Translated from *Uspekhi Khimii* **59** 375–400 (199) U.D.C. 541.6

The structure and properties of complexes simulating molecular recognition

O A Raevskii
Institute of Physiologically Active Compounds, USSR Academy of Sciences, Chernogolovka

ABSTRACT. **The main trends in the evolution of the concept of molecular recognition are examined. The possibility of using thermodynamic parameters for a quantitative characterisation of recognition and the effect of their relationships on the degree of recognition are analysed. Complexes, containing one or more hydrogen bonds, between macrocyclic compounds and metal ions, alkyl and alkylammonium derivatives, and various anions are characterised in detail as model compounds. The problem of competitive complexation in an aqueous medium is discussed. The role of computer-aided simulation of molecular recognition in applied studies relating to the design of biologically active compounds is assessed. The suitability of such a combination of experimental and theoretical studies, in which the models for molecular recognition could be constantly improved, is noted.**

Contents

I. Introduction 219
II. Thermodynamics of molecular recognition 219
III. Complexes with hydrogen bonding 221
IV. Synthetic macrocyclic biomimetics 224
V. Simulation of equilibria in aqueous solution 227
VI. Computer-aided molecular simulation 229

I. Introduction

The phenomenon of molecular recognition may be regarded as fundamental for many biological processes—in the functioning of the immune system, the formation of substrate–enzyme and hormone–receptor complexes, the transmission of nerve impulses, and in the sense of taste and smell, etc.[1] Identification of the mechanism and nature of the interactions between the molecules in such systems is not only a fundamentally important scientific development but also should assist in the creation of a firm basis for resolving the extremely urgent applied tasks of obtaining specific biologically active compounds with pesticidal and pharmacological action.[2–4] It is obvious therefore that research workers have given very considerable attention to the problem of molecular recognition; the exponential rise in the number of publications on this topic in recent decades is an indication of this.

A considerable contribution to this scientific development has been made by the following Nobel prize-winners: E Fischer (the complementarity principle of substrate–receptor complexes), P Ehrlich (the "lock and key" mechanism in the action of the immune system), C Landsteiner (the specificity of serological reactions), L Pauling (establishment of the cooperation effect of weak forces, including van der Waals forces, electrostatic interactions, and hydrogen bonding between complementary structures), F H C Crick and J D Watson (the molecular structure of desoxyribonucleic acid), K Pedersen, J M Lehn, D Cram (the synthesis, structure, and properties of macromolecular polydentate ligands that can act as "hosts").

In the present review the task is to analyse the current trends in the development of the theory and experiment concerning the simulation of molecular recognition. The examination of material is carried out according to the order of complexity of the systems (from systems of several low molecular-weight compounds to complexes of ions and low molecular-weight compounds with synthetic macromolecular receptors and then natural molecular receptors). This type of presentation of the material is helpful for an assessment and analysis of the relative contributions of different noncovalent interactions to molecular recognition. It should be emphasised here that the degree of molecular recognition of chemical compounds increases by an order on going from a selective donor–acceptor interaction or hydrogen bonding of molecules, containing one electron-donor and one acceptor site, to selective complexation of metal ions with polydentate ligands, then to selective catalytic reactions, and finally to enzyme reactions.

The very large number of articles that have been devoted to molecular recognition make it necessary to leave out whole aspects of this topic from the present publication. However, the detailed reviews and books that have been published in recent years on the conformational analysis of macromolecules,[5–7] quantum-chemical calculations of biological systems,[8–10] and protein engineering[11] compensate for these omissions to a considerable extent. The classes of complexes reviewed have also been limited. Since the main focus of attention in this review is the nature of the noncovalent interactions in substrate–receptor systems, there is a detailed analysis of H-complexes, complexes of polydentate macrocyclic ligands, and mixed ternary complexes involving transition metal ions and aminocarboxylic and aminophosphonic acids.

II. Thermodynamics of molecular recognition

In considering the binding of a physiologically active compound A to a receptor R, the resulting concentration of complex AR can be given by the relationship†

$$[AR] = \frac{K_A [A] R_T}{1 + K_A [A]},$$

† R H Davies *Int. J. Quantum. Chem., Quantum Biol. Symp.*, 1987, (14) 221.

where K_A is the binding constant and R_T is the total number of molecules of receptor. The observed response E of the system to the introduction of compound A into it is a given function of the concentration of complex AR. In this case two possible relationships linking the binding [AR] with the observed response can occur. Thus, if the response is given by the equation

$$E = \frac{K_E\,[\mathrm{AR}]}{1 + K_E\,[\mathrm{AR}]},$$

where K_E is a given factor, when

$$E = \frac{K_A\,[\mathrm{A}]\,\tau}{1 + (\tau + 1)\,K_A\,[\mathrm{A}]},$$

where $\tau = K_E R_T$, or

$$E = \varepsilon\,\frac{K_A'\,[\mathrm{A}]}{1 + K_A'\,[\mathrm{A}]},$$

where $\varepsilon = \tau/(\tau + 1)$ and $K_{A'} = (\tau + 1)K_A$. Thus the three parameters—the binding constant K_A, the factor K_E, and the total number of molecules of receptor R_T—determine the recognitive and effector functions of a physiologically active compound A in its effect on a receptor.

The thermodynamic criteria of molecular recognition during competitive complexation in multicomponent solutions have been clearly stated in two articles.[12,13] A recognizer A_0 must bind with molecules of a particular type B_0, namely, with the correct effector, in a solution that contains N types of molecules (B_0, B_1, $B_2 \ldots B_{N-1}$) at a random series of concentrations. The probability of all the false recognitions can be denoted as P_f. Thus, the degree of recognition can be characterised by the number N of possible effectors amongst which A_0 correctly recognises B_0 and by the probability P_f of a false recognition.

For the simplest example—complexation between A_0 and B_i in the ratio 1:1—we obtain

$$\mathrm{A_0} + \mathrm{B}_i = \mathrm{A_0B}_i,$$
$$[\mathrm{A_0B}_i]/[\mathrm{A_0}]\,[\mathrm{B}_i] = K_{0i} = \exp\,(-\Delta G_{0i}/RT),$$
$$i = 0,\ 1,\ 2,\ \ldots\ N-1.$$

The probability in this case of a false recognition is equal to:

$$P_f = \frac{\sum_{i=1}^{N-1} [\mathrm{A_0B}_i]}{[\mathrm{A_0B_0}] + \sum_{i=1}^{N-1} [\mathrm{A_0B}_i]} = \frac{\sum_{i=1}^{N-1} [\mathrm{B}_i]\,K_{0i}}{[\mathrm{B_0}]\,K_{00} + \sum_{i=1}^{N-1} [\mathrm{B}_i]\,K_{0i}}$$

Hence there will be a greater degree of recognition the higher the ratio of the binding constant for correct recognition to that for false recognitions, namely, the greater the difference in free energies ($\Delta\Delta G$) which represents the difference between the concentrations of A_0B_0 and A_0B_i. The degree of recognition decreases, and hence the value of P_f increases, if the total number of molecules B_i, and B_0 in particular, is comparable to or less than the number of molecules of recogniser A_0. If the number of molecules A_0 exceeds the total number of molecules B_i and all constants K_{0i} are of a single order, virtually all of the molecules B_i will be bound in complexes A_0B_i and P_f will reach a value equal to unity.

Thus, highly specific recognition requires a large value of $\Delta\Delta G$, characterising the clearly defined capacity of the recogniser A_0 to interact with the correct (B_0) as opposed to false (B_i) effector.

It is evident from the description above that in order to develop quantitative models of molecular recognition, it is necessary to have information about the interactive capacity of very diverse electron-donating and electron-withdrawing sites. In this case the thermodynamic representation of complexation as a whole requires a determination not only of the difference in free energies but also that of the enthalpy (ΔH) and also entropy (ΔS), given by

$$\Delta G = \Delta H - T\Delta S.$$

Four possible combinations of ΔH and ΔS values, resulting in different bonding of the molecules in complexes, have been selected by Raevskii.[14]

1. There is a positive change in enthalpy and negative change of entropy on formation of a complex. In this case at all temperatures and absolute values of ΔH and ΔS the equilibrium will be displaced towards the initial compounds and complexation will not occur.

2. The change is enthalpy and entropy are positive. In this situation the final result of the reaction for complexation is determined by the ratio of the absolute values of ΔH and $T\Delta S$. There will be perceptible binding when $|T\Delta S| \gg |\Delta H|$.

3. The change in enthalpy and entropy are negative. In this case perceptible binding will occur when $|\Delta H| \gg |T\Delta S|$.

4. The change in enthalpy is negative and the change in entropy is positive. In this case for all ratios of the absolute values of ΔH and ΔS there is a maximum quantity of complex (in comparision with the first three examples).

These four possible combinations of the values of ΔH and ΔS do occur in molecular recognition processes. Highly specific recognition will occur when there are maximum absolute negative values of the difference between the enthalpy change of the recogniser interaction with the correct effector and that of its interaction with false effectors ($\Delta\Delta H$) and when there are maximum positive values of the difference in entropy change for these interactions ($\Delta\Delta S$).

It should be noted that in the vast number of publications on the thermodynamics of molecular recognition, the change in entropy is either neglected[15] or considered to be proportional to the enthalpy change, which is determined experimentally or from the change in calculated energy.[16] However, in a number of cases a significant contribution of the entropy factor to binding has been established. This relates in particular to the hydrophobic interaction which plays a significant role in biological systems. A clear example of the predominant contribution of entropy to the free energy is given by Hobza et al.[17] The significant influence of the relationship between ΔH and ΔS on the binding of ligands with receptors is convincingly shown in the study of Weiland et al.[18] It is clear from these results (Table 1) that the binding of agonists with an adrenergic receptor of turkey erythrocytes takes place with a large negative entropy, while antagonists display a relatively low enthalpy but an entropy that favours complexation. This in the complexation of *L*-adrenaline and practolol with this receptor with the same values of ΔG, binding occurs in the first case mainly because of a negative enthalpy while in the second case it occurs as a result of a positive entropy.

It is clear from these examples that a more fundamental understanding of the nature of molecular recognition is only possible when the entropy change is taken into account. Theoretical approaches to a calculation of entropy change are described in a review.[9] It is noted in this publication that for real biological systems the best method of determining the entropy change is to carry out calculations using molecular dynamics or the Monte Carlo method. However, considering the rather complex nature and high cost of such calculations, simple empirical approaches to a

determination of the changes in the thermodynamic properties of complexation should not be disregarded. Thus, a correlation between the difference in free energy and the number of covalent and ionic interactions has been reported,[19] and the difference in free energy has been calculated using a simple additive scheme.[20] In such approaches the noncovalent interactions have been divided into a number of components: hydrophobic, van der Waals, hydrogen bonding, dipole–dipole, ion–dipole, ionic, intensified ionic interactions, charge transfer, etc.[21] In order to assess the importance of the contributions of these components to the overall thermodynamic properties of specific interactions, a study of model systems in which one of the types of interaction is the predominant one is of particular importance. It is for this reason that in the present review a considerable amount of space is given to simple complexes with hydrogen bonding, complexes of macrocyclic ligands, and mixed complexes. It appears that a detailed study of such systems should favour a more precise simulation of substrate–receptor complexes.

Table 1. Thermodynamic properties of the binding of ligands with the β-adrenergic receptor of turkey erythrocytes at 310 K.

