# Chemistry of HCN. 1. Formation and Reactions of *N*-(Aminomethylidene)diaminomaleonitrile, an HCN Pentamer and Precursor to Adenine

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*N*-(Aminomethylidene)diaminomaleonitrile (**10**) was shown to be a crucial and previously undetected intermediate in the formation of adenine (**4**) from diaminomaleonitrile (DAMN, **2**) and formamidine acetate. Although stable in neutral 1-butanol at 100 °C, **10** was selectively converted in alcohols with various reagents to any one of several products. Two of these products, 4-amino-5-cyanoimidazole (**8**) and 4-aminoimidazole-5-carboxamidine (**11**), appear to form by attack of the $NH_2CH$= substituent at the adjacent nitrile. This mechanism is discussed relative to published proposals for the nonphotolytic prebiotic and preparative pathway(s) to adenine from DAMN or HCN.

Studies of the prebiotic chemistry of HCN[1,2] have established that oligomerization of hydrogen cyanide to adenine (**4**) in aqueous ammonia proceeds through at least four detectable and measurable intermediates: formamidine (**3**), diaminomaleonitrile (DAMN, **2**), 4-amino-5-cyanoimidazole (**8**), and 4-aminoimidazole-5-carboxamidine (**11**). Kinetic data for the oligomerization of HCN to DAMN are available[1] and many details of the photoisomerization of DAMN to **8** in water[3,4] and methanol[5] are established, but no similar information exists for nonphotolytic conversions of DAMN to **8** or **11** in water, alcohols, or liquid ammonia. Without knowledge of such details and in lieu of additional intermediates, published proposals about the nature of nonphotolytic pathways to adenine from HCN in liquid $NH_3$ have been necessarily speculative. It was suggested[6] that, once DAMN is formed in 1:8 HCN–$NH_3$, either (1) HCN is eliminated to give aminomalononitrile (**1**), a known precursor[7] to **4** when synthesized separately and combined with formamidine, or (2) DAMN undergoes N-formiminylation to **6** which converts to the *E* isomer **7**, from which **8** is formed. These two possibilities are shown as Schemes I and II. It is important to note that isolation or detection of **6** as the crucial intermediate in HCN/$NH_3$ reactions has not been reported, and the retrograde reaction of **2** → **1** has recently been shown[8] to be of little or no consequence.

From the observations of Yamada et al.[9] on the formation of doubly labeled adenine from $H^{13}C^{15}N$ and $NH_2CHO$ at 160 °C, it may be inferred[10] that **1**, the HCN trimer, reacts as it is formed with formamide to give **4**. Such a pathway appears to be specific to the conditions and reagents employed and only peripherally related to prebiotic studies in water wherein DAMN (**2**) is formed and then elaborated to **8** and **11**.

Because DAMN may be converted to adenine in 55% yield by reaction with formamidine acetate and $NH_4OAc$ in methanol,[11] these conditions were adapted for study to better understand the particulars involved. The reagents are most conveniently handled in alcohols and give a higher yield than the 24% obtained from liquid ammonia[6] and HCN. As a result we report substantive evidence that a crucial intermediate, *N*-(aminomethylidene)diaminomaleonitrile (**10**), forms directly from the reaction of DAMN with formamidine acetate in alcohols at 50-100 °C.

Scheme I[6]

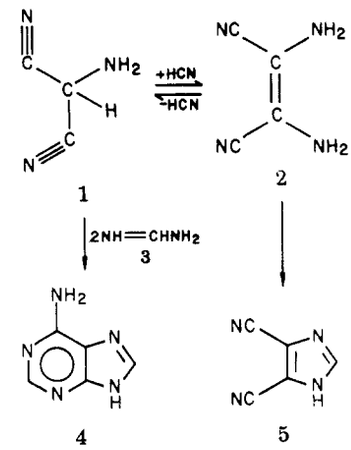
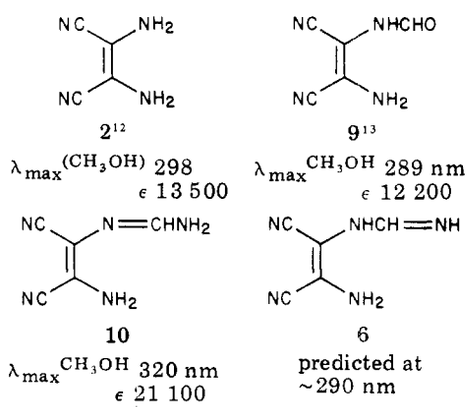



Table I. Comparison of UV Spectra for 2 and Its Derivatives




(1) R. A. Sanchez, J. P. Ferris, and L. E. Orgel, *J. Mol. Biol.*, **30** 223 (1967).
(2) J. Oro and A. P. Kimball, *Arch. Biochem. Biophys.*, **96**, 293 (1962).
(3) J. P. Ferris and L. E. Orgel, *J. Am. Chem. Soc.*, **88**, 1074 (1966).
(4) J. P. Ferris and J. E. Kuder, *J. Am. Chem. Soc.*, **92**, 2527 (1970).
(5) T. H. Koch and R. M. Rodehurst, *J. Am. Chem. Soc.*, **96**, 6707 (1974).
(6) Y. Yamada, I. Kumashiro, and T. Takenishi, *J. Org. Chem.*, **33**, 642 (1968).
(7) J. P. Ferris and L. E. Orgel, *J. Am. Chem. Soc.*, **88**, 3829 (1966).
(8) J. P. Ferris and E. H. Edelson, *J. Org. Chem.*, **43**, 3989 (1978).
(9) H. Yamada, M. Hirobe, K. Higashiyama, H. Takahashi, and K. Suzuki, *J. Am. Chem. Soc.*, **100**, 4617 (1978).
(10) This mechanism is our suggestion, since the authors of ref 9 neither give a yield nor present an explicit mechanistic proposal.
(11) Y. Yamada, M. Sakurai, and I. Kumashiro, U.S. Patent 3671649 (1972). See particularly example 6 which employs DAMN, formamidine acetate, and $NH_4OAc$ in the ratio 1:2:0.5 in $CH_3OH$ at 130 °C and gives an isolated 47% yield (55% by UV assay) of adenine.

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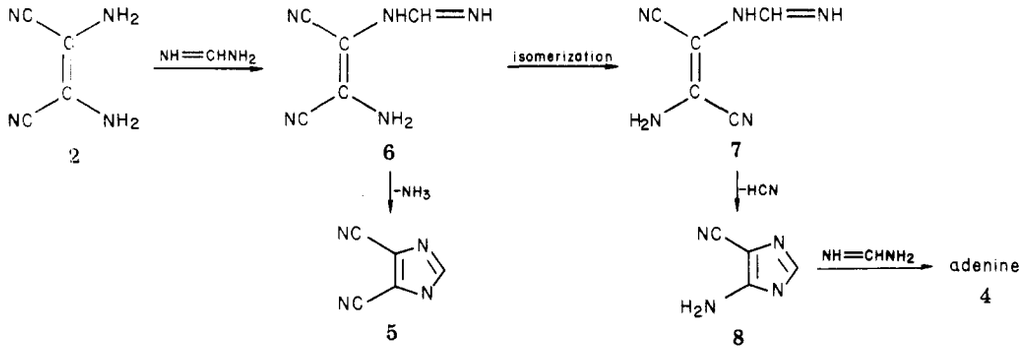
Scheme II[6]




This intermediate was first detected as a transient species by TLC and eventually isolated in 2.1% yield by preparative high-pressure LC. Observation of a reaction mixture by $^{13}C$ NMR spectroscopy and comparison of the amidine carbons for **3** and **10** at 158.95 and 154.15 ppm, respectively, led to an estimation of up to 5% of **10** present when the reaction temperature was kept at 90–95 °C for a few minutes and before isolation. Above 100 °C **10** is rapidly and exothermally converted in the reaction mixture to adenine and other materials. Stored as an amorphous powder in a nitrogen atmosphere it undergoes slow darkening and decomposition with a half-life of about 2.2 years at room temperature.

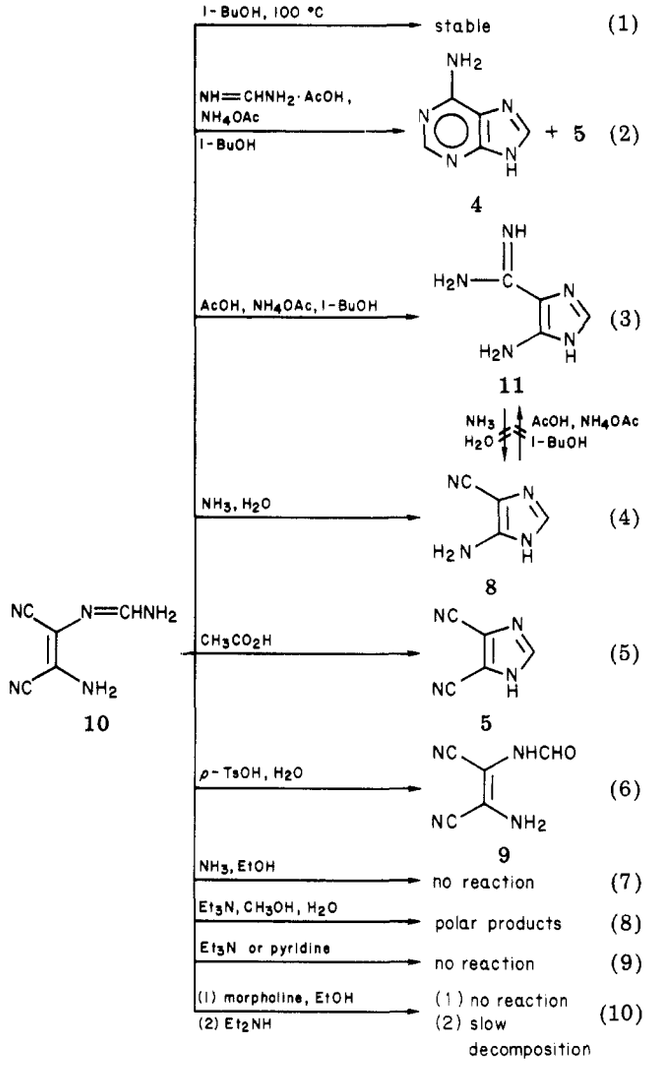
Of the analytical methods available only consideration of UV absorption wavelengths distinguished conclusively between structures **6** and **10**. The maximum at 320 nm agrees only with **10**, since **6** would absorb in the UV region similarly to *N*-formyldiaminomaleonitrile (**9**). The ultraviolet maxima of these related compounds are compared in Table I.[12,13]

