The Use of Molecular Descriptors in the Design of Gadolinium (III) Chelates as MRI Contrast Agents

Alessandro Maiocchi*

Milano Research Centre, Bracco Imaging spa, Via E. Folli 50, 20134 Milano, Italy

Abstract: Nuclear Magnetic Resonance Imaging (MRI) is a very useful tool in modern medical diagnostics, especially when gadolinium(III)-based contrast agents are administered to the patient with the aim of increasing the image contrast between normal and diseased tissues. The main purpose of this review is to show that a new generation of these contrast agents could be developed by making greater use of soft modelling techniques such as QSAR/QSPR after a suitable description of their molecular structure.

Keywords: Magnetic Resonance Imaging, MRI, Gadolinium, QSAR, QSPR, Molecular Descriptors.

INTRODUCTION

Nuclear Magnetic Resonance Imaging (MRI) [1,2] has become a powerful tool in modern medical diagnostics due to continuing improvements both in the relevant technology and in the development of a new class of pharmaceuticals able to enhance, after administration, the image contrast between normal and diseased tissues. These pharmaceuticals, commonly referred to as contrast agents, are based on metal complexes of paramagnetic ions, which enhance the nuclear magnetic relaxation rates of the bulk water protons in the tissues where they are distributed. Due to the high magnetic moment (seven unpaired electrons, see below for details) and the relatively long electronic relaxation time of the metal ion, Gd(III) ion complexes are currently the most used contrast agents in clinical MRI diagnostics. Nowadays, several Gd(III) complexes are available in the market and many others are undergoing clinical trials (Table 1).

It has been estimated that today at least 30% [3] of clinical MRI procedures make use of contrast agents, and a further increase in their use is expected, justifying greater interest on the part of several pharmaceutical companies in the research and development of Gd(III)-based contrast agents.

The design of new Gd(III) chelates as MRI contrast agents requires the optimization of several properties: (1) suitable paramagnetic properties; (2) high thermodynamic stability and/or kinetic inertness of the complex in vivo; (3) low toxicity; (4) specific bio-distribution in vivo; (5) excretability; (6) relevant water solubility.

Table 1. Clinically Relevant Gd(III) Complexes[a]

name	brand name	company
$[Gd(DTPA)(H_2O)]^{2-}$	Magnevist[b]	Schering (Germany)
$[Gd(DOTA)(H_2O)]^{-}$	Dotarem[b]	Guerbet (France)
$[Gd(DTPA\text{-}BMA)(H_2O)]$	Omniscan[b]	Nycomed-Amersham (U.K.)
$[Gd(HP\text{-}DO3A)(H_2O)]$	Prohance[b]	Bracco (Italy)
$[Gd(DO3A\text{-}butrol)(H_2O)]$	Gadovist[b]	Schering(Germany)
$[Gd(DTPA\text{-}BMEA)(H_2O)]$	OptiMark[c]	Mallinckrodt (U.S.)
$[Gd(BOPTA)(H_2O)]^{2-}$	MultiHance[b]	Bracco (Italy)
$[Gd(EOB\text{-}DTPA)(H_2O)]^{2-}$	Eovist[b]	Schering(Germany)
MS-325	AngioMARK[c]	Epix/Mallinckrodt (U.S.)

[a]Molecular structures are shown in Fig. (**1**). [b]Approved. [c]In clinical trials

There is a general consensus about the particular relevance of all these properties, and many academic and industrial researchers have put great effort into the search for their optimization, as is clearly demonstrated by the large amount of data and structures already reviewed [3-5].

However, despite the large amount of information available and the multi-parametric character of the properties, which have to be tuned in the design of Gd(III) complexes, only a few works contain a true multivariate approach aimed at correlating the molecular structures with some relevant

*Address correspondence to this author at the Milano Research Centre, Bracco Imaging spa, Via E. Folli 50, 20134 Milano, Italy; E-mail: Alessandro.Maiocchi@Bracco.com

1389-5575/03 $41.00+.00

properties. On the contrary, multivariate techniques (see article by Migliavacca in this issue), such as data reduction methods (e.g., PCA, Cluster Analysis) [6-9] and linear or non-linear regression methods (e.g. MLR, PLS, ANNs) [10-12], in conjunction with the use of a suitable numerical coding of the molecular structures, commonly referred to as molecular descriptors [13], are a well-established practice in the medicinal chemistry framework.

In the present review, we have tried to discuss only those contributions belonging to MRI literature which deal with the evaluation of those parameters or molecular features, which can affect a certain property in a series of Gd(III) complexes, thus giving some insights into the best molecular descriptors to be used in the search for a suitable multivariate structure-property relationship.

As we believe that in order to generate a suitable structure-property relationship, it is essential to have some fundamental knowledge of the phenomenon being studied, a brief introduction about the already recognized rules or parameters affecting the property values is given at the beginning of each property-related section of the manuscript.

THE STRUCTURE OF Gd(III) COMPLEXES AND THEIR MOLECULAR DESCRIPTORS

The first step in any attempt to either analyze the similarities among a series of compounds or search for some suitable QSAR/QSPR models is the calculation of a set of molecular descriptors.

However, the calculation of any n-dimensional molecular descriptor requires a clear representation of the molecular structure at the corresponding n-dimensional level (see article by Gasteiger in this issue). While this prerequisite may be quite obvious when working with organic compounds, the same cannot be said for metallo-organic compounds. Indeed, having a metal-ion and a polydentate ligand to sketch the 2D molecular structure of the resultant complex, it is necessary to know both the number of coordination sites available on the metal-ion and which donor atoms are in the ligand. Moreover, the 3D description of a metal-ligand complex requires some additional information: the geometry of the coordination cage of the metal ion and how the donor sites of the ligand occupy the coordination sites around the metal ion. Thus, before starting with the calculation of some

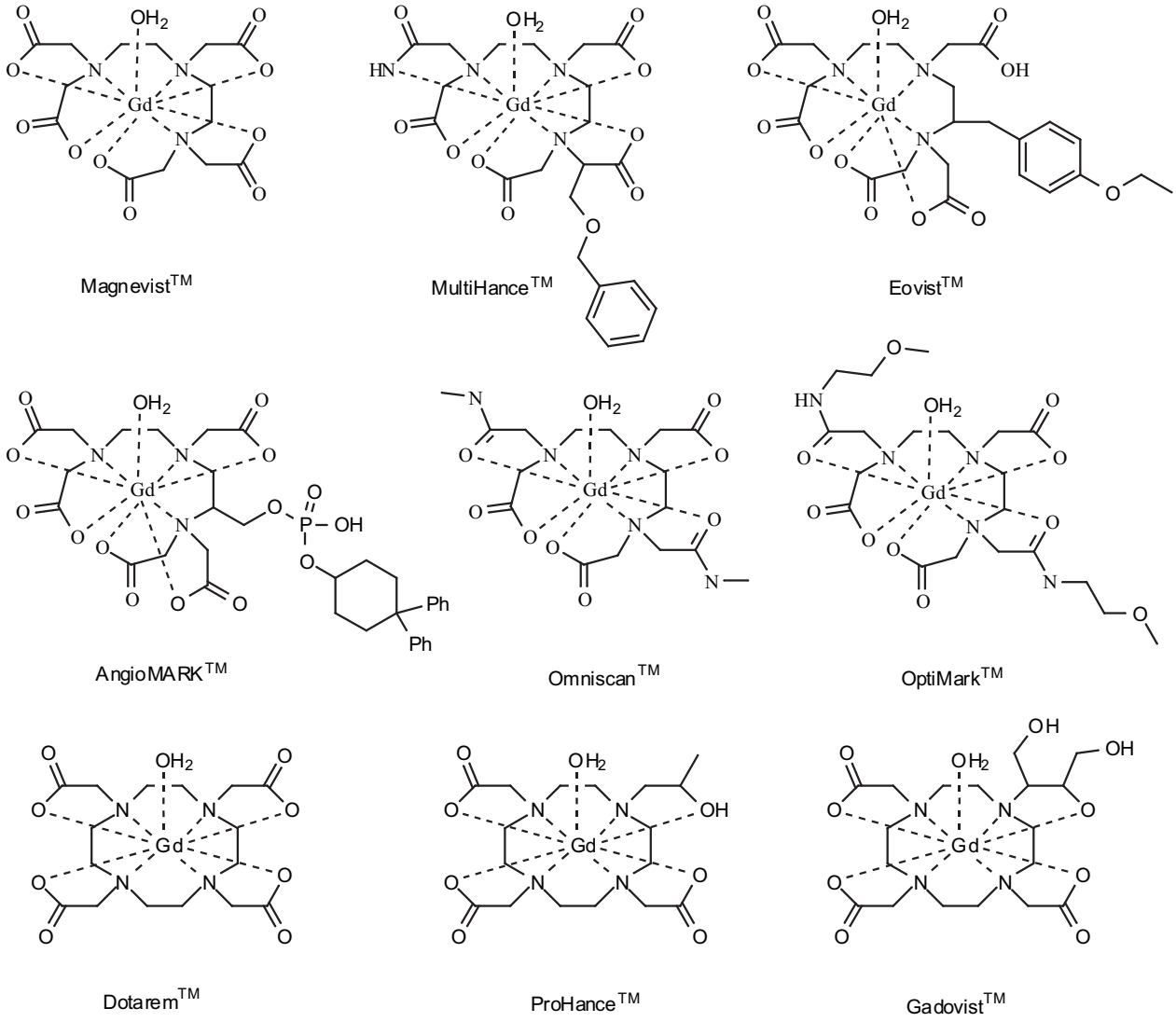


Fig. (1). Molecular structures of Gd(III) complexes commercially available or under clinical trial.

molecular descriptors, especially in the case of Gd(III) complexes, a clear understanding of their structural properties is strongly required.