Ligands	ΔG, kcal mol^{-1}	ΔH, kcal mol^{-1}	ΔS, kcal mol^{-1} K^{-1}
Agonists:			
L-isoprenaline	−9.39	−13.39	−12.9
L-noradrenaline	−7.91	−18.86	−35.3
L-adrenaline	−7.50	−12.75	−16.9
Partial agonists:			
soterenol	−8.23	−7.84	+1.26
metaproterenol	−6.78	−10.83	−13.06
terbutaline	−6.19	−4.13	+6.65
Antagonists:			
L-propranolol	−12.51	−3.85	+27.9
pindolol	−11.85	−5.08	+21.8
zinterol	−9.13	−3.06	+19.6
metoprolol	−8.36	−0.66	+24.8
sotalol	−8.21	−2.15	+19.5
practolol	−7.46	+3.93	+36.7

III. Complexes with hydrogen bonding

1. Complexes with a single H-bond

The exceptional role of hydrogen bonding in the formation of specific substrate–receptor complexes has been quite well covered in the literature.[22–32] There is a large number of studies that contain information on an experimental determination of the thermodynamic properties of H-complexes. However, calculation methods are of greatest value for the simulation of molecular recognition, making possible an assessment of interactions in hypothetical complexes. Two approaches—quantum-chemical and empirical—can be selected here in relation to the calculation of the energy of an H-bond. At the present time quantum-chemical calculations are only applicable to small H-complexes[33–35] and this is a serious obstacle preventing their widespread application. For this reason in current methods of calculating H-complexes there is a predominance of empirical approaches—additive,[36–38] multiplicative,[39–42] and additive-multiplicative.[43,44] By using a multiplicative approach in the form of a "coefficient rule"[39] it was possible in two studies[45,46] to develop a single scale of hydrogen bonding, which led to the possibility of a quantitative comparison of electron-donating and electron-withdrawing capacities of different active sites.[47–49] The practical application of a single scale of electron-donating and electron-withdrawing coefficients in order to predict the enthalpy values of the hydrogen bonding of various molecules is only made possible by the development of a relatively simple system for calculating their values. In studies by Martynov and Raevskii[50–52] equations that relate the values of these factors with constants representing the electronic effect of the substituents form the basis of such a system. The algorithm of a programmed calculation of the values of electron-donating and electron-withdrawing factors has been reported.[53]

It should be emphasised that only the enthalpies of hydrogen bonding are discussed in the publications reviewed.[33–53] At the same time, as has already been stressed in the previous section, a knowledge of both the enthalpy and entropy change is needed for the thermodynamic treatment of complexation. At the present time, even though calculations of the change in entropy on formation of an H-bond have been carried out (see, for example, the study of Carsky and Urban[54]) they are complex and for that reason are not to be found very often in the literature. From this point of view there is interest in extending a simple multiplicative approach, as proposed[55] in a discussion of the experimental data on the complexation of electron-withdrawing phenols with various electron-donating compounds, to the problem of calculating the change in entropy.

The applicability of a multiplicative approach to the calculation of thermodynamic properties of hydrogen bonding of very diverse electron-donating and electron-withdrawing compounds has been specially tested.[56] In this study[56] data on the thermodynamic properties of 174 systems was analysed, 27 compounds acted as electron donors (dioxan, tetrahydrofuran, acetone, dimethylformamide, benzaldehyde, cyclohexanone, various sulphoxides, sulphodioxides, phosphoryl compounds, pyridine, acetonitrile, thiophosphoryl and selenophosphoryl derivatives), while 23 compounds acted as electron acceptors (substituted phenols, alcohols, carboxylic acids, and their halogen-substituted derivatives, amines, nitroamines, chloroform). By using this collection of data for determination of the values of electron-donating (E_j) and electron-withdrawing (E_i) factors, 174 nonlinear equations of the form:

$$\Delta H_{ij} = \Delta H_{11} E_i E_j,$$

were drawn up, where the values E_i of twenty-two acceptors (for phenol it is assumed that $E_i = -1.00$) and the values E_j of twenty-six donors (for diethyl ether $E_j = 1.00$) acted as unknown quantities. The solution of these equations led to a determination of the $E_i(E_j)$ values of all the compounds used in this study. It transpired that the electron-donating and electron-withdrawing capacity of the compounds varied over a wide range (E_i from −0.25 to −1.70 and E_j from 0.70 to 1.70) and were considerably dependent on the nature of the substituents at the active site. In this case the ranges of the $E_i(E_j)$ values for the different forms of active sites showed considerable overlap, which provides the opportunity for developing compounds that are capable of forming the required complexes from examples of different classes. A clear picture of the convergence of the experimental values of enthalpy and those calculated from $E_i(E_j)$ values is given by Fig. 1*a*. When

represented mathematically the correlation between ΔH_{calc} and ΔH_{exp} appears as the following:

$$\Delta H_{calc} = (-0.23 \pm 0.65) + (0.99 \pm 0.03)\Delta H_{exp},\ \text{kJ mol}^{-1}$$

$$(n = 174;\ R = 0.98;\ SD = 1.82;\ F = 5307)$$

Comparison of the data from particular systems showed that only in one out of 174 cases did the discrepancy between the experimental and calculated values of ΔH exceed 5.0 kJ mol^{-1}. In 18 systems the discrepancy was 3.0 – 4.5 kJ mol^{-1}. In the remaining 155 systems the calculated values were no different from the experimental values within the limits of experimental error. Thus the multiplicative principle for calculating the enthalpy of a hydrogen bond was borne out by studying a wide variety of electron-donating and electron-withdrawing compounds. It should also be emphasised that since the range of enthalpy change in the systems studied was 5.0 – 65.0 kJ mol^{-1}, the coefficient rule represents weak, medium, and strong H-complexes to the same extent.

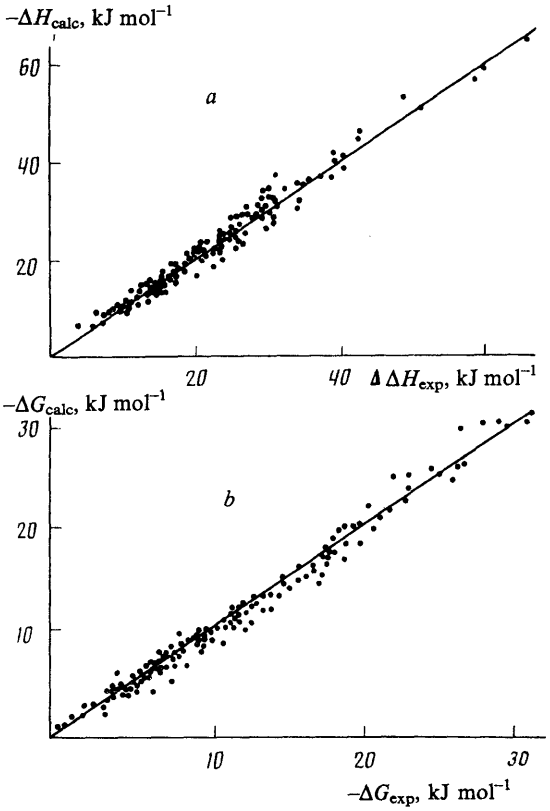


Figure 1. Graphical correlation of the experimental values and those calculated by an empirical multiplicative scheme for the enthalpy (*a*) and free energies (*b*) of H-complexes.[56]

In the same study[56] the coefficient rule was extended for the first time to free energy. For this the following equation was used:

$$\Delta G_{ij} = \Delta G_{11} C_i C_j,$$

where C_i and C_j are electron-withdrawing and electron-donating free-energy factors and ΔG_{11} is the free energy for complexation between phenol and diethyl ether in CCl_4 at 298 K. Formulation of 174 nonlinear equations of this type led to the derivation of the $C_i(C_j)$ values, which made it possible to calculate values of ΔG with an error no greater than the experimental error (Fig. 1*b*).

$$\Delta G_{calc} = (0.006 \pm 0.27) + (1.01 \pm 0.02)\Delta G_{exp}\ \text{kJ mol}^{-1}$$

$$(n = 174,\ R = 0.99,\ SD = 0.96,\ F = 8733).$$

Thus, by using the coefficient rule on the basis of the multiplicative principle it was possible to carry out accurate calculations of the thermodynamic parameters of hydrogen bonding. In order to demonstrate this, the experimental values of the difference in enthalpy and free energy and those calculated from the coefficient rule are given in Table 2 together with the binding constants for certain systems not included in the collection of data from which the values of the factors were calculated.

Table 2. Experimental and calculated values of the thermodynamic parameters of some H-complexes (solvent—CCl_4, 298 K, 1:1 complexes).[56]

Electron acceptor	Electron donor	ΔH, kJ mol^{-1}		ΔG, kJ mol^{-1}		log K, litres	
		exp.	calc.	exp.	calc.	exp.	calc.
C_6H_5OH	CH_3CN	14.2	12.5	4.50	4.16	0.79	0.73
n-ClC_6H_4OH	CH_3CN	15.1	13.5	4.73	5.12	0.83	0.90
CF_3CH_2OH	$HC(O)N(CH_3)_2$	25.5	23.2	9.18	10.04	1.61	1.76
CF_3CH_2OH	$CH_3C(O)N(CH_3)_2$	26.8	24.9	10.49	11.18	1.84	1.96
CCl_3COOH		33.0	34.1	14.08	10.8	2.47	1.89
		14.2	15.6	3.19	4.99	0.56	0.88

Apart from the collection of data used in this study[56] there is quite a considerable amount of information in the literature on the thermodynamic properties of hydrogen bonding of a wide variety of compounds. However, these data are incomplete and therefore for the time being cannot be used for a systematic analysis of electron-donating and electron-withdrawing functions of the active sites. Nevertheless, it may be assumed that the growing scientific interest in a quantitative measurement of intermolecular interaction and the improvement in measuring techniques and mathematical simulation will lead in the near future to a considerable expansion of experimental data that can be used to develop accurate calculation methods of the thermodynamic properties of hydrogen bonding.

It should be emphasised that the random collision of molecules is too "unreliable" a process for specificity to be exhibited.[57] It is necessary to define the direction of interaction. Hence a determination of the steric configuration of the interacting atoms in the molecular complexes is of considerable importance. The study of Murray-Rust and Glusker[58] is worthy of special mention here in relation to H-complexes. In this study, in order to determine the directivity of H-bonds involving sp^2- and sp^3-hybridised orbitals of

oxygen atoms, a systematic analysis of the structural data on several thousand of such compounds available on the Cambridge crystallographic data bank was carried out. Fairly strict requirements [effective localisation of the atoms, a small *R*-factor (less than 0.1), etc.] were applied to the selected data. In this case it was assumed that an intermolecular H-bond occurs when the distances between two electron-donating sites (one of which contains a hydrogen atom capable of forming a hydrogen bond) are less than 3.0 Å. Classification of the structural data on H-complexes of various cyclic ethers and ketones, epoxide compounds, acyclic ethers, ketones, esters, and other oxygen-containing compounds made it clear that hydrogen bonding occurs in all cases occurs preferentially in certain directions and that probably the most important factor is that zones of increased electron density from the unshared electron pairs (UEPs) are distributed along these directions. Thus, for ethers and epoxides hydrogen bonding occurs in a plane orthogonal to the plane of C–O–C, whereas for ketones and esters it occurs in the plane of CC(=O)Y, where Y = C, O. Murray-Rust and Glusker[58] concluded from this that the directivity of proton-donating groups towards UEPs is a characteristic feature of H-complexes.