It is particularly important in considering the subsequent chemistry of **10** to understand that DAMN, with a recent exception,[14] has been viewed as a diamine and/or a dinitrile rather than as an $\alpha$-amino nitrile in mechanisms explaining the formation of adenine or of imidazoles. This has led previous workers to postulate mechanisms for the appearance of imidazoles **8** and **11** which avoided chemical interaction between a derivatized amino group and the neighboring nitrile (e.g., Schemes I and II).

When the reaction temperature of formamidine acetate, **2**, and $NH_4OAc$ in 1-butanol was taken to 105 °C for only 5 min, it was possible to isolate an 18% yield of amidine **11**, but there was no **8** present by TLC with a limit of detection of well below 0.5% yield. Any mechanism for ring closure of **10** must account for the complete absence of **8** and for the collection of sublimed ammonium cyanide in up to 66% yield when the internal pot temperature is above 100 °C. Scheme III illustrates the mechanism favored to account for these observations, although the precise roles played by acetic acid, acetate ion, and ammonium ion still remain obscure.

That **10** is the touchstone for understanding much of the prebiotic and preparative nonphotolytic chemistry of DAMN is shown by examination of the reaction products from **10** and various reagents (Chart I). Thus, **8**, **11**, and **5** may each be generated from **10** free of other imidazoles by proper selection of conditions. The variety of products resulting from **10** demonstrates its sensitivity to pH, solvent, and nucleophiles.

Chart I




By itself **10** is stable when heated for a few minutes at 100 °C in 1-butanol, but with formamidine acetate and $NH_4OAc$, conversion to adenine occurs. The necessity of $NH_4OAc$ for cyclization of **10** to **11** is apparent in eq 3 when compared to eq 4 and 5. The rapid conversion in the presence of aqueous ammonia (eq 4) suggests that **10** may also be the prebiotic precursor of **8** in the dilute aqueous ammonia solutions at pH 9 studied by Sanchez, Ferris, and Orgel.[1]

Because **8** and **11** are not interconvertible with either $NH_3$ or AcOH and $NH_4OAc$, they *must* form by a different chemistry from **10**. Mechanistically, formation of **8** from **10** in aqueous ammonia might proceed by any of several pathways, and present information does not allow a clear

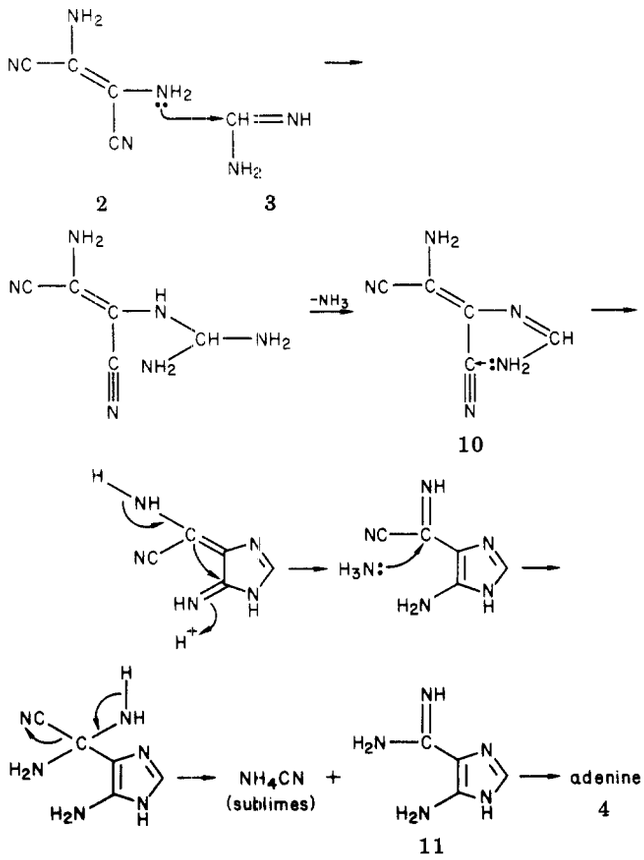
(12) Purchased from Aldrich Chemical Co., Inc.
(13) H. Brederek and G. Schmötzer, *Justus Liebigs Ann. Chem.*, **600**, 95 (1956).
(14) J. P. Ferris, P. C. Joshi, and J. G. Lawless, *Biosystems*, **9**, 81 (1977).

