Its ^1H NMR spectrum reflects the dissymmetry introduced by the methyl substituent. Compared to **1** (R = H), the C-7 signal for **13** at 171.3 ppm is shifted upfield by 5.8 ppm. The shielding is fully accommodated by the monoalkyl substitution plan at C-8, since C-7 in **1** (R = CH_3) is seen at 165.7 ppm.[1c] In fact, the incremental Δppm values of 5.6–5.8 indicate that **13** is strain-free as suggested by molecular models.

In summary, a convenient synthesis of annulated fulvenes and their conversion to previously unknown 1,7-cyclohexenonorbornadienes has been demonstrated. We believe the pathway from **9** to **4** to be as illustrated. This protocol and both classes of product molecules are expected to play useful roles in future synthetic and mechanistic investigations.

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Biosynthesis of Aristeromycin. Elucidation of the Origin of the Adenine and Cyclopentane Rings

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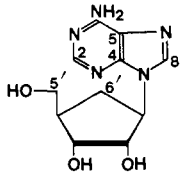
Aristeromycin (**1**) is a novel, carbocyclic analogue of adenosine obtained from the fermentation broth of *Streptomyces citricolor*.[1] The compound exhibits a number of interesting biological properties[2,3] including inhibition of AMP synthesis in mammalian cells, inhibition of cell division and elongation in rice plants, and inhibition of the enzyme *S*-adenosylhomocysteine hydrolase.[4] The unusual structure of aristeromycin and its important biological activity have prompted us to carry out the biosynthetic investigations that are reported here.



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The initial phase of our studies was concerned with the origin of the cyclopentane ring in **1**. Since a carbohydrate origin appeared likely, both [U-^{14}C]-D-glucose and [1-^{14}C]-D-ribose were evaluated as precursors (Table I, experiments 1 and 2). Very low incorporations were observed in each instance. These experiments were carried out using the fermentation medium reported in the literature.[1] Since this medium contains glucose, soluble starch, and cornsteep liquor, it seemed likely that the low incorporation of glucose observed in experiment 1 could be attributed to dilution of the labeled precursor. Accordingly, a series of experiments were carried out to measure aristeromycin production in replacement cultures. After many trials, it was discovered that excellent production of the antibiotic could be obtained by fermentation in the "normal" medium for 48 h followed by fermentation in a replacement medium lacking both glucose and soluble starch. When [1-^{14}C]glucose was administered to *S. citricolor* growing in replacement culture, there was a sixfold increase in the specific incorporation figure (experiment 3). This observation was followed up by administration of (1-^{13}C)-D-glucose to *S. citricolor* under the same conditions. Examination of the noise-decoupled ^{13}C NMR spectrum of the resulting aristeromycin revealed a fourfold enrichment of the signal due to C-5′ of aristeromycin (experiment 4).

Table I. Administration of Labeled Precursors to *S. citricolor*

expt	precursor ($^3H/^{14}C$)	fermentation conditns	spec incorpn, % ($^3H/^{14}C$)	labeling pattern
1	[U-^{14}C]-D-glucose	normal	0.01	
2	[1-^{14}C]-D-ribose	normal	0.0003	
3	[1-^{14}C]-D-glucose	replmnt	0.06	
4	(1-^{13}C)-D-glucose	replmnt	4	C-5′
5	(6-^{13}C)-D-glucose	replmnt	2	C-6′
6	sodium [^{14}C]formate	normal	0.04	
7	[1-^{14}C]glycine	normal	0.24	
8	sodium (^{13}C)formate	normal	0	
9	(1,2-$^{13}C_2$)glycine	normal	7 at C-4, C-5	C-4, C-5 $^1J_{CC}$ = 65 Hz
			13 at C-2, C-8	C-2, C-8
10	(^{15}N,2-^{13}C)glycine	normal	10 at C-5	C-2, C-5, C-8
			9 at C-2, C-8	
11	[2-^3H,8-^{14}C]-adenosine (5.98)	normal	0.39 (4.90)	

If D-glucose is incorporated intact into the cyclopentane moiety of aristeromycin, the discovery that C-1 of glucose corresponds to C-5′ of the antibiotic indicates that C-6 of D-glucose should reside at either C-3′ or C-6′ of aristeromycin. In order to differentiate between these two possibilities, (6-^{13}C)-D-glucose was synthesized[5] and administered to replacement cultures of *S. citricolor*. Examination of the ^{13}C NMR spectrum of the antibiotic isolated in this experment showed a twofold enhancement in the signal due to C-6′ of aristeromycin (experiment 5). From this result, one can conclude that the formation of the cyclopentane moiety of aristeromycin proceeds with formation of a carbon–carbon bond between C-2 and C-6 of glucose. The details of this cyclization process remain to be elucidated.

The second phase of our investigation focused upon the origin of the adenine ring in **1**. The biosynthesis of the adenine ring of purine nucleosides has been extensively studied in both avian liver and bacterial systems.[6] As a result of these studies, it is generally accepted that C-2 and C-8 of the adenine moiety are derived from formate, while C-4 and C-5 are derived from C-1 and C-2 of glycine, respectively. The C-7 nitrogen atom is also derived from the nitrogen atom of glycine. A preliminary evaluation of this pathway in *S. citricolor* was carried out by administration of [^{14}C]formate and [1-^{14}C]glycine. Radioactive aristeromycin was obtained in both experiments (experiments 6 and 7) and glycine was clearly the more efficient precursor. Experiments 6 and 7 were followed by evaluation of (^{13}C)formate and (1,2-$^{13}C_2$)glycine as aristeromycin precursors. The results from these studies were surprising. No visible enrichment was apparent in the ^{13}C NMR spectrum of aristeromycin derived from (^{13}C)formate (experiment 8). On the other hand, (1,2-$^{13}C_2$)glycine yielded aristeromycin that was highly enriched at C-2 and C-8 as well as having lower enrichment at C-4 and C-5 (experiment 9). The coupling between C-4 and C-5 observed in this experiment confirms the expected intact incorporation of glycine into this position of the purine nucleus.[7] The derivation of C_1 units from glycine observed in the experiment is unusual, but not without precedent. Some 37 years ago, Karlsson and Barker[8] reported that C-2 and C-8 of uric acid were labeled by both [^{14}C]formate and [2-^{14}C]glycine

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in pigeons. More recently, Herbert and Mann[9] found that both the *N*-formyl and *O*-methyl groups of tuberin were labeled by [2-^{14}C]glycine, but the *N*-formyl group was not labeled by formate or methionine. In order to confirm that C-2 of glycine is the source of C-2 and C-8 in the adenine ring of **1**, as well as to define the origin of C-5 and N-7, (^{15}N,2-^{13}C)glycine was utilized as a precursor (experiment 10). The results of this experiment verify that C-5 of the adenine ring of **1** derives from C-2 of glycine and that C-2 of glycine serves as the source of C-2 and C-8 of the purine. However, no coupling was observed for the labeled carbon at C-5 suggesting complete loss of the ^{15}N label of glycine by transamination in vivo. Finally, we examined the possibility that the adenine ring of **1** is derived by catabolism of adenosine to yield free adenine[10] which is then joined to the cyclopentane moiety. Administration of [2-^{3}H,8-^{14}C]adenosine (experiment 11) showed that the adenine ring of adenosine is incorporated largely intact into the adenine ring of aristeromycin. The high incorporation figure suggests that the major route to the adenine ring of **1** may proceed via adenosine.

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Formation of $HCr(CO)_3^-$ from the Remarkable Reaction of Hydride Ion with Benzenechromium Tricarbonyl. Gas-Phase Reactions of a Novel 14-Electron Metal Anion Complex

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We wish to report here on the novel ligand displacement reaction that occurs when benzenechromium tricarbonyl (**1**) reacts with hydride ion in the gas phase. In this paper we describe our studies of the mechanism of this reaction and provide a preliminary account of the reactivity of the 14-electron metal ion product, $HCr(CO)_3^-$.