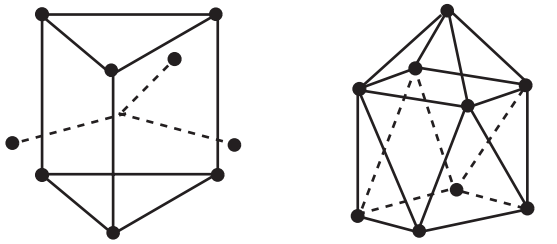


Fig. (2). The idealized geometries of enneacoordinated complex: Tricapped Trigonal Prism and Capped Square Antiprism.

The coordination cage of lanthanide ions is characterized by high coordination numbers, varying from nine for lighter lanthanides to eight for the heavier ones. This behaviour has been ascribed to the occurrence of so-called lanthanide contraction, a steady reduction of the ionic radii with increasing of the atomic number. The principal cause of this contraction is the electronic effect due to the low shielding ability of the 4f electrons with respect to an increased nuclear charge. The most common coordination number (CN) for Gd(III) ion is nine, even though a coordination of eight has been occasionally observed [14,15]. For the enneacoordination, (CN=9) there are two possible idealized coordination geometries: the tricapped trigonal prism (TTP) and the capped square antiprism (CSAP) (Fig. (**2**)). Based on a "points-on-a-sphere" repulsive model [16], the TTP geometry generates the most stable polytopal form (a spatial arrangement of a ligand about a central atom, where the ligand defines the vertices of a polyhedron) while the CSAP geometry is slightly less stable. Both coordination geometries have been recognized in the crystal structures determined for Gd(III) complexes with polydentate chelates, although distorted polyhedra were frequently observed especially when chelates with some steric constraints were used. In some cases, unequivocal assignment of the coordination cage is not possible and, because of the close relationship between the two coordination geometries, a particular enneacordinated structure may be described equally well by both TTP and CSAP geometries.

In Fig.(**3**) are shown the crystallographic structures of $[Gd(DTPA)(H_2O)]^{2-}$ [17] and $[Gd(DOTA)(H_2O)]^{-}$ [18], which adopt, in an enneacoordinated state, the TTP and the CSAP geometries respectively. In the case of $[Gd(DTPA)(H_2O)]^{2-}$, the coordination sites are occupied by three amine nitrogen atoms and five monodentate carboxylic oxygen atoms, while for $[Gd(DOTA)(H_2O)]^{-}$, four amine nitrogen atoms and four monodentate carboxylic oxygen atoms are coordinated to the metal ion. Moreover, the two structures have another coordination site occupied by a water molecule, which is a fundamental structural feature for the effectiveness of all Gd(III)-based MRI contrast agents (more details will be given in the next section).

From the analysis of crystallographic data available [3], it is possible to conclude that the above coordination cages are only slightly modified by bulky substituents regardless of their position, both on the carboxylic arms and on the ethylendiamine moieties. In addition, there is much evidence that, in the solid state and in aqueous solution, the oxygen atoms in amide [19-21] and hydroxyalkyl [22] groups can bind the Gd(III) ion without inducing severe distortions in the geometries of its coordination cage.

Thus, using the structural features of $[Gd(DTPA)(H_2O)]^{2-}$ and $[Gd(DOTA)(H_2O)]^{-}$ as templates, it becomes possible to build Gd(III) complexes having quite different coordination cages derived from the substitution of the carboxylic pendant arms with several other donor groups in any possible combination. So far, having the essential topological information on the structure of the resultant Gd(III) complexes makes it quite straightforward to calculate several 2D molecular descriptors.

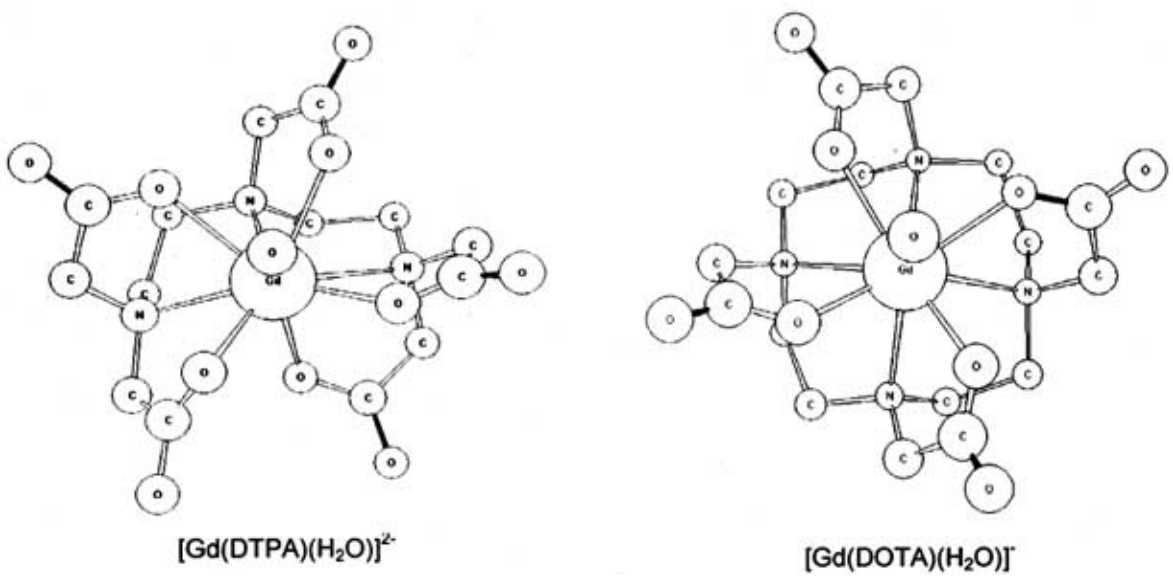


Fig. (3). Crystallographic structures of $[Gd(DTPA)(H_2O)]^{2-}$ and $[Gd(DTPA)(H_2O)]^{-}$ complexes.

Recently, Todeschini and co-workers have distributed a Web version of their *Dragon* software [23] for the calculation of almost 1500 molecular descriptors (see article by Tetko in this issue). The complete set of the available molecular descriptors is partitioned in 18 logical blocks that can be generated on the basis of a three-dimensional molecular structure. One specific feature of *Dragon* is the internal parameterization of the atom of gadolinium that enables the calculation of five blocks of 2D molecular descriptors as follows: (1) topological descriptors [24-26], (2) molecular walk counts [27,28], (3) BCUT descriptors [29,30], (4) Galvez topological charge indexes [31], (5) 2D autocorrelation functions [32-34]. To roughly evaluate the information content of all these 2D molecular descriptors, a database of 111 Gd(III) complexes containing linear and macrocyclic ligand motifs with several donor groups and substituents was analyzed[1]. After the calculation of all the available 2D descriptors, three filters were applied to remove constant variables, nearly constant variables and variables highly correlated ($r > 0.950$). The results obtained are summarized in Table 2.

Table 2. 2D Molecular Descriptors Calculated and Left Behind after Filtering

2D Molecular descriptors	Calculated	After filtering	Cumulated sum[a]
Topological descriptors	266	62	62
Molecular walk counts	21	2	64
BCUT descriptors	64	14	78
Galvez charge indices	21	14	92
2D Autocorrelation	96	61	153

[a]The cumulated sum represents the total number of survived 2D molecular descriptors after filtering.

Among the classes of 2D descriptors calculated, the molecular walk counts seem to be the most redundant, and therefore negligible, while the others, having a higher information content, could be used for further QSAR/QSPR studies.

The calculation of 3D descriptors requires a spatial molecular model of the complex (see article by Gasteiger in this issue). Nowadays, several computational techniques have been used with the aim of predicting the geometries of Gd(III) complexes, such as molecular mechanics, semi-empirical, density functional and *ab initio* methods [35-38]. All of these methods have proven to be able to reasonably predict the three-dimensional structures of either linear or macrocyclic Gd(III) complexes. However, if the goal of a certain study is to discriminate among the relative population of some conformational isomers, it has been proven that more computationally demanding methods are required: in the case of the $[Gd(DOTA)(H_2O)]^-$ complex, only the introduction of solvent effects into *ab initio*-type calculations, by using the polarizable continuum model [39], has enabled the reproduction of the conformational equilibrium between the two main isomers experimentally observed in aqueous solutions [40].

From a practical point of view, the calculation of 3D molecular descriptors of Gd(III) complexes can be done on a routinely basis, simply using the structure geometries derived from molecular mechanics calculations after a suitable force field parameterization of Gd(III) ion; if the 3D molecular descriptors used in a certain study require additional information about electronic distribution properties of the Gd(III) complex, quantum-mechanical methods should be used.

It is important to point out that the average dimension of Gd(III)-based contrast agents, especially those under current scrutiny, is growing and, consequently, quantum-mechanical methods could be very time-consuming even at the high computational performance levels of the hardware platforms available on the market. Moreover, if a QSAR/QSPR model is routinely applied to predict a certain property for a large number of Gd(III) complexes, the use of quantum-mechanical methods is really too expensive and the cost of this choice will be a very low effectiveness of the whole Gd(III) complex design procedure, in particular where discovery programs are based on short-time strategies.

