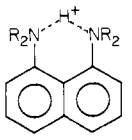
Table I. Thermodynamics of Deprotonation of Aromatic Amines in Two Alkali Superbase Systems and the Gas Phase at 25 °C

		K^+DMSYL^-/Me_2SO				KAPA	gas phase	
compd	pK_a[a]	$\Delta G°$ [a,e]	ΔH_D[b,e]	$\Delta H°_i$[b,e]	$\Delta S°_i$[f]	ΔH_D[c,e]	$\Delta G°_{acid}$[d,e]	$\delta\Delta G°_{acid}$[e]
II	24.8	+33.8	−12.3 ± 0.8	35.7 ± 1.1	6.3 ± 3.7	−28.7 ± 0.5	346.6 ± 2	−13.2 ± 0.5
isopropyl mercaptan			−28.5 ± 0.3	19.5 ± 0.9			349.3 ± 2	−10.5 ± 0.5
m-chloroaniline	28.5	+38.9	−11.0 ± 0.5	37.0 ± 0.9	−6.3 ± 2.7		353.8 ± 2	6.0 ± 0.5
I	29.2	+39.8	−8.5 ± 0.4	39.5 ± 0.9	−1.1 ± 3.0	−28.7 ± 0.5	351.9 ± 2	−7.9 ± 0.5
aniline	30.7	+42.0	−7.1 ± 0.2	40.9 ± 0.8	−3.7 ±	−14.5 ± 0.8	359.8 ± 2	0

[a] pK_a's for aniline and *m*-chloroaniline are from ref 1b. [b] Arnett, E. M.; Small, L. E. *J. Am. Chem. Soc.* 1977, *99*, 808. $\Delta H°_i$ and ΔH_D are defined in this reference. [c] For earlier work on ΔH_D's in KAPA see ref 7. [d] $\Delta G°_{acid}$ for isopropylmercaptan and *m*-chloroaniline are from ref 2. $\Delta G°_{acid}$ is the standard Gibbs free energy change for the reaction $AH = A^- + H^+$ at 298 K. [e] Values in kcal/mol. [f] Values in gibbs/mol. $\Delta S_i = [(\Delta H_i - 1.364)/298.15] \times 10^3$

Sponge is a reasonable driving force for its enhanced basicity since neither unmethylated II nor the mono-, di-, or trimethylated homologues are nearly as basic although they should all be capable of forming a peri hydrogen bonded ammonium ion (IV). However, if relief of electron repulsion is a driving force for protonation, it is hard to see how it can also promote deprotonation.



IV

As noted before,[8] a fairly close parallel is found between enthalpies and free energies of ionization for many weak acids in $DMSYL^-/Me_2SO$ and the compounds in Table I fall close to the $\Delta G°_i/\Delta H°_i$ correlation line. The entropy differences between I, II, and aniline are probably statistically significant but do not merit interpretation in such a complex system.

A second point of interest concerning the interplay of deprotonation and homoconjugation is shown in the pattern of acidities for I, II, and aniline in the superbase KAPA[9] which is strikingly different from that in K^+DMSYL^-. Since KAPA is the stronger base by 10^5–10^6 times,[10] we propose that both I and II lose two protons in KAPA in contrast to aniline which loses but one. Since the dianion of II should not be able to enjoy stabilization from an internal hydrogen bond, there is no reason why the difference between the ΔH_D of I and II seen in $DMSYL^-/Me_2SO$ should be repeated in KAPA.

Intramolecular hydrogen-bonding stabilization of oxyanions by neighboring hydroxyl or carboxylate groups has been well demonstrated in dipolar aprotic media,[11] and we have found a difference of 4 pK_a units between catechol and hydroquinone in Me_2SO. A rather delicate trade-off between the acid–base and hydrogen bond donor and acceptor properties of the intramolecular functions vs. those of the external medium may be required as shown by our results in Me_2SO and KAPA and those of Kolthoff and Chantooni.[11a-c]

The gas-phase acidities were obtained by using pulsed ion cyclotron resonance to produce monodeprotonation in every case as shown by monitoring of the M − 1 peak from the parent. The method of stair-step comparisons[7] was required to cover the large differences between aniline, I, and II. Clearly, the acidity differences seen in Me_2SO are increased greatly in the gas phase. We attribute the greater gas-phase acidity difference between I and aniline to the larger polarizable π system of the former which should be more effective for anion stabilization in the gas phase than in solution where polarization of the medium can attenuate charge dispersal.