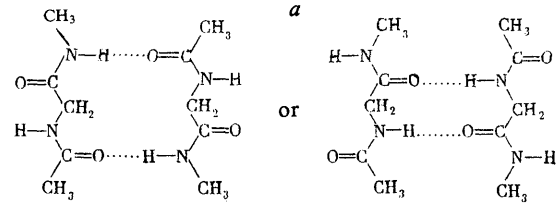
2. Complexes with several H-bonds

A study of the interaction of molecules, each of which contains several sites capable of forming H-bonds, is of particular importance in relation to molecular recognition. It is in these cases that there is a theoretical possibility for complexes with very large values of binding constants to occur both as a result of the summation of the enthalpies from each H-bond and as a result of the change in entropy in the conformational reorganisation of the interacting molecules providing the most convenient relative orientation of the atoms participating in the formation of H-bonds. Unfortunately, while there is quite an extensive amount of data on the thermodynamics, structure, and physicochemical properties of complexes with a single H-bond, the literature information on complexes with several H-bonds is very sparse. It is therefore useful to analyse the data from the few studies devoted to an experimental physicochemical investigation of such complexes.

A detailed study of the thermodynamics of the complexation of compounds, each containing two active sites, was carried out by Hopman.[59] In this study the thermodynamic parameters for autoassociation of 1-isobutyl-6-methyluracil (IBMU), γ-butyrolactam (BL), δ-valerolactam (VL), and ε-caprolactam (CL) were determined using UV-spectrophotometric titration, and their interaction with 2-aminopyridine (AP) and 2-aminopyrimidine (APM) was studied. Table 3 summarises the results obtained, which lead to the conclusions that: 1) while autoassociates of IBMU are least in strength, complexes of IBMU with AP and APM have a substantial value of the free energy difference because of the lower absolute value of the negative change in entropy in the series of compounds studied; 2) while the strength of complexes of BL with AP and APM are virtually the same, the free energy difference of the latter is twice as large as the former; 3) replacing the solvent (cyclohexane by benzene) has little effect on the enthalpy values of the reactions of both homo- and hetero-association (however, the free energy is markedly reduced). The results given above indicate with certainty that in the case of the hydrogen bonding of several sites a significant role is played by the steric structure of the interacting molecules. Unfortunately, the method selected in this study[59] can shed hardly any light in relation to this (as regards details about the effect of structure on complexation).

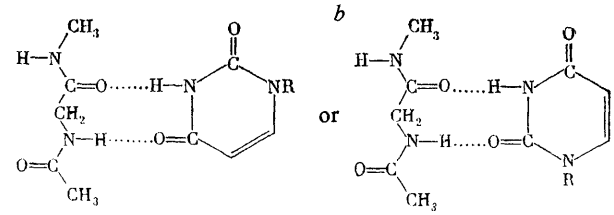
In one article[60] IR spectroscopy, which is more informative for determining structure, was used to study similar systems. In this case complexation between acetylglycine *N*-methylamide (AGMA) and 1-cyclohexyluracil (CHU) was studied. From the enthalpy value determined for homoassociation of AGMA and also the observed pattern of change in intensities of the bands with temperature and concentration, Carmona and Garcia-Ramos[60] were able to deduce that the homoassociates are dimers formed from each other:

a



The following structure was proposed for the heteroassociates:

b



It is noted here that since the ΔH values of heteroassociation do not constitute twice the value of ΔH for a single bond of this type, this suggests that steric hindrance has a considerable effect on complexation.

Table 3. Thermodynamic parameters for complexation of some compounds with two sites capable of forming hydrogen bonds (solvent — cyclohexane).[59]

System*	$-\Delta H$, kJ mol^{-1}	$-\Delta S$, J mol^{-1} K^{-1}	$-\Delta G$, kJ mol^{-1}
BL+BL	37.4	65.7	18.0
VL_VL	40.0	72.0	18.8
CL+CL	37.6	74.0	15.8
CL+CL	35.7**	101.0**	5.9**
IBMU+IBMU	18.3	24.7	11.0
BL+AP	25.2	68 8	4.9
BL+APM	25.8	55.0	9.8
VL+AP	23.0	58.6	5.7
VL+APM	23.3	54.6	7.2
CL+AP	25.3	62.9	6.7
CL+APM	25.5	48.9	11.0
CL+APM	24.3**	71.3**	3.3**
IBMU+AP	29.0	58.4	11.8
IBMU+APM	26.8	49.4	12.2

*For notation, see text; **solvent — benzene.

In another article[61] the possibility of forming simultaneously two H-bonds of the type NH...N and/or N–H...O is examined. 2-Aminopyridine and 2-pyridone were taken as research compounds.

In this study calculations on the energy of formation of H-complexes using the CNDO/2 method were carried out together with the collection of experimental data on the thermodynamic parameters of complexation. It was established that the enthalpy of dimerisation of 2-aminopyridine is approximately twice as high as the usual values of the enthlapy of complexation for compounds that each contain one nitrogen site and one-H-bond. Thus, formation of an eight-membered ring with four nitrogen atoms is very favourable in this case in terms of enthalpy. The enthalpy value for the formation of a 2-aminopyridine-2-pyridone complex was approximately half the sum of the energies of formation of 2-aminopyridine and 2-pyridone dimers. This is clearly a reflection of the multiplicative principle for electron-donating and electron-withdrawing functions as described in the previous section.

There are other publications in the literature related to a study of complexes with two H-bonds.[62–68] Unfortunately, there is no detailed information in these publications about the molecular structure of the complexes. This makes it very difficult to interpret the results and does not offer any idea about the nature and directivity of the H-bonds.

The progress in organic synthesis has had a considerable influence on studies relating to simulation of molecular recognition. As an example, reference may be made here to the studies of Rebek et al.[69,70] In these studies the problem was to synthesise a compound which would interact in a complementary manner with diamines and would thus serve as a model for the respective receptor. A model receptor containing two carbonyl groups and an acridine nitrogen atom was synthesised as this type of compound. A very polar environment of the hydrophobic skeleton of the molecules, which assists transport of this compound through biological membranes, should be regarded as a characteristic of its structure. The complex of this model receptor with purine has three H-bonds and very much resembles complementary biological systems. Thus, it is known that purine and pyrimidine derivatives, forming part of the polymeric molecule, DNA, act as monomer units in the storage of genetic information.

It is known that in the system of H-bonds that occur in DNA, the bases of one of the chains are joined to the bases of the other chain in a very specific manner, forming two H-bonds in a thymidine–adenine pair and three H-bonds in a cytosine–guanine pair. With regard to the problem of molecular recognition, particular attention should be given to studies relating to a determination of the stability of complexes of the different pairs of bases. The available literature data on the enthalpy of interaction of such bases[22,71,72] is given in Table 4. This material suggests that the guanine–cytosine complex which occurs in DNA is very much more stable than all of the others. As a reflection of this, using very simple calculations that took into account the strength of H-bonds, Pauling and Pauling[15] established that the probability of the wrong nucleotide entering a chain was approximately one in ten million.

It should be mentioned that at the present time there are many publications relating to a determination of the role of hydrogen bonding in substrate–receptor complexes. However, the use of this material for a discussion of the role of hydrogen bonding alone is complicated because of the occurrence of other types of interaction. Nevertheless, such studies provided a better understanding of the complex processes (the mechanism of the action of α-chymotrypsin,[73] proton transfer along hydrogen-bonded residues formed by hydroxyl groups in the side-chains of amino acids,[74–76] etc.).

It should also be noted that in the simulation of substrate–receptor complexes it is not possible to take into account hydration of the molecules that can affect both the interactive ability of the active sites and the conformational state of the partners. During the last ten years a great deal of attention has been given to this problem.[77–84] However, at the present time there is still much that remains unclear about the nature of the interaction of water molecules even with very simple organic compounds.

Table 4. ΔH values for homo- and hetero-association reactions of certain bases.[22,71,72]

Base pairs	$-\Delta H$, kJ mol^{-1}	
	gas phase	nonpolar medium
Adenine–uracil	58.0	24.8
Uracil–uracil	38.0	17.2
Adenine–thymine	52.0	—
Thymine–thymine	36.0	—
Guanine–cytosine	84.0	40.0–46.0
Cytosine–cytosine	64.0	25.2

IV. Synthetic macrocyclic biomimetics

The confirmation of the cyclic structure of valinomycin and the determination in the mid-1960's of the mechanism of this and other antibiotics led to the possible idea of synthesising macromolecules that are capable of selectivity binding metals that play an important part in biological processes and of transporting them through biological membranes. In 1967 Pedersen[85] reported the synthesis and complexing properties of such compounds. The first publications of Pedersen literally started an avalanche in the development of studies on the synthesis and use of these compounds both in fundamental chemical and biological studies and in a wide variety of applied fields.[86] Many thousands of the widest variety of ligands that can be classed as crown compounds have been synthesised to date. These are mainly coronands (containing classical crown ethers with oxygen, nitrogen, sulphur, and phosphorus atoms), cryptands (bicyclic polydentate ligands), podands (open-ring crown compounds), podand-coronands, oxocrowns, spherands, etc.[87] In the present section the complexing ability of such polydentate sytems towards a wide variety of ions and molecules will be discussed very briefly.

1. H-Complexes of macrocyclic compounds

18-Crown-6 is the most frequently used electron-donating compound for such complexes. In particular, its complexes with 4-nitroaniline,[88] 3-nitroaniline,[89] urea,[90] thiourea,[91] 4-nitro-1,2-benzdiamine,[92] and 2,4-dinitrophenylhydrazine[93] have been reported and studied.

Complexes of macrocycles involving the hydrogen atoms of methyl groups are of particular interest. Thus, Rogers and Green[94] reported the results of X-ray diffraction analysis of a complex between 18-crown-6 and nitromethane. This complex consists of a single crown ether molecule and two nitromethane molecules. In this case one nitromethane molecule is positioned above and the other molecule is positioned below the plane of the macrocycle, and each hydrogen atom of both methyl groups of the two nitromethane

molecules is connected by weak hydrogen bonds to one of the six oxygen atoms. The existence of H-bonds has also been established spectroscopically for this complex. A characteristic H-bond vibration has been detected in the far-infrared region.[95] Complex formation of this type has also been established for 18-crown-6 with the hydrogen atoms of the methyl groups of dimethylacetylene dicarboxylate,[96] bis(dimethyl) sulphone,[97] and dimethyl sulphate.[98] Complexes of other macrocyclic compounds with methyl and methylene groups have also been reported.[99,100] It appears that complexes of this type have still not been studied sufficiently. At the same time, the importance of noncovalent interactions of the type C–H...X in biological systems may be very considerable, since in substrate–receptor complexes there are many CH groups adjacent to electronegative substituents and having appreciable electron-withdrawing properties on account of this.

2. Complex formation with alkyloammonium cations

The interaction of macrocycles with ammonia and alkylammonium compounds, in the same way as complexation between macrocycles and methyl groups, may be classed as "tetrahedral" recognition.[101] However, in this case, in addition to H-bonds, $N^+...O^{\delta-}$ electrostatic interactions also occur. The ratio between these two forms of interaction is 0.75:0.25.[102] In this case the strength and stability of the H-bonds depends to a considerable extent on the steric structure of the complexes. A group situated in the polar part of the molecule can affect the nature of complexation. The interaction of such groups with macrocyclic ligands can either destabilise the complex or increase its stability through hydrophobic and dipole–dipole interactions, H-bond formation, etc.

Complexes of the ammonium ion may be regarded as the simplest examples of this type of tetrahedral recognition. This ion forms complexes of medium stability with a large number of macrocyclic polyethers and cryptands. In the case of using a cryptand with four nitrogen atoms and six oxygen atoms as a ligand, an exceptionally stable complex is formed as a result of the considerable structural and energy complementarity.[103,104] As a result of such strong binding of NH_4^+ in a complex the pK_a value increases by six units. In the opinion of Lehn,[101] such effects of a change in pK_a values also exist in the active areas of enzyme molecules.