MOLECULAR DESCRIPTORS AND THE RELAXIVITY OF Gd(III) COMPLEXES

The so-called *proton relaxivity* is a measure for the ability of a paramagnetic substance to accelerate the nuclear magnetic relaxation of water protons in the media where this paramagnetic substance has been dissolved. A Gd(III) complex can be considered a possible candidate as an MRI contrast agent only if it shows a high proton relaxivity at the magnetic field commonly used in clinical MRI (0.5-1.5 T).

A detailed description of the relaxation mechanism of water protons in the presence of Gd(III) complexes is beyond the scope of the present review: only a brief description will be given here to enable the reader to understand the main principles governing the nuclear magnetic relaxation phenomena. In this way, it will be possible to recognize why some molecular descriptors may be more suitable than others in the development of quantitative structure-property relationships also having a close connection with the basis of the relaxation theory. However, several excellent references covering more detailed aspects of the general relaxation theory are available in literature [41-43].

Basically, the proton relaxivity of a paramagnetic metal complex can be described as the result of the sum of two independent components: inner-sphere and outer-sphere relaxation.

The inner-sphere relaxation arises from the water molecules in the first coordination sphere. These water molecules are in contact with the metal ion in the complex and are in fast exchange with the bulk water molecules. The increase in the relaxation rates of bulk water molecules is dependent upon the concentration of the paramagnetic ion, the number of water molecules in the first coordination

[1] Maiocchi A. Unpublished results, 2002

sphere, the rate of water exchange and the rate of relaxation of the protons on the coordinated water molecule.

The outer-sphere relaxation is the contribution to the relaxation of water protons arising from the diffusion of the bulk water in the surroundings of the paramagnetic molecule and from those water molecules which occupy the nearest space to the paramagnetic molecule for a relatively long time, the so-called "second coordination sphere" (see Fig.(**4**)).

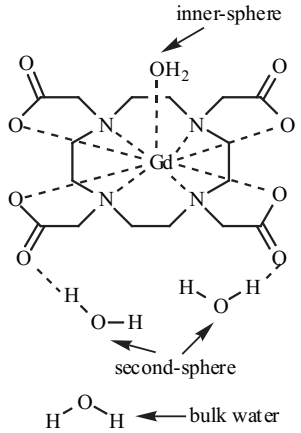


Fig. (4). The three types of water molecules in the relaxation theory: inner-sphere, second-sphere and bulk water.

For commercially available Gd(III)-based contrast agents, the inner and outer-sphere relaxation mechanisms contribute approximately to the same extent to the overall relaxation enhancement at the imaging fields.

The relaxation rate enhancement due to the paramagnetic substance is proportional to the concentration of the paramagnetic species, while the degree of relaxation enhancement provided by the paramagnetic ion is commonly referred to as relaxivity, r_i. The relaxation rates of water protons in the presence of a Gd(III) complex can be described by equation 1 where $1/T_{i,obs}$ (s^{-1}) are the observed relaxation rates of water protons, $1/T_{i,d}$ (s^{-1}) are the relaxation rates of water protons in the absence of any paramagnetic substance, and [Gd] is the concentration of the Gd(III) complex (mM).

$$\frac{1}{T_{i,obs}} = \frac{1}{T_{i,d}} + r_i\,[Gd] \qquad (1)$$

where i=1,2

The subscript i has been used in equation 1 to take into account that Gd(III) complex affects both the longitudinal and transverse relaxation rates, $1/T_1$ and $1/T_2$ respectively, of the water protons.

The relaxivity r_i are experimental quantities, which are characteristic of each Gd(III) complex and depend both on the applied magnetic field and the temperature. The most common experimental conditions used to evaluate the relaxivity of a new Gd(III)-based contrast agents are as follows:

T=39°C, B=0.47 T (20 MHz), and buffered water (pH= 7.4) or saline solution (NaCl) as solvent.

In the search for new Gd(III)-based contrast agents, which are more effective, or in other words with higher relaxivity, the inner-sphere contribution to the overall relaxivity can play an important role because while it can be considerably increased by inducing suitable structural modifications to the Gd(III) complexes, the outer-sphere contribution cannot be modified to the same extent.

The quantification of the inner-sphere component can be done by using the Solomon-Bloembergen model [44,45]. At a fixed magnetic field strength, it can be simplified as follows [46]:

$$r_1^{IS} \propto \frac{Cq\tau_c}{a^6} \qquad (2)$$

where,

$$\frac{1}{\tau_c} = \frac{1}{\tau_s} + \frac{1}{\tau_m} + \frac{1}{\tau_r} \qquad (3)$$

In the above equations, the IS superscript refers to inner-sphere contribution, C is a constant, q is the number of water molecules coordinated to the paramagnetic metal ion, a is the water proton–metal distance and τ_c is the overall correlation time, which is, in turn, a result of the sum of three contributions: the electronic relaxation time τ_s, the life time of the water molecule in the complex τ_m and the molecular rotational correlation time τ_r.

According to the brief and simplified description of the relaxation theory given above, the first molecular descriptor that must be considered in order to derive quantitative structure-relaxivity models is the number q of water molecules bound in the inner coordination sphere of the Gd(III) ion.

The most simple method to calculate the q value is based on a preliminary assumption about the nature of the coordination cage of the Gd(III) complex. As already discussed, the Gd(III) ion is generally nine-coordinated and the chelate occupies a fraction of the binding site at the metal centre depending upon the postulated number of donor atoms available on its structure. Under the above simple assumption, the q value can be calculated as follows:

$$q = 9\text{-}N_{DA} \qquad (4)$$

where N_{DA} is the total number of postulated donor atoms on the chelate.

The postulated donor atoms must be both electron-rich atoms and occupy a vertex in at least one five-membered ring containing the Gd(III) ion. In this way, the calculation of the q value is based on a topological perception of the Gd(III) complex and requires *a priori* hypothesis about the connectivity of the Gd(III) ion in the absence of water molecules. This method can be applied successfully for almost all the Gd(III) complexes derived from polyamino-polycarboxylic chelates also containing oxygen donor atoms belonging to amide, hydroxyalkyl, phosphate and phosphinate moieties.

A second method to derive the number of water molecules in the inner coordination sphere has been proposed under the hypothesis that the q value would be proportional to the solvent-accessible surface area (SASA) of the Gd(III) ion [47]. The authors derived a linear relationship between the Connolly surface area [48] and the number q of

water molecules coordinated to the Gd(III) ion, iteratively removing waters from the crystal structure of $Gd(H_2O)_9$ [49]. To validate the method, the linear relationship was used to determine the q values for the following Gd(III) complexes by which the crystal structures were available: $Gd(EDTA)(H_2O)_3$ [50], $Gd(DTPA)(H_2O)$ [51], $Gd(DTPA\text{-}BEA)(H_2O)$ [52], $Gd(DOTA)(H_2O)$ [53], Gd(TTHA) [54] . To demonstrate the general validity of the proposed method, the authors have applied their procedure using the three-dimensional structures of the above Gd(III) complexes obtained by structure minimization, using an extended version of the TAFF force field [55] where the non-bonding interaction parameters for the Gd(III) ion were added. The three-dimensional molecular models obtained after a genetic algorithm conformational search were in reasonable agreement with the crystal structures, and the predicted q values were in complete agreement with the experimentally determined values.

Quite recently, a third method for the evaluation of the q value has been proposed under the hypothesis that the right number of water molecules coordinated to the Gd(III) ion must minimize the strain energy of the resultant complex [56,57]. The proposed method is based on the so-called "coordination scan" technique, in which the strain energy of a Gd(III) complex is calculated after structure minimization using the TAFF force field, under a systematic variation of the Gd(III)-ligand bond lengths. This procedure is repeated with various numbers of water molecules coordinated to the metal ion to evaluate the steric strain induced on the complex by binding additional coordination sites onto the metal ion. Thus, a plot of the complex strain energy versus the metal ionic radius can be generated for each coordination number. The lowest strain energy value in each curve indicates the preferred ionic radius for the metal ion in the given coordination state. It is expected that, for Gd(III) ion, the radius for a six-coordinate environment is 0.938, for a seven-coordinate one is 1.00, for an eight-coordinate one is 1.053, and for a nine-coordinate one is 1.107Å [58]. The preferred coordination number, and thus the q value, is chosen according to the closeness of the preferred ionic radius of each curve to the expected one, for a given coordination state, providing that the strain energy at this ionic radius is the lowest among those calculated. The authors have claimed the successful prediction of q values for several Gd(III) complexes and details were given on the nature of the curves obtained with the "coordination scan" technique for GdDTPA with q=0 and q=1.