The difference between ΔH_D for I and II is nearly the same in Me_2SO as in the gas phase which is consistent with the above argument since I and II have π systems of nearly equal size. The enormous difference between the gas-phase and liquid-phase values for deprotonation of the mercaptan relative to the aromatic system is again to be expected on the basis of polarizability.

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Registry No. I, 2243-62-1; II, 479-27-6; isopropylmercaptan, 75-33-2; *m*-chloroaniline, 108-42-9; aniline, 62-53-3; K^+DMSYL, 15590-26-8; KAPA, 54856-92-7.

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Chemical Excision of Apurinic Acids from RNA. A Structurally Modified Yeast $tRNA^{Phe}$

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Apurinic (apyrimidinic) acids are of considerable importance in the chemistry and biochemistry of nucleic acids. Such species can be formed chemically from DNA and RNA at high temperature and extreme pH[1] and by the action of mutagens;[2] their potential role in cell lethality[3] and mutagenesis[2] is suggested strongly by the existence of endonucleases that mediate incision (and permit subsequent repair) at such lesions.[4] Apurinic acids are also key intermediates in nucleic acid sequencing,[5] can be formed selectively from certain modified nucleosides in RNA's,[6]

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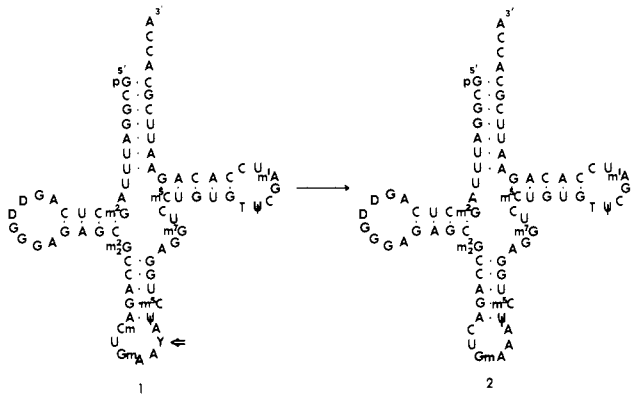
and have been employed for site-specific fluorescent labeling of RNA's.[7]

Much of the interest in apurinic acids derives from their lability in the presence of, e.g., acid and base, and especially from the nucleic acid strand scission that accompanies their transformation.[5,8] Although the products of these processes have not been characterized in detail, for RNA they are believed to include a 3′-oligomer terminating with a 5′-phosphate group and a 5′-oligomer terminating with 4,5-dihydroxy-2-oxovaleraldehyde[9] (v, Scheme I). There is no method available for concomitant excision of this apurinic acid moiety from the 5′-RNA oligomers, in spite of the potential utility of the product in RNA modification schemes.[10]

Presently, we analyze the problem of apurinic acid excision from RNA. Experiments based on this analysis affirm the intermediacy of iv in Scheme I and provide a generally applicable procedure for effecting the requisite transformation. A key feature is believed to involve stabilization of an otherwise disfavored enolic intermediate via H bonding to the ring N atom of 2-aminopyridine. The utility of this procedure for RNA modification is illustrated by depurination of nucleoside 37 in yeast $tRNA^{Phe}$ (**1**) and conversion of the depurinated tRNA to a species (**2**) having only six nucleotides in the anticodon loop.



Depurinated RNA undergoes strand scission in aqueous aniline–HCl (pH 4.5), a process that undoubtedly involves initial formation of C-1′ imine(-enamine) iii.[11] The acidity of C-2′H in the derived imine would be expected to facilitate base-catalyzed strand scission, as shown. Following strand scission, C-4′H in the newly formed enol (iva) would bear a vinylogous relationship to C-2′H in iii and should, in principle, provide access to the 5′-oligomer lacking the 4,5-dihydroxy-2-oxovaleraldehyde moiety via a second β elimination. However, consistent with the expectation that iv would exist largely as keto tautomer ivb,[12] the product of this β elimination is not observed.

