

Vapor Spectra and Heats of Vaporization of Some Purine and Pyrimidine Bases¹

by Leigh B. Clark, Gary G. Peschel, and Ignacio Tinoco, Jr.

*Chemistry Department and Lawrence Radiation Laboratory, University of California, Berkeley, California
(Received May 18, 1965)*

The vapor spectra of some purine and pyrimidine bases are presented. Vapor pressures of adenine and 9-methyladenine have been determined in order to obtain absolute intensities. Heats of vaporization are also determined from the spectral data.

Introduction

The spectra of the purine and pyrimidine bases found in nucleic acids have been extensively studied in aqueous solution where strong hydrogen bonds may occur.^{2,3} These spectra show no vibronic structure and even the number of electronic absorption bands present are not established. In less polar solvents such as acetonitrile,^{4,5} methylcyclohexane,⁵ and trimethyl phosphate³ some resolution becomes apparent. We expected that vapor phase spectra might give even better resolution, therefore we have investigated the main nucleic acid bases and their more volatile methyl derivatives. However, the vapor spectra closely resemble the solution spectra and do not show any new bands. The vapor phase studies do give the spectra of the isolated bases, and the temperature dependence of the spectra provides heats of vaporization.

Experimental Section

Adenine, cytosine, guanine, uracil, and 1,3-dimethyluracil were California Biochemical Corp. Cfp grade products. The latter two were recrystallized from water. 9-Methyladenine and 9-methylhypoxanthine were purchased from Cyclo Chemical Corp. and were recrystallized three times from water. Trimethyl phosphate was from Victor Chemical Co.; methylcyclohexane and acetonitrile were Matheson Spectrograde.

A double-windowed, quartz, 10-cm. absorption cell was covered with a layer of thin copper sheeting. Heat was supplied *via* resistance wire wound onto the cell body and insulated with asbestos. The temperature was measured by a copper-constantan thermocouple placed in contact with the cell.

Approximately 20 mg. of sample was inserted into the cell through a side arm. The cell was then evacuated (pressure $<10^{-5}$ mm.) for about 12 hr. The temperature of the cell was then slowly raised over a period of several hours until it was evident that the base itself was evaporating. The cell was allowed to cool and sealed, and the side arm was wound with heating wire so that its temperature would be a few degrees above that of the cell body.

The spectra were recorded with a Cary Model 15 spectrophotometer. An unheated double-windowed cell was used as a blank. Temperature increments of successive scans varied from 10 to 20°; spectra were recorded both upon heating and cooling to test for decomposition.

The vapor pressures of adenine and 9-methyladenine were measured by the Knudsen effusion method. The values (mm.) obtained are $5.1 \pm 0.5 \times 10^{-3}$ at 175° and 24×10^{-3} at 200° for adenine, and $0.92 \pm 0.05 \times 10^{-2}$ at 140°, $10.4 \pm 0.5 \times 10^{-2}$ at 170°, and $29.5 \pm 0.4 \times 10^{-2}$ at 185° for 9-methyladenine.

Results and Discussion

Vapor Spectra. The vapor spectra of adenine, 9-methyladenine, 9-methylhypoxanthine, uracil, and 1,3-dimethyluracil are given in Figures 1–3. These spectra are in general very similar in shape to those seen in

(1) Supported in part by Public Health Service Research Grant GM 10840 and by the U. S. Atomic Energy Commission.

(2) D. Voet, W. B. Gratzner, R. A. Cox, and P. Doty, *Biopolymers*, **1**, 193 (1963).

(3) L. B. Clark and I. Tinoco, Jr., *J. Am. Chem. Soc.*, **87**, 11 (1965).

(4) R. Haselkorn, Ph.D. Thesis, Harvard University, 1959.

(5) E. Charney and M. Gellert, *Biopolymers, Symp.*, **1**, 469 (1964).

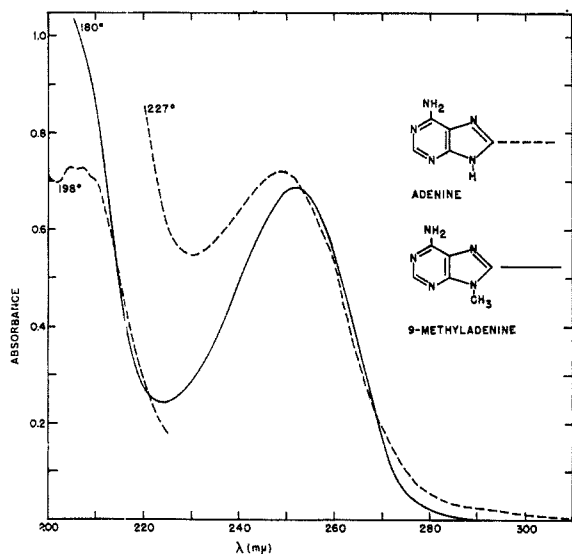


Figure 1. The vapor phase spectra of adenine and 9-methyladenine.