Our experiments have been carried out at 300 ± 2 K in a flowing afterglow apparatus which has been described in detail previously.[1] Hydride is produced as the major observed ion from electron impact on trace amounts of NH_3 added past an electron gun. A fast flow of helium buffer gas carries hydride ions the length of a 100 cm × 7.3 cm i.d. flow reactor where they interact with $C_6H_6Cr(CO)_3$ added to the system through a heated helium flow inlet. Hydride rapidly reacts with **1** to yield a proton-abstraction product and a benzene-displacement product, **2**, in roughly equal amounts (eq 1).[2] Production of $HCr(CO)_3^-$ can

$$H^- + \underset{\mathbf{1}}{C_6H_6Cr(CO)_3} \rightarrow C_6H_5Cr(CO)_3^- + H_2$$
$$\rightarrow \underset{\mathbf{2}}{HCr(CO)_3^-} + C_6H_6 \qquad (1)$$

be shown to be unique to hydride since OH^- and other strongly basic anions react with **1** by exclusive proton abstraction.[3]

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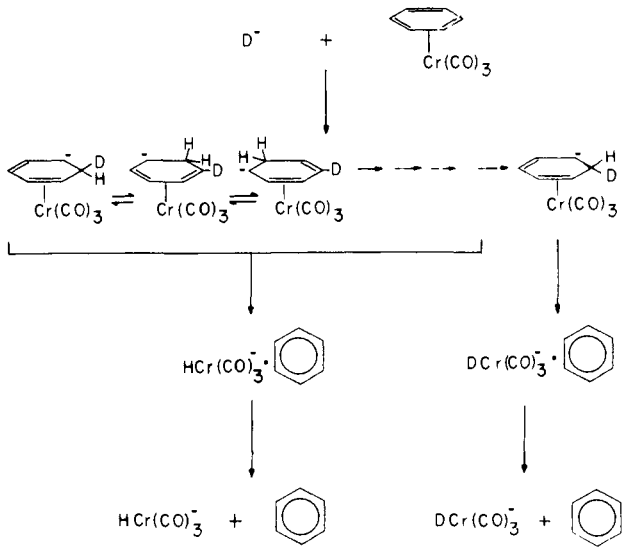
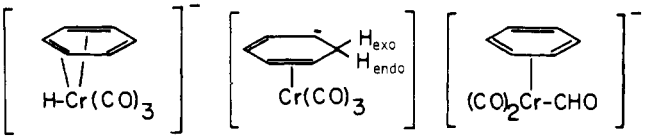
(2) An accurate rate and product distribution for eq 1 could not be determined due to the low volatility of **1** and the unavoidable presence of NH_2^- and OH^- in the system, respectively.

Scheme I



In analogy with nucleophilic addition mechanisms postulated for metal arenes in solution,[4–7] we can envisage hydride addition to complex **1** in (at least) three different ways. Direct H^- attachment to the metal requires "slippage" of the η^6-benzene ligand to η^4 (A); subsequent or simultaneous expulsion of the hydrocarbon



A B C

from the energy-rich adduct then produces $HCr(CO)_3^-$. Alternatively, initial hydride addition to the benzene ring or a carbonyl ligand may occur to produce a chromium–cyclohexadienyl anion B or chromium–formyl anion intermediate C, respectively. Intramolecular hydride migration with accompanying benzene loss may then give rise to the observed product. In solution, nucleophilic addition to compound **1** has been shown to occur predominantly to the exo face of the ring.[8–12] In order to distinguish among the conceivable intermediates shown above, we have carried out experiments using D^- produced from ND_3. If reaction 1 proceeds exclusively via intermediate A or C, then only $DCr(CO)_3^-$ would be produced. However, if reaction proceeds through intermediate B and the endo hydrogen is preferentially transferred to the metal, then only $HCr(CO)_3^-$ would be observed. A mixture of both $DCr(CO)_3^-$ and $HCr(CO)_3^-$ would appear if ring attachment occurs, but hydrogen migration is unselective. The reaction of D^- with **1** under the conditions described above affords both $HCr(CO)_3^-$ and $DCr(CO)_3^-$ in an observed ratio of 7:1. No

(3) The ΔH_{acid} of **1** is 371.5 ± 5 kcal mol^{-1}. The conjugate base anion, $C_6H_5Cr(CO)_3^-$, undergoes five H/D exchanges with D_2O, suggesting a π-arene, as opposed to a σ-phenyl structure. Lane, K. R.; Squires, R. R., unpublished results.
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