To facilitate a study of the transformation iva $\rightleftharpoons$ ivb, we prepared methyl guanylyl(3′→5′)-7-methylguanosine 3′-phosphate (**3**).[13] Depurination of **3** according to known methods[6a,6b,14] provided model "oligomer" **4**, containing a single, defined apurinic site. Incubation of **4** in 0.5 M aniline–HCl at pH 4.5 (5 days, 45 °C, N_2) effected facile conversion to vi (Scheme II, X = CH) but provided little 3′-GMP.[12,15] In an effort to influence the equilibrium concentration of vib, and thereby provide access to 3′-GMP via β elimination, the experiment was repeated in the presence of 2-aminopyridine. After 5 days of incubation at 45 °C, 3′-GMP was isolated (polyethyleneimine TLC, 1 M LiCl) in 98% yield;[16] its formation must reflect stabilization of vib (X = N) by H bonding of the enolic hydrogen to the ring N atom of 2-aminopyridine.[17]

The suitability of this procedure for the excision of apurinic acids from RNA was studied by using purified yeast $tRNA^{Phe}$, which contains an acid-labile nucleoside at position 37 [i, R = $CH_2CH_2CH(COOCH_3)NHCOOCH_3$].[6d,6e,18] Following depurination of nucleoside 37, treatment with a solution containing 0.5 M 2-aminopyridine and 0.25 M aniline, pH 5.5, at 45 °C (60–72 h, N_2) effected clean strand scission; polyacrylamide gel electrophoresis revealed the disappearance of intact $tRNA^{Phe}$ and the appearance of two sharp new bands of similar mobility, having molecular weights about half that of the original tRNA. Purified bacterial alkaline phosphatase removed all of the terminal phosphate groups from the tRNA fragments. After replacement of the 5′-phosphates by incubation with polynucleotide kinase and ATP,[19] the anticodon loop was "resealed" quantitatively by incubating the reannealed half-molecules in the presence of RNA ligase and ATP.[20] Consistent with the absence of a nucleotide in the anticodon loop, tRNA **2** had slightly greater mobility on polyacrylamide gels than did tRNA **1**.[21]

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(10) Interestingly, the 5′-oligomers resulting from cleavage at apurinic sites in DNA can be so modified,[5,8] presumably because the initially formed α,β-unsaturated valeraldehyde is susceptible to a second β elimination. See: Bayley, C. R.; Brammer, K. W.; Jones, A. S. *J. Chem. Soc.* **1961**, 1903.

(11) An analogous imine has been formed by admixture of ethidium bromide to a sample of depurinated $tRNA^{Phe}$.[7]

(12) The representation of intermediate iv in Scheme I (and vi in Scheme II) is oversimplified. It must be an equilibrium mixture of enol and keto species in both cyclic and acyclic forms.

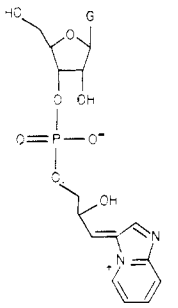
(13) Prepared by *N,N′*-dicyclohexylcarbodiimide-mediated condensation of 2′,5′-di-*O*-(1-methoxyethyl)guanosine 3′-phosphate (Mackey, J. K.; Gilham, P. T. *Nature (London)* **1971**, *233*, 551) and methyl 2′-*O*-(1-methoxyethyl)-7-methylguanosine 3′-phosphate. The latter was prepared via 2′-*O*-(1-methoxyethyl)guanosine cyclic 3′,5′-phosphate by successive treatments with snake venom and dimethyl sulfate (pH 6.5–7.5); the overall yield was approximately 70%. Deblocking of the dinucleotide was carried out in dilute aqueous acetic acid.

(14) (a) Wintermeyer, W.; Zachau, H. G. *FEBS Lett.* **1975**, *58*, 306. (b) Hecht, S. M.; Arcari, P.; Adams, B. L. *J. Am. Chem. Soc.* **1977**, *99*, 3890. (c) Arcari, P.; Hecht, S. M. *J. Biol. Chem.* **1978**, *253*, 8278.

