The Interaction of Oxomolybdenum(VI) with Nucleic Acid Bases and Nucleosides

PHOTINI PIPERAKI, NIKOS KATSAROS*

Chemistry Department, Nuclear Research Center "Demokritos", Aghia Paraskevi Attikis, Athens, Greece

and DIMITRIS KATAKIS

Chemistry Department, University of Athens, Athens, Greece

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The reactions have been studied of the nucleic acid bases, adenine, cytosine, guanine, purine, hypoxanthine, mercaptopurine and the nucleosides, guanosine, cytidine and adenosine with sodium molybdate in aqueous solutions. Compounds of the type $(AH^+)_4Mo_8O_{26}\cdot 4H_2O$ *(where A = adenine, cytosine, guanine, hypoxanthine, purine, mercaptopurine, adenosine and cytidine) were isolated at pH values around 4 and compounds of the type* $(AH^+)_2Mo_6O_{19}$ *(where A = adenosine, cytidine or guanosine) at around pH 2. The compounds prepared were characterized by elemental analysis, infrared, Raman, electronic and* ^1H *NMR spectroscopy, conductivity measurements and pH titrations.*

Introduction

Interactions of transition metal ions with nucleic acids, their constituent bases and nucleosides have recently been the subject of considerable research [1–3]. Metal ions participate in biological functions of nucleic acids and under various conditions can stabilize the secondary and tertiaty macromolecular structures, or even bring about the decomposition of the primary stucture [4–6]. Both metal ions and certain nucleotides can serve as cofactors for various enzymatic reactions [7]. The occurrence of a variety of trace metals in nucleic acids, ribonucleic acids, and viruses have been reported [8–14]. The significance of these trace metals cannot be fully explained unless the nature of the metal binding to the nucleotide unit is understood. Heavy metal adducts of nucleic acids also proved useful in an X-ray structure determination of t-RNA [15], and have been investigated in attempts to sequence nucleic acids by electron microscopy techniques [16, 17].

The goal of the present study was to synthesize and characterize by physical and chemical methods a series of stable oxomolybdenum(VI) compounds with nucleic acid bases and nucleosides, in order to investigate the nature of the interactions in these systems.

Experimental

Materials

Nucleic acid bases and nucleosides were obtained from Sigma Chemical Company and used without further purification, after verifying that the measured melting points corresponded with those reported in the literature. Sodium molybdate and molybdenum trioxide were from Fluka A.G., DMSO was purified by vacuum distillation after drying over calcium hydride.

Microanalyses

Carbon, hydrogen and nitrogen analyses were performed by Dr. H. Mantzos of the National Hellenic Research Foundation (N.H.R.F.) in Athens. Molybdenum was determined gravimetrically as molybdenum (VI) oxinate [18] and sodium by flame photometry, after the compounds had been decomposed by heating with concentrated nitric and sulfuric (1:1) acids.

Preparations

$(Cytosine\ H^+)_4Mo_8O_{26}$ was prepared by adding to a hot aqueous solution of cytosine (1 mmol), 2.6 g (2 mmol) of molybdenum trioxide, MoO_3. The mixture was refluxed for one hour and then filtered. The filtrate was evaporated to one-third of its volume and upon cooling gave a white precipitate. The preci-

*Author to whom correspondence should be addressed.

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pitate was filtered again, washed repeatedly with water and dried in vacuum over P_2O_5. Then it was twice recrystallized from hot water.

(Adenine-H^+)$_4Mo_8O_{26}$, (guanine-H^+)$_4Mo_8O_{26}$, (hypoxanthine-H^+)$_4Mo_8O_{26}$, (mercaptopurine-H^+)$_4$-Mo_8O_{26} and (purine-H^+)Mo_8O_{26}. These compounds were prepared by dissolving 4.4 g (2 mmol) of Na_2-$MoO_4 \cdot 2H_2O$ in 20 ml 1 N HCl solution and adding to this dropwise an aqueous solution of the nucleic acid base (1 mmol). Cooling the mixture to 15 °C precipitated microcrystalline compounds which were filtered off, washed with water and dried under high vacuum over P_2O_5. The compound (cytosine-H^+)$_4Mo_8O_{26}$ was also prepared by this method. (Adenosine-H^+)$_4Mo_8O_{26}$ and (cytidine-H^+)$_4$-Mo_8O_{26}: these compounds were prepared by mixing an aqueous solution of adenosine or cytidine (1 mmol in 10 ml of 0.5 NaOH) with a solution of Na_2-MoO_4 (2 mmol in 20 ml of 0.5 NaOH). The resulting solution was cooled to 15 °C and acidified by adding dropwise and under constant stirring 1 N HCl, to pH ~ 3.8–4. Usually turbidity occurred around pH = 3.8 when the precipitation process began, which may last for a day or two. The precipitates were filtered off, washed with water and acetone and dried in vacuum over P_2O_5.