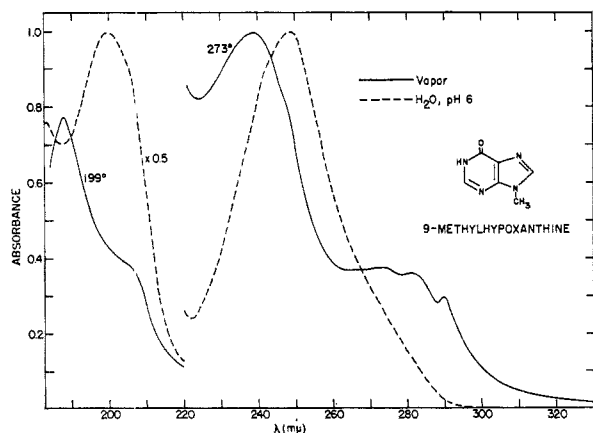


Figure 2. The spectrum of 9-methylhypoxanthine in the vapor phase and in neutral solution in water.

aqueous solution. Only in 9-methylhypoxanthine is a new band seen which is not found in the aqueous spectrum (Figure 2); however, this band is present in trimethyl phosphate solution.³ The main electronic band of each compound is shifted to the red by about 10 $m\mu$ in the aqueous solution spectrum compared with the vapor spectra. This is commonly found for $\pi-\pi^*$ transitions; it is interpreted as a greater London attraction of the solvent for the excited state than the ground state of the molecule.⁶ Table I gives the wavelength maxima for these components in the vapor and in solvents of increasing polarity. The main band is seen to shift uniformly with solvent polarity (from methylcyclohexane to water).

No bands which could be attributed to $n-\pi^*$ transi-

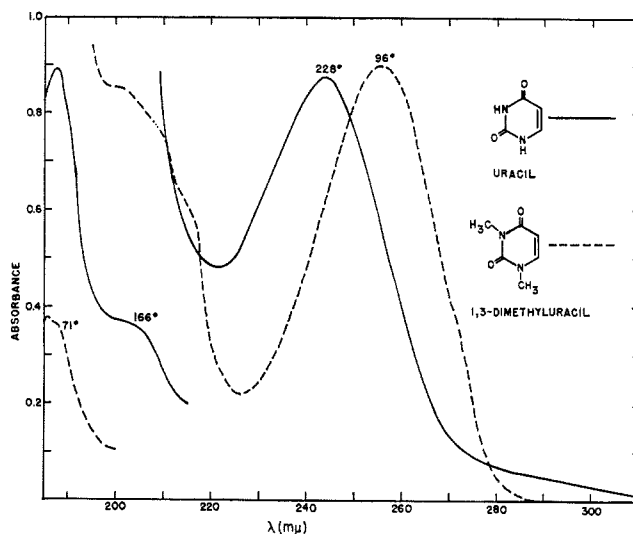


Figure 3. The vapor phase spectra of uracil and 1,3-dimethyluracil.

tions were found. These bands would be expected to shift in the opposite direction to the $\pi-\pi^*$ (the solvent prefers the ground state of the $n-\pi^*$ transition) and be easier to see in the gas phase. Their absence indicates that the $n-\pi^*$ transitions remain hidden under the $\pi-\pi^*$ transitions, or that their intensity is less than one-twentieth that of the $\pi-\pi^*$.

Extinction coefficients were obtained for adenine and 9-methyladenine from the vapor pressure data. At the maxima these are λ 249 $m\mu$ (ϵ 12×10^3 l./mole cm.) for adenine at 227° and λ 252 $m\mu$ (ϵ 7.7×10^3 l./mole cm.) for 9-methyladenine at 171°. A comparison of oscillator strengths of the gas phase and the aqueous bands shows a marked hyperchromism in solution. For adenine the oscillator strength increases from 0.24 in the gas phase to 0.28 in water, while for 9-methyladenine it increases from 0.16 to 0.28.