(15) As judged by the spectral (UV, NMR) characteristics of the isolated intermediates and their chromatographic properties relative to authentic standards. The intermediate assigned structure vi was isolated by chromatography on a small DEAE-cellulose column and shown to contain an aniline moiety.

(16) The isolated 3′-GMP was characterized by comparison with authentic 3′-GMP (polyethyleneimine TLC, paper chromatography) and by conversion to a product identical with guanosine upon treatment with alkaline phosphatase. The formation of 3′-GMP was found to proceed optimally in the presence of both 2-aminopyridine and aniline; the more strongly basic nature of the latter appears to facilitate the requisite β eliminations.

(17) It is also conceivable that 3′-GMP may have arisen via intermediate vii, although the facility with which this species could form is unclear.



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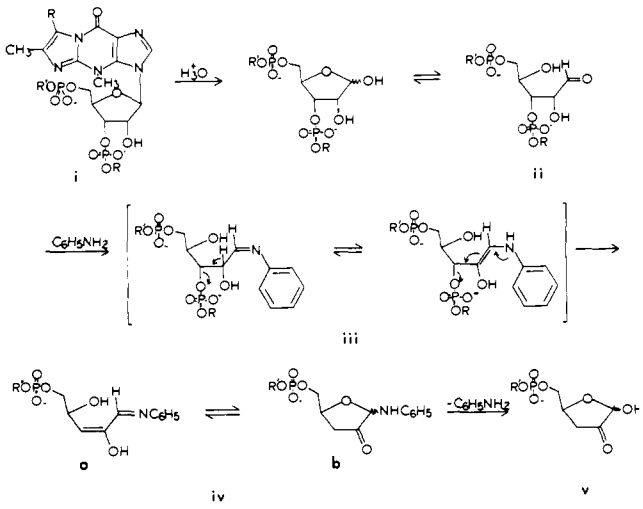
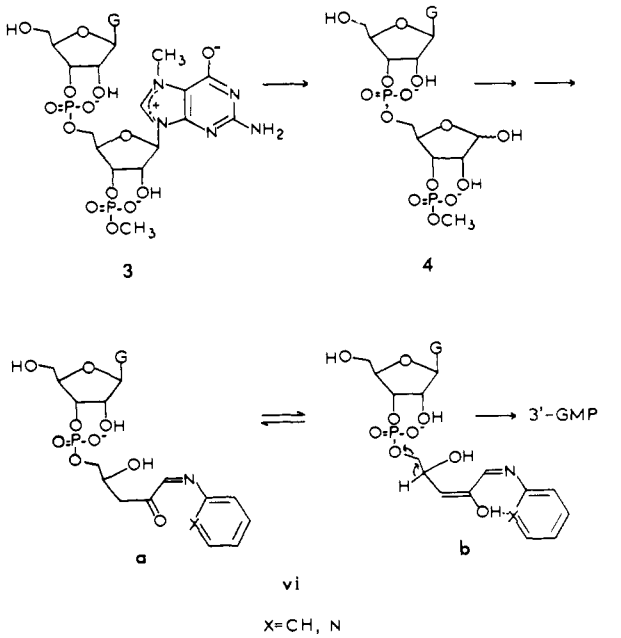
(20) A similar transformation has been reported previously (Kaufmann, G.; Littauer, U. Z. *Proc. Natl. Acad. Sci. U.S.A.* **1974**, *71*, 3741), albeit in extremely low yield.

(21) When the polynucleotide kinase reaction was carried out by using γ-^{32}P-labeled ATP, subsequent ligation of the fragments produced a tRNA-like species containing alkaline phosphatase-resistant radiolabel. The structure of **2** was verified by nucleotide sequence analysis.

Scheme I



Scheme II



The chemical transformations described herein provide a basis for the modification of intact RNA's and for the characterization and manipulation of species following alteration, e.g., by mutagens. In the present case the alterations afforded a highly unusual tRNA species (**2**) having only six nucleotides in the anticodon loop. This tRNA is of considerable interest for biochemical studies, e.g., of the structural features in $tRNA^{Phe}$ that permit interaction with[22] and conformational activation of[23] its cognate aminoacyl-tRNA synthetase. Diminution in the size of the anticodon loop was found to have a dramatic effect on certain biochemical properties of $tRNA^{Phe}$.[24]

Acknowledgment. We thank Professor Glenn McGarvey for a helpful discussion during the course of this work. This investigation was supported by PHS Research Grant GM27815.