For Gd(III) complexes having low molecular weights, such as those available on the market today, the main parameter which determines the value of the overall correlation time of proton relaxation τ_c (eq.3), and thus the value of the proton relaxivity, is the rotational correlation time τ_r which commonly assumes values lower than 200 ps. Since fast rotation is a significant limiting factor in the efficiency of the Gd(III) complexes, several approaches have been followed to slow down the molecular tumbling by increasing the dimension of the designed complexes. The relationship between the rotational correlation time τ_r and the dimension of the Gd(III) complex can be derived from the Debye-Stokes equation in the case of a spherical molecule:

$$\tau_r = \frac{4 \cdot \pi \cdot \eta \cdot r^3_{eff}}{3 \cdot k \cdot T} \qquad (5)$$

where η is the microviscosity around the molecule, and r is the effective radius of the spherical molecule. Tweedle and co-workers [59,60] have shown that the relaxivity not only of monomeric but also multimeric Gd(III) complexes correlates quite well with their molecular weights. The use

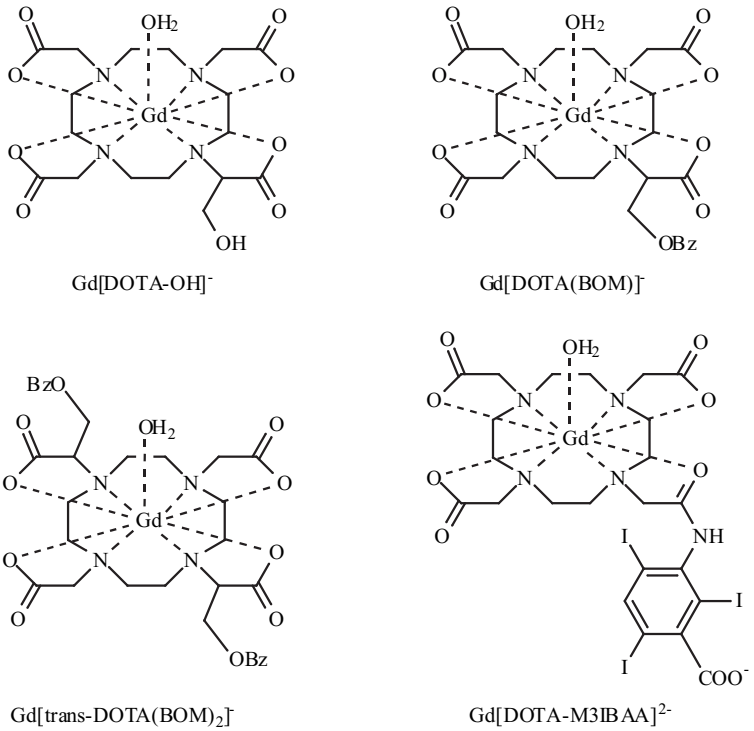


Fig. (5). Gd(III) complexes derived by ligands with increasing molecular weights [3].

of molecular weight as a size descriptor to account for the proton relaxivity dependency on the dimension of the Gd(III) complexes has become a common practice among researchers active in the field. However, it should be underlined that the relationship between the atomic radii and the atomic weights is not linear and, for this reason, the use of molecular weight can lead to considerable deviations from the expected relationship with the proton relaxivity, especially in the presence of heavier halogen atoms. An example of this occurrence is given for the Gd(III) complexes shown in Fig. (**5**): in this case, the relevant increases in molecular weight do not lead to a significant improvement in the relaxivity accordingly to the molecular Van der Waals volumes (V_{VDW}) calculated (Table 3).

Table 3 Experimental Relaxivities of Some Gd(III) Complexes with an Increasing Molecular Weight

Compound	MW (g mol^{-1})	VDW(Å)	r_1(mM^{-1}s^{-1})[a]
Gd[DOTA-OH]$^-$	605.7	488.9	3.72
Gd[DOTA(BOM)]$^-$	695.8	598.0	4.15
Gd[*trans*-DOTA(BOM)$_2$]$^-$	816.0	737.0	4.95
Gd[DOTA-M3IBAA]$^{2-}$	1072.5	678.0	5.01

[a] The relaxivities were measured at magnetic field of 0.5 T, T=39°C, pH=7.4, in saline solution (NaCl 0.15 M).

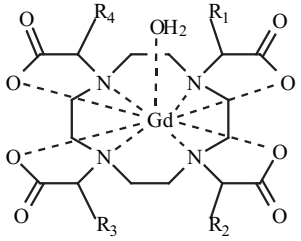
For the above reason, a more consistent approach with the Debye-Stokes theory should make use of molecular size descriptors, such as molecular volumes [61-64] and/or surfaces [48,65]. There are also several topological indexes which have been proven to encode explicitly or implicitly information about the molecular size [66,67] and, in turn, they may be used to develop more useful quantitative structure-relaxivity relationships.

Another important parameter, which affects the proton relaxivity of a Gd(III) complex, is the exchange rate of the water molecules in the inner coordination sphere of the Gd(III) ion. For enneacoordinated complexes, it has been demonstrated that the exchange process between a coordinated water molecule and the bulk waters follows a dissociative mechanism [68-71]. The activation energy of the dissociative mechanism is influenced by the stereoelectronic properties of the coordination cage. In particular, the presence of neutral donor atoms, such as amide or hydroxyl oxygens reduces the shielding effect over the positive charges of the Gd(III) ion, which increases its electrostatic interaction with the water molecule in the complex. Moreover, the water exchange process can be influenced by the steric crowding on the coordination cage as has been observed when amide moieties, which form shorter Gd-O bonds, participate in the complexation of the Gd(III) ion [72].

All those molecular descriptors that can encode both the variation of the charge distribution on the coordination sites of the Gd(III)-complexes and the steric crowding around the Gd(III) ion should be available among the descriptors used to search for a structure-relaxivity model. Depending upon the number of complexes in the data set and the computational tools available, we can use a rough estimate of charge distribution around the Gd(III) ion simply using the formal charge of the coordination cages (not the formal charges of the whole complexes). Otherwise, it is possible to use the sum of more accurate (at least in principle) atomic point charges derived from quanto-mechanical calculations. Similarly, the variation of the steric crowding in the coordination cage of Gd(III) ion could be roughly estimated by the count of amide moieties bound to the Gd(III) ion or, more accurately, from the sum of the bond lengths over all the donor atoms derived from calculated three-dimensional molecular models of the Gd(III)-complexes.

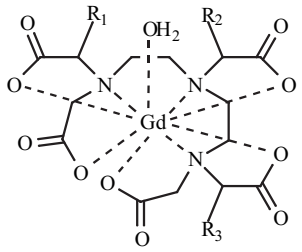
Nowadays, to the best of our knowledge, there is only one example of a multivariate quantitative structure-

Table 4 Molecular Structures and Experimental Proton Relaxivities Used to Search for Quantitative Structure-Relaxivity Relationships

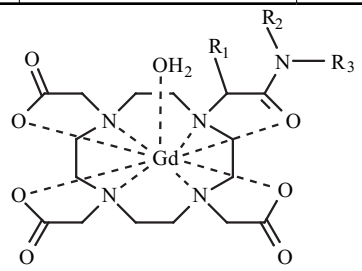


Compound	R_1	R_2	R_3	R_4	r_1(mM^{-1} s^{-1})
1	H	H	H	H	3.56
2	CH_2OH	H	H	H	3.72
3	CH_2OCH_2Ph	H	H	H	4.15
4	Me	Me	Me	Me	3.46
5	CH_2OH	CH_2OH	CH_2OH	H	3.91
6	CH_2OCH_2Ph	CH_2OCH_2Ph	CH_2OCH_2Ph	H	5.66

(Table 4). contd.....

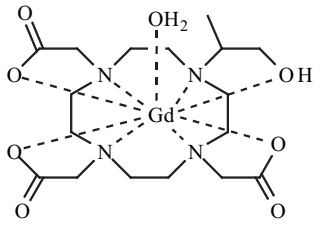
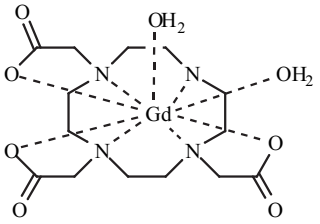
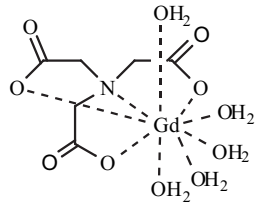
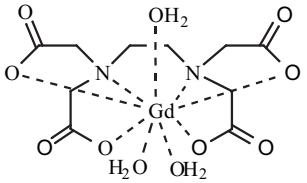
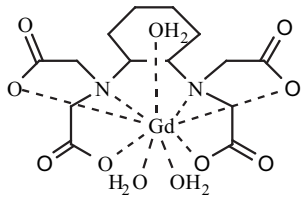


Compound	R_1	R_2	R_3	$r_1(mM^{-1} s^{-1})$
7	H	H	H	3.76
8	CH_2OH	H	H	3.79
9	CH_2OCH_2Ph	H	H	4.39
10	$CH_2OCH_2C_6H_{11}$	H	H	4.23
11	CH_2OCH_2(4-NH_2Ph)	H	H	4.60
12	CH_2OCH_2(4-OEtPh)	H	H	5.10
13	CH_2OH	H	CH_2OH	4.16
14	CH_2OCH_2Ph	H	CH_2OCH_2Ph	5.24
15	CH_2OCH_2Ph	CH_2OCH_2Ph	CH_2OCH_2Ph	6.78
16	CH_2OCH_2Ph	CH_2(4-OHPh)	CH_2OCH_2Ph	6.61



Compound	R_1	R_2	R_3	r_1 $(mM^{-1}s^{-1})$
17	CH_2OCH_2Ph	$CH(CH_2OH)_2$	H	4.49
18	CH_2OH	$CH(CH_2OH)_2$	H	4.03
19	CH_2OCH_2Ph	CH_2CH_2OH	H	4.33
20	CH_2OH	CH_2CH_2OH	H	3.84
21	CH_2OH	$CH_2CHOHCH_2OH$	H	3.95
22	CH_2OCH_2Ph	$CH_2CHOHCH_2OH$	H	4.54
23	CH_2OH	CH_2 $(CHOH)_4CH_2OH$	Me	4.53
24	CH_2OCH_2Ph	CH_2 $(CHOH)_4$ H_2OH	Me	5.19
25	Me	$CH_2CH_2OCH_2CH_2OH$	H	3.96
26	$=CH_2$	$CH_2CH_2OCH_2CH_2OH$	H	4.05
27	CH_2OH	$CH_2CH_2OCH_2CH_2OH$	H	4.23
28	CH_2CH_2OMe	$CH_2CH_2OCH_2CH_2OH$	H	4.34
29	CH_2OCH_2Ph	$CH_2CH_2OCH_2CH_2OH$	H	4.48
30	Me	$C(CH_2CH_2CH_2OH)_3$	H	4.68
31	$=CH_2$	$C(CH_2CH_2CH_2OH)_3$	H	4.78
32	CH_2OH	$C(CH_2CH_2CH_2OH)_3$	H	4.81
33	CH_2OCH_2Ph	$C(CH_2CH_2CH_2OH)_3$	H	5.36

(Table 4). contd.....

Compound	Structure	r_1 ($mM^{-1}s^{-1}$)
34		3.33
35		4.76
36		7.70
37		5.87
38		6.10

relaxivity relationship available[2]. In this study, a data set of 38 monomeric Gd(III) complexes, with their proton relaxivity measured at 0.5 T (Table 4), was analysed with the aim of developing a general model able to predict the proton relaxivity starting from the molecular structures.

Two separate chemical spaces were generated depending upon the level of complexity in the structure description being topological or topographical. All the molecular descriptors were calculated with a prototype of the *Dragon* software developed by Todeschini and co-workers [23]. The three-dimensional structure of the Gd(III) complexes was obtained by molecular mechanics simulation, using an extended version of the TAFF force field [73]. For the sake of simplicity, only the results obtained using the whole set of WHIM descriptors (Weighted Holistic Invariant Molecular descriptors)[74] will be summarized here, even though quantitative models with high prediction power were obtained using topological descriptors as well. The search for the best models was done using the OLS (Ordinary Least Square) algorithm, evaluating all possible models ranging from one to four independent variables (molecular descriptors). The best five models for each model size were selected according to the LOO (Leave-One-Out) cross-validation procedure and further validated under the LMO (Leave-More-Out) cross-validation procedure with the aim of analyzing the intrinsic stability of the best models when a higher level of perturbation, up to 50% of the data available, was applied to the original data set. All the retained best models for each model size, together with their most relevant statistical parameters and the selected molecular descriptors, are shown in Table 5.

[2] Calabi, L; Cosentino, U.; Maiocchi, A.; Marengo, E.; Todeschini, R.; Uggeri, F. *Book of Abstracts*; 11th European Symposium on QSAR: Computer-Assisted Lead Finding and Optimization, September 1-6, 1996, Lausanne, Switzerland, P-27.A

Table 5. Statistical Parameters for the Best Structure-Relaxivity Models[a]

Model size	Q^2_{LOO}	r^2	*s*	Model descriptors[b]
1	0.341	0.381	0.737	q
2	0.879	0.912	0.278	q, **A**-m
3	0.907	0.938	0.233	q, **A**-m, **V**-u
4	0.933	0.960	0.188	q, **A**-p, **V**-m, **V**-v

[a] Q^2_{LOO}: leave-one-out cross-validated explained variance; r^2 squared correlation coefficient; s: standard estimate of the error

[b] q: water molecules in the inner coordination sphere; $A=\lambda_1\lambda_2 + \lambda_1\lambda_3 + \lambda_2\lambda_3$; $V=T + A + \lambda_1\lambda_2\lambda_3$; $T=\lambda_1 + \lambda_2 + \lambda_3$; the lower-case letters define the weighting scheme used (u unweighted , m atomic masses, v van der Waals volumes, p atomic polarizabilities

It is interesting to note that the selected molecular descriptors belong to the category of the non-directional WHIM descriptors, also called "WHIM size". For this type of descriptor, any information related to the individual principal axis of the molecule is lost (this is not true for all the other defined WHIM descriptors), thus encoding only size-dependent information. Therefore, from the best models obtained, two main independent contributions to the proton relaxivity of the Gd(III) complexes can be recognized: the number of water molecules in the inner coordination sphere and the molecular size of the complexes. The agreement between the relaxation theory and the quantitative structure-relaxivity relationships obtained clearly demonstrates that it is possible to approach the prediction of proton relaxivity of the Gd(III)-based contrast agents via multivariate soft-modelling without any loss of relevant information. Nowadays, the main directions of research into new Gd(III)-based contrast agents with higher proton relaxivity are based on the development of more complicated molecular systems where the Gd(III) complexes are incorporated in macromolecular entities, such as linear polymers [75,76], dendrimers [77], micelles [78] or bio-molecules [79]. The increasing complexity of the molecular systems under current scrutiny is a challenging perspective where multivariate quantitative structure-relaxivity relationships could be a valuable tool in gaining new insights into the future of MRI research programs.

MOLECULAR DESCRIPTORS AND THERMODYNAMIC STABILITIES OF Gd(III) COMPLEXES

Common MRI diagnostic procedures using a metal-based contrast agent require an intravenous administration of solutions containing a relevant amount of contrast agent in order to induce a useful increase in the image contrast. A typical dose used for approved Gd(III) chelates is 0.1-0.3 mmol/kg total body weight. The free Gd(III) ion is too toxic (LD_{50}i.v. =0.1-0.4 mmol/kg in mouse) at the concentrations needed for MRI studies, and, for this reason, it must be administered in the form of stable complexes to avoid any release of the free ion before complete excretion. It is important to note that the dissociation of the complexes also generates unchelated ligands, which are generally more toxic than the complexes themselves. Indeed, it is compulsory for safe *in vivo* use to design Gd(III) complexes with high thermodynamic stability and kinetic inertness. Commonly, the thermodynamic stability of a metal-ligand complex is measured by the equilibrium constant K_{ML} of the complexation reaction as in Eqs. (6)-(8):

$$M + L \rightleftharpoons ML \quad (6)$$

$$K_{ML} = \frac{[ML]}{[M]\cdot[L]} = e^{-\Delta G_c^0/RT} \quad (7)$$

$$\Delta G_c^0 = \Delta H_c^0 - T\Delta S_c^0 \quad (8)$$

where M, L, and ML are the concentration of the metal ion, ligand and complex, respectively. The complexation reaction requires the desolvatation of both the metal ion and the ligand, the complexation process and the solvatation of the complex. Thus, the thermodynamic stability of a metal-ligand complex is the resultant of the sum of electronic effects (e.g., metal-donor atom bonding), steric effects (eg. ligand preorganization and size fitting), entropic contributions (e.g., chelate and macrocycle effects) and solvent dependencies (e.g., solvatation and ion-pairing).

To date, these concepts have not been brought together in a general theory for an accurate prediction of the metal-ligand complex stability despite the large amount of data now available [80]. Nonetheless, some approaches have been proposed with the aim of finding suitable molecular descriptors for either ligands or metal-complexes enabling the development of structure-stability relationships at least for congeneric series of compounds [81-84]. In the case of Gd(III) complexes, only few examples of structure-stability relationships are available in literature and most of them are based both on the use of the pK_a's (protonation constants) of the free ligands and on some descriptors obtained by molecular mechanics calculation.

Since the formation of a Gd(III) complex with protonated ligands is essentially a competition process between the metal ion and the proton, it is often possible to find a significant correlation between the protonation and the stability constants, providing that the ligands in the series have the same donor atoms and their hybridization state remains unchanged. Moreover, deviations from linearity are expected if atoms adjacent to the donor atoms are substituted with bulky residues or also if the same donor atoms form rings of different sizes with the metal ion after complexation. To the best of our knowledge, the first example of correlation between the pK_a's and stability constants was reported by Larsson in 1934 [85]. More recently, several authors extended these previous findings to Gd(III) complexes formed with both linear and macrocyclic polyamino-polycarboxylic ligands [86-88]. The results confirmed that it is possible to obtain a reasonable correlation when the ligand basicity is calculated as the sum of all those protonation constants ΣpK_a that are needed to result in a neutral ligand (e.g., one for glycine, two for IMDA, three for NTA, four for DOTA, five for DTPA) as shown in Fig.(**6**). An example of such correlation between $\log K_{ML}$ and ΣpK_a obtained for a series of polyamino-polycarboxilic ligands is shown in Fig.(**7**). The high correlation observed should not be overrated; it depends on the high similarity of either the donor atoms or the topology of the coordination cages in the selected ligands.

Nevertheless, the existence of such correlation strongly suggests the use of the ΣpK_a as an indirect measure of the average strength of the metal-ligands bonds after complexation. To date, the prediction of these pK_a's for complex polyprotonated systems continues to be an open issue (see article by Petrauskas *et al.* in this issue), as most of the works available in literature refer to singly or doubly ionized species. Indeed, even very recently, Li Xing and co-workers [90] have proposed a multivariate Partial Least Squares (PLS) model for the prediction of pK_a's using a training set of 384 bases and 645 acids. The novelty of their approach is in the molecular descriptors used, which are ionizing centres fingerprints, obtained representing the surrounding atoms of the ionizing centre in a hierarchical tree containing five levels, each level being defined by the topological distance from the ionizing centre. Each atom has been described taking into account its hybridization state or its belonging to some chemical groups. So far, for each ionizing centre, a string of 189 bins (33 atom types/groups x 5 levels + 24 atom types for the ionizing centre) has been built and then used as it is, in a PLS model calculation. From the validation data presented, it is not clear if the proposed model can be used with systems having more than two ionizing centres. The protonation process of polyprotic species is complicated by the relative basicity of the ionizing centres, which can result in an unpredictable sequence of microscopic protonation steps. The knowledge of these protonation steps is a piece of essential information needed to assign experimentally determined pK_a[1]s to a specific ionizing centre. To date, the best approaches to the prediction of pK_a[1]s, as well as complexation constants, seem to be those based on information platforms coupling a collection of experimental thermodynamic data, with quantitative relationships between some structural features of the ligands and their properties. An example of such information platforms can be described by the ACD/pK_a DB software [91], where both microscopic or macroscopic protonation constants can be predicted from the 2D representation of the molecular structure of a polyprotic compound. Using ACD/pK_a DB for the ligands shown in Fig.(**7**), the calculated ΣpK_a correlates quite well with the experimental one (r = 0.937), while the correlation with the stability constants $\log K_{ML}$ of the Gd(III) complexes is slightly lower (r = 0.835) with respect to the correlation calculated with the experimental ΣpK_a.

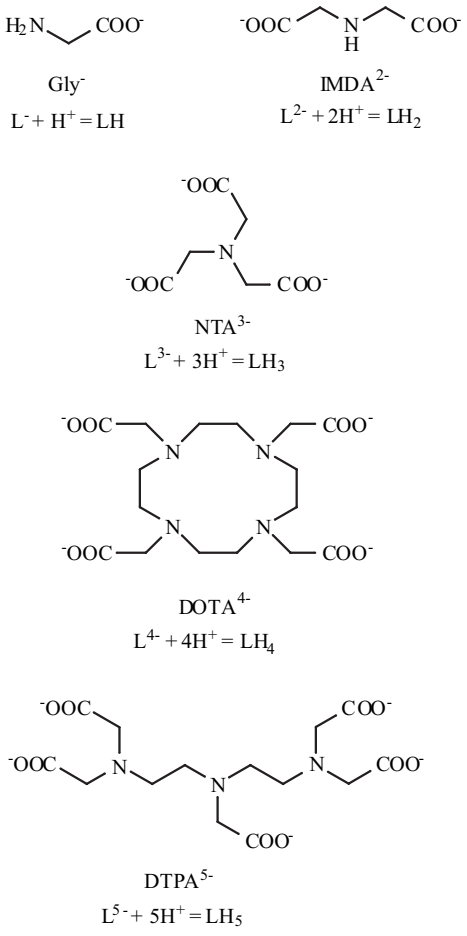


Fig. (6). Calculation of the number of protonation steps required to obtain a neutral form of some polyamino-polycarboxylic ligands.

Obviously, the ΣpK_a cannot be the only descriptor used in the development of a quantitative structure-stability relationship: many other independent effects are at work and, for each of them, we have to select an appropriate combination of molecular descriptors. It is important to point out that molecular descriptors, which depend on the structural property of the whole molecule may be useless: the complexation process, for each metal ion, is mainly related to the structural properties of the ligand in the surroundings of the donor atoms, as is clearly demonstrated by the comparison of the stability constants of a series of ligands shown in Table 6.

On the contrary, some simple descriptors, such as the counts of five and six-membered rings involving the metal ion in the complex, can give more direct information on the variation among the metal complex structures of the chelate ring effect. Similarly, a descriptor, such as the maximum topological dimension of a cycle (e.g., the count of the vertexes in the cycle) available in the ligand, containing at least two donor atoms, can account for possible variations of the macrocycle effects. As each metal ion has its own preference about the size of the coordination cage, the expected relationship between the dimension of the macrocycle in the ligand and the stability constant is non-linear: indeed the stability of the complex should have a maximum for a given macrocycle dimension. The calculation of the ligand cavity size can also be achieved with computational methods, such as molecular mechanics, which have already been extensively reviewed [92-93]. With the aim of correlating the stability constants of Gd(III) complexes with their structures, several authors have proposed the use of molecular descriptors derived from molecular mechanics calculations. Reichert *et al.*, have shown that the difference in the strain energy ΔE_{coord}, between the fully solvated complex and the desolvated complex, correlates with the thermodynamic stability constant log K_{ML} [55, 56]. In particular, an interesting correlation (r = 0.91) has been found for a series of nine polyamino-polycarboxilic Gd(III) complexes having only one water molecule in the inner coordination sphere q=1. The introduction of other coordinating groups, like amides or esters in place of carboxylic moieties, greatly reduces the quality of the correlation observed. However, a certain degree of correlation has also been shown for Gd(III) complexes

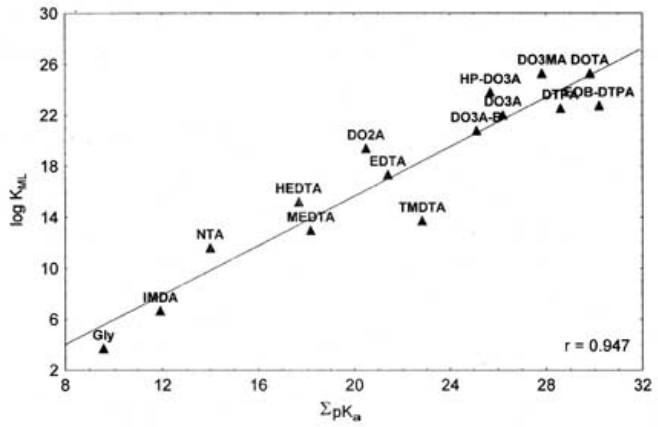


Fig. (7). Correlation between the logarithm of metal-ligand stability constants ($logK_{ML}$) and the sum of protonation constants (ΣpK_a) for several linear and macrocyclic polyamino-policarboxylic ligands. All structures and thermodynamic data are derived from reference [89].

having q = 2 and q = 3. A major drawback of the proposed method, which can partly explain the observed deviations from the observed linear correlation, is the lack of electrostatic effects in the applied force field.

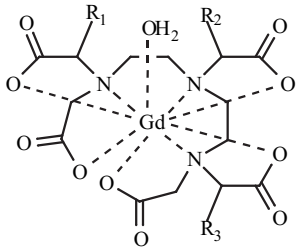
Again, in the framework of molecular mechanics calculations, Fossheim *et al.* have attempted to build a structure-stability relationship, using an estimation of the reaction energy of complexation [94,95]. According to the proposed approach, the reaction energy of complexation in aqueous solution, $E_{R,aq}$, can be calculated as follows:

$$E_{R,aq} = E_{R,g} + E_{H1} + E_{H2} \quad (9)$$

$$E_{R,g} = E_{GdL} - E_L \quad (10)$$

where $E_{R,g}$ is the reaction energy of complexation *in vacquo*, E_{H1} is the difference in the hydration energy of carboxylate or related donor groups in free ligands and in

Table 6. The Logarithm of Experimental Metal-Ligand Stability Constants ($logK_{ML}$) of Some GdDTPA Analogues



Compound	R_1	R_2	R_3	$logK_{ML}$ [a]
$[Gd(DTPA)(H_2O)]^{2-}$	H	H	H	22.55
$[Gd(BOPTA)(H_2O)]^{2-}$	CH2OCH2Ph	H	H	22.58
$[Gd(DIBOPTA)(H_2O)]^{2-}$	CH2OCH2Ph	H	CH2OCH2Ph	21.72
$[Gd(TRIBOPTA)(H_2O)]^{2-}$	CH2OCH2Ph	CH2OCH2Ph	CH2OCH2Ph	22.18

[a] Experimental conditions: 0.1M TMANO3, 25°C. Method: potentiometric.

complexes, and E_{H2} is the hydration of the Gd(III) ion in the complex due to the presence of water molecules in the inner coordination sphere.

From the above calculated reaction energies, the following order of complex stability was derived:

$Gd(DOTA)^{-} > GdDTPA^{2-} > Gd(DO3A) > Gd(OTTA)$ [94].

Since this result was in close agreement with the experimental data, in a second attempt, Fossheim *et al.* extended their method to another group of nine Gd(III) complexes in order to explore the performance of the method with complexes having a higher molecular diversity [95]. A fairly good correlation between the experimental $\log K_{ML}$ and the reaction energies of complexation ($r = 0.93$) has been achieved using six complexes of the original training set. To summarize, the results obtained using molecular descriptors derived from molecular mechanics energies calculation do not seem to be completely convincing owing to the limited number of complexes in the training set of the proposed quantitative structure-stability relationships, and the relatively high computational cost required for their calculation.

To the best of our knowledge, the first and only example of multivariate quantitative structure-stability relationship derived for a series of Gd(III) complexes has been recently published by Qi and co-workers [96]. In this work, the stability constants of 28 Gd(III) complexes derived from either linear or macrocyclic chelates have been subjected to correlation analysis with a set of 23 molecular descriptors derived directly from the structures of the chelates. Among the molecular descriptors used, 10 have been derived from semi-empirical calculation while the others have been calculated from the topology of the ligand structures (11 Kier-Hall connectivity indices [24] and 3 indices proposed by one of the authors in some previous works [97,98]). Multiple linear regression analysis and neural networks have been applied to derive quantitative structure-stability models. In the former, the Leaps-and-bounds variable selection algorithm [99] has been used to search for the best models while, in the case of neural network, the input layer has been filled with the best subset of variables derived from the previous multiple linear regression analysis. At the end of their analysis, the authors have proposed two models (one from each type of regression method applied) with the following five descriptors: the two Kier-Hall connectivity indices, $^{5}x_{pc}$ and $^{6}x_{pc}$; the molecular total energy; the ionization potential; and the HOMO-LUMO energy gap. Unfortunately, the proposed models have not been validated using cross-validation or related procedures. Moreover, the physical meanings of the models remain quite obscure too. Hence, at this time, it seems quite difficult to assess the validity of the proposed approach.