Table II. TLC[a] Behavior for DAMN, Adenine, and Related Intermediates

| compd | $R_f$ in solvent system[b] A | B | C | D | response to $I_2$ vapor | UV[c] |
|---|---|---|---|---|---|---|
| diaminomaleonitrile (2) | 0.53 | 0.91 | 0.91 | 0.85 | + | + |
| *N*-formyldiaminomaleonitrile (9) | 0.35 | 0.63 | 0.83 | 0.61 | + | + |
| *N*-(aminomethylidene)diaminomaleonitrile (10) | 0.53 | 0.63 | 0.85 | 0.72 | + | − |
| 4,5-dicyanoimidazole (5) | 0.49 | 0.80 | 0.90 | 0.77 | − | + |
| 4-amino-5-cyanoimidazole (8) | 0.36 | 0.55 | 0.77 | 0.51 | + | + |
| 4-aminoimidazole-5-carboxamidine (11) | 0 | 0.03 | 0.32 | 0.06 | + | + |
| adenine (4) | 0.37 | 0.53 | 0.63 | 0.54 | − | + |

[a] Quanta/Gram Q1F silica gel plates, 20 × 20 cm, 250-μm coating. [b] Solvent system: A, 8:1 $CHCl_3$-$CH_3OH$; B, 6:6:1 $CHCl_3$-EtOAc-AcOH; C, 4:1:1 1-BuOH-AcOH-$H_2O$; D, 12:12:2:2:1 $CHCl_3$-EtOAc-$CH_3OH$-AcOH-$H_2O$. [c] Hand-held 254-nm UV light.

Scheme III




choice. Any proposal must utilize water as a reagent or catalyst, since $NH_3$ in ethanol (eq 7) is not effective for **10** → **8**. Any mechanism must also show loss of HCN and be consistent with the ineffectiveness of $Et_2NH$, morpholine, $Et_3N$, and pyridine as substitutes for $NH_3/H_2O$ (eq 8–10).

This does not represent an exhaustive study of the chemistry of **10**, since reaction in new combinations of solvents and reagents would probably give additional products. But, whatever the exact details may be for converting **10** to adenine either in unphotolyzed or prebiotic solutions or in preparative reactions, it is a curious circumstance that the following sequence and interplay of HCN oligomers is probably operative in both instances: tetramer ($C_4H_4N_4$, DAMN) → pentamer ($C_5H_5N_5$, **10**) → tetramer ($C_4H_4N_4$, **8**; or $C_4H_4N_4 + NH_3$, **11**) → pentamer ($C_5H_5N_5$, adenine).

## Experimental Section

**Approaches to Identification and Assay of Products from 10.** Because of the low yield and the effort required to prepare and purify **10**, many experiments using it were necessarily run using as little as 10 mg. Judicious use of TLC on fluorescent silica gel plates combined with prior knowledge of each compound's response to iodine vapor and to UV light was sufficient for positive identification of products. Where a corroborating assay or firm yield was needed, isolation by preparative TLC (Analtech GF silica gel, 20 × 20 cm, 1000-μm thickness) was employed and combined with mass spectra, UV, and/or quantitative LC.

**Analytical Procedures.** The TLC behavior of the various compounds in four solvent systems is summarized in Table II. The most generally useful mixtures were B and D.

Melting points are uncorrected and were taken on a Thomas-Hoover apparatus. Proton magnetic resonance spectra with tetramethylsilane ($Me_4Si$) as an internal standard were taken on Hitachi Perkin-Elmer R-24A, Varian A-60A, and JEOL C-60HL spectrometers by the authors or by Dr. A. Douglas and Messrs. R. Reamer and R. Zerfing. The carbon magnetic resonance spectra were taken on a Varian XL-100 spectrometer at 25.2 MHz with $Me_4Si$ as an internal standard. Elemental analyses were performed by Mr. J. Gilbert and his staff. Quantitative assay for adenine (**4**) and 4,5-dicyanoimidazole (**5**) by high-pressure LC was carried out at 254 nm by Mr. M. Davis with a Du Pont 848 chromatograph. The solid support was Water's $C_{18}$ reversed-phase μBondapak, and the liquid phase was 0.01 M $KH_2PO_4$ in 9:1 $H_2O$–$CH_3OH$ adjusted to pH 7 with NaOH which separates **4** and **5** at about 625 and 300 s, respectively.

