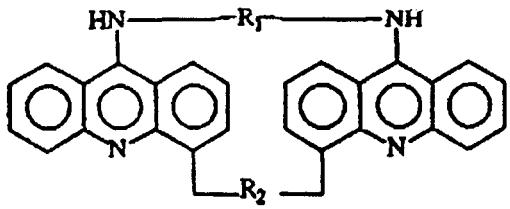
Polyfunctional intercalation has the potential of providing a mechanism for high-affinity binding to DNA. Such compounds could have interesting antitumor and antiviral properties. A topologically novel bifunctional DNA intercalator, **2**, has been synthesized and its DNA binding studied in comparison with the binding of 9-aminoacridine, **1**, and spermine diacridine, **3**. Compound **2** shows a high affinity for calf thymus DNA as indicated by large increases in the helix-coil transition temperature. Viscometric analysis of helix extension using sonicated calf thymus DNA gave results characteristic of a double intercalator. Metachromic shifts in the absorption spectra of **2** were observed upon addition of DNA. Macrocycle, **2** reverses the supercoils in a closed circular supercoiled plasmid (pOP1Δ6). These data, and other experiments, support the conclusion that **2** binds to DNA as a bifunctional intercalator. DNA binding and molecular modeling studies of these polyfunctional DNA intercalators will be discussed.



1 $R_1 = H, R_2 = H$
2 $R_1 = (CH_2)_3NH(CH_2)_4NH(CH_2)_3$, $R_2 = CH_2S(CH_2)_2NHCO(CH_2)_2CONH(CH_2)_2SCH_2$
3 $R_1 = (CH_2)_3NH(CH_2)_4NH(CH_2)_3$, $R_2 = H$

Structure and Molecular Modeling of the Anti-infective Drug Pentamidine

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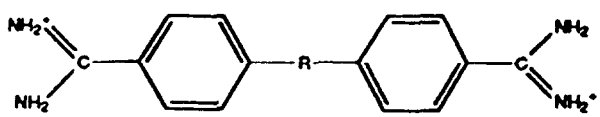
Pentamidine (1a) is valuable for the treatment of *Pneumocystis carinii* infections in AIDS patients as well as trypanosomiasis. The molecule is terminated by the same aromatic amidine moieties as the DNA-binding drug berenil (1b), but the central chain is longer.



a: R = –O–$(CH_2)_5$–O–
b: R = –NH–N=N–

The crystal structure of berenil has been determined,[1] and its interaction with A–T rich regions of DNA has been investigated by graphic modeling and molecular mechanics.[1] The present study comprises a determination of the crystal structure of (1a) as the hydrated isethionate salt and an investigation of the conformational preferences and electronic properties of three important structural features of the pentamidine molecule, namely the junction between amidinium groups and benzene rings, the ether linkages and the central chain.

The crystals have unit cell dimensions a = 25.959, b = 14.105, c = 8.781 Å, β = 102.44° and exhibit symmetry consistent with space group C2/c. The structure has been refined to R = 0.06, with typical standard deviations of 0.006Å and 0.5° for nonhydrogen bond distances and angles. As in (1b), molecules of (1a) adopt a flat extended conformation and have twofold symmetry imposed by a crystallographic axis through the central methylene C atom. The planes of the amidinium groups and benzene rings intersect at 27° in (1a) but only 8° in (1b). However, the latter angle increased upon refinement using molecular mechanics of (1b) with DNA.[1] Our *ab initio* STO-3G calculations for the phenylamidium cation show that the energy is relatively insensitive to twists up to ca. 35°, with a minimum at 27°.

The C–O–C angle is 117.9° with no significant twist about either bond. This corresponds with one of the two energy minima found by STO-3G calculations for anisole.[2]

Superposition of the middle three atoms of (1a) onto the central chain of (1b) places the aromatic rings in similar regions of space with their inner edges in (1a) aligned with their outer edges in (1b). Thus, the vector from end to end of the central chain is 5.20 Å longer in (1a) than in (1b). For a polymer to have an isohelical fit with the minor groove of a DNA helix at a distance of 5.0 Å from the helix axis, a chain length of 4.61 Å between contacts with successive bases is required.[3] The bending and twisting needed to enable (1a) to conform to a helical template should reduce the length of the chain vector to around this value, thus enabling it to span one more base-pair than (1b).

This work was supported by the Science and Engineering Research Council under the Molecular Recognition Initiative.

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Accurate Redox Potentials from Theoretical Calculations

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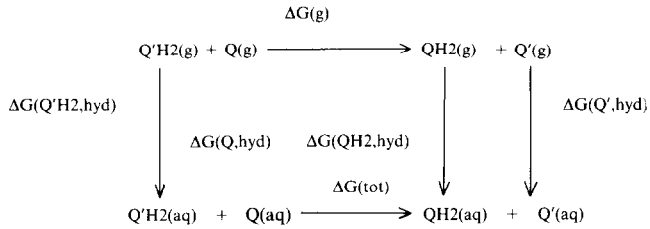
The ability to calculate electrode potentials would be advantageous in many areas of molecular design because many molecules are activated by enzymatic oxidation or reduction. Previous theoretical approaches to this problem have usually neglected the solvent and have often neglected the reduced molecules, too, using the LUMO energy as a guide to the electrode potential. However, the free-energy perturbation method has recently been used to calculate free energies of hydration with astonishing accuracy, and combined with high-level quantum mechanical calculations, it is possible to treat the problem accurately using the following thermodynamic cycle to evaluate differences in free energy in aqueous solution.



$$\Delta G(tot) = \Delta G(QH2,hyd) - \Delta G(Q,hyd) + \Delta G(Q',hyd) - \Delta G(Q'H2,hyd) + \Delta G(g)$$

Here, Q and Q′ are, for example, two quinone molecules. The gas phase reactions have been treated with a variety of basis sets of 6-31g* (or superior) quality, at both the RHF and MP2 levels. Zero-point, enthalpy and entropy corrections to the *ab initio* energies have been made using semiempirical MO theory. The hydration free-energy components have been calculated using the free-energy perturbation method during computer simulations in which QH2 is mutated to Q during about 40–50 ps of molecular dynamics.

The results for the difference in electrode potentials between parabenzoquinone and ortho-benzoquinone are accurate to 20 mV, and this level of accuracy has been reproduced for a growing range of compounds. However, the work is significant beyond its potential use in the design of bioreductive compounds because it shows that free energies can now be calculated for reactions in solution, as well as for reactions in the gas phase.

REFERENCE

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The Use of Molecular Modeling in Anticancer Drug Design

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DNA is generally regarded as the most sensitive and appropriate molecular target for the action of cytotoxic drugs, and the majority of current antitumor agents indeed interact with DNA.

The full description of structural features of intercalation (the binding mechanism utilized by clinically important agents such as adriamycin and actinomycin) has not yet been obtained from X-ray crystallographic analyses. We have used computer modeling to extend crystallographic data so as to produce a model for an intercalation site in the middle of an oligonucleotide sequence. This has subsