Contribution from the Institute of Physical Chemistry of the Roumanian Academy, Dumbrava Rosie St., 23, Bucharest, Roumania.

Coordination Chemistry of Cu^{2+} and Mn^{2+}. I. I.R. Spectroscopic Evidence of Cu^{2+} and Mn^{2+} Ions Interaction with the Purinic Bases (Guanine and Theophylline)

D. Crăciunescu and Al. Fruma

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In the complexes of Cu^{2+} and Mn^{2+} with theophylline a weakening occurs in the C=N bands (1555-1590 cm^{-1}), which together with the modifications in the («additional») 930-950 cm^{-1} band, account for the changes in the stereochemistry of theophylline. No enolisation of the carbonyl at C_6 occurs, and from the modifications in intensity of the bands C=N and = NH, we conclude that there is a stronger bonds in [Mn(theophylline)$_2$] than in [Cu(theophylline)$_2$] and [Ag(theophylline)]. We report for the first time the I.R. spectra of the complexes: [$CuCl_2$(guanine)$_2$], [Cu(guanine)$_2$] . SO_4 . H_2O and [$CuSO_4$(guanine)$_2$]. H_2O.

Introduction

The first compounds of theophilline with metal ions (Mn^{2+}, Cu^{2+}, Zn^{+2}, and Ag^{2+}) were reported by A.F. Schutz and Umschweif, and by R. Klimek.[2] Owing to their marked biological importance a series of purinic base complexes has recently been used in terapeutics as diuretics.[3,4] The compound [Cu(theophylline)$_2$], where theophylline is $C_7H_7O_2N_4$, has been shown to have a catalytic effect on the decomposition of H_2O_2: in this reaction the catalytic effect is due to the release of Cu^{2+}.[5]

J. Bayer and E. Posgoy[6] assume similar structures for the complexes of theophylline, guanine, xanthine, and hypoxanthine with Hg^{2+},[6] as in Figure 1.

Recently, R. Weiss and H. Venner[7,8,9] have synthesised complexes of Cu^{2+} with purinic bases (theophylline, guanine, hypoxanthine, purine, adenine) and have determined their stability as a function of pH.

Theophylline complexes are of the type [M(theophylline)$_2$] (Figure 1) and those of guanine (2-amino,6-hydroxypurine) of the types $Pu_2MCl_2(H_2O)_n$, $Pu_2MSO_4(H_2O)_n$, $PuMSO_4(H_2O)_n$, where M is Cu^{2+} and Pu = purinic compound. The structures are assumed on the basis of the chelation possibilities between N_7 and the substituent for C_6 (Figure 2).

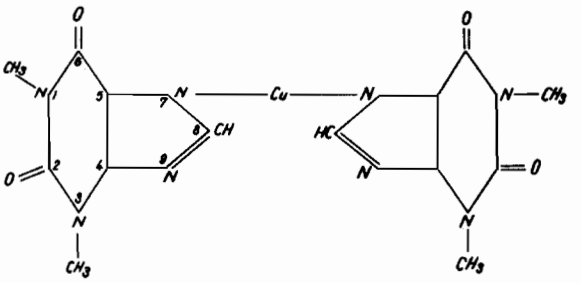

Figure 1. Structure of [Cu(theophylline)$_2$].

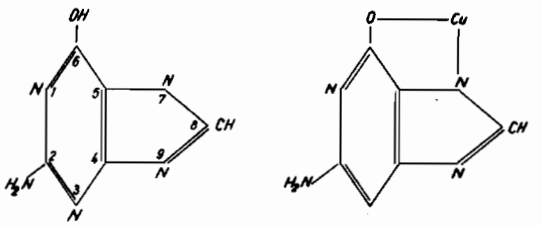

Figure 2. Guanine(2-amino, 6-hydroxypurine); coordination around the Cu^{2+} ion.

The tendency of purinic bases to form complexes with Cu^{2+} decreases with increase in number of substituents in the purinic cycle. Whereas in the case of guanine the authors clearly assume the mode of complex formation (Figure 2), in the case of theophylline chelation has been confirmed by spectroscopic observations. This chelation may occur either by the enolisation of carbonyl and the creation of a stable chelate ring, or by the simple substitution of the hydrogen atom from N_7. We have endeavoured to explain in this sense the observations made by T. Tu and J. A. Reinosa.[10]

The spectra were obtained with a UR-10, Carl Zeiss Jena apparatus, by the KBr disc method. The substances used were obtained from Schuchardt, München.