**Evolution of $NH_4CN$ during Preparation of Adenine.** The following experimental procedure is adapted from the preparative procedure of Yamada et al.[11] Ethylene glycol was substituted for lower, more volatile alcohols to avoid contamination of the sublimate with solvent. In the absence of DAMN neither formamidine acetate nor $NH_4OAc$, alone or in combination, evolved $NH_4CN$. Although 1 mol of formamidine acetate is employed here, a similar amount of $NH_4CN$ is evolved when 2 mol of formamidine is used.

In ethylene glycol (50 mL) were placed DAMN[12] (10.8 g, 0.1 mol) and formamidine acetate (20.8 g, 0.1 mol). This mixture was heated at 121–124 °C for 1.25 h during which time a white, crystalline sublimate was collected on the surface of a dry ice condenser. This was quickly transferred under nitrogen into a tared vial. The yield of $NH_4CN$ was 2.98 g (68%); mp rapid sublimation at 30–30.5 °C; $^1H$ NMR ($Me_2SO$-$d_6$, $Me_4Si$ standard) δ 4.1 (s, $NH_4$); $^1H$ NMR ($D_2O$ exchange), 4.6 (s, HOD); $^{13}C$ NMR ($Me_2SO$-$d_6$, $Me_4Si$ standard) δ 120.2 (CN).

Anal. Calcd for $CH_4N_2$: C, 27.26; H, 9.15; N, 63.59. Found: C, 27.47; H, 8.98; N, 63.35.

By TLC adenine was the only heterocyclic product.

Analytical results from older literature: mp 36 °C;[15] the $^{13}C$ NMR for HCN (neat) is 83.7 ppm upfield from a $CS_2$ internal standard[16] which is itself 192.8 ppm downfield from $Me_4Si$; hence HCN with a $Me_4Si$ standard should have a shift of approximately 192.8–83.7 = 109.1 ppm, differing by only 11.1 ppm from that found above for $NH_4CN$.

***N*-(Aminomethylidene)diaminomaleonitrile (10).** Diaminomaleonitrile (10.8 g, 0.1 mol) and formamidine acetate (20.8 g, 0.2 mol) were placed in ethanol (300 mL) and heated at reflux under nitrogen for 40 min. The dark solution was immediately

(15) J. Gay-Lussac in "Beilsteins Handbuch der Organischen Chemie", Hauptwerke, Band II, 40-41, Syst. No. 156.
(16) G. Olah and T. E. Kiovsky, *J. Am. Chem. Soc.*, **90**, 4666 (1968).

concentrated under vacuum at ≤35 °C to a damp residue (41.5 g). The residue was extracted by stirring for 5 min with ethyl acetate (50 mL) at 50 °C followed by filtration. This extraction cycle on the residue was repeated five times and the combined filtrates were stirred for 15 min with 60–200-mesh silica gel (50 g, J. T. Baker). The silica gel was filtered and washed with ethyl acetate (150 mL). The filtrates were concentrated under vacuum at ≤30 °C to a brown residue (10 g) which was stirred with warm (50 °C) ethyl acetate (30 mL). The insolubles (2.5 g) were filtered and washed with ethyl acetate (10 mL). The combined filtrates were applied to a 1-in. stainless steel, preparative high-pressure LC column (6 ft × $^7/_8$ in. i.d.) holding 60–200-mesh silica gel (455 g). Ethyl acetate was the eluent and 100-mL fractions were collected every 30 s. Five fractions of nearly single-spot **10** were combined and concentrated to dryness. The residue was slurried in diethyl ether, filtered, and dried under vacuum to give 0.56 g of **10** as a light brown solid with a trace impurity by TLC: darkens at 125 °C, shrinks at 180 °C, and does not melt up to 300 °C; TLC (Quantum Q1-F silica gel plates, 5:1 EtOAc–$CHCl_3$, viewed by UV light and $I_2$ vapor) $R_f$ 0.75 for **10** and $R_f$ 0.5 for the impurity; UV max ($CH_3OH$) 320 nm (ε 21 100); $^1H$ NMR (300 MHz, $Me_2SO$-$d_6$) δ 7.63 (d of d, 1, CH), 7.29 (s, 2, NH), 7.26 (s, 2, NH); $^{13}C$ NMR ($Me_2SO$-$d_6$) δ 152.63 (s, $NH_2CH{=}N$), 116.71 (s, C-3), 116.03 (s, C-4), 115.00 (s, C-1), 106.99 (s, C-2); mass spectrum $m/e$ 135 ($M^+$).

Anal. Calcd for $C_5H_5N_5$: C, 44.44; H, 3.73; N, 51.83. Found: C, 44.93; H, 3.75; N, 51.11.

**Shelf Stability of Solid 10.** After storage for 9 months under nitrogen **10** was assayed by UV spectroscopy in methanol. A small amount of black, methanol-insoluble material was present and the UV absorption at 320 nm had decreased to ε 17 522. From this a half-life of 2.2 years was calculated.