The effect of substituents in the amine group on the stability of complexes may be assessed from the results of Izatt et al.[105] It was shown that a substantial change in the nature of the substituents in NH_3Alk has little effect on the stability constants of its complexes with 18-crown-6. The effect of the structure of the macrocyclic ligand on the stability of complexes with a t-butylammonium cation has been studied in detail.[106] It was found that there was a considerable effect from the nature of the anion, the size of the ring, and the type of functional groups in the ring on the stability of the complexes. How strongly these structural features can affect the stability of such complexes is suggested by data on the discrimination of enantiomers of racemic salts by chiral macrocyclic ligands.

The concept of "chiral recognition" in the chemistry of macrocyclic compounds was developed by Cram et al.[107] using the analogy of the stereospecificity exhibited by high molecular residues in their interaction with low molecular substrates. In order for recognition of a chiral "guest" to be made possible, the "host" ligand must satisfy at least two conditions: 1) to have a cavity suitable for complexation; and 2) to have the conditions for energetically more stable interactions with only one of the two enantiomeric "guest" molecules to occur. A schematic representation of an example of "chiral" recognition is given in Fig. 2. The enantiomeric resolution achieved for the racemate of an ammonium salt[108] was obtained because in the (*R*) "host–guest" complex four strong noncovalent interactions in the cavity of the macrocycle occur whereas in the (*S*) "host–guest" complex there is no such steric correspondence.

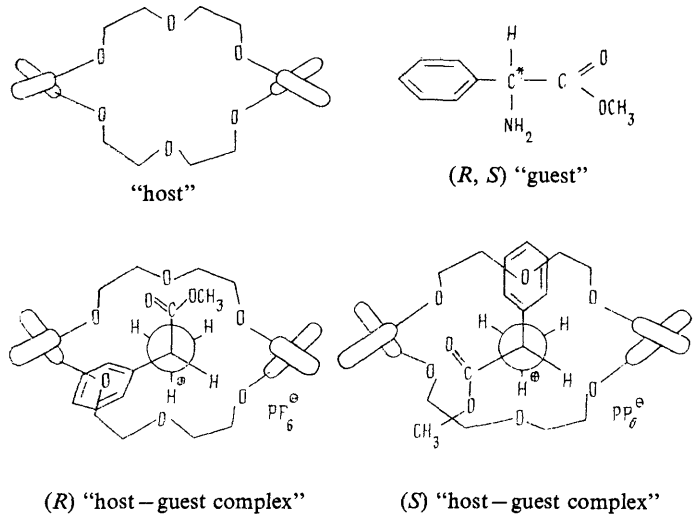
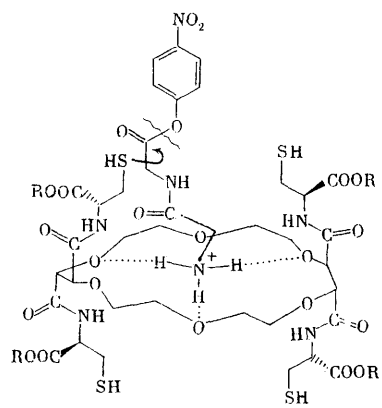


Figure 2. Schematic representation of an example of "chiral" recognition.[10]

In the simulation of enzyme systems, the process of cleavage of an ester bond, which occurs widely in metabolism, is often discussed. In the case of macrocyclic compounds it was possible to carry out this process using a tetra-*L*-cystinyl derivative which bound *p*-nitrophenyl esters of amino acids and peptides; *p*-nitrophenol was obtained as a result:[109]



In this case precise "chiral" recognition occurred between enantiomeric dipeptide esters. Many similar examples of the simulation of biological systems have been reported to date.[101,110,111] They have very considerable value for an understanding of the mechanisms of complex biochemical processes.

3. Complex formation with metal ions

The property of crown ethers that has been studied in the greatest detail is the "spherical" recognition of ions of alkali and alkaline earth metals. At the present time this topic is covered by a very large number of publications, which cannot be examined in detail within the scope of a short review. Therefore only the most prominent examples of a physicochemical study of such systems are given here. The X-ray studies of complexes of macrocycles with metal ions will be selected primarily (see several reviews[112–115]).

Let us consider 1:1 complexes between calcium and macrocycles as an example. The structure of a complex between calcium thiocyanate and 18-crown-6 has briefly been reported.[116] According to these findings, a coordination number of 8, which is typical for calcium, occurs for the metal in this complex, and the metal ion is situated inside the cavity of the macrocycle and lies at approximately the same distance from all six oxygen atoms of the macrocycle. The complex molecule has D_{3d} symmetry.

Quite a different situation has been identified in the complex of calcium nitrate with the same ligand.[117] In this case the complex molecule has C_2 symmetry. The calcium atom is also situated inside the cavity of the macrocycle. The nitrate groups act as bidentate ligands. The dihedral angles in two pairs of COCCOC residues have a *trans-gauche-trans*-conformation while in the third pair there is approximately a *trans-gauche-gauche*-conformation, with the dihedral angle in the CCOC fragment deviating by 37° from that which is usual for a *gauche*-conformation.

On the whole, the available data provide a basis for the conclusion that the conformational state of macrocyclic ligands in complexes is determined by the nature, not only of the cation, but also the anion.[118] This question has been studied in greater detail in the case of X-ray and spectroscopic analysis of benzo-15-crown-5 and its complexes with different calcium salts.[119] It was shown that the nature of the bonding between the electron-donating sites of the macrocycle and the calcium ion is determined by the combined interactions between the cation, anion, and macrocyclic ligand and electronically neutral molecules of water or solvent. This conclusion has been fully confirmed[120,121] with respect to the complexation of other macrocycles with calcium. Thus, in the established molecular structure of the complex between calcium and benzo-12-crown-4 that was isolated from acetonitrile solution, containing this ligand and calcium isocyanate as well as water of crystallisation,[120] the coordination sphere of calcium is formed by the four oxygen atoms of the macrocycle, the two nitrogen atoms of the isocyanate groups, an oxygen atom from one water molecule, and the nitrogen atom of a single acetonitrile molecule. In the case of the complex between calcium thiocyanate and dibenzo-24-crown-8,[121] the immediate environment of calcium is made up by four oxygen atoms of the macrocycle (two ether and two anisole), two nitrogen atoms of anionic isothiocyanate groups, and an oxygen atom of a coordinated water molecule. The four other oxygen atoms of the macrocycle do not participate in coordination. It appears that these types of calcium complexes of macrocycles can be regarded as models of bioorganic complexes.[122] It is possible to conclude from a number of publications[123–130] that there is evidence for competitive occupation of the coordination sphere during complexation of other metals with macrocyclic ligands. However, it should be noted in this case that a metal–ligand composition of not only 1:1 but also 1:2,[131] 2:1 [132,133] and 2:2 [134] is typical for such complexes.

Data on the thermodynamic properties of complexation of macrocyclic ligands with metal salts in solution are of considerable value in relation to molecular recognition. Considerable attention was given to this problem in the 1970's and 1980's. Several detailed reviews[135] and also the study of Buschmann‡ should be mentioned first of all. Of the original studies, mention may be made of the publication of Solov'ev at al.,[140] in which the thermodynamic properties of the complexes of two crown ethers (benzo-12-crown-4 and benzo-15-crown-5) with thiocyanate salts of lithium, sodium, potassium, calcium, and ammonia were obtained and analysed. It was established that in complexation between benzo-12-crown-4 and lithium and sodium, cation–ligand complexes occur with a composition of both 1:1 and 1:2. In the case of benzo-15-crown-5, 1:2 complexes are not formed for these salts. In complexation involving benzo-15-crown-5, the enthalpy of interaction with sodium, potassium, and calcium increases by 25% compared to benzo-12-crown-4, which may be regarded as evidence for the participation of an additional electron-donating site in complexation with benzo-15-crown-5. The entropy data are also very interesting. Thus, in complexes of lithium with benzo-12-crown-4 and benzo-15-crown-5 with composition 1:1, the enthalpy of interaction is virtually the same. At the same time, the value of the stability constant for the complex with benzo-15-crown-5 is higher by a factor of three. The small negative value of the entropy is thought to be the cause of this phenomenon. It is worth noting that in the case of the 1:2 complex between calcium and benzo-15-crown-5, the entropy change is even positive, resulting in a significant value of the stability constant even though there is a small enthalpy.

It is possible to assess how the structure of macrocyclic compounds affects their ability to bind metal ions from the data of Table 5.[141] It can be seen from Table 5 that the maximum binding results when there is a correspondence between the size of the cavity of the macrocycle and the diameter of the cation (complexation between potassium and 18-crown-6 and between caesium and

Table 5. Stability constants of 1:1 complexes of macrocyclic compounds with alkali metals in methanol at 298 K.[141]

	log *K*, litres mol^{-1}		
	Na^+	K^+	Cs^+
Tetramethyl-12-crown-4	1.41	—	—
Dicyclohexano-14-crown-4	2.18	1.30	—
Cyclohexano-15-crown-5	3.71	3.58	2.79
18-Crown-6	4.32	6.10	4.62
Cyclohexano-18-crown-6	4.09	5.89	4.30
Dicyclohexano-18-crown-6	4.08	6.01	4.61
Dibenzo-18-crown-6	4.36	5.00	3.55
21-Crown-7	—	4.41	5.02
Dibenzo-21-crown-7	2.40	4.30	4.20
24-Crown-8	—	3.48	4.15
Dibenzo-24-crown-8	—	3.49	3.78
Dibenzo-30-crown-20	2.00	4.60	—
Dibenzo-60-crown-20	—	3.90	—
Pentaglyme	1.52	3.20	—
Valinomycin*	0.67	4.90	—

*From the results of Lehn.[142]

‡ H J Buschmann *Stereochemistry of Organometallic and Inorganic Compounds* (Edited by I Bernal), Elsevier, Amsterdam–New York, 1987, p. 109.

21-crown-7). Data on an acyclic analogue — pentaglyme — and a natural ionophore — valinomycin[142] — are also given in Table 5. Comparison of these data shows that binding with macrocyclic ligands has a considerable advantage over binding with open ligands. The very high selectivity of the natural ionophore towards the potassium ion compared to the sodium ion attracts attention.

The discovery of the complexing ability of macrocyclic ligands towards alkali and alkaline earth metal was the basis for research into synthetic ionophores. Extensive studies, including a number cited here,[143–150] which led to both a better understanding of ion transport processes through biological membranes and the preparation of compounds of practical importance, have been carried out in this area up to the present time.

4. Complex formation with anions

In contrast to the coordination chemistry of cations and particularly metal ions, complexation involving anions has not been studied in sufficient detail. This situation is evidently at variance with the important role that is played by anions in chemistry and biology. That is obviously why interest in research in this area has increased markedly in recent years.[151,152]

Charged ammonium and electronically neutral residues containing strong electron-withdrawing atoms can act as atomic groups capable of binding anions. However, particularly stable and selective anionic complexes are obtained when macrocyclic molecules are used as receptors. In these cases the anionic substrate can enter the cavity of the receptor and can be stabilised in this position through different types of interactions. The structure of such complexes is analysed in detail in a study by Lehn et al.[153] A cryptand (BT) with six secondary and two tertiary amine nitrogen atoms and three ether oxygen atoms was used as the receptor. In the formation of a complex between this ligand and a fluoride ion, the latter occupies an asymmetrical position inside the cavity, probably because of the small size of the anion, and it coordinates tetrahedrally with four protonated nitrogen atoms (two from one chain and one each from two other chains). In the case of complexes with chlorine and bromine ions, the anions are positioned almost exactly on the axis joining the tertiary amine nitrogen atoms and are equidistant from them. In this case the anions are coordinated octahedrally with the six protonated secondary amine atoms through hydrogen bonds of the type NH ... X. In the azide complex the N_3^- ion lies on the N ... N axis, the two terminal atoms are at a distance of 3.24 and 3.21 Å from the tertiary amine nitrogen atoms of the ligand. In this case the terminal nitrogen atoms of the N_3^- ion form hydrogen bonds of virtually the same length (2.96 ± 0.04 Å) with the protonated secondary amine groups. These coordinate bonds form two tetrahedra. In both the free ligand and the three complexes studied an *endo,endo*-conformation occurs for the macrocyclic skeleton, with the tertiary nitrogen atoms turned towards the cavity.

Complex formation of this macrocyclic ligand with chloride, bromide, iodide, and azide anions, as well as with a number of organic and inorganic anions has been studied in solution by means of potentiometric titration (Table 6). It was established that stable complexes are formed with substrates that have the highest charge density such as the small-sized fluoride anion and polyphosphate ions. However, it follows from the literature data that when other polyammonium macrocycles are used as receptors, the binding of adenosine monophosphate (AMP^{2-}), adenosine diphosphate (ADP^{3-}), and adenosine triphosphate (ATP^{4-}) can be even more effective.[154]

The synthesis of the so-called co-receptors, which can bind either polyfunctional substrates or two or more small substrates at the same time[155] should be regarded as an important area of the chemistry of molecular receptors. Thus, polyammonium macrocycles that can effectively bind dicarboxylate anions have been obtained.[156] In this case there was a well-defined selectivity for formation of a complex between these macrocycles and dicarboxylates in whose molecules the coordination sites are separated by a different number of methylene bonds.

The significant change in ratio of the different free and bonded molecules when the solvent is varied for complexation of polyammonium macrocycles with anions is worthy of note. This evidently occurs as a result of the differing ability of partially and fully protonated structures to bind anions.

It should be mentioned in conclusion to this section that apart from the macrocyclic compounds described above, which are used as models of biological complexes related to the transport of substances and enzyme processes, synthetic porphyrins and nonporphyrinic tetraazomacrocyclic ligands have been widely used. The use of macrocyclic compounds as models of molecular recognition has also been described in reviews.[148,157–159]

Table 6. Stability constants of complexes of the two protonated forms of the macrocyclic BT ligand ($BT\text{-}6H^+$) and ($BT\text{-}5H^+$).[153]

Anion	log *K*, litres mol^{-1}		Anion	log *K*, litres mol^{-1}	
	BT-6H+	BT-5H+		BT-6H+	BT-5H+
F^-	4.10	3.20	$CH_2(CO_2)_2^{2-}$	3.10	2.00
Cl^-	3.00	1.95	SO_4^{2-}	4.90	2.90
Br^-	2.60	1.60	HPO_4^{2-}	5.50	2.75
I^-	2.15	1.55	AMP^{2-}	3.85	2.65
N_3^-	4.39	2.65	ADP^{3-}	5.85	4.15
NO_3^-	2.80	1.65	ATP^{4-}	8.00	5.40
HCO_2^-	2.30	1.50	$P_2O_7^{4-}$	10.30	6.45
$(CO_2)_2^{2-}$	4.95	3.35	—	—	—

V. Simulation of equilibria in aqueous solution

When the role of molecular recognition in specific biological processes is considered, the effect of the aqueous medium on the nature of complexation must also be taken into account. In a physiological medium a large number of chemical equilibria occur, in which hydrogen and metal ions and many biologically active compounds such as amino acids, peptides, proteins, carboxylic acids, organic phosphates, nucleic acids, and others participate. In this case there is competition between hydrogen and metals for the ligands. It should be emphasised here that even in a three-component system (one bivalent metal M^{2+} and two different bidentate ligands A and B) the formation of various species such as MA, MA_2, MB, MB_2, MAB is possible. In relation to molecular recognition there is particular interest in mixed complexes MAB. The probability of formation of the latter is twice as high compared to MA_2 and MB_2 complexes. Moreover, electrostatic and steric

interactions and hydrogen bonding,§ which is favourable for stable complexation, can occur in such mixed complexes.

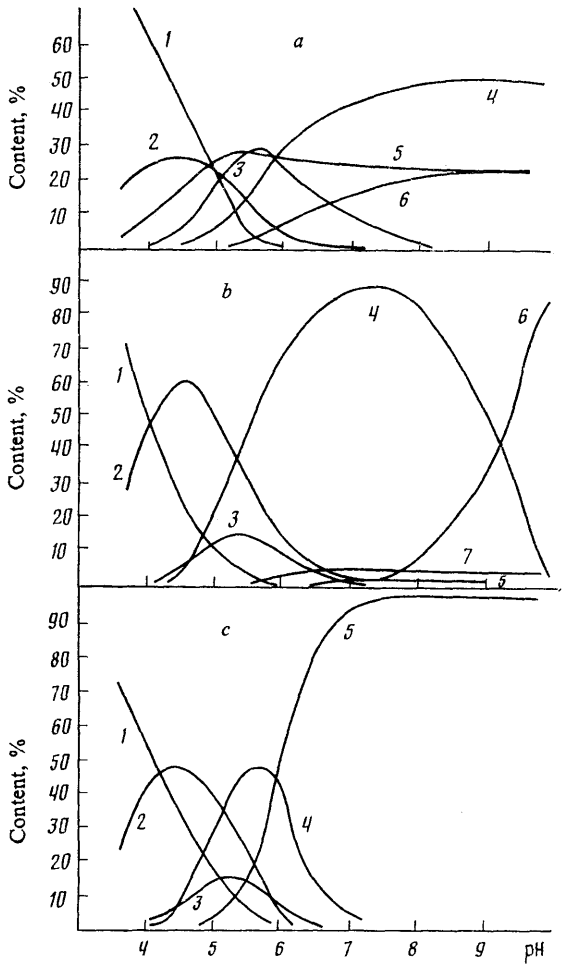


Figure 3. Graphical representation of the dependence of the composition of the following reaction systems: *a*, α-methylphenylaminophosphonic acid (MPAP) + *L*-phenylalanine (PA); the curves correspond to the change in Cu content (in %) in solution of: copper ion (curve *1*), Cu(PA) (curve *2*), Cu(MPAP) (curve *3*), Cu(PA)(MPAP) (curve *4*), $Cu(PA)_2$ (curve *5*), $Cu(MPAP)_2$ (curve *6*); *b*, MPAP + *L*-tyrosine (Tyr); the curves correspond to the change in Cu content (in %) in solution of: copper ion (curve *1*), Cu(TyrH) (curve *2*), Cu(MPAP) (curve *3*), Cu(TyrH)(MPAP) (curve *4*), $Cu(MPAP)_2$ (curve *5*), Cu(Tyr)(MPAP) (curve *6*), $Cu(Tyr)_2$ (curve *7*); *c*, MPAP + *L*-dihydroxyphenylalanine (Dopa); the curves correspond to the change in Cu content (in %) in solution of: copper ion (curve *1*), Cu(Dopa-H) (curve *2*), Cu(MPAP) (curve *3*), Cu(Dopa)(MPAP) (curve *4*), Cu(Dopa)(MPAP) (curve *5*); the content of $Cu(MPAP)_2$ and $Cu(Dopa)_2$ was less than 3%.

§ V V Lukachina *Ligand–ligandnoe Vzaimodeistvie i Ustoichivost' Raznoligandnykh Kompleksov* (Ligand–ligand Interaction and the Stability of Heteroligand Complexes), Naukova Dumka, Kiev, 1988, p. 181.

Another important characteristic of complexation in an aqueous medium is the change in the number of different complex species when the medium is varied. This results from the possible interaction of ligands with hydrogen ions when there is a change in their electron-donating properties. Since the pH values of the medium differ considerably in different parts of the organism, a study of such systems is particularly important for an understanding of molecular recognition under natural conditions.

The structure and stability of mixed complexes have been discussed in detail in three reviews.[160–162] Data on the composition of complex species in systems containing copper and two amino acids are displayed in the form of graphs in Fig. 3. A study is made on four types of system, in which one of the amino acids, namely, α-methylphenylaminophosphonic acid (MPAP), is present in all four cases, while the second varies, leading to substantial changes in the stability of the resulting complexes.[163] In the system MPAP–PA–Cu (Fig. 3*a*) a mixed complex is formed in quantities not exceeding its statistical proportion, while in the system MPAP–Dopa–Cu (Fig. 3*c*) it is evidently in excess of all other complexes. There has been a growing emphasis on studying such systems in recent years aimed at a better understanding of the complexation processes in a physiological medium and the potential practical application of the selective nature of bonding.[164–172]

To what extent competitive complexation can affect the composition and distribution of complex species in a physiological medium can be assessed from the results of calculation of a model plasma in the absence of EDPA and when this ligand is added.[160] It follows from the results in Table 7 that the addition of EDTA leads to its complete binding of zinc and copper, has a considerable effect on the type of calcium complexes, and in actual fact has no effect on the distribution of magnesium. The calculations carried out clearly demonstate that because of such a complex action on the plasma,

Table 7. Distribution of complexes in a plasma model.[160]

Type of species		Content, %		Type of species		Content, %	
metal	ligand	without EDTA	with EDTA	metal	ligand	without EDTA	with EDTA
Ca	Globulin	24.0	15.0	Zn	Histidine, cysteine	3.4	—
Ca	Albumin	16.0	13.0				
Ca	Bicarbonate	7.6	4.3	Zn	Cysteine, histidine	2.1	—
Ca	Citrate	2.4	2.0				
Ca	Phosphate	0.8	0.5	Zn (free)		4.7	0.0
Ca (free)		49.0	28.0	Zn	EDTA	—	99.99
Ca	EDTA	—	38.0	Mg	Bicarbonate	12.0	12.0
Zn	Cysteine, histidine	19.0	0.0	Mg	Phosphate	4.3	4.2
				Mg	Citrate	2.4	3.0
Zn	$(Cysteine)_2$	16.0	0.0	Mg	Lactate	0.6	0.6
Zn	$(Histidine)_2$	13.0	0.0	Mg (free)		80.0	78.0
Zn	Albumin	13.0	0.0	Mg	EDTA	—	1.5
Zn	Glycine, histidine	6.2	—	Cu	Histidine, cysteine	44.0	0.7
Zn	Histidine	3.6	—	Cu	Histidine, cysteine	38.0	0.6
Zn	Citrate	3.6	—				
				Cu	$(Histidine)_2$	16.0	0.3
				Cu	EDTA	—	98.3
				Other species		0.3	

Note. Metals and EDTA were at the following concentrations (in moles): $[Ca] = 2.5 \times 10^{-3}$, $[Mg] = 1.1 \times 10^{-3}$, $[Zn] = 46 \times 10^{-6}$, $[Cu] = 1.1 \times 10^{-6}$, $[EDTA] = 1.0 \times 10^{-3}$.

the use of EDTA as a pharmacological compound that assists the binding of free calcium can have very unfavourable results. It seems that such calculations should become an essential element of studies that precede the pharmacological insertion of ligands which effectively bind biometals.

In examining complex equilibrium systems in solution, we should not omit the progress made in programming[179] using the Newton method,[174] Simplex method,[175] Monte Carlo method,[176] and others.

VI. Computer-aided molecular simulation

The general principles of this method have been presented in sufficient detail in several studies.[177-184] Within the scope of this review it should be noted that the most important feature of any molecular computer model is its energy. In principle, quantum mechanics can provide a basis for the calculation of the molecular energies of biologically active compounds.[8-10,184,185] However, the complexity of the research subjects makes it necessary in practice to use various approximations with the calculation of force field parameters based on experimental data. Vibrational spectra are used to determine the force constants. Information on the bond lengths and bond angles obtained from X-ray diffraction analysis, gas electron diffraction, neutron diffraction, and microwave spectroscopy is used for a determination of the geometric parameters. The parameters of the interactions between nonbonded atoms are often determined from thermodynamic data (heats of mixing, evaporation, etc.).

In the computer-aided molecular simulation of a specific compound using systems that comprise a minicomputer, computer graphics, and also a data bank of structural and physicochemical properties, the design of the initial structure is carried out at the first stage. Then calculations are carried out, leading to an optimisation of the molecular geometry and a determination of the possible electronic and steric structure of the molecule and its energy properties. In this case, since the molecules of biologically active compounds usually contain several axes of internal rotation and hence have many stable conformations that are similar in energy, special routines of systematic scanning of such conformations are used.

A significant contribution to a more fundamental understanding of the structure of molecules under real conditions is provided by the computer-aided simulation of the dynamic properties of chemical systems based on the Monte Carlo method[8,186,187] and that of molecular dynamics,[188-191] which make it possible, in principle, to calculate also the entropy.[192,193]

Computer-aided simulation of molecular recognition can be carried out using calculations on the competitive interaction of several ligands (L_I, $L_{II} \ldots L_N$) with a receptor (R). If the entropy contribution to this process is neglected, by analogy with the study of Fruhbeis et al.[184] we can write:

$$\Delta E(L_I) = E(RL_I) - E(R) - E(L_I),$$
$$\Delta E(L_{II}) = E(RL_{II}) - E(R) - E(L_{II}),$$
$$\ldots\ldots\ldots\ldots\ldots\ldots$$
$$\Delta E(L_N) = E(RL_N) - E(R) - E(L_N).$$

A negative ΔE value is necessary for the formation of a complex, and for highly selective binding of L_I with the receptor, the following condition must be fulfilled:

$$|\Delta E(L_I)| \gg |\Delta E(L_{II})|, \ \ldots \ |\Delta E(L_N)|.$$

Thus, in order to assess the recognition between molecules L and R, it is necessary to have information on the steric structure and energetics not only of the molecules themselves but also for the corresponding complexes. According to Humphries,[194] three aspects need to be considered in the scanning of stable conformations of a complex: the relative orientation of L to R, the optimum conformations of L in contact with R, and the optimum conformations of R in contact with L. All these questions have now been successfully resolved using computer docking of molecules.[182,184,195,196] This is assisted by quite a well developed contemporary mathematical facility for molecular computer graphics, which makes it possible to carry out numerous manipulations on the display screen: to represent the molecules as a combination of bonds (Dreiding models) or as overlapping spheres of atoms. The image can be rotated, fragmented, superimposed, put in perspective, etc.[197,198]

It is very important to emphasise that the methodological developments in computer-aided molecular design as described above have been applied very quickly to the development of biologically active compounds. There are a number of review articles on this topic.[2,19,199-202] As an example we can examine a recent publication,[203] in which the computer analysis of basic structures for interaction with papain and carbonic anhydrase was carried out. The X-ray diffraction data on these enzymes were taken from the study of Bernstein et al.[204] Compounds selected from the widely used Cambridge databank of X-ray diffraction data were investigated as interaction partners. Docking of the active sites of the pairs of molecules was carried out using a programmed procedure described by Kuntz et al.[205] Amongst the identified compounds whose active sites were effectively docked with papain were: 3-iodophenyl hippurate (IPH) (a known substrate of this enzyme), a derivative of trimethoxybenzamide (TMB), and three macrocyclic compounds—2,6-pyridine-24-crown-8 (PC8), 10,10-ethylene-bis(1,4,7-trioxa-10-azocyclododecane) (TACD), and 2,2-bis(4,4-benzophenono(2,6)naphthalenophane) (BPN). The calculated energies of these ligands and their complexes with papain are given in Table 8. The results in Table 8 are an indication of the high stability of these complexes.

Table 8. Energies of complexation of a number of compounds with papain (calculated value of the free energy of papain is equal to 266.2 kJ mol^{-1}).[203]

Ligand	E_L, kJ mol^{-1}	E_C, kJ mol^{-1}	E_I, kJ mol^{-1}
PC8	22.5	−2784.0	−141.3
TACD	60.2	−2712.7	−117.7
TBA	145.6	−2555.1	−135.6
BPN	82.2	−2696.0	−113.0
IPH	24.7	−2741.2	−100.7

Note. For ligand notation, see text; E_L—energy of free ligand; E_C—energy of complex; E_I—energy of interaction.

In recent years studies of computer-aided methods for the simulation of molecular recognition have been increasingly extended to a determination of the relationships between the structure of

compounds and the biological activity which they exhibit. In particular, simulation has been carried out for the inhibition of dihydrofolate reductase,[206,207] and elastase,[208] the binding of DNA,[209] adrenergic and muscarinic receptor systems,[210,211] the complexation of ligands with a serotonin receptor[212] and cytochrome,[213] the mimetics of α-chymotrypsin,[214] the inhibition of photosynthesis,[215] the reactions of ribonucleic acid with synthetase[216] and different drugs with tubulin.[217]

The simulation of bioorganic complexes in an aqueous medium may be regarded as a close approximation to reality. Thus, the results of studies on the effect of hydration on ionic permeability processes have been correlated.[218] In these studies complexes of valinomycin, nonactin, and dialkyl-substituted dibenzo-18-crown-6 with potassium[219–221] and antibiotic X 537 A with serotonin, dopamine, noradrenaline, and adrenaline[222] have been investigated. The ionic channels formed in a lipid membrane by gramicidin A [223] and amphotrycin B [224] have also been simulated.

— o0o —

In view of the progress made in the 1980's in the fine organic synthesis of model compounds, physicochemical techniques, mathematical methods for analysis of the properties of complex equilibrium systems, and computer-aided molecular design it is possible to agree with the view that "we are starting to recognise the molecular parameters which control the reactions of ligands with macromolecules".[2] However, in order that the concept of molecular recognition should become a firm principle in the directed search for compounds with given properties, further development of fundamental studies in many areas is necessary. This primarily concerns studies to determine the thermodynamic properties of H-complexes in different solvents in order to identify the principles concerning the effect of the medium on complexation. The extremely sparse amount of information at present on the detailed molecular structure and thermodynamics of complexes with several H-bonds should also be mentioned.

Determination of the role of the entropy factor in the binding of substrate–receptor complexes is of particular importance for a more fundamental understanding of molecular recognition. At the present time the entropy effect is neglected in the vast majority of studies. However, there are findings which suggest that a decisive role is actually played by this factor, for example, in the complexation of different ligands with similar enthalpy values. Hence, the usefulness of experimental studies to determine the entropy as well as calculations, particularly those using the Monte Carlo method and molecular dynamics, is evident.

In the final section of this review the growing contribution of computer-aided molecular simulation to the study of molecular recognition was noted. For all the evident merits of this method, it should not be forgotten that it is based on fairly incomplete knowledge of the nature of intra- and inter-molecular interactions. On this basis it seems reasonable to combine the different types of studies so that the models of molecular recognition can be constantly improved through the application of accurate new experiments as well as theoretical developments.

References

1. J Avery *Int. J. Quant. Chem.*, 1984, **26** 843
2. C Hansch *Khim.-farm. J.*, 1980, **14** 15
3. C Hansch, J M Blaney *Drug Design: Fact or Fantasy* (Edited by J M Blaney), Academic Press, 1984, p. 185
4. N C Cohen *Trends Pharm. Sci.*, 1985, **4** 503
5. V G Dashevskii *Konformatsionnyi Analiz Makromolekul* (Conformational Analysis of Macromolecules), Izd. Nauka, Moscow, 1987
6. E Osawa, H Husso *Angew. Chem. Int. Ed. Engl.*, 1983, **22** 1
7. B Beagley *Molecular Structure by Diffraction Methods* (Edited by L E Sutton and M R Truter), Chemical Society, London, 1978, **6** 63
8. E Glementi *J. Phys. Chem.*, 1985, **89** 4426
9. G Naray-Szabo, P R Surjan *Theoretical Chemistry of Biological Systems* (Edited by G Naray-Szabo), Elsevier, Amsterdam, 1986, p. 1
10. A A Ovchinnikov, A I Boldyrev *Usp. Khim.*, 1986, **55**(4) 539 [English translation in *Russ. Chem. Rev.*, 1986(4)]
11. A V Finkel'shtein, N P Kirpichnikov, O B Ptitsyn, K G Skryabin *Vestn. Akad. Nauk SSSR*, 1988, 102
12. Z Simon *Angew. Chem. Int. Ed. Engl.*, 1974, **13** 719
13. R C Hayward *Chem. Soc. Rev.*, 1983, **12** 285
14. O A Raevskii *Teor. Eksp. Khim.*, 1986, **22** 450
15. L Pauling, P Pauling *Chemistry* (Translated into Russian from English), Izd. Mir, Moscow, 1978, p. 458
16. G Naray-Szabo *J. Am. Chem. Soc.*, 1984, **106** 4584
17. P Hobza, F Mulder, C Sandorfy *J. Am. Chem. Soc.*, 1982, **104** 925
18. G A Weiland, K P Minneman, P B Molinoff *Nature (London)*, 1979, **281** 114
19. C R Beddel *Chem. Soc. Rev.*, 1984, **13** 279
20. P R Andrews, D J Craik, J L Martin *J. Med. Chem.*, 1984, **27** 1648
21. H Kubinyi *Chem. unser. Zeit.*, 1986, 191
22. D Hadzi *Spectroscopy of Biological Molecules* (Edited by C Sandorfy et al.), Reidel Publishing Co., New York, 1984, p. 61
23. G Wippf, A Dearing, P K Weiner *J. Am. Chem. Soc.*, 1983, **105** 997
24. G Naray-Szabo, A Kapur, P G Mezey, L Polgar *J. Mol. Struct.*, 1982, **90** 137
25. G Naray-Szabo *J. Mol. Struct.*, 1986, **138** 197
26. C Sandorfy, R Buchet, P Mercier *Spectroscopy of Biological Molecules* (Edited by Sandorfy et al.), Reidel Publishing Co., New York, 1984, p. 563
27. H Merz, G Zundel *Biochem. Biophys. Res. Commun.*, 1981, **101** 540
28. W F Anderson, D H Ohlendorf, Y Takeda, B W Mattews *Nature (London)*, 1981, **290** 754
29. C Ceccarelli, G A Jeffrey, R Taylor *J. Mol. Struct.*, 1981, **70** 255
30. V P Pankov, R G Zhbankov *Vnutri- i Mezhmolekulyarnye Vzaimodeistviya v Uglevodakh* (Intra- and Inter-molecular Interactions in Carbohydrates), Izd. Nauka i Tekhnika, Minsk, 1988, p. 357
31. D L Ollis, S W White *Chem. Rev.*, 1987, **87** 981
32. J A Gerlt *Chem. Rev.*, 1987, **87** 1079

33. V P Bulychev, N D Sokolov *Vodorodnaya Svyaz'* (Hydrogen Bonding) (Edited by N D Sokolov), Izd. Nauka, Moscow, 1981, p. 10
34. K Morukuma, K Kitaura *Chemical Application of Atomic and Molecular Electrostatic Potentials* (Edited by P Politzer, et al.), Plenum Press, New York, 1980, p. 215
35. A A Bliznyuk, A A Voityuk *Zh. Strukt. Khim.*, 1988, **29** 31
36. M A Spackman *J. Chem. Phys.*, 1986, **85** 6579
37. M A Spackman *J. Chem. Phys.*, 1986, **85** 6587
38. M A Spackman *J. Phys. Chem.*, 1987, **91** 3179
39. A V Iogansen *Teor. Eksp. Khim.*, 1971, **7** 302
40. A V Iogansen *Teor. Eksp. Khim.*, 1971, **7** 312
41. A V Iogansen *Vodorodnaya Svyaz'* (Hydrogen Bonding) (Edited by N D Sokolov), Izd. Nauka, Moscow, 1981, p. 112
42. V A Terent'ev *Termodinamika Donorno-aktseptornoi Svyazi* (The Thermodynamics of Donor-Acceptor Bonding), Izd. Sarat. Univ., Saratov, 1981
43. R M Guidry, R S Drago *J. Am. Chem. Soc.*, 1973, **95** 759
44. M K Kroeger, R S Drago *J. Am. Chem. Soc.*, 1981, **103** 3250
45. O A Raevskii, V P Novikov *Khim.-farm. Zh.*, 1982, **16** 583
46. O A Raevskii, V V Avidon, V P Novikov *Khim.-farm. Zh.*, 1982, **16** 968
47. O A Raevskii *Vvedenie v Konstruirovanie Biologicheski Aktivnykh Veshchestv* (Introduction to the Design of Biologically Active Compounds), D I Mendeleev MKhTI, Moscow, 1984, p. 80
48. O A Raevskii, V Yu Grigor'ev *Zadachi po Kursu "Konstruirovanie Biologicheski Aktivnykh Veshchestv"* (Problems from the Course "The Design of Biologically Active Compounds"), D I Mendeleev MkhTI, Moscow, 1986, p. 80
49. O A Raevskii, V Yu Grigor'ev, V P Solov'ev, I V Martynov *Dokl. Akad. Nauk SSSR*, 1988, **298** 1186
50. I V Martynov, O A Raevskii *Dokl. Akad. Nauk SSSR*, 1982, **265** 664
51. I V Martynov, O A Raevskii *Zh. Vses. Khim. Obshch. im D I Mendeleeva*, 1983, **28** 716
52. I V Martynov, O A Raevskii *Vestn. Akad. Nauk SSSR*, 1983, 93
53. A M Sapegin, O A Raevskii, V V Chistyakov, I V Martynov *Khim.-farm. Zh.*, 1987, **21** 1098
54. P Carsky, M Urban *Ab Initio Calculation Methods. Applications in Chemistry*, Springer Verlag, Berlin, 1980, p. 247
55. O A Raevskii, V Yu Grigor'ev, V P Solov'ev, I V Martynov *Dokl. Akad. Nauk SSSR*, 1988, **299** 1433
56. O A Raevskii, V Yu Grigor'ev, V P Solov'ev *Khim.-farm. Zh.*, 1988, **28** 1216
57. G Duga, C Penny *Bioorganic Chemistry* (Translated into Russian), Izd. Mir, Moscow, 1983
58. P Murray-Rust, J P Glusker *J. Am. Chem. Soc.*, 1984, **106** 1018
59. R E W Hopman *Ber Bunsenges. Phys. Chem.*, 1979, **83** 702
60. P Carmona, J V Garcia-Ramos *Spectrochim. Acta, Part A*, 1984, **40** 155
61. K Inuzuka, A Fujimoto *Spectrochim. Acta, Part A*, 1984, **40** 623
62. M T Cung, M Marraud, J Neel *H. Chim. Phys. et Phys.-chim. Biol.*, 1976, **73** 213
63. G G Hammes, A C Park *J. Am. Chem. Soc.*, 1969, **91** 956
64. A Fujimoto, K Inuzuka, R Shiba *Bull. Chem. Soc. Jpn.*, 1981, **54** 2802
65. D Gerritzen, H H Limbach *J. Phys. Chem.*, 1980, **84** 799
66. S Ram, K Ram, J S Yaday *J. Chim. Phys. et Phys.-chim. Biol.*, 1984, **81** 577
67. L L Kishtis *Molekulyarnaya Spektroskopiya. Sbornik Statei* (Collected Articles on Molecular Spectroscopy) (Edited by S F Bureiko, et al.), Izd. LGU, Leningrad, 1983, No. 6, p. 216
68. L L Kishtis, V I Shabshinskas, V I Balavichyus *Zh. Fiz. Khim.*, 1986, **60**(2) 478 [English translation in *Russ. J. Phys. Chem.*, 1986(2)]
69. J J Rebek, B Askew, N Islam, et al. *J. Am. Chem. Soc.*, 1985, **107** 6736
70. J J Rebek, D Nemeth *J. Am. Chem. Soc.*, 1986, **108**, 5637
71. J Kyogoku, R C Lord, A Rich *Biochim. Biophys. Acta* 1969, **179** 10
72. I K Yanson, A B Teplitzky, L F Sukhodub *Biopolymers*, 1978, **18** 1149
73. D M Blow *Acc. Chem. Res.*, 1976, **9** 145
74. E W Knapp, K Schulten *Z. Chem. Phys.*, 1980, **46** 215
75. H Merz, G Zundel *Biochem. Biophys. Res. Commun.*, 1981,**101** 540
76. J M Leclercq, P Dupuis, C Sandorfy *Croat. Chem. Acta*, 1982, **55** 105
77. H Berndt, J S Khitkowski *Theoretical Chemistry of Biological Systems* (Edited by G Naray-Szabo), Elsevier, Amsterdam, 1989, p. 349
78. G Naray-Szabo *J. Mol. Struct.*, 1986, **138** 197
79. S Fraga *J. Mol. Struct.*, 1983, **94** 251
80. S Scheiner *Biopolymers*, 1983, **22** 731
81. D L Beveridge, M Mezei, F T Mehrotra, et al. *Ann. N. Y. Acad. Sci.*, 1981, **367** 108
82. E Clementi, G Corongiu *J. Chem. Phys.*, 1980, **72** 3979
83. G Corongiu, E Clementi *Biopolymers*, 1981, **20** 551
84. D C Rapaport *Mol. Phys.*, 1983, **50** 1151
85. C J Pedersen *J. Am. Chem. Soc.*, 1967, **89** 2495
86. M Hiraoka *Crown Compounds, Properties and Use* (Translated into Russian), Izd. Mir, Moscow, 1986, p. 363
87. E Weber, F Vögtle *Host-Guest Complex Chemistry. Vol. 1. Topics in Current Chemistry*, Interscience, New York—Berlin, 1981, Vol. 98, p. 3
88. G Weber *Z. Naturforsch., B*, 1982, **36** 896
89. G Weber *Z. Naturforsch., B*, 1982, **37** 1250
90. S Harkema, G J Hummel, K Daasvatn, D N Reinhoudt *J. Chem. Soc., Chem. Commun.*, 1981, 368
91. G Weber *J. Incl. Phenom.*, 1983/4, **1** 339
92. G Weber *Acta Crystallogr., Sect. B*, 1982, **38** 629
93. R Hilgenfeld, W Saenger *Z. Naturforsch. B*, 1981, **36** 242
94. R D Rogers, L M Green *J. Incl. Phenom.*, 1986, **4** 77
95. W P McKenna, E M Eyring *Appl. Spectrosc.*, 1986, **40** 16
96. I Goldberg *Acta Crystallogr., Sect. B*, 1975, **31** 754
97. J A Bandy, M R Truter, F Vögtle *Acta Crystallogr., Sect. B*, 1981, **37** 1568
98. G Weber *J. Mol. Struct.*, 1983, **98** 333
99. F Vögtle, W H Müller, E Weber *Chem. Ber.*, 1980, **113** 1130
100. F Vögtle, H Sieger, W Müller *Host-Guest Complex Chemistry. Vol. 1. Topics in Current Chemistry*, Interscience, New York—Berlin, 1981, Vol. 98, p. 109
101. J M Lehn *Angew. Chem. Int. Ed. Engl.*, 1988, **27** 90
102. J H Timko, S S Moore, R C Newcomb, et al. *J. Am. Chem. Soc.*, 1977, **99** 4207
103. E Graf, J M Lehn, J Moigne *J. Am. Chem. Soc.*, 1982, **104** 1672

