was present earlier and for a shorter period than the corresponding mannono epimer. This trend, which is indicative of greater reactivity in the glucono series, is illustrated in Figure 2 for the pH 9.3 reaction.

The ratio of mannonate to gluconate formed in the Kiliani reaction displayed a marked pH dependence. This ratio was found to be approximately 70:30 from pH 5.1 to 9.7, while at pH 10.1 it decreased slightly. In pH 11.6 buffer a dramatic reversal was seen as the mannonate-to-gluconate ratio became 32:68. Only a slight increase in the proportion of gluconate was seen at pH 12.0 and 12.5. The reactions conducted by Isbell et al.[3] in unbuffered aqueous salt solutions were repeated with pH monitoring and ^{13}C NMR determination of product distribution. When the maximum pH attained was 10.3, the final mannonate-to-gluconate ratio was 56:44; thus, the reversal of the product epimer distribution occurs not much above pH 10.

The mannonate-to-gluconate ratios reported by Isbell et al.[3] for the reaction of sodium cyanide with D-arabinose in the presence of various salts appear to be solely a function of pH. Stepiński and Świderski,[11] however, have demonstrated a marked effect of metal ions, especially cadmium, on the product ratio for the reaction of sodium cyanide with D-erythrose. To ascertain whether metal ions had any influence in our aqueous-buffer approach, reactions between sodium [^{13}C]cyanide and D-arabinose were performed in pH 8.1 buffer containing calcium or cadmium ions, those most likely to have a sizable effect.[3,11] The resulting epimer distributions were within the previously observed range for pH 8.1 reactions.

Two possible causes of the pH dependence of the mannonate-to-gluconate ratio were investigated. This knowledge might allow control of the ratio so a maximum amount of a desired epimer could be produced. One possibility was reversibility of the cyanohydrin-forming reaction; i.e., below ca. pH 10 primarily D-mannononitrile (**2b**) is produced as the kinetic product, while above ca. pH 10 reversibility generates mostly D-gluconononitrile (**2a**) as the thermodynamic product. D-[1-^{13}C]Gluconononitrile (**2a**) was allowed to hydrolyze in pH 5.1, 6.9, 8.1, 10.1, and 11.6 buffers containing 1 equiv of [^{13}C,^{15}N]cyanide. Reversibility of the cyanohydrin-forming reaction would be indicated in ^{13}C NMR spectra by the appearance of C-1 doublets for the nitrogen-containing species as a result of cyanide exchange with the medium. This phenomenon was observed over the entire pH range studied without any major singlet-to-doublet ratio variation. Thus, cyanohydrin-forming reaction reversal did not occur to a greater extent above pH 10.1.

Another possible explanation for the aldonate epimer ratio differences was that above ca. pH 10 deprotonation at C-2 of some species occurs, leading to epimerization. At pH 11.6 D-[1-^{13}C]mannonamide (**9b**) and D-[1-^{13}C]gluconamide (**9a**) gave only the single corresponding aldonates. Thus, any species able to undergo epimerization at high pH must precede the aldonamides in the course of the reaction (Scheme I). In the series of experiments described above for testing for reversibility, the buffers contained 50% D_2O. Deprotonation and, therefore, possible epimerization would be indicated by incorporation of deuterium at C-2 at some stage, which could be detected by an upfield shift of approximately 0.1 ppm in the C-1 resonances of the aldonates.

(10) Witanowski, M.; Stefaniak, L.; Januszewski, H. In ref 7a, pp 166–167.

(11) Stepiński, J.; Świderski, J. *Rocz. Chem.* **1976**, *50*, 1991–1993.

Over the pH 6.9–11.6 range, final ^{13}C NMR spectra showed closely spaced pairs of peaks, representing C-1 resonances of the C-2 deuterio and C-2 protio aldonates, without significant differences in their ratios. Therefore, deprotonation potentially resulting in epimerization did not increase noticeably above pH 10.1.

Although two possible controlling factors have been ruled out, the cause of the pH dependence of the mannonate-to-gluconate ratio remains unknown. Presumably, cyanohydrin formation occurs via attack of cyanide on D-*aldehydo*-arabinose. Conformational equilibria influenced by pH might determine which face of the aldehydo group is preferentially attacked. We have not studied this possibility.

Experimental Section

^{13}C NMR spectra were obtained with a Varian CFT-20 NMR spectrometer system equipped with an 8-mm variable-temperature probe. A 3 vol % [1-^{13}C]acetic acid in water–deuterium oxide (1:1) solution in a coaxial tube was used to obtain lock and provide a reference peak. At 27 °C the $H_3C^{13}CO_2H$ peak was observed at 177.5 ppm when referenced to dioxane (5 vol % in 0.4 M pH 10.1 bicarbonate buffer) as 67.4 ppm. Two sets of parameters were used commonly: spectral width, 500/1603 Hz (ca. 160–185/ca. 105–185 ppm); pulse width, α = 64/39°; number of transients, 250/800 (34/34 min). The free induction decays consisted of 8192 data points. All spectra were proton decoupled. Mannonate-to-gluconate ratios were determined from peak heights, which were shown to be proportional to peak areas. Any errors in the ratios due to differences in T_1 for mannonate and gluconate were ruled out by comparing peak height ratios to those in a small-pulse-width, long-pulse-delay experiment.