(1) A. P. Schutz and B. Umschweif, *Biochemische Zeitschrift, 268,* 326 (1934).
(2) R. Klimek and J. K. Parnas, *f. für Physiologische Chemie, 218,* 30 (1933).
(3) Th. Greener and H. Gold, *J. Pharmacol. Exptl. Therap., 113,* 140 (1955).
(4) O. de Walter, *Austrian Patent,* 18.37.61 Nov. 10, 1955.
(5) K. Anisimov, *Zhur. Fiz. Khim., 23,* 1427 (1951).
(6) J. Bayer and E .Posgoy, *Pharm. Zentralhalle, 101,* 476 (1962).
(7) R. Weiss and H. Venner, *Z. für Physiologische Chemie, 340,* 34, 138 (1965).
(8) H. Venner and E. Weiss, *Z. für Physiologische Chemie, 333,* 169 (1959).
(9) H. Venner and R. Weiss, *Z. für Physiologische Chemie, 317,* 82 (1959).
(10) Anthony T. Tu and J. A. Reinosa, *Biochemistry, 5,* 3375 (1969).

Results and Discussion

The theophylline compounds [Mn(theophylline)$_2$] and [Cu(theophylline)$_2$] were prepared according to the methods of Venner[7] and Klimek.[2] In the spectrum of [Cu(theophylline)$_2$], the weakening of the NH (2500-3500 cm^{-1}) bands was evident. This fact supports a structure (Figure 1) in which a bond is formed by substitution of the hydrogen from N. The weakening and flattening of the NH band appears clearly at 3330 cm^{-1}. On the other hand, the characteristic band of the carbonyl grouping is at about 1690 cm^{-1} and is not affected by complexation, showing only slight widening. Our formulation, in agreement with the suggestion of Tu and Reinosa,[10] assumes the absence of interaction with the carbonyl grouping and is supported by the fact that the spectrum shows no absorption in the range of the O-H frequencies and, therefore, no possibility of enolisation for the carbonyl at C_6 (1680-1720 cm^{-1} range).[11]

In the 1555-1590 cm^{-1} range the maxima are somewhat stronger than for pure theophylline. This is accounted for by the modification in the energy of the C=N bond of the purinic cycle upon coordination. According to Blout,[11] the bands at 1610 cm^{-1} correspond to the C=C bond stretching in the purinic cycle. Bands in the region 1590-1610 cm^{-1} may also be assigned to the C=C bond stretching. According to the observations of Tu and Reinosa,[10] in analogous complexes with inozine and guanozine the phenomenon of enolization was related to the evident weakening of the C=O band to notable changes in the 1680-1720 cm^{-1} range. The mechanism explaining the changes in the I.R. spectrum of guanine is shown in Figure 3.

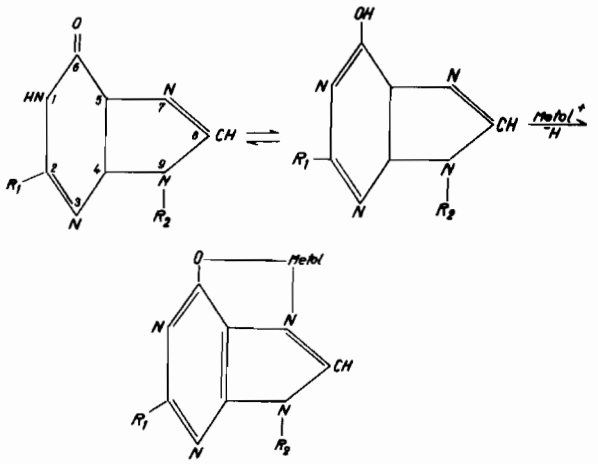

Figure 3. Enolisation phenomenon in the guanine case.

In theophylline the positions N_1, N_3 are methylated and the occurrence of enolization can only be considered as very improbable. The fact that the carbonyl band 1690 cm^{-1}, is only slightly widened in [Cu(theophylline)$_2$] is an additional evidence that the replaced hydrogen atom derives from N_7. The observation is also confirmed by the changes in bands assigned to the NH bond stretching in the 2500-3 200 cm^{-1} range of absorption.

In the complex we again find a slight attenuation of the free theophylline (940 cm^{-1} and 950 cm^{-1}) bands,[12] as well as a small shift of the maximum from 1200 cm^{-1} to 1210 cm^{-1}. The weak modification in the 900-950 cm^{-1} additional band, after complexation, could be attributed to the stereochemical modifications in theophylline. We draw the conclusion that the existence of a hydrogen bond between the carbonyl at C_6 and the hydrogen at N_7 in free thephylline is unlikely because according to the hypothesis of Tu and Reinosa,[10] it would certainly break when complexation occurs (Figure 4).

