72. *Polarography of the Complexes of Cadmium(II) with Histamine, Antihistamine, and Some Related Molecules: a Thermodynamic Analysis.*

By A. C. Andrews and J. Kirk Romary.

A thermodynamic analysis of the cadmium(II) complexes of several molecules related to the histamine–antihistamine interaction systems has been conducted polarographically, at 0°, 25°, and 45° in systems having an ionic strength of 0·10 with potassium nitrate. The stabilities of the complexes have been related to the structures of the respective ligands. The free energies of formation for cadmium(II)–histamine complexes are compared with those for cadmium(II)–antihistamine complexes. The results support the theory of competitive binding at metal-ion receptor sites as a possible explanation of antihistaminic activity.

The thermodynamic properties of antihistaminic compounds have hitherto received little attention. Recently, a thermodynamic analysis of complexes of copper(II), cobalt(II), and nickel(II) with a series of commercial antihistamines has been reported.[1] Smith [2] has extended these studies to include iron(II), iron(III), mercury(II), zinc(II), and silver(I). It was of interest further to extend the thermodynamic investigation of these allergically associated molecules to include complex formation with the cadmium(II) ion. The ligands studied include Antistine [2-(*N*-benzylanilinomethyl)imidazoline hydrochloride], Benadryl (*NN*-dimethyl-2-diphenylmethoxyethylamine hydrochloride), ephedrine (2-methylamino-1-phenylpropan-1-ol), Neohetramine {2-[*N*-(2-dimethylaminoethyl)-*N*-4-methoxybenzylamino]pyrimidine hydrochloride}, histamine, histidine, and histidine methyl ester. Stable complexes were formed in all cases. However, thermodynamic data have not been reported for Neohetramine because, in this case, the electrode reaction was irreversible, thus precluding an exact thermodynamic treatment.

[1] Andrews, Lyons, and O'Brien, *J.*, 1962, 1776.
[2] Smith, Doctoral Diss., Kansas State Univ., 1960.

Experimental

Materials.—Reagent-grade cadmium nitrate and potassium nitrate were used without purification. Stock solutions of cadmium nitrate were analysed gravimetrically by conversion into cadmium sulphate. Chemicals used as ligands were supplied by various manufacturers as the best grade available. Ephedrine was obtained from K & K Laboratories; histamine from Eastman; and histidine and its methyl ester from Nutritional Biochemicals Corp. The antihistamines were graciously presented by the following manufacturers: Antistine, Ciba Pharmaceutical Co; Benadryl, Parke-Davis and Co.; Neohetramine, Neparo Chemical Co. Chemical analysis of the antihistamines has been previously reported.[1] Triton X-100 (Röhm and Haas Co.) was used as a maximum suppressor in the histidine and histidine methyl ester systems.

Apparatus.—Polarographic current–voltage curves were obtained with a Sargent model XXI polarograph. Potential measurements were made with respect to the saturated calomel electrode (S.C.E.) by means of a Rubicon potentiometer no. 2730. Measurements of pH were made with Leeds and Northrup pH meter no. 7663AL. Glass electrode no. 1199-30 was used at 25° and 45°. Measurements at 0° were made with glass electrode no. 1194-44.

Cylinder nitrogen, purified by an alkaline-pyrogallol train, was used to remove air-oxygen from the cell solution. During electrolysis a nitrogen atmosphere was maintained over the solution. A constant and reproducible drop time was obtained by means of a capillary stand-tube affixed to a millimeter scale. In 0·10M-potassium nitrate at 25°, at an applied potential of −1·000 v, the capillary characteristics of the dropping mercury electrode (D.M.E.) were: $m = 2{\cdot}44$ mg. sec.$^{-1}$ and $m^{2/3}t^{1/6} = 2{\cdot}28$ mg.$^{2/3}$ sec.$^{-1/2}$. In all systems the cadmium concentration was 5×10^{-4}M.

Discussion

The reversibilities of the electrode processes were established by plots of $\log[(i_d/i) - 1]$ against the potential of the dropping electrode.[3] In each case these plots were linear with slopes in agreement with the theoretically expected slope. Further, the half-wave potential values obtained from these plots were corrected for IR drop.

The number of ligands co-ordinated to a central cadmium(II) ion was calculated from the slope of plots of $E_{\frac{1}{2}}$ against log concentration of free ligand, $[\mathrm{li}]_f$, according to the following equation:[3,4]

$$\Delta E_{\frac{1}{2}}/\Delta \log [\mathrm{li}]_f = -p(2{\cdot}303\ \boldsymbol{RT}/n\boldsymbol{F}). \quad (1)$$

where p is the number of ligands co-ordinated. The concentration of free ligand was calculated from the pH of the solution, the pK of the ligand, and the effective concenreation of ligand by means of the relation

$$\mathrm{p}K - \mathrm{pH} = \log\{([\mathrm{li}]_c/[\mathrm{li}]_f) - 1\}, \quad (2)$$

where $[\mathrm{li}]_c$ is the effective concentration of ligand and was obtained by subtracting the concentration of ligand combined with cadmium from the total molar concentration of ligand present.

Values for the base dissociation constants of the ligands, obtained by potentiometric titration, are listed in Table 1. The experimental values for p are given in Table 2. Although ligand concentrations as low as 10^{-3}M were used, no evidence was found for complexes lower than 2 : 1.

The over-all stability constants (formation constant), K_{st}, were calculated by using the relation:

$$(E_{\frac{1}{2}})_c - (E_{\frac{1}{2}})_s = -(2{\cdot}303\boldsymbol{RT}/n\boldsymbol{F}) \log (K_{st} f_s k_c / f_c k_s) - p(2{\cdot}303\boldsymbol{RT}/n\boldsymbol{F}) \log [\mathrm{li}]_f. \quad (3)$$

In the latter equation, f is the activity coefficient and k is a constant which is proportional

[3] Lingane, *Chem. Rev.*, 1941, **29**, 1.
[4] Li and Chen, *J. Amer. Chem. Soc.*, 1958, **80**, 5678.

TABLE 1.

Ionisation constants for protonated ligands.

	pK for H_2li			pK for Hli		
	0°	25°	45°	0°	25°	45°
Antistine	2·30	2·37	3·15	10·81	10·10	9·43
	2·37 *	2·45 *		11·09 *	10·13 *	
Benadryl				9·65	9·10	8·64
				9·67 *	9·12 *	
Ephedrine				10·47	9·56	8·94
				10·45 †	9·60 †	8·95 †
Histamine	6·59	6·13	5·63	10·71	9·88	9·10
	6·63 ‡	6·13 ‡		10·5 ‡	9·88 ‡	
Histidine	6·65	6·08	5·66	9·97	9·18	8·63
Histidine methyl ester	5·62	5·30	5·12	7·82	7·32	6·94

* Ref. 1. † Ref. 2. ‡ Mickel and Andrews, *J. Amer. Chem. Soc.*, 1955, **77**, 5291.

to the square root of the diffusion coefficient of the ion. The subscripts c and s refer to the complex and simple ion, respectively. The ratio, $f_s k_c / f_c k_s$, is often assumed to be unity. Here, however, the ratio k_c/k_s was determined experimentally from the ratio of the observed diffusion currents of the complex and simple metal ions as shown by

$$(i_d)_c/(i_d)_s = (D_c/D_s)^{\frac{1}{2}} = k_c/k_s, \quad (4)$$

where D represents the diffusion coefficient. This equation was obtained from the ratio of the Ilkovic equation [5] for the complex ions to that for the simple ions.

The values of the stability constants are given in Table 2. Also included in Table 2 are the values of $\Delta G°$, $\Delta H°$, and $\Delta S°$ for the complex formation reactions. The enthalpy

TABLE 2.

Thermodynamic quantities for cadmium(II) complexes.

	Temp.	$\bar{p}$ (exptl.)	log K_{st} (log K_1K_2)	$-\Delta G°$ (kcal./mole)	$-\Delta H°$ (kcal./mole)	$\Delta S°$ (e.u.)
Antistine	0°	1·9	10·08	12·6		
	25	1·8	8·73	11·9	17·7	−19
	45	1·7	8·10	11·8		
Benadryl	0	1·9	7·86	9·8		
	25	2·0	7·28	9·9	8·6	4
	45	1·8	6·89	10·0		
Ephedrine	0	1·8	6·94	8·7		
	25	1·9	6·49	8·8	10·8	−8
	45	1·8	5·55	8·1		
Histamine	0	1·8	9·60	12·0		
	25	1·9	8·57	11·8	11·8	1
	45	1·9	8·30	12·1		
Histidine	0	1·8	11·40	14·2		
	25	1·8	10·20	13·9	14·2	0
	45	1·8	9·90	14·4		
Histidine methyl ester	0	1·8	8·34	10·4		
	25	1·9	7·42	10·1	11·2	−3
	45	1·8	7·10	10·3		

values were obtained by least-squares for log K_{st} against $1/T$. The entropy values were calculated by the Gibbs–Helmholtz equation. It is estimated that the values for these functions are correct within ±1·0 kcal./mole for $\Delta G°$ and $\Delta H°$ and ±1·0 e.u. for $\Delta S°$.

The structure of the Antistine molecule seems to indicate a possibility of chelation with metal ions to form five-membered rings. The magnitudes of log K_{st} appear to support this. They are of the same order as those for cadmium complexes known to have chelate structures as a consequence of nitrogen co-ordination. Cadmium(II) complexes of histamine and histidine methyl ester are examples of such chelates. Benadryl and

[5] Ilkovic, *Coll. Czech. Chem. Comm.*, 1934, **6**, 498.

ephedrine, having lower stabilities, probably form monodentate complexes; consideration of their structures leads to the same conclusion; the bulky nature of these molecules would be expected to produce steric resistance to the formation of complexes having orders greater than 2 : 1. From Table 2 it is seen that the log K_{st} of the cadmium(II) complex of histidine is 1·6—1·8 units greater than for the corresponding histamine complex. Such an increase could indicate some degree of carboxyl involvement in the histidine complex. An earlier observation [6] of complex formation between cadmium(II) and histidine was reported at a single temperature (25°) and an ionic strength of 0·15. The value of log K_1K_2 was reported as 11·10 but thermodynamic values were not given.

It has been suggested by Andrews, Lyons, and O'Brien [1] that the free energies of formation for the complexes involving a given metal ion may possibly be used as an index of the effectiveness of the various antihistaminic substances. It seems reasonable, for purposes of comparison, that the free energy should be related to the number of co-ordinate covalent bonds formed. The $\Delta G°$ for the binding of one histamine molecule represents the formation of two co-ordinate bonds. This should be compared with the $\Delta G°$ due to the binding of one bidentate or two monodentate antihistamine ligands. On this basis the order of spontaneity of complex formation is found to be Benadryl > ephedrine > Antistine > histamine. It is noted that Idson,[7] in his summary of antihistamine drugs, has also rated Benadryl as the highest in antihistaminic activity of all compounds in its class, as determined by clinical studies.

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Department of Chemistry, Kansas State University,
Manhattan, Kansas, U.S.A. [*Received, April 17th*, 1963.]

[6] Li and Manning, *J. Amer. Chem. Soc.*, 1955, **77**, 5225.
[7] Idson, *Chem. Rev.*, 1950, **47**, 307.