104. H Metz, J M Rozalsky, R Weiss *J. Chem. Soc., Chem. Commun.*, 1976, 533
105. R M Izatt, N E Izatt, B F Rossiter, et al. *Science*, 1978, **199** 994
106. M Newcomb, J M Timko, D M Walba, D J Cram *J. Am. Chem. Soc.*, 1977, **99** 6392
107. D J Cram, J M Cram *Science*, 1974, **183** 803
108. L R Sousa, G D Sogah, D H Hoffman, D J Cram *J. Am. Chem. Soc.*, 1978, **100** 4569
109. J M Lehn, C Sirlin *J. Chem. Soc., Chem. Commun.*, 1978, 949
110. J P Behn, J M Lehn *J. Chem. Soc., Chem. Commun.*, 1978, 143
111. D J Cram *Chemical Future. Proceedings of the 29th IUPAC Conference at Cologne*, Pergamon Press, Oxford, 1984, p. 3
112. N S Poonia, A V Bajaj *Chem. Rev.*, 1979, **79** 389
113. N S Poonia *Progress in Macrocyclic Chemistry* (Edited by R M Izatt, J J Christensen), Wiley Interscience Publishers, New York, 1979, Vol. 1, p. 115
114. D J Cram, K N Truebold *Host-Guest Complex Chemistry. Vol. 1. Topics in Current Chemistry*, Interscience, New York–Berlin, 1981, Vol. 98
115. D Parker *Adv. Inorg. Chem. Radiochem.*, 1983, **27** 1
116. J D Dunitz, P Seiler *Acta Crystallogr., Sect. B*, 1974, **30** 2750
117. V V Tkachev, L O Atovmyan, V E Zubareva, O A Raevskii *Koord. Khim.*, 1987, **13** 264
118. O A Raevskii, S V Trepalin, V E Zubareva, D G Batyr *Koord. Khim.*, 1986, **12** 723
119. O A Raevskii, V V Tkachev, L O Atovmyan, V E Zubareva *Teor. Eksp. Khim.*, 1988, **24** 242
120. O A Raevskii, V V Tkachev, L O Atovmyan, et al. *Koord. Khim.*, 1988, **14** 803
121. V V Tkachev, O A Raevskii, L O Atovmyan, et al. *Izv. Akad. Nauk SSSR, Ser. Khim.*, 1987, 2225
122. D G Gorenstein *Chem. Rev.*, 1987, **87** 1047
123. G Shoham, W N Lipscomb, V Olsher *J. Am. Chem. Soc.*, 1983, **105** 1247
124. G Shoham, W N Lipscomb, V Olsher *J. Chem. Soc., Chem. Commun.*, 1983, 208
125. J D Owen *Acta Crystallogr., Sect. C*, 1983, **39** 579
126. N S Poonia *Curr. Sci. (India)*, 1981, **50** 881
127. R Gandour, F Fronczek, V J Gatto, et al. *J. Am. Chem. Soc.*, 1986, **108** 4078
128. C Homany, K Clinger, M L Hackort, N S Poonia *J. Incl. Phenom.*, 1986, **4** 61
129. F R Fronczek, V J Gatto, R A Schultz, et al. *J. Am. Chem. Soc.*, 1983, **105** 6717
130. D Owen *J. Chem. Soc., Dalton Trans.*, 1978, 1418
131. F P Remmortere, F P Boer *Inorg. Chem.*, 1974, **13** 2071
132. S Doughty, J F Stoddart, H M Colquhoun, et al. *Polyhedron*, 1985, **4** 567
133. S Bugen, J Dale, P Groth, J Krane *J. Chem. Soc., Chem. Commun.*, 1982, 1172
134. J D Owen *J. Chem. Soc., Perkin Trans. 2*, 1983, 407
135. J J Christensen, D J Eatough, R M Izatt *Chem. Rev.*, 1974, **74** 351
136. J D Lamb, R M Izatt, J J Christensen, D L Eatough *Coordination Chemistry of Macrocyclic Compounds* (Edited by C A Melson), Plenum Press, New York, 1979, p. 245
137. F Jong, D N Reinhoudt *Adv. Phys. Org. Chem.*, 1980, **17** 279
138. R M Izatt, J S Bradshaw, A Nielsen, et al. *Chem. Rev.*, 1985, **85** 271
139. J Inoue, T Hakushi *J. Chem. Soc., Perkin Trans. 2*, 1985, 935
140. V P Solov'ev, N N Strakhova, O A Raevskii *Izv. Akad. Nauk SSSR, Ser. Khim.*, 1988, 1112
141. N K Frensdorff *J. Am. Chem. Soc.*, 1971, **93** 600
142. J M Lehn *Structure and Bonding*, Springer Verlag, Berlin, 1973, Vol. 16, p. 1
143. D H Haynes, J Duncan, T Wiens, B C Pressman *J. Membr. Biol.*, 1974, **18** 28
144. H Kirch, J M Lehn *Angew. Chem. Int. Ed. Engl.*, 1975, **14** 555
145. B A Tashmukhamedov, A I Gagel'gans, A K Tashmukhamedova, et al., *Sintez, Membanoaktivnye Svoistva i Biologicheskie Effekty Kraun-efirov* (The Synthesis, Membrane-active Properties, and Biological Effects of Crown Ethers), Izd. Fan, Tashkent, 1987, p. 261
146. W E Morf, D Ammann, R Bissig, et al. *Progress in Macrocyclic Chemistry* (Edited by R M Izatt, J J Christensen), Wiley Interscience Publishers, New York, 1979, Vol. 1, p. 1
147. T M Fyles, A Malik-Diemer, D M Whitfield *Can. J. Chem.*, 1981, **59** 1734
148. A V Bogatskii *Bioorg. Khim.*, 1983, **9** 1445
149. N V Lukoyanov, O A Raevskii, A S Shtepanek, T N Kudrya *Biol. Membrany*, 1984, **1** 379
150. N V Lukoyanov, G I Van'kin, V A Panarin, O A Raevskii *Biol. Membrany*, 1985, **2** 71
151. J M Lehn *Science*, 1985, **227** 849
152. E Kumura *Topics in Current Chemistry*, Interscience, New York–Berlin, 1985, Vol. 128, p. 113
153. B Dietrich, J Giulchem, J M Lehn, et al. *Helv. Chim. Acta*, 1984, **67** 91
154. M W Hosseini, J M Lehn, M P Mertes *Helv. Chim. Acta*, 1983, **66** 2454
155. J M Lehn *Biomimetic Chemistry* (Edited by N Ise, et al.), Elsevier, Amsterdam, 1983, p. 163
156. M W Hosseini, J M Lehn *Helv. Chim. Acta*, 1986, **69** 587
157. V L Goedkin *Coordination Chemistry of Macrocyclic Compounds* (Edited by C A Melson), Plenum Press, New York, 1979, p. 603
158. L J Boucker *Coordination Chemistry of Macrocyclic Compounds* (Edited by C A Melson), Plenum Press, New York, 1979, p. 641
159. R L Kellog *Recherches*, 1984, **15** 819
160. D D Perrin, R P Agarwal *Metal Ions in Biological Systems* (Edited by H Sigel), Marcel Dekker, New York, 1973, Vol. 2, p. 168
161. A E Martell *Metal Ions in Biological Systems* (Edited by H Sigel), Marcel Dekker, New York, 1973, Vol 2, p. 208
162. H Sigel *Angew. Chem. Int. Ed. Engl.*, 1975, **14** 394
163. E P Nabirkina, T P Ignat'eva, O A Raevskii, Yu P Belov *Izv. Akad. Nauk SSSR, Ser. Khim.*, 1989, 2482
164. C A McAuliffe *Inorg. Biochem.*, 1982, **3** 1
165. R Sahai, S S Kushiwaha *J. Indian Chem. Soc.*, 1984, **61** 205
166. J R Sorensen *Metal Ions in Biological Systems* (Edited by H Sigel), Marcel Dekker, New York, 1982, Vol. 7, p. 77
167. P R Reddy, V B Rao *Polyhedron*, 1985, **4** 1603
168. Ya D Fridman, M S Dzhusueva, N V Dolgashova *Zh. Neorg. Khim.*, 1985, **30**(9) 2286 [English translation in *Russ. J. Inorg. Chem.*, 1985(9)]
169. Ya D Fridman, S V Alikeeva, N V Dolgashova, T T Nemal'tseva *Zh. Neorg. Khim.*, 1986, **31**(5) 1232 [English translation in *Russ. J. Inorg. Chem.*, 1986(5)]
170. O Yamauchi *Inorg. Chim. Acta*, 1985, **100** 165

171. P G Daniele, P Amico, O Zerbinati, G Ostacoli *Ann. Chim. (Rome)*, 1986, **76** 393
172. R R Amirov, Z A Saprykova *Zh. Obshch. Khim.*, 1987, **57** 1526
173. *Matematika v Khimicheskoi Termodinamike* (Mathematics in Chemical Thermodynamics), Izd. Nauka, Novosibirsk, 1980, p. 192
174. V P Novikov, T I Ignat'eva, O A Raevskii *Zh. Neorg. Khim.*, 1986, **31**(6) 1474 [English translation in *Russ. J. Inorg. Chem.*, 1986(6)]
175. D M Himmelblau *Applied Nonlinear Programming* (Translated into Russian), Izd. Mir, Moscow, 1975, p. 448
176. W Conley *Int. J. Math. Educ. Sci. Technol.*, 1981, **12** 609
177. A J Morffew *J. Mol. Graph.*, 1983, **1** 83
178. W G Richards, L Mangold *Endeavour. New Ser.*, 1983, **7** 2
179. I G Vinter *Chem. Br.*, 1985, **21** 32
180. D N J White, J K Tyler, M R Lindley *Comput. Chem.*, 1986, **10** 193
181. M J C Crabbe *Biochem. Soc. Trans.*, 1984, **12** 1095
182. N Pattabiraman, M Levitt, T E Ferrin, R Langridge *J. Comput. Chem.*, 1985, **6** 432
183. K Muller *Computer Aided Molecular Design*, Elsevier, Basel, 1985, p. 45
184. H Fruhbeis, R Klein, H Wallmeier *Angew. Chem. Int. Ed. Engl.*, 1987, **26** 403
185. V B Luzhkov, G N Bogdanov *Usp. Khim.*, 1986, **55**(1) 3 [English translation in *Russ. Chem. Rev.*, 1986(1)]
186. K Binder (Editor) *Monte-Carlo Methods in Statistical Physics*, Springer Verlag, Berlin, 1979
187. G Corangiu, E Clementi *Biopolymers*, 1982, **21** 763
188. W M Evans, G J Evans *Adv. Chem. Phys.*, 1985, **43** 377
189. M Levitt *J. Mol. Biol.*, 1983, **168** 595
190. S Harvey, M Prabhakaran, B Mao, J A McComman *Science*, 1984, **223** 1189
191. A T Hagler, F Dauber, D J Osguthorpe, J Hempel *Science*, 1985, **227** 1309
192. M Mezei, P K Mehrotra, D L Beveridge *J. Am. Chem. Soc.*, 1985, **107** 2239
193. R M Levy, M Karplus, J Kuschik, D Perahia *Macromolecules*, 1984, **17** 1370
194. R L Humphries *J. Theor. Biol.*, 1981, **91** 477
195. C H Hassall *Chem. Br.*, 1985, **21** 39
196. R L des Jarlais, R P Sheridan, J C Dixon, et al. *J. Med. Chem.*, 1986, **29** 2149
197. W G Richards *J. Mol. Graph.*, 1984, **2** 21
198. D P Hiujsman, A Van Delft, C A C Kuip *Comput. Graph.*, 1987, **11** 326
199. E F Meyer *Medical Chemistry* (Edited by E J Ariens), Academic Press, New York, 1980, Vol. 11, p. 267
200. C Humblet, G R Marshall *Drug. Dev. Res.*, 1981, **1** 409
201. N C Cohen *Advances in Drug Research* (Edited by B Testa), Academic Press, London, 1985, Vol. 14, p. 41
202. D E Walters, R A Pearlstein, C P Krimmel *J. Chem. Educ.*, 1986, **63** 869
203. R L des Jarlais, R P Sheridan, G L Seibel, et al. *J. Med. Chem.*, 1988, **31** 722
204. F C Bernstein, T F Koetzle, G J Williams, et al. *J. Mol. Biol.*, 1977, **112** 535
205. I D Kuntz, J M Blaney, S J Oatley, et al. *J. Mol. Biol.*, 1982, **161** 269
206. J Labanowski, I Motoc, C B Naylor, et al. *Quant. Struct. Activ. Relat.*, 1986, **5** 138
207. C A Reynolds, W G Richards, P J Goodford *Anti-cancer Drug Design*, 1987, **1** 281
208. T Fujita *Quant. Struct. Activ. Relat.*, 1987, **6** 54
209. D A Collier, S Neidle *J. Med. Chem.*, 1988, **31** 847
210. G M D Kelder, T Bultsma, H Timmerman, B Rademaker *J. Med. Chem.*, 1988, **31** 1069
211. F I Carroll, P Abraham, K Parham, et al. *J. Med. Chem.*, 1987, **30** 805
212. M F Hibert, H W Gittos, D N Middlemiss, et al. *J. Med. Chem.*, 1988, **31** 1087
213. G H Loew, J Collins, B Luke, et al. *Enzyme*, 1986, **36** 54
214. C A Venanzi, J D Bunce *Enzyme*, 1986, **36** 79
215. W Draber *Z. Naturforsch., C*, 1987, **42** 713
216. S Brunie, P Mellot, C Zewler, et al. *J. Graphics*, 1987, **4** 1
217. N L Liebman *Ann. N. Y. Acad. Sci.*, 1986, **466** 788
218. V E Khutorskii *Abstract of Doctorate Thesis in Chemical Sciences*, Institute of Organic Chemistry, Academy of Sciences of the Ukrainian SSR, Kiev, 1988, p. 32
219. V E Khutorskii, N N Makovskii *Bioorg. Khim.*, 1981, **7** 1465, 1982
220. V E Khutorskii, A V Krupko, V V Pen'kovskii *Biol. Membrany*, 1985, **2** 1217
221. V E Khutorskii, A V Krupko *Biofizika*, 1987, **32** 482
222. V E Khutorskii, A A Kamenchuk, I N Alieva *Biol. Membrany*, 1987, **4** 756
223. V E Khutorskii *Biol. Membrany*, 1984, **1** 244
224. V E Khutorskii, A A Kamenchuk *Biol. Membrany*, 1988, **5** 173