***p*-Toluenesulfonic Acid Salt of 10.** In THF (3 mL) 27 mg (0.2 mmol) of **10** and 38.7 mg (0.204 mmol) of *p*-TsOH·$H_2O$ were stirred for 0.75 h at 25 °C. A small amount of insoluble material was filtered off. Diethyl ether (1.5 mL) was added to the filtrate and white crystals were separated over 30 min. Three milliliters more of ether was added slowly, and the product was filtered, washed with ether, and dried to give 4.97 mg (81%): darkens at 125 °C, melts at 150–155 °C; UV max ($CH_3OH$) 320 nm (ε 19 000); $^1H$ NMR ($Me_2SO$-$d_6$) δ 2.3 (s, 3, $CH_3$), 7.35 (q, 4, ArH), 7.9–8.3 (m, 3, 1 amidine CH and 2 NH), 9.3–10.1 (m, 2, NH); $^1H$ NMR ($CD_3OD$ exchange) δ 4.05 (s, 5, $CD_3OH$).

Anal. Calcd for $C_{12}H_{13}N_5O_3S$: C, 46.89; H, 4.26; N, 22.79; S, 10.44. Found: C, 46.82; H, 4.30; N, 22.09; S, 10.70.

This salt, just as **10**, slowly darkens after several months of storage.

**Stability of 10 in 1-Butanol at 100 °C.** Compound **10** (10 mg) was placed in 1-butanol (1.5 mL) and heated at 100 °C for 1 h. Inspection by TLC (5:1 EtOAc–$CHCl_3$) showed a slight diminution in spot intensity and no evidence of new, mobile materials. The sample was diluted with methanol and at UV max 320 nm gave ε 15 025, equivalent to 71.3% of unconverted **10**.

**Conversion of 10 to Adenine (4).** Under nitrogen a mixture of **10** (27 mg, 0.2 mmol), formamidine acetate (20.8 mg, 0.2 mmol), and $NH_4OAc$ (15.4 mg, 0.2 mmol) was placed in 1-butanol (1 mL) and heated at 100 °C for 2 h. By TLC **10** was wholly converted in 30–45 min, and the major product was **4** with a smaller amount of **5**. By LC the amount of adenine was 6.8 mg (25%) and that of **5** was 2.6 mg (11%).

**Isolation of 4-Aminoimidazole-5-carboxamidine Dihydrochloride (11·2HCl).** In 1-butanol (160 mL) were placed DAMN (21.6 g, 0.2 mol), formamidine acetate (41.6 g, 0.4 mol), and $NH_4OAc$ (8 g, 0.104 mol). With stirring this mixture was heated rapidly and held at 105 °C for 5 min. It was cooled and TLC (system C, Table II) showed adenine and amidine **11**. No **8** was observed. The reaction mixture was chromatographed through 1 kg of 60–200-mesh silica gel (J. T. Baker) and eluted with a slight vacuum, using 8:1:1 2-PrOH–AcOH–$H_2O$. Cuts were made at 250-mL intervals, and cuts 3–9 were combined. Evaporation to dryness under vacuum gave 60 g of a viscous residue containing both **11** and **4**. This residue was rechromatographed through 800 g of silica gel (7:1 $CHCl_3$–$CH_3OH$), and 20–25-mL fractions were collected. Fractions 41–80 gave 5.6 g of crude solids containing 2.2 g (8.1%) of adenine measured by quantitative LC. Fractions 136–260 were combined, concentrated under vacuum, slurried in 2-PrOH (150 mL), and filtered through Celite to remove salts. The filtrate was reconcentrated to give 24.25 g of dark solids. This residue was dissolved in a solution of EtOH (150 mL) and concentrated HCl (30 mL). After 15–20 min the resulting brown solid was filtered, washed with ethanol and ether, and dried: yield of **11**·2HCl 7.13 g (18%); mp 256–257 °C dec (lit.[17] mp 242–244 °C (dec); UV max ($H_2O$) 284 nm (ε 10 800) (lit.[18] UV max ($H_2O$) 287 nm (ε 10 700); IR (Nujol), no C≡N at 2200 $cm^{-1}$; $^1H$ NMR ($Me_2SO$-$d_6$) δ 8.65 (s, 1, $C_2$-H), 8.85 (s, 3, $NH_2$ and $N_3$-H), 10.6 (s, 5, amidine and HCl); $^1H$ NMR ($D_2O$ exchange) δ 8.4 (s, 1, $C_2$-H), 4.8 (s, 8 HOD); $^{13}C$ NMR ($Me_2SO$-$d_6$) δ 99 ($C_5$), 133 ($C_2$), 144 ($C_4$), 155 (amidine); mass spectrum $m/e$ 125 ($M^+$), 108 ($M^+$–$NH_3$, 125–17).

Anal. Calcd for $C_4H_9N_5Cl_2$: C, 24.26; H, 4.58; N, 35.36; Cl, 35.81. Found: C, 24.33; H, 4.51; N, 35.25; Cl, 34.97.

**Limit of Detection of 8.** One microliter of a stock solution of **8** (1.35 mg/mL of $CH_3OH$) applied to a TLC plate and eluted with solvent system D gave a readily observed spot in iodine vapor. This concentration corresponds to a 0.1% yield of **8**, if it were formed in the preceding experiment.

**Conversion of 10 to 4-Aminoimidazole-5-carboxamidine (11).** In 1-butanol (2 mL) **10** (54 mg, 0.4 mmol), glacial acetic acid (24 μL, 25.2 mg, 0.419 mmol), and $NH_4OAc$ (62.5 mg, 0.811 mmol) were heated at 100–105 °C for 2 h. By TLC (system B, Table II) the major product was **11** with a trace of adenine and complete conversion of **10**. The reaction mixture was separated on two preparative TLC plates (system C, Table II). The band corresponding to **11** was removed and washed with ethanol to give 42 mg of residue after evaporation of the filtrate. By UV analysis (UV max 280 nm, 0.1 N HCl) this contained 12.5 mg (25% yield) of **11**: mass spectrum $m/e$ 125 ($M^+$).

**Stability of 11·2HCl in aqueous $NH_3$.** 4-Aminoimidazole-5-carboxamidine dihydrochloride (**11**·2HCl; 22.3 mg, 0.113 mmol) was dissolved in 28% $NH_4OH$ (1.5 mL, 27.5 mmol). By TLC no **8** or other new materials were detected after 30 min. Ammonia and most of the water were removed at a vacuum pump. The residue was dissolved in methanol and appeared unchanged by TLC and UV analysis (UV max 284 nm (ε 10 000). For the starting material ε was 10 800.

**Conversion of 10 to 4-Amino-5-cyanoimidazole (8).** Compound **10** (9.2 mg, 0.068 mmol) was dissolved in concentrated $NH_4OH$ (1 mL of 28–30%) and sampled at ambient temperature for TLC (system B, Table II). At 5 min a small amount of **10** was present, but at 7 min it was wholly converted to material at $R_f$ 0.25. The latter product, in turn, was slowly replaced by **8** until it was the sole product observed after 40 min of reaction. The TLC spot of the final product turned yellow on standing for several hours, a characteristic of authentic **8**. Neither **11** nor **5** was evident. Assayed by UV analysis, the yield of **8** was 71% (UV max 234 nm, 0.1 N HCl in $CH_3OH$). The UV spectrum also showed the very pronounced and characteristic shoulder for **8** at 250–254 nm.[19]

**Stability of 8 to AcOH/$NH_4OAc$/1-BuOH.** 4-Amino-5-cyanoimidazole[19] (**8**; 100 mg, 0.93 mmol), AcOH (110 μL, 115 mg, 1.92 mmol), and $NH_4OAc$ (286 mg, 3.72 mmol) were placed in 1-butanol (3 mL) and heated from 25 to 100 °C within 3 min. Samples for TLC were taken at 5, 30, and 60 min, and no trace of amidine **11** or any other new product was detected; only **8** was present. Compound **8** was diluted with 0.1 N in methanol for UV analysis and observed were the expected maximum at 230 nm (ε 10 638; expected 10 380) and the shoulder at 250 nm (ε 7776; expected 7520). A solution of known concentration of **11** was used to estimate that a 1% yield of **11** would have been detected by TLC.

**Conversion of 10 to 4,5-Dicyanoimidazole (5).** In 1-butanol (2 mL) **10** (54 mg, 0.4 mmol) and glacial acetic acid (24 μL, 25.2 mg, 0.419 mmol) were heated at 100–105 °C for 2 h. By TLC (system B, Table II) **5** was the major product, a trace of minor product at $R_f$ 0.5 (adenine?) was present, and **10** was completely converted. The mixture was evaporated under vacuum and the residue dissolved in methanol (1 mL). This solution was applied

(17) E. Shaw, *J. Biol. Chem.*, **185**, 439 (1950).
(18) L. F. Cavalieri, J. F. Tinker, and G. B. Brown, *J. Am. Chem. Soc.*, 71, 3973 (1949).
(19) Y. Yamada, I. Kumashiro, and T. Takenishi, *Bull. Chem. Soc. Jpn.*, **41**, 241 (1968).

to a preparative TLC plate (system B), the band corresponding to **5** was removed, and 19.8 mg of solids was eluted with ethanol. By UV analysis (UV max 244 nm, 0.1 N HCl) this contained 9.7 mg (21% yield) of **5**:[20,21] mass spectrum $m/e$ 118 ($M^+$).

**Conversion of 10 to *N*-Formyldiaminomaleonitrile (9).** In methanol (50 mL) were dissolved **10** (6.3 mg, 0.0467 mmol) and *p*-toluenesulfonic acid monohydrate (8.82 mg, 0.047 mmol). A portion of this solution was diluted further with methanol, and the UV spectrum showed a broader peak with a maximum at 310 nm. After 20–30 min a portion of the original 50-mL solution was diluted and showed a sharp maximum at 289 nm ($\epsilon$ 12900) compared to 289 nm ($\epsilon$ 12200) for authentic **9**.[13] In a more concentrated (1 mL) methanolic solution **10** (~6 mg) is wholly converted as observed by TLC (solvent A) in ≤1 h to **9** under the influence of equimolar *p*-TsOH·$H_2O$.

**Stability of 10 to $NH_3$ in EtOH.** Compound **10** (9.8 mg) was dissolved in 10% $NH_3$ in ethanol (1.5 mL). After 40 min at ambient temperature no change was observed by TLC (solvent mixture A).

**Stability of 10 to $Et_3N/CH_3OH/H_2O$.** Compound **10** (9.5 mg, 0.0705 mmol) was dissolved in 2 mL of 1:1:1 $Et_3N$ (~4.8 mmol)–$H_2O$–$CH_3OH$ and stirred at 25 °C. Over 30 min two new materials were detected by TLC (solvent mixture B) at $R_f$ 0.15 and 0.25, but nothing corresponding to **8** or **11** was observed. No attempt was made to isolate the products which proved stable over 18 h after $NH_4OH$ (1 mL) was added to the solution.

**Stability of 10 to Pyridine and $Et_3N$.** Compound **10** (9.6 mg, 0.0711 mmol) was dissolved in pyridine (1 mL) and samples for TLC were withdrawn. No change in the TLC was observed for up to 3 h. Triethylamine (1.5 mL) was added and no change was observed by TLC after 30 min at ambient temperature.

**Stability of 10 to Morpholine and Diethylamine.** Compound **10** (10 mg, 0.074 mmol) was dissolved in a mixture of water (1 mL) and ethanol (0.5 mL) and after 2 h no change was noted by TLC. Morpholine (29.6 mg, 0.34 mmol) was added and stirred for 4 h without any change. Diethylamine (1 mL, 0.71 g, 9.7 mmol) was added with immediate darkening and conversion of **10** to material at the origin by TLC (system B).

**Registry No.** **2**, 1187-42-4; **4**, 73-24-5; **5**, 1122-28-7; **8**, 5098-11-3; **9**, 53144-01-7; **10**, 71749-37-6; **10** *p*-toluenesulfonate salt, 71749-38-7; **11**, 7269-66-1; **11**·2HCl, 71749-39-8; hydrocyanic acid, 74-90-8; ammonium cyanide, 12211-52-8; formamidine acetate, 40730-94-7.

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# Bicyclic Guanidino Ketones

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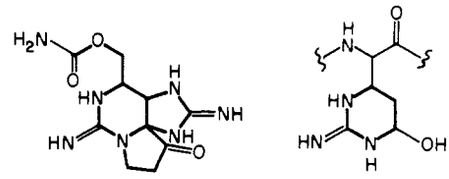
Syntheses of bicyclic [3.3.1] guanidino ketones are described by methods that permit the preparation of compounds substituted at the endo- and exocyclic nitrogen as well as at C-4. The primary synthetic method involved the synthesis of substituted and protected 3-aminopiperidones and their subsequent cyclization with *S,S*-dimethyl *N*-(*p*-toluenesulfonyl)dithiocarbonimidate to the substituted and protected bicyclic guanidino ketones. The ketones, blocked as the ethylene ketals, were deblocked by transketalization with cyclohexanone while the guanidine, blocked by tosylation, was deblocked by liquid HF.

Recent reported examples of natural products containing the guanidino functionality as part of a ring system include the puffer fish poison tetrodotoxin,[1] the paralytic shellfish poison saxitoxin (**1**),[2-4] the peptide antibiotics capreomycin,[5] viomycin (**2**),[6] and tuberactinomycin,[7] the antifungal agent stendomycin,[8] and the alkaloids of *Alchornea jovanensis* (**3**).[9] Since the available methods for the preparation of functionalized cyclic guanidines are limited, it was of interest to develop such methods and to prepare a series of functionalized cyclic guanidines in order to study



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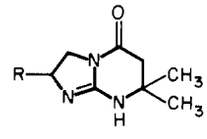

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their chemistry and to examine their biological activity. We present at this time the synthesis of a number of bicyclic [3.3.1] guanidino ketones.

The first compound sought was the monocyclic guanidino ketone 1,3-dimethyl-2-imino-5-oxohexahydropyrimidine hydrochloride (**8**) (Scheme I), and its preparation was attempted by using the ethyl ester of sarcosine (**5a**). Good yields of diester **6a** and the cyclic salt **7a** were obtained, but deblocking and decarboethoxylation gave only a poor yield of **8**. An alternative procedure, whereby

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