(Adenosine-H^+)$_2Mo_6O_{19}$, (guanosine-H^+)$_2Mo_6O_{19}$ (cytidine-H^+)$_2Mo_6O_{19}$· the nucleoside (1 mmol) was dissolved in 15 ml of 0.5 N NaOH and a solution of 2 mmol Na_2MoO_4 in 20 ml 0.5 N NaOH was added (in the case of guanosine 0.2 N NaOH solution was used). This solution was acidified by adding dropwise 1 N HCl solution under constant stirring to pH = 2–3. Usually turbidity appeared around pH ~ 3 and precipitation occurred upon cooling to 15 °C. The precipitates were filtered off, washed with water and acetone and dried under high vacuum over P_2O_5.

^1H NMR Measurements

The ^1H NMR spectra were measured in D_2O or d_6-DMSO using an XL-100-15 NMR spectrometer. The chemical shifts are related to TMS for DMSO and DSS for D_2O as internal standards at ambient temperatures.

Conductivity Measurements

The measurements were carried out in water and DMSO at 25 °C by means of an E365B conductoscope, Metrohm Ltd., Herisau, Switzerland. Λ_m values are expressed in ohm^{-1} cm^2 mol^{-1}.

Electronic Spectra

UV–visible spectra were recorded on a Cary-14 spectrophotometer in aqueous solutions.

Raman Spectra

The Raman spectrum of the compound (cytosine-H^+)$_4Mo_8O_{26}$ was obtained on a Joben-Yvon Ramanor HG 25 instrument with an Ar Laser from Spectra Physics Model 165-03.

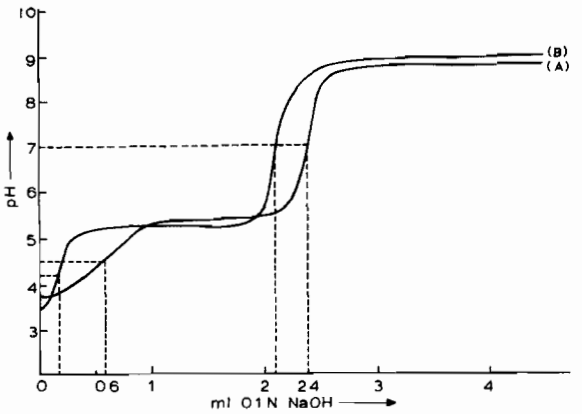

Fig. 1.

pH Titrations

pH Titrations were performed on an E436 Potentiograph Metrohm Ltd., Herisau, Switzerland. Titration of molybdate solutions with base is an effective method for determining oxomolybdenum anions, as has been recently shown by Burns *et al.* [19]. The end point corresponds to a sharp break in the titration curve (Fig. 1) that occurs at pH = 7, when the molybdate solution is neutralized with a strong base (0.1 N NaOH). Pertinent equations that give useful stoichiometric relations are.

$$Mo_8O_{26}^{4-} + 12OH^- \xrightarrow{pH=7} 8MoO_4^{2-} + 6H_2O \quad (1)$$

$$Mo_6O_{19}^{2-} + 10\ OH^- \xrightarrow{pH=7} 6MoO_4^{2-} + 5H_2O \quad (2)$$

The nucleic acid or nucleoside molybdates that precipitated around pH = 4 are formulated as (AH$^+$)$_4Mo_8$-O_{26} (type I) while those precipitated around pH = 2 are formulated as (AH$^+$)$_2Mo_6O_{19}$ (type II) and their neutralization to pH = 7 can be described by equations (3) and (4) respectively.

$$4AH^+ + 4OH^- \rightarrow 4A + 4H_2O$$

$$Mo_8O_{26}^{4-} + 12OH^- \rightarrow 8MoO_4^{2-} + 6H_2O$$

$$(AH)_4Mo_8O_{26} + 16OH^- \rightarrow 4A + 8MoO_4^{2-} + 10H_2O \quad (3)$$

$$2AH^+ + 2OH^- \rightarrow 2A + 2H_2O$$

$$Mo_6O_{19}^{2-} + 10OH^- \rightarrow 2A + 6MoO_4^{2-} + 7H_2O$$

$$(AH)_2Mo_6O_{19} + 12OH^- \rightarrow 2A + 6MoO_4^{2-} + 7H_2O \quad (4)$$

TABLE I. Analytical Data for Nucleic Acid Base or Nucleoside-Molybdate Complexes.

Complexes	Color	M.p.[a] °C	Analysis Found (Calcd.) %			
			C	H	N	Mo
(cytosine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White	220 D	11 37 (11.26)	1.35 (1.40)	10.18 (9.86)	45.20 (45.07)
(adenine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White	210 D	13.48 (13.36)	1.87 (1.55)	15.74 (15.59)	43.10 (42.76)
(guanine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White-Yellow	210 D	13.30 (12.90)	2.21 (2.58)	15 28 (15.05)	41.75 (41.29)
(purine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White	150 D	14.11 (13.79)	1.36 (1.37)	13.15 (12.87)	44.20 (44.13)
(mercaptopurine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	Yellow	210 D	12.98 (12.90)	1.40 (1.50)	12.26 (12.04)	41.40 (41.29)
(hypoxanthine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White	250 D	13.48 (13.33)	1.63 (1.55)	12.60 (12.44)	42.98 (42.66)
(adenosine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White	160 D	20.76 (20.65)	3.30 (3 18)	11.99 (12.04)	33.15 (33.04)
(cytidine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	White	170 D	19 59 (19.38)	3.18 (3.32)	7.62 (7.54)	34.87 (34.47)
(adenosine $H^+)_2Mo_6O_{19}\cdot 2H_2O$	White	50 D	16.42 (16.62)	2.69 (2.62)	9.71 (9.65)	39.52 (39.72)
(cytidine $H^+)_2Mo_6O_{19}\cdot 2H_2O$	Yellow	50 D	15.42 (15.60)	2.36 (2.31)	5.80 (6.06)	41.49 (41.61)
(guanosine $H^+)_2Mo_6O_{19}\cdot 2H_2O$	Yellow	50 D	16.22 (16.39)	2.24 (2.32)	9.45 (9.59)	39.22 (39.34)

Complexes	Molar Conductance $Ohm^{-1}\ cm^2\ mol^{-1}$ (C = 1 × 10^{-3} at 25 °C)
(cytosine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	365 in H_2O
	69 in DMSO
(adenine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	57 in DMSO
(guanine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	47.6 in DMSO
(purine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	46.2 in DMSO
(mercaptopurine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	44.2 in DMSO
(hypoxanthine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	51.6 in DMSO
(adenosine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	374 in H_2O
	45.4 in DMSO
(cytidine $H^+)_4Mo_8O_{26}\cdot 4H_2O$	378 in H_2O
	45.9 in DMSO
(adenosine $H^+)_2Mo_6O_{19}\cdot 2H_2O$	205 in H_2O
	25.8 in DMSO
(cytidine $H^+)_2Mo_6O_{19}\cdot 2H_2O$	207 in H_2O
	32.6 in DMSO
(guanosine $H^+)_2Mo_6O_{19}\cdot 2H_2O$	209 in H_2O
	29.9 in DMSO

[a]D = decomposition.

Thus as shown by equation (3) sixteen equivalents of base react with one mol of $(AH)_4Mo_8O_{26}$, and according to equation (4) 12 equivalents of base react with one mol of $(AH)_2Mo_6O_{19}$. These relations permit the determination by pH titration of the formula weight of the molybdate species present. A typical calculation is shown for (cytosine $H^+)_4$-Mo_8O_{26}. Data for the other molybdates are given in Table V.

1.4 ml × 0.1 meq/ml × 1 mmol/16 meq = 0.015 mmol of the complex present. Molecular weight (in mg) 1 mmol × mg sample/mmol

TABLE II. Infrared Data of Nucleic Acid Base or Nucleoside Molybdates (cm^{-1}).