^{15}N NMR spectra were obtained with a Varian XL-100 spectrometer (10.2 MHz) equipped with a 12-mm variable-temperature probe and interfaced to a Nova 1210 computer. Lock was obtained with deuterium oxide in a coaxial tube. The carrier frequency was referenced to a 4.2 M [^{15}N]ammonium sulfate solution in deuterium oxide as −18 ppm. Parameters used included spectral width, 5000 (−67 to 423 ppm) or 6666 Hz (−116 to 537 ppm); pulse width, α = 36°; pulse interval, 1.000 s; number of transients, 6700–55 500 (1.9–15.4 h). The free induction decays consisted of 8192 data points. All spectra were proton decoupled.

D-(−)-Arabinose obtained from Eastman Organic Chemicals was recrystallized from ethanol–water with Norit treatment prior to use. D-[1-^{13}C]Glucono-1,5-lactone,[12] D-[1-^{13}C]mannono-1,4-lactone,[12] D-[1-^{13}C]gluconamide,[13] D-[1-^{13}C]mannonamide,[13] D-[1-^{13}C]gluconononitrile,[14] sodium [^{13}C]cyanide,[15] sodium [^{13}C,^{15}N]cyanide,[16] and potassium [^{15}N]cyanide[17] were prepared by published procedures. All other chemicals were commercial products, which were used as received. Buffers used were phthalate (pH 5.1), phosphate (pH 6.9 and 11.6), Tris (pH 8.1), *p*-hydroxybenzoate (pH 9.3), and bicarbonate (pH 10.1). Reaction solutions (1.5–2.5 mL) were 0.40 M in buffer and 0.08 M in both D-arabinose and labeled sodium cyanide unless otherwise stated. Reactant concentrations were doubled for the ^{15}N NMR studies. Isotopic enrichment in cyanide was 91 mol % ^{13}C and/or 99 mol % ^{15}N. Reactions were maintained at 5, 10, 15, or 25 °C to give suitable reaction rates. Spectral data accumulations were carried out consecutively immediately following the start of each reaction, and intermittently as the reactions slowed.

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(14) Prepared from D-[1-^{13}C]glucose by the procedure of Wohl, A.; Wollenberg, O. *Justus Liebigs Ann. Chem.* **1933**, *500*, 281–286. Attempts to prepare D-[1-^{13}C]mannononitrile from D-[1-^{13}C]mannose by this procedure were unsuccessful.
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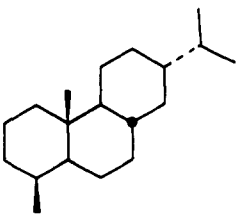
Intramolecular Diels–Alder Route to Angularly Substituted Perhydrophenanthrenes. Synthesis of (±)-Fichtelite

Douglass F. Taber* and Samir A. Saleh

Contribution from the Department of Pharmacology, School of Medicine, Vanderbilt University, Nashville, Tennessee 37232. *Received May 21, 1979*

Abstract: A central issue in the synthesis of the steroids and a variety of other complex carbocyclic natural products is stereocontrolled construction of the angularly substituted *trans,anti,trans*-perhydrophenanthrene nucleus. We report a new approach to this problem, the key to which is induction of relative chirality in the course of the intramolecular Diels–Alder reaction. Thus, triene **3** on heating is transformed into the crystalline tricyclic ketone **2**. Ketone **2** is carried on in five steps to (±)-fichtelite (**1**).

Fichtelite, a crystalline hydrocarbon, was first isolated by Bromeis in 1841 from pine trunk remains found in a peat bed in the Fictelgebirge region of Bavaria.[1] The relationship of fichtelite to the resin acids was suspected early on.[2] The presence of an angular methyl group, and hence gross structure **1**, was demonstrated by Ruzicka in 1935.[3] This was supported by a careful molecular-weight determination.[4] This early work was capped by a courageous synthesis by Bogert and Sterling of a mixture of hydrocarbons having the gross structure **1**. Modern analytical techniques would probably have confirmed the presence of authentic fichtelite in that mixture.



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(2) For a summary of this work, see: Simonsen, J. S.; Barton, D. H. R. "The Terpenes", Vol. III; Cambridge University Press: New York, 1952; p 337.
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(5) (a) Sterling, E. C.; Bogert, M. T. *Science* **1938**, *87*, 196. (b) *J. Org. Chem.* **1939**, *4*, 20.

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