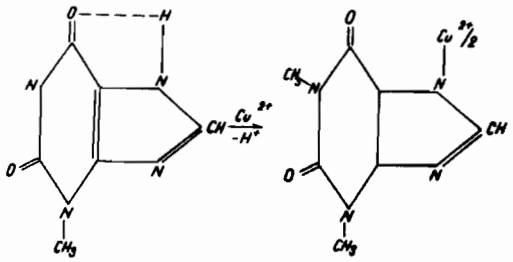

Figure 4. Destruction of the hydrogen bond in theophyyline after coordination around Cu^{2+}

The I.R. spectroscopic study of the interaction of the Mn^{2+} ion with theophylline confirms our assertions that the only metal-nitrogen bond is formed by substitution of the hydrogen atom at N_7. In [Mn(theophylline)$_2$] the 2500-3000 cm^{-1} range (NH frequency)[10] shows more important changes than those found in the spectrum [Cu(theophylline)$_2$]. We interpret these changes keeping in mind that the Mn^{2+}-N_7 coordinative bond is stronger than it was for Cu^{2+}-N_7 and that it exceeds the strength of the AgN_7 bond in [Ag(theophylline)]. In support of these assertions we note the disappearance of some maxima in free theophylline (2620, 2720, 2790, 2800, 3200 cm^{-1}) after complexation with Mn^{2+}. The C=N (1555-1590 cm^{-1}) bond, according to Blout,[11] is weakened more than in [Cu(thephylline)$_2$]. The 1690 cm^{-1} carbonyl band is not altered, displaying only a very weak widening. The «additional» band in the purines (Blout[11] and Willits[12]) at 930-950 cm^{-1} displays, after complexation, maxima at 910 and 940 cm^{-1}. In general, we notice the fact that the bands are more intense in the 900-1200 cm^{-1} range. We interpret the more important changes in the «additional» range, characteristic of the purinic cycle vibrations, as showing possible changes in the stereochemistry of theophylline after complexation, as in the case of [Cu(theophylline)$_2$]. The presence of more marked changes in the «additional» range of [Mn,theophylline)$_2$] could be due to a disturbance in the purinic cycle stereochemistry, which would be more important after complexation with Mn^{2+}. The more important changes in the range of the NH frequencies in [Mn(theophylline)$_2$] (2500-3000 cm^{-1}) suggests that the Mn-N_7 bond is stronger than the Cu-N_7 bond.

(11) E. R. Blout and M. Fields, *J. Amer. Chem. Soc.*, *72*, 479 (1950).

(12) C. H. Willits and J. C. Decius, *J. Amer. Chem. Soc.*, 9, 2579 (1955).

Table I. Wavenumbers (cm^{-1})

Theophylline	930	950	1555	1590	1690	2620	2720	2790	2800	3330			
[Cu(theophylline)$_2$]	940	990	1210	1555	1590	1600	1690	3330					
[Mn(theophylline)$_2$]	910	940	1555	1590	1690	2500-3000							
			weak			weak							
Guanine	840	940	950	1980	1530-1608	1650	1490	2701	2910	3120	3330		
[$CuCl_2$(guanine)$_2$]	580	930	950	980	970 intense	1050	1180	1210 intense	1470	1490	1590	2910	3330 weak
[Cu(guanine)$_2$]SO_4 . H_2O	970	1030	1060	1530 split	1590	2930	3085	3120 weak	3330 weak	3340			
[$CuSO_4$(guanine)$_2$] . H_2O	570	620	690	750 intense	1440	1510	1570	1700	2930	3120 weak	3160	3320	3190

The complexes of guanine with transitional metals (Co^{2+}, Fe^{2+}, Cu^{2+}) are interesting from the standpoint of biological activity and also because they are well suited for through physico-chemical studies (Mössbauer effects,[13] NMR spectra[14]).

The [$CuCl_2$(guanine)$_2$], [Cu(guanine)$_2$]SO_4 . H_2O and [$CuSo_4$(guanine)$_2$] . H_2O complexes have been synthesised by the methods of Weiss and Venner.[7] The bands were assigned starting from the data recorded by Blout[11] and C. H. Willits[12] which show that the vibration frequencies of NH and OH in guanine are 3330 and 3120 cm^{-1} respectively, and that the C=H frequencies appear in the 2910-2702 cm^{-1} range. The characteristic bonds of purinic cycles absorb at 1590-1600 cm^{-1} ,C=N) and at 1750 cm^{-1} (C=C). We have also compared the 930-950 cm^{-1} range of additional bands (Blout).[11]

According to H. Venner and R. Weiss[8,9] two complexes of the same [$CuCl_2$(guanine)$_2$] constitution are obtained, one immediately after complexation and a second, somewhat lighter in colour, which crystallizes from the mother liquor after a few days. Our I.R. determinations do not reveal any notable differences between the two compounds, and we are therefore unable to specify whether they are isomeric species or not. The second compound (recrystallised after a few days) shows somewhat weaker bands in the 500-700 cm^{-1} and 950-930 cm^{-1} ranges. The bands at 2910 cm^{-1} (C=N) are split and show an increase in intensity, and the bands at 1700 cm^{-1} (C=C) show the same aspect of shift and change.

The band at 1590 cm^{-1} is much more marked and more intense than in pure guanine. We could attribute this effect, as in the complexes with theophylline, to the changes of the C=N bond vibration energies after complexation. It is more difficult to account for the other changes in the 800-1200 cm^{-1}, 1150-1410 cm^{-1}, and 1450-1600 cm^{-1} ranges. They could be explained by a disturbance of the vibrations of the various bonds in purine after complexation. Details of these changes are as follows: a displacement of the maximum placed at 840 cm^{-1} in pure guanine to 780 cm^{-1} and appearance of new bands at about 1050 cm^{-1}.

In the 1150-1400 cm^{-1} range a maximum appears at 1180 cm^{-1} and another, somewhat weaker, at 1210 cm^{-1}. In the 1400-1600 cm^{-1} range some bands appear 1470, 1490, 1530 (weak), and 1570 cm^{-1}, generally split with respect to guanine. In the range of «additional» bands (Blout)[11] at 930-950 cm^{-1} characteristic for adenine, guanine, hypoxantine, and xantine, we note a displacement of the maximum from 930 cm^{-1} to 970 cm^{-1}. The changes in the «adidtional» band are attributable to some changes induced by complexation in the stereochemistry of the guanine molecule. We have not been able to account more definitely for these changes.

In the 600-700 cm^{-1} range the maxima for the [Cu(guanine)$_2$]SO_4 . H_2O complex are less intense than in pure guanine owing to absorption by the SO_4^{2-} ion. The «additional» band displays a new feature: the peak at 930 cm^{-1} has disappeared while the peak at 970 cm^{-1} is intact. The explanation is similar to the one given in the case of [$CuCl_2$(guanine)$_2$].

The NH band is weaker (3330 cm^{-1}) and displays a weak satellite at 3430 cm^{-1}, suggesting a link through nitrogen. The O-H band at 3120 cm^{-1} is somewhat lower, having a maximum at 3125 cm^{-1}, and the C-H band (2910-2702 cm^{-1}) displays a series of maxima at 2930 cm^{-1} and 3285 cm^{-1}, stronger and better outlined than in guanine. The changes in the absorption range of C=N (1590 cm^{-1}–1608 cm^{-1}) account for changes in the vibrational energy of the C=N bond; the maximum at 1590 cm^{-1} decreases considerably, and the maxima at 1530 and 1650 cm^{-1} are split.

Finally, marked changes appear in the 900-1000 cm^{-1} range. The maximum at 1000 cm^{-1} is split and two maxima are weakly outlined at 1030 and 1060 cm^{-1}.

The [$CuSO_4$(guanine)$_2$] . H_2O complex displays a series of peaks in the 600-700 cm^{-1} range, attributed to SO_4^{2-}. These maxima are much more intense than for [Cu(guanine)$_2$]SO_4 . H_2O, thus constituting a spectroscopic confirmation of the presence of SO_4^{2-} within the coordinating sphere of the complex (Venner's formulation[7]). The marked maxima are at 570-620 cm^{-1}, 690-730 cm^{-1}, 750 cm^{-1}. In the range of the NH frequency (3330 cm^{-1} according to Blout[11]), a series of displacements and strengthening of the maxima in guanine appear from 3330 to 3320 cm^{-1}.

At 3340 cm^{-1} the compound displays another maximum. In the region of hydroxyl, 3120 cm^{-1}, a weak descending tendency of the rather flat maximum displayed by pure guanine at 3130 cm^{-1} is observed.

The band corresponding to the C–H bond (2910-2702 cm^{-1} [11]) in pure guanine is displaced to 2960 cm^{-1}. In the 2800-3200 cm^{-1} range, maxima appear at 3160 and 3190 cm^{-1}. The C=C frequency is unchanged and on the whole, the splitings in this band

(13) R. A. Stukan, *Biofizica, 10*, 343 (1965).
(14) A. Veillard, *J. Chim. Phys., 59*, 1056 (1962).

are likewise very pronounced. The 1400-1600 cm^{-1} range exhibits a few changes at 1440-1510-1570 cm^{-1} and a displacement of the band from 1490 cm^{-1} in pure guanine to 1510 cm^{-1} in the complex.

Conclusions

When studying the I.R. spectra of the [Cu(theophylline)$_2$] and [Mn(theophylline)$_2$] we notice, in agreement with the suggestions of A. Tu and J. Reinosa,[10] that the phenomenon of coordination does not involve the carbonyl in position C_6 and that the metal-nitrogen bond is formed by substitution of the hydrogen atom at N_7.

The study of the [Cu(guanine)$_2$]$SO_4 \cdot H_2O$ and [$CuSO_4$(guanine)$_2$] $\cdot H_2O$ complexes indicates that the SO_4^{2-} ion is bonded differently in the coordination sphere of the two complexes (600-700 cm^{-1} range).

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