Cytosine, 3-methylcytosine, and guanine showed evidence of decomposition in the vapor spectra. Sharp ammonia bands as well as other unidentified bands were observed to develop with time at constant temperature. The long wave length absorption band in all three compounds, however, did not display the time dependence associated with decomposition products and therefore most probably may be attributed to the base compounds themselves. Thus, in spite of the difficulty due to decomposition, certain information may be obtained with moderate confidence.

The λ_{\max} for both cytosine and 3-methylcytosine is about 285–290 $m\mu$. Guanine shows a long wave length

(6) H. C. Longuet-Higgins and J. A. Pople, *J. Chem. Phys.*, **27**, 192 (1957).

Table I: Absorption Maxima in Solvents of Different Polarity^a

Compd.	Vapor	Methylcyclohexane	Acetonitrile	Trimethyl phosphate	H ₂ O, pH 7
Adenine	252, 207	Insol.	Insol.	260, 208	260, 207 ^b
9-Methyladenine	249, ?	258, 212	259	260, 210	260
Hypoxanthine	...?...	Insol.	269 (sh) (249, 244)	250, 196	249, 280 (sh)
9-Methylhypoxanthine	(290, 281, 273) 239, 205 (sh), 198	Insol.	270 (sh) (251, 245)	(249, 244), 198, 184	~280 (sh), 249, 200, 183
Uracil	244, 205 (sh), 187	Insol.	...?...	258, 203, 182	260, 202, ? ^b
1,3-Dimethyluracil	256, 205 (sh), 187	262, 203, 185	...?...	264, 206, 185	265, 204, 187
Cytosine	290 (sh), ~260 ^c	Insol.	...?...	277, 204, 186	267, 230, 209 (sh), 196 ^b
3-Methylcytosine	290 (sh), 259 ^c	Insol.	...?...	...?...	271

^a The abbreviation insol. means that a saturated solution of the compound did not give a usable spectrum in a 10-cm. cell. A shoulder is designated by sh. ^b Data from ref. 2. ^c The temperature dependence of these peaks indicated that the shoulder at 290 m μ is the absorption maximum of the compound, while the 260-m μ peak is due to decomposition.

structured band (maxima at 293 and 284 m μ) which is very similar to that found in 9-methylhypoxanthine.

Heats of Vaporization. In order to determine heats of vaporization from the spectral data one needs to measure the slope of a plot of $\log p'$ vs. T^{-1} , where p' is a quantity linear in the vapor pressure. Since the extinction coefficient will in general be temperature sensitive (band broadening), the optical density at constant wave length will not be a good measure of pressure changes. However, the band area or relative oscillator strength should be more nearly insensitive to temperature variations and has been used in the evaluation of

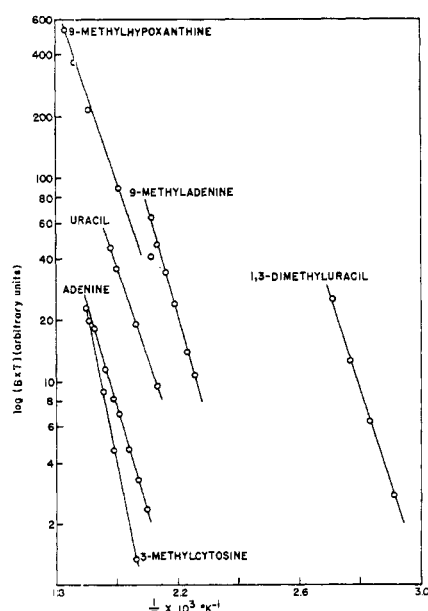


Figure 4. Heats of vaporization obtained from plots of $\log p'$ vs. $1/T$. The quantity p' , which is proportional to pressure, is the product of the area of an absorption band and the absolute temperature ($B \times T$).

the heats of vaporization from the spectra. The product of the band area and the temperature ($B \times T$) is proportional to the vapor pressure.

Figure 4 shows plots of $\log BT$ vs. T^{-1} for the main band of adenine, 9-methyladenine, 9-methylhypoxanthine, uracil, and 1,3-dimethyluracil. The area of the band, B , is evaluated from an absorbance vs. energy plot. The slope is constant for these lines and corresponds to the heat of vaporization of the solid. For cytosine, 3-methylcytosine, and guanine only the absorbance at one wave length was used in the plots, because decomposition products obscured the low wave length part of the spectrum. A straight line was obtained for 3-methylcytosine (Figure 4), but the data for guanine and cytosine do not produce straight lines.