Cytosine	$(cytH^+)_4Mo_8O_{26}$	Cytidine	$(cytH^+)_4Mo_8O_{26}$	$(cytH^+)_2Mo_6O_{19}$	Adenine	$(AdenH^+)_4Mo_8O_{26}$	Adenosine	$(adenH^+)_4Mo_8O_{26}$	$(adenH^+)_4Mo_6O_{19}$	Assignments
3360s 3160s	3100sb 3190s	3445m	3200mb	3200mb	3310w 3254m	3320w 3260m 3200m	3320mb 3100mb	2700–3600mb	2700–3600mb	OH, NH_2, NH^+ CH aromatic or aliphatic stretchings
1652s 1617s	1710s 1650s	1647s 1603s	1725s 1680s	1725s 1680s	1623s	1670s	1667s	1690s	1690s	C=O and C=C with C=N + δNH_2
1450s 1225m	1440m 1210s	1525s 1495s 1380sb 1290s 1200s 1100s 1060s	1520s 1400s 1270s 1200s 1150mb	1520s 1400s 1270sb 1200m 1150mb	1576m 1440m 1410vs 1320s 1300vs 1242s 1115m 930m 710s	1580m 1470m 1435m 1390vs 1315w 1235m 1125m	1650s 1603s 1460m 1290s 1200s 1100m 1025m	1680s 1600s 1490m 1310m 1220s 1005m	1680s 1600s 1490m 1310m 1230s 1005m	ring and other ligand vibrations
	945s, 920s 890s, 730sb		935s, 890s 730sb	935s, 790s 610m		935s, 910s 890sb		930s, 920sb 890s	930s, 790s 590s	Mo–O vibrations
Guanine	$(guaH^+)_4Mo_8O_{26}$	guanosine	$(guaH^+)_2Mo_6O_{19}$	hypoxanthine	$(hypH^+)_2Mo_8O_{26}$	mercapt	$(merH^+)_2Mo_8O_{26}$	purine	$(purH^+)_2Mo_8O_{26}$	Assignments
1660s	1695s	1730s 1635s, 1605s 1690s	1650s, 1605s 1710s	1650s	1700s	1620s	1625s	1590s	1605s	C=O and C=C with C=N + δNH_2
1550m 1465m 1410m 1370s 940s	1545m 1450m 1400m 1350m 945s	1670w 1540s 1490s 1390s	1545s 1520w 1380s	1450s 1400s 1350m 1330s 1260s 1200s 1120s 950s	1445s 1400s 1370m 1330m 1260m 1200s 1130m 950s	1580s 1530m 1420s 1350s 1280s 1220s	1550s 1405s 1340s 1290s 1210s	1560m 1450s 1420m 1390s 1320m 1260m	1575m 1470m 1400s 1315m 1250m	ring and other ligand vibrations
						1015s	1040s			C=S
890s, 825s, 650s	940s, 920s		950s, 780s 604m, 433m		940vs, 920sh 890s, 550vs		950s, 920sh 570sb, 890s		950vs, 920vs 890mb, 800s	Mo–O vibrations
					450s				635w, 550sb	

sample = 1 mmol × 2.5 4 mg/0.015 mmol = 1693 mg.

Results and Discussion

The compounds prepared, their elemental analyses and other physical properties are listed in Table I. The octamolybdates had no definite melting points, and started to change color at about 200 °C (the hexamolybdate compounds with nucleosides changed color at ~50 °C) and decomposed. The complexes were not very soluble in water or DMSO. However, their solubilities in these solvents allowed nmr and conductivity measurements. They are insoluble in all other common organic solvents. They are also slightly photosensitive and should not be exposed to sunlight for a long period of time. In fact, all attempts to prepare molybdate compounds with nucleic acid bases, nucleosides or nucleotides besides the ones reported here gave products that were either highly photosensitive or had no definite composition, being most probably mixtures.

The results of chemical analysis established that the molybdenum–nucleic acid base or nucleoside ratio is 2:1 and 3:1 for type I and type II complexes respectively. This ratio suggests the hexamolybdate formulation for the latter and rules out the hepta-molybdate ($Mo_7O_{24}^{6-}$) anion. However, several molybdates exist having a 2:1 molar ratio [35]. Thus from the analytical results alone the nature of the oxomolybdenum residue cannot be determined.