CONCLUSIONS

During the last two decades, a lot of work has been done to understand the basic principles governing the relevant responses to design suitable Gd(III)-based contrast agents. The results obtained were encouraging, as is demonstrated by the number of Gd(III) complexes with low-molecular weight already commercially available. Furthermore, their actual correspondence to medical needs has been demonstrated by the relevant role they have taken in MRI clinical practice. Nevertheless, by analyzing the chemical structures of these commercial Gd(III) complexes, significant molecular similarity can be clearly perceived. All of them are extra-cellular contrast agents and only the Gd(III) complex Multihance™ has shown a certain degree of hepatospecificity due to its partial excretion from the body through the hepatic route. In practice, most of the clinically relevant Gd(III) complexes have been developed from quite common design criteria where the search for high thermodynamic stability of the complex, biological inertness and reasonable relaxivity have dominated the scene. Only with the Gd(III) complex AngioMARK™, now undergoing clinical trials, has a new concept in the design of a contrast agent with higher relaxivity and a significant increase of its concentration in the blood stream (owing to strong binding with albumin) been concretized, opening towards MRI exams of vascular structures. In the near future, research programs for new MRI Gd(III)-based contrast agents should be able to produce complexes with higher relaxivity and tissue specificity, interacting more closely and selectively with biological material. Thus, the complexity of the phenomena governing the effectiveness of the Gd(III) complexes will grow as well, and more factors will be at work. In this scenario, it is expected that the interpretation of the facts observed on the sole basis of hard theories and models will be more time-consuming and less effective, owing to reduced control over the many factors involved in the phenomena observed. However, approaches based on soft modelling like QSAR/QSPR can provide an alternative methodology to understand how some structural variations can modify certain experimental responses, thus providing a practical tool with which to achieve multifactorial optimization. Nowadays, there are no technical reasons to avoid the use of such multivariate methodologies in the design of Gd(III)-based contrast agents. All the required computational tools are available, as has been shown in the present overview; probably the only additional requirement to expand their use, as in other more classical medicinal chemistry fields, is cultural.

ACKNOWLEDGMENTS

The author wishes to express his thanks to Dr. Luisella Calabi for helpful discussion. Dr. Massimo Mabilia is kindly acknowledged for providing the pK_a calculations with the ACD/pK_a DB software.

ABBREVIATIONS

ANNs	=	Artificial neural networks
BCUT	=	Burden – CAS - University of Texas eigenvalues
CN	=	Coordination number
CSAP	=	Capped Square antiprism
HOMO	=	Highest occupied molecular orbital
LMO	=	Leave-more-out
LOO	=	Leave-one-out

LUMO = Lowest unoccupied molecular orbital

MRI = Magnetic resonance imaging

PCA = Principal component analysis

PLS = Partial least squares regression

QSAR = Quantitative structure-activity relationship

QSPR = Quantitative structure-property relationship

SASA = Solvent accessible surface area

TAFF = Tripos force field

TTP = Tricapped trigonal prism

WHIM = Weighted holistic invariant descriptors

REFERENCES

[1] Vlaardingerbroek, M.T.; den Boer,J. A. *Magnetic Resonance Imaging: Theory and Practice*; Springer-Verlag: Berlin, **1996.**

[2] Mansson, S.; Bjørnerud, A. In *The Chemistry of Contrast Agents in Medical Magnetic Resonance Imaging*; Merbach, A. E., Tòth, È. Eds.; Wiley & Sons, Chichester, **2001**, pp.1-43.

[3] Caravan, P.; Ellison, J. J.; McMurry, T. J.; Lauffer, R. B. *Chem. Rev.* **1999**, *99*, 2293.

[4] Aime, S.; Botta, M.; Fasano, M.; Terreno, E. *Chem. Soc. Rev.* **1998**, *27*, 19.

[5] Lauffer, R. B. *Chem. Rev.* **1987**, *87*, 901.

[6] Jolliffe I. T. *Principal Component Analysis*, Springer-Verlag: New York, **1986.**

[7] Franke, R.; Gruska, A. In *Chemometrics Methods in Molecular Design*; Mannhold, R.; Krogsgaard-Larsen, P., Timmerman H., Eds.; VCH, Weinheim, **1995,** pp.113.

[8] Brereton, G. R. *Multivariate Pattern Recognition in Chemometrics,* illustrated by case studies, Elsevier: Amsterdam, **1992**.

[9] Downs, G. M.; Willet, P. In *Advanced Computer-Assisted Techniques in Drug Discovery*; Mannhold, R.; Krogsgaard-Larsen, P., Timmerman H., Eds.; VCH, Weinheim, **1995,** pp.111.

[10] Draper, N. R.; Smith, H. *Applied Regression Analysis, 2nd Ed.*;Wiley, New York, **1981.**

[11] Wold, S. In *Chemometrics Methods in Molecular Design*; Mannhold, R.; Krogsgaard-Larsen, P., Timmerman H., Eds.; VCH, Weinheim, **1995,** pp.196.

[12] Manallack, D. T.; Livingstone, D. J. In Advanced *Computer-Assisted Techniques in Drug Discovery*; Mannhold, R.; Krogsgaard-Larsen, P., Timmerman H., Eds.; VCH, Weinheim, **1995,** pp.293.

[13] Todeschini, R; Consonni, V. *Handbook of Molecular Descriptors*; Mannhold, R.; Krogsgaard-Larsen, P., Timmerman H., Eds.; WILEY-VCH, Weinheim, **2000.**

[14] Bukowska-Strzyzewska, M; Tosik, A. *Acta Cristallogr. Sect. B*, **1982**, *38*, 265.

[15] Rogers, R.D.; Kurihara, L. K. *Inorg. Chim. Acta*, **1987**, *130*, 131.

[16] Guggenberger, L. J.; Muetterties, E. L. *J. Am. Chem. Soc.*, **1976**, *98*, 7221.

[17] Inoue, M. B.; Inoue, M. *Inorg. Chim. Acta*, **1995**, *232*, 203.

[18] Chang, C. A.; Francesconi, L. C.; Malley, M. F.; Kumar, K.; Gougoutas, J. Z.; Tweedle, M. F.; Lee, D. W.; Wilson, L. J. *Inorg. Chem.*, **1993**, *32*, 3501.

[19] Ehnebon, L.; Pedersen, B. F. *Acta Chem. Scand.*,**1992**, *46*, 126.

[20] Konings, M. S.; Dow, W. C.; Love, D. B.; Raymond, K. N.; Quay, S. C.; Rocklage, S.M. *Inorg. Chem.,* **1990**, *29*, 1488.

[21] Alderighi, L.; Bianchi, A.; Calabi, L.; Dapporto, P.; Giorgi, C.; Losi, P.; Paleari, L.; Paoli, P.; Rossi, P.; Valtancoli, B.; Virtuani, M. *Eur. J. Inorg. Chem.*, **1998**, 1581.

[22] Kumar, K.; Chang, C. A.; Francesconi, L. C.;Dischino, D. D.; Malley, M. F.; Gougoutas, J. Z.; Tweedle, M. F. *Inorg. Chem.*, **1994**, *33*, 3567.

[23] Todeschini, R.; Consonni, V.; Mauri, A.; Pavan, M. *DRAGON Web Version 3.1*, **2003**, Italy, WebSite: http://www.disat.unimib.it/chm/.

[24] Kier, L. B.; Hall, L. H. *Molecular Connectivity in Structure-Activity Analysis*; Research Studies Press: Letchworth, **1986.**

[25] Bonchev, D. Information *Theoretic Indices for Characterization of Chemical Structures*; Research Studies Press: Letchworth, **1983.**

[26] Silipo, C.; Vittoria, A. *In Comprehensive in Medicinal Chemistry*; Hansch C., Sammes, P.G., Taylor, J. B., Eds.; Pergamon Press: Oxford, **1990**,Vol. *4*, pp. 154-200.

[27] Rücker, G ; Rücker, C. *J. Chem. Inf. Comp. Sci.*, **2000**, *40*, 99.

[28] Rücker, C ; Rücker, G. *J. Chem. Inf. Comp. Sci.*, **1994**, *34*, 534.

[29] Burden, F.R. *Quant. Struc.-Act. Relat.*, **1997**, *16*, 309.

[30] Pearlman, R. S.; Smith, K. M. In *3D-QSAR and Drug Design: Recent Advances*; Kubinyi, H., Martin, Y., Folkers, G., Eds.; Kluwer Academic: Dordrecht, **1997**; pp 339-353.

[31] Rios-Santamarina, I.; Garcìa-Domenech, R.; Gàlvez, J.; Cortijo, J.; Santamaria, P.; Marcillo, E., *Bioorg. Med. Chem. Lett.*, **1998**, *8*, 477.

[32] Broto, P.; Moreau, G.; Vandicke, C. *Eur. J, Med. Chem.*, **1984**, *19*, 66.

[33] Broto, P.; Moreau, G.; Vandicke, C. *Eur. J, Med. Chem.*, **1984**, *19*, 71.

[34] Broto, P.; Moreau, G.; Vandicke, C. *Eur. J, Med. Chem.*, **1984**, *19*, 79.

[35] Villa, A.; Cosentino, U.; Pitea, D.; Moro, G.; Maiocchi, A. *J. Phys. Chem. A*, **2000**, *104*, 3421.

[36] Cundari, T.R.; Moody, E. W.; Sommerer, S. O. *Inorg. Chem.*, **1995**, *34*, 5989.

[37] Cundari, T.R.; Saunders, L. C. *J. Chem. Inf. Comput. Sci.*, **1998**, *38*, 523.

[38] Beech, J.; Drew, M. G. B.; Leeson, P. B. *Struct. Chem.*, **1996**, *7*, 153.

[39] Amovilli, C.; Barone, V.; Cammi, R.; Cances, E.; Cossi, M.; Mennucci, B.; Pomelli, C. S.; Tomasi, J. *Adv. Quantum Chem.*, **1998**, *32*, 227.