The heats of vaporization obtained are given in Table II. For adenine and 9-methyladenine, heats of vaporization were also calculated from the temperature dependence of the vapor pressure. The values obtained (shown in Table II in parentheses) are within 12% of

Table II: Heats of Vaporization and Heats of Solution in Water

	ΔH_{vap} , kcal./mole	ΔH_{soln} , kcal./mole
Adenine	24 (26) ^a	8 ^b
9-Methyladenine	26 (29) ^a	6.860 \pm 0.042 ^c
9-Methylhypoxanthine	20	
Uracil	20	
1,3-Dimethyluracil	22	
3-Methylcytosine	36	
1-Methylthymine	..	5.3 \pm 0.030 ^c

^a The values in parentheses are calculated from the temperature dependence of the vapor pressure. ^b Data from ref. 8. ^c Data from ref. 7.

the spectral value. Heats of solution in water have been measured for 9-methyladenine and 1-methyl-thymine⁷; a heat of solution for adenine can also be calculated from the data of T'so, *et al.*⁸ These values (Table II) lead to a large negative ΔH (~ -20 kcal./mole) for the process: purine or pyrimidine in gas phase \rightarrow purine or pyrimidine in aqueous solution.

Knowledge of these numbers is necessary to an understanding of the forces which determine the stability of nucleic acids in solution.⁹

(7) S. J. Gill, D. B. Martin, and M. Downing, *J. Am. Chem. Soc.*, **85**, 706 (1963).

(8) P. O. P. T'so, I. S. Melvin, and A. C. Olson, *ibid.*, **85**, 1289 (1963).

(9) H. DeVoe and I. Tinoco, Jr., *J. Mol. Biol.*, **4**, 500 (1962).

Uranium Monoselenide. Heat Capacity and Thermodynamic

Properties from 5 to 350°K.^{1a}

by Yoichi Takahashi and Edgar F. Westrum, Jr.^{1b}

Department of Chemistry, University of Michigan, Ann Arbor, Michigan 48104 (Received May 20, 1965)

The low temperature heat capacity of USe was determined by adiabatic calorimetry and found to have a normal sigmoid temperature dependence except near the 160.5°K. transition from the ferromagnetic to the paramagnetic state. The heat capacity (C_p), entropy (S°), enthalpy function ($[H^\circ - H^\circ_0]/T$), and Gibbs energy function ($-[G^\circ - H^\circ_0]/T$) at 298.15°K. in cal./(g.f.m. °K.) are 13.10, 23.07, 10.39, and 12.68, respectively.

Introduction

The presence of several phases, such as USe, U₃Se₄, U₂Se₃, U₃Se₅, α -, β -, and γ -USe₂, and USe₃, has been recognized in the uranium-selenium system,² but few thermodynamic properties have been reported. Recent investigations of magnetic³ and electrical⁴ properties have shown uranium monoselenide (USe) to be of particular interest because of its ferromagnetism below 200°K. and low electrical resistivity and high thermoelectric power at room temperature. Its thermal properties are of interest also in comparison with those of other uranium compounds such as US, UC, UN, and UP, all of which also possess the NaCl structure.

Experimental Section

Preparation and Characterization of the Sample. The sample of uranium monoselenide was prepared at the Battelle Memorial Institute⁴ by allowing uranium turnings (containing 0.01 wt. % spectrographically detected

impurities) to react with the vapor of rectifier grade (99.999% pure) selenium in evacuated, sealed quartz capsules. The resulting finely divided selenide preparations were consolidated, melted, and homogenized to monoselenide under an argon atmosphere at about 2000° in a tantalum crucible. Determination of uranium and selenium gave values within $\pm 0.2\%$ of the stoichiometric ratio. Tantalum was not detected by spectrochemical analysis sensitive to 0.001%, and oxygen contamination was less than 0.1% by weight.

(1) (a) This research was supported in part by the United States Atomic Energy Commission and by the Selenium-Tellurium Development Association, Inc. (b) To whom correspondence concerning this work should be addressed.

(2) (a) R. Ferro, *Z. anorg. allgem. Chem.*, **275**, 320 (1954); (b) P. Khodadad, *Bull. soc. chim. France*, 133 (1961).

(3) W. Trzebiatowski and W. Suski, *Bull. Acad. Polon. Sci., Ser. sci. chim.*, **10**, 399 (1962).

(4) L. K. Matson, J. W. Moody, and R. C. Himes, *J. Inorg. Nucl. Chem.*, **25**, 795 (1963).