Infrared Spectra

Infrared data of the compounds in the solid state are recorded in Table II. The three strong absorptions that occur in the 1000–800 cm^{-1} region are assigned to molybdenum–oxygen stretching frequencies [19–21]. The strong band around 950 cm^{-1} is attributed to terminal Mo–O stretching vibration, while the two broader bands at 920 and 890 cm^{-1} are Mo–O stretching vibrations from two different Mo–O–Mo bridging structures. The same trio of absorptions was more or less consistent in all the nucleic acid or nucleoside-molybdates of type I, therefore, it was suggested that the same molybdate ion was present in these compounds. A similar, characteristic pattern of absorptions also appeared in the 1000–700 cm^{-1} region in the spectra of all the nucleoside–molybdate complexes of type II, indicating that the same kind of oxomolybdenum ion is present in these compounds. Sodium and ammonium octamolybdates show a triplet in the region 1000–800 cm^{-1} [21], similar to those of the compounds of type I reported here. Literature values [41] for the hexamolybdate ion compare quite well with the absorptions at the 1000–700 cm^{-1} region observed for the compounds of type II. In the spectrum of cytosine there is a strong absorption at 1650 cm^{-1}, which is assigned to the C=O stretching mode. In the cytosine–molybdate compound an additional band appears at 1710 cm^{-1}. It has been found that in many complexes and in cytosinium chloride, where cytosine is protonated at the N-3 position, the carbon–oxygen stretching frequency shifts to higher wavenumbers (due to π-electron localization) so the C=O bond has more pronounced double bond character [25, 26]. Similarly cytidine exhibits strong bands at 1645 and 1600 cm^{-1}, which are assigned to C=O and C=C stretching modes. Both of these bands shift (by 80 cm^{-1}) in the corresponding nucleic acid base- or nucleoside-molybdates to higher frequencies (1725 and 1680 cm^{-1} respectively), in accordance with the spectra of protonated cytidine [17].

Guanine exhibits an absorption around 1660 cm^{-1} which is attributed to the C=O stretching mode. In the protonated guanine at the N-7 position of the puring ring this band shifts to higher wavenumbers, around 1670 cm^{-1}. In the guanine-molybdate compound this band appears at 1695 cm^{-1}, indicating protonation at the N-7 position. We also observed that the C=O band in guanosine-molybdate shifts to higher frequencies with respect to the C=O band of the free ligand, indicating again protonation [25, 27] of the guanosine. Similarly the C=O stretching mode in hypoxanthine which appears at 1650 cm^{-1} shifts to 1700 cm^{-1} in its molybdate compound. Mercaptopurine in mercaptopurine-molybdate is also in its protonated form since the bands at 1620, 1580 and 1530 cm^{-1} assigned to C=S and C=N stretching modes are also shifted to higher wavenumbers. Finally in the free ligands adenine, adenosine, and purine the C=N ring stretching vibration (which is coupled with the NH_2 bending motion at ~1600 cm^{-1}) shifts to higher frequencies in the corresponding molybdate compounds, again indicating protonation.

In the adenine, adenosine, purine, guanine, guanosine and hypoxanthine-molybdates a strong broad absorption around 3200 cm^{-1} was identified as being due to NH^+ stretching vibration. This provides additional evidence that these bases and nucleosides are in their protonated form. Thus, the infrared spectra of the nucleic acid base and nucleoside-molybdates indicate that the organic moieties exist in their protonated forms.

IH NMR

The ^1H NMR data of the nucleic acid base (or nucleoside) molybdates are presented in Table III. In all cases the observed shifts relative to the free ligands in D_2O or d_6-DMSO are less than 0.5 ppm, indicating that there is no direct interaction between the metal ion and the ligands [28–31]. The small shifts observed are in accordance with literature values [28] reported for the protonated nucleic acid

TABLE III. Chemical Shifts (ppm) of Nucleic Acid Base or Nucleoside Molybdates in d_6-DMSO.