[40] Cosentino, U.; Villa, A.; Pitea, D.; Moro, G.; Barone, V.; Maiocchi, A. *J. Am. Chem. Soc.*, **2002**, *124*, 4901.

[41] Banci, L.; Bertini, I.; Luchinat, C. *Nuclear and Electron Relaxation*, VCH: Weinheim, **1991.**

[42] Koenig, S.H.; Brown III, R.D. *Prog. Nucl. Magn. Reson. Spectrosc.*, **1990**, *22*, 487.

[43] Kirsch, J. E. *Top. Magn. Reson. Imaging*, **1991**, *3*, 1.

[44] Kowalewski, J.; Nordenskioeld, L.; Benetis, N.; Westlund, P.O. *Prog. Nucl. Magn. Reson. Spectrosc.*, **1985**, *17*, 141.

[45] Bertini, I.; Luchinat, C. *Coord. Chem. Rev.*, **1996**, *150*, 1.

[46] Sosnovsky G. *Appl. Magn. Reson.*, **1992**, *3*, 131-149.

[47] Castonguay L. A.; Treasurywala A. M.; Caulfield, T.J.; Jaeger, E. P.; Kellar, K. E. *Bioconjugate Chem.*, **1999**, *10*, 958.

[48] Connolly, M. L. *J. Appl. Cryst.*, **1983**, *16*, 548.

[49] Chatterjee, A.; Maslen, E. N.; Watson, K.J. *Acta Cristallogr. Sect. B*, **1988**, *44*, 381.

[50] Templeton L. K.; Templeton, D. H.; Zalkin, A.; Ruben, H.W. *Acta Cristallogr. Sect. B*, **1982**, *38*, 2155.

[51] Gries, H.; Miklautz, H. *Physiol. Chem. Phys. Med. NMR*, **1984**, *16*, 105.

[52] Smith, P. H.; Brainard, J. R.;Morris, D. E.; Jarvinen, G. D.; Ryan, R. R. *J. Am. Chem. Soc.*, **1989**, *111*, 7437.

[53] Dubost, J. P.; Leger, J. M.; Langlois, M. H.; Meyer, D.; Schaefer, M. *C. R. l'Academie. Sci. Ser. II Univers.*, **1991**, *312*, 349.

[54] Chen, D.; Squattrito, P. J.; Martell, A. E.; Clearfield, A *Inorg. Chem.*, **1990**, *29*, 4366.

[55] Clark, M.; Cramer III, R. D.; Opdenbosch *J. Comp. Chem*. **1989**, *10*, 982.

[56] Reichert, D. E.; Hancock, R. D.;Welch, M. *J. Inorg. Chem.*, **1996**, *35*, 7013.

[57] Reichert, D. E.; Welch, M. J. *Coord. Chem. Rev.*, **2001**, *212*, 111.

[58] Shannon R. D. *Acta Cristallogr. Sect. A*, **1976**, *32*, 751.

[59] Zhang, X.; Pillai, K. M. R.; Shukla, R.; Ranganathan, R. S.; Tweedle, M. F. *Invest. Radiol. Suppl. 2*, **1994**, *29*, S69.

[60] Ranganathan, R. S.; Fernandez, M. E.; Kang, S. I.; Nunn, A. D., Ratsep, P.; Pillai, K. M. R.;Zhang, X.; Tweedle, M. F. *Invest. Radiol.*, **1998**, *33*, 779.

[61] Bondi A. *J. Phys. Chem.*, **1964**, *68*, 441.

[62] Gavezzotti A. *J. Am. Chem. Soc.*,**1983**, *105*, 5220.

[63] Rohrbaugh, R. H.; Jurs, P. C. *Anal. Chim. Acta.*, **1987**, *199*, 99.

[64] Pearlman R.S. *In Physical Chemical Properties of Drugs*; Yalkowsky S. H., Sinkula A. A., Valvani S. C. Eds.; Marcel Dekker: New York, **1980**; pp. 321-347.

[65] Pacios, L. F. *Computers Chem.*, **1994**, *18*, 377.

[66] Labanowski, J. K.; Motoc, I.; Dammkoehler, R. A. *Computers Chem.*, **1991***, 15*, 47.
[67] Todeschini, R.; Cazar, R. Collina, E. *Chemom. Intell. Lab. Syst.*, **1992**, *15*, 51.
[68] Tòth, È.; Pubanz. D.; Vauthey, S.; Helm, L.; Merbach, A. E. *Chem. Eur. J.*, **1996**, *2*,1607.
[69] Lammers, H.; Maton, F.; Pubanz, D.; Van Laren, M. W.; Van Bekkum, H.; Merbach, A. E.; Muller, R. N.; Peters, J. A. *Inorg. Chem.,* **1997**, *36*, 2527.
[70] Tòth, È.; Burai, L.; Brücher, E; Merbach, A. E. *J. Chem. Soc. Dalton Trans.*, **1997**, 1587.
[71] Tòth, È; Connac, F.; Helm, L.; Adzamli, K.; Merbach, A. E. *Eur. J. Inorg. Chem.*, **1998**, 2017.
[72] Pubanz, D.; Gonzales G.; Powell, D.H.; Merbach, A. E. *Inorg. Chem.*, **1995**, *34*, 4447.
[73] Cosentino, U.; Moro, G.; Pitea, D.; Villa, A.; Fantucci, P. C.; Maiocchi, A.; Uggeri, F. *J. Phys. Chem. A*, **1998**, *102*, 4606.
[74] Todeschini R.; Gramatica P. *Quant. Struct.-Act. Relat.*, **1997**, *16*, 113.
[75] Sieving, P. F.;Watson, A. D.; Rocklage, S. M. *Bioconjugate Chem.*, **1990**, *1*, 65.
[76] Aime S.; Botta, M.; Crich, S.G.; Giovenzana, G.; Palmisano, G.; Sisti, M. *Bioconjugate Chem.*, **1999**, *10*, 192.
[77] Dong, Q.; Hurst, D. R.;Weinmann, H. J.; Chenevert, T. L.; Londy, F. J.; Prince, M.R. *Invest. Radiol.*, **1998**, *33*, 699.
[78] André, J. P.; Tòth, È.; Maecke, H. R.; Merbach, A. E. *Chem. Eur. J.*, **1999**, *5*, 2977.
[79] Aime S.; Fasano M.; Terreno, E. ; Botta, M. In *The Chemistry of Contrast Agents in Medical Magnetic Resonance Imaging*; Merbach, A. E., Tòth, È. Eds.; Wiley & Sons, Chichester, **2001**, pp.193-241.
[80] Smith R. M.; Martell, A. E. *Critically Selected Stability Constants of Metal Complexes Database. Version 6.0*, National Institute of Standards and Technology: Gaithersburg, **2001**.
[81] Shi, Z. G.; McCollough, E. A. Jr. *J. Inclusion. Phenom.*, **1994**, *18*, 9.
[82] Gakh, A. A.; Sumpter, B. G.; Noid, D. W.; Sachleben, R. A.; Moyer, B. A. *J. Inclusion. Phenom.*, **1997**, *27*, 201.
[83] Solov'ev, V. P.; Varnek, A.; Wipff, G. *J. Chem. Inf. Comput. Sci.*, **2000,** *40*, 847.
[84] Comba, P. *Coord. Chem. Rev.*, **1999**, *185-186*, 81.
[85] Larsson, E., *Z. Physik, Chem. Leipzig*, **1934**, *A169*, 207.
[86] Choppin, G.R. J. *Less-Common Met.*, **1985**, *112*, 193.
[87] Kumar, K.; Chang, C. A.; Tweedle, M. F. *Inorg. Chem.*, **1993**, *32*, 587.
[88] Kumar, K.; Tweedle, M. F.; Malley, M. F. ; Gougoutas, J. Z. *Inorg. Chem.*, **1995**, *34*, 6472.
[89] Bianchi, A.; Calabi, L.; Corana, F.; Fontana, S.; Losi, P.; Maiocchi, A.; Paleari, L.; Valtancoli, B. *Coord. Chem. Rev.*, **2000**, *204*, 309.
[90] Xin, L.; Glen, R.C. *J. Chem. Inf. Comput. Sci.* , **2002**, *42*, 796.
[91] *ACD/pK_a DB, Version 6.00*, Advanced Chemistry Development Inc., 90 Adelaide Street West, Toronto, Ontario, Canada. WebSite: www.acdlabs.com.
[92] Hancock, R.D.; *Acc. Chem. Res.*, **1990**, *23*, 253.
[93] Comba, P.; Hambley, T. W. *Molecular Modelling of Inorganic Compounds*, VCH; Weinheim, **1995.**
[94] Fossheim, R.; Dahl, S. G. *Acta Chem. Scand.*, **1990**, *44*, 698.
[95] Fossheim, R.; Dugstad, H.; Dahl, S. G. *J. Med. Chem.* **1991**, *34*, 819.
[96] Qi, Y.; Zhang, Q.; Xu, L. *J. Chem. Inf. Comput. Sci.*, **2002**, *42*, 1471-1475.
[97] Yao, Y. Y.; Xu, L.; Yang, Y. Q.;Yuan, X. S. *J. Chem. Inf. Comput. Sci.*, **1993**, *33*, 590.
[98] Xu, L.; Yao, Y. Y.; Wang, H. M. *J. Chem. Inf. Comput. Sci.*, **1995**, *35*, 45.
[99] Furnival, G. M.; Wilson, R. W. Jr. *Technometrics*, **1974**, *16*, 499.