Registry No. Nucleoside Y, 55196-46-8.

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A Mixed-Metal Nitrido Carbonyl Cluster Compound. Synthesis and X-ray Structure of the $[PtRh_{10}N(\mu\text{-}CO)_{10}(CO)_{11}]^{3-}$ Anion

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Two of the newly growing fields in the chemistry of carbonyl cluster compounds concern the studies of novel mixed-metal species and clusters containing new types of interstitial atoms. We have recently reported the synthesis and structure of the first cluster compounds containing interstitial nitrogen atoms, namely, the $[M_6N(CO)_{15}]^-$ anions (M = Co and Rh)[1] and we wish to describe here the characterization of the first example of a mixed-metal cluster compound containing an interstitial nitrogen atom, the anion $[PtRh_{10}N(CO)_{21}]^{3-}$. This species exhibits many interesting features because (i) it is an unusual example of an 11-metal cluster compound, (ii) it is a new step in the chemistry of mixed platinum–rhodium clusters which were up to now limited to five[2] or six[3] metal species, (iii) it is the compound with the highest metal-to-interstitial atom ratio presently known, and (iv) it is a good example for comparison of the bonding properties of interstitial nitrides vs. carbides[4] in large metal atom clusters.

The $[PtRh_{10}N(CO)_{21}]^{3-}$ anion has been first obtained as a byproduct in the synthesis of $[Rh_6N(CO)_{15}]^-$ from K_3RhCl_6 containing traces of platinum salts. It is now prepared by reaction of $[Rh_6N(CO)_{15}]^-$ with $[PtRh_4(CO)_{14}]^{2-}$:[2]

$$[Rh_6N(CO)_{15}]^- + [PtRh_4(CO)_{14}]^{2-} \xrightarrow[N_2,\ \text{reflux}]{\text{acetone}} [PtRh_{10}N(CO)_{21}]^{3-} + \text{other products}$$

The reaction is slow and, due to the concomitant decomposition of the starting anions,[5] gives a mixture of products from which the mixed-metal anion has to be separated by fractional precipitation of the potassium salts.[6] The corresponding cesium and bulky organic cation salts can be obtained by metathesis in water and aqueous alcohols, respectively.

The IR spectrum of the $[N(CH_3)_4]^+$ salt in CH_3CN solution shows bands at 1988 vs, 1952 m, 1812 m, 1800 sh, and 1770 sh cm^{-1}.

The nitrogen nature of the interstitial atom is proved by the synthetic reaction itself and further by the reaction of the

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(5) It is very probable that the real Pt furnishing species is the $[PtRh_4(CO)_{12}]^{2-}$ anion, which is quickly formed from the $[PtRh_4(CO)_{14}]^{2-}$ anion by loss of CO[2] and which is more stable under our reaction conditions. The $[Rh_6N(CO)_{15}]^-$ anion also undergoes slow thermal decomposition to give new nitrido species.[11]

(6) In a typical synthesis $(PPN)_2[PtRh_4(CO)_{14}]$ (0.29 g) and $K[Rh_6N(CO)_{15}]$ (0.15 g) are refluxed in acetone (20 mL) under nitrogen until the IR band of the bridging COs at 1873 cm^{-1} of the $[Rh_6N(CO)_{15}]^-$ anion disappears (~25 h). The mixture of products is then transformed into the sodium salts by addition of $NaB(C_6H_5)_4$ (0.24 g), evaporation to dryness in vacuum, and extraction with water (10 mL). The excess $NaB(C_6H_5)_4$ is eliminated by addition of 3% aqueous KCl (10 mL); the solution is filtered, briefly pumped in vacuum to eliminate residual traces of acetone, and treated with solid KCl up to an 8% KCl concentration. The potassium salt precipitates as a powder which is filtered, washed first with 6.5% aqueous KCl (3 mL) and then with saturated aqueous KBr, and finally vacuum dried; yield 10–20%. The dark violet solutions of the anion are quickly oxidized in air.

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