Compound	H_2	H_8	H_5	H_6
Cytosine			5.62	7.34
$(cyt.H^+)_4Mo_8O_{26}$			5.75	7.50
cytidine			5.90	8.18
$(cyt.H^+)_4Mo_8O_{26}$			6.12	6.35
$(cyt.H^+)_2Mo_6O_{19}$			6.20	8 72
Adenine	8.08	8.12		
$(aden.H^+)_4Mo_8O_{26}$	8.26	8.28		
Adenosine	8.14	8.34		
$(aden.H^+)_4Mo_8O_{26}$	8.52	8.50		
$(aden.H^+)_2Mo_6O_{19}$	8.52	8.46		
guanine		7.70		
$(guan.H^+)_4Mo_8O_{26}$		7.98		
guanosine		7.94		
$(guan.H^+)_2Mo_6O_{19}$		8.00		
hypoxanthine	7.98	7.82		
$(hp.H^+)_4Mo_8O_{26}$	8.10	7.96		
purine	8.92	9.34		
$(pur.H^+)_4Mo_8O_{26}$	8.90	9.32		
mercaptopurine	8.38	8.18		
$(merc.H^+)_4Mo_8O_{26}$	8.32	8.14		

bases or nucleosides. The observed shifts are due to the pH of the solution and the bulkiness of the anion.

Electronic Spectra

The ultraviolet spectra in aqueous solutions (summarized in Table IV) also confirm that the nucleic acid bases and nucleosides appear in their protonated forms [30–34]. Furthermore, additional evidence is provided by comparison of the pH values of the compounds in H_2O and the corresponding pk of protonation of the organic ligands [34].

Reaction with Ph_3BzCl

When the nucleic acid base or nucleoside-molybdates were reacted in aqueous solutions with triphenylbenzylphosphonium chloride (abbreviated as Ph_3BzCl) which contains a bulky cation, white precipitates were immediately formed. Elemental analysis showed a 2:1 and a 3:1 molybdenum to triphenylphosphonium ion mol ratio for the molybdate compounds of type I and II respectively. In the infrared spectra of these compounds the P–Cl stretching mode at $580m^{-1}$ is absent and a characteristic set of absorptions between 1000–700 cm^{-1} indicates the presence of an isopolymolybdate anion. This reaction indicates that the compounds prepared are probably of ionic nature.

TABLE IV. Electronic Spectra and pH Values of Nucleic Acid Base or Nucleoside Molybdate Complexes in H_2O.

Compound	λ_{max}, nm	$\epsilon \times 10^{-3}$ M^{-1} cm^{-1}	pH	pK
Cytosine	266	6.8		4.45
$(cyt.H^+)_4Mo_8O_{26}$	275	10.6	3.7	
cytidine	270	8.2		4.15–4.22
$(cyt.H^+)_4Mo_8O_{26}$	278	10.8	3.4	
$(cyt.H^+)_2Mo_6O_{19}$	278	10.9	3.4	
Adenine	259	14.6		4.15
$(aden.H^+)_4Mo_8O_{26}$	261	14.8	3.6	
Adenosine	260	12.5		3.5–3.6
$(aden.H^+)_4Mo_8O_{26}$	256	12.2	3.4	
$(aden.H^+)_2Mo_6O_{19}$	256	12.0	3.4	
guanine	274	8.0		3.2–3.4
$(guan.H^+)_4Mo_8O_{26}$	272	7.8	3.4	
guanosine	250	14.4		2.5
$(guan.H^+)_2Mo_6O_{19}$	255	12.0	2.5	
purine	264	8.2		2.52
$(pur.H^+)_4Mo_8O_{26}$	260	6.6	2.5	
hypoxanthine	250	11.2		2.5
$(hyp.H^+)_4Mo_8O_{26}$	246	11.2	2.5	
mercaptopurine	323	18.1		–
$(mercap.H^+)_4Mo_8O_{26}$	321	17 2	3.2	

TABLE V. Nucleic acid or Nucleoside-Molybdate Molecular Weights by Titration.

Compound	Weight taken in grams	ml 0.1 N NaOH used	Found M.W.	Calcd. M.W.
$(cyt.H^+)_4Mo_8O_{26}$	0.0254	2.40	1693	1704
$(aden.H^+)_4Mo_8O_{26}$	0.0600	5.30	1811	1800
$(guan.H^+)_4Mo_8O_{26}$	0.0615	5.50	1789	1796
$(hypo.H^+)_4Mo_8O_{26}$	0.0427	3.70	1846	1860
$(mercap.(H^+)_4Mo_8O_{26}$	0.0357	3.1	1842	1860
$(pur.H^+)_4Mo_8O_{26}$	0.0261	2.4	1740	1736
$(adenosineH^+)_4Mo_8O_{26}$	0.0580	4.0	2320	2324
$(cytidineH^+)_4Mo_8O_{26}$	0.0350	2.5	2240	2228
$(adenosine\ H^+)_2Mo_6O_{19}$	0.0258	2.1	1474	1468
$(cytidineH^+)_2Mo_6O_{19}$	0.0268	2.25	1429	1402
$(guanosineH^+)_2Mo_6O_{19}$	0.0272	2.22	1470	1482

Conductivity Measurements

The conductivity data of the compounds in H_2O and DMSO are included in Table I. The equivalent conductance values at 1×10^{-3} molar concentrations are very close for all molybdates of type I, and fit quite well the values expected for 4:1 electrolytes [37, 38]. Similarly, the equivalent conductance values for type II molybdates at 1×10^{-3} molar concentrations are close to each other and in agreement with those expected for 2:1 electrolytes. The somewhat lower values observed in comparison to the true 4:1 and 2:1 electrolytes are most probably due to the bulkiness of the ions and possible ion-pair formation.

pH Titrations

A neglected approach to the analysis of compounds containing polymolybdate ions is the simple titration with a base. Condensed molybdates like octa-, hexa-, or tetramolybdates are treated as weak acids and can be titrated with base to give the stable normal molybdate, at pH = 7. Furthermore, in the compounds prepared the nucleic acid bases or nucleosides are in their protonated forms, so an additional amount of base is required to neutralize the protons of the nucleic acid bases or of the nucleosides since their corresponding pk values lie below pH = 7 [34]. The pertinent neutralization reactions are given in the experimental section. Formula weights determined by this method (Table V) agree quite well with the calculated ones for an octa- (type I) and hexa-molybdate (type II) anion. Since the initial investigations of the condensations of normal molybdate ion by Jander and Rosenheim [38], practically every conceivable isopolymolybdate has been proposed to be formed when an aqueous molybdate solution is acidified. Anionic species ranging from $Mo_2O_7^{2-}$ to a gigantic anion containing twenty-four molybdenum atoms have been suggested [37]. The conclusions of recent work [41] are that normal molybdate upon acidification forms two condensation products with no detectable intermediates: a heptamolybdate, $Mo_7O_{24}^{6-}$, and an octamolybdate, $Mo_8O_{26}^{4-}$. Higher molybdates are still formed at higher acid concentrations, but this is not conslusive yet: cationic molybdyl ion MoO_2^{2+} begins to form at pH = 1 and below [35]. The classic method of preparing isopolymolybdates is crystallization of alkali and alkaline earth salts (including ammonium, tetraalkylammonium salts *etc.*) from acidified aqueous metalate solutions. However, when normal molybdate reacts with organic ligands in aqueous or non-aqueous solvents a wide variety of oxomolybdenum species are formed [37]. Compounds of organic bases and molybdate ions are probably the more common type of organomolybdates. Organic bases containing one or more nitrogen atoms form generally insoluble adducts with molybdates in which the nitrogen atoms are usually protonated. Van Slyke [36] utilized the basic behavior of aminoacids about fifty years ago when he devised a scheme for separating the more basic diaminoacids from the less basic or neutral monoamino acids. Several investigations on amino acid molybdates and various other organic base molybdates have been reported [37–45]. However, there is considerable controversy over the form of the isopolymolybdate [20, 34–45]. The nature of the oxomolybdenum species does not depend only on the pH of the solution but depends also on the nature of the organic moiety. Further systematic studies are required in order to determine which ligands favor the precipitation of a particular form of condensed molybdate in a specific solvent.

The crystal structure determination [46] of cytosine-molybdate established the following formulation $(cytosine\text{-}H^+)_4Mo_8O_{26}^{4-}$ with discrete $Mo_8O_{26}^{4-}$

anions [46]. The physical measurements of nucleic base or nucleoside molybdates of type I agree quite well with those of (cytosine-H^+)$_4$ $Mo_8O_{26}^{4-}$. Therefore, it is reasonable to suggest that all molybdate compounds of type I contain the same anion, both in the solid state and in solution. Regarding the nucleoside molybdates of type II, the anion can be formulated as $Mo_6O_{19}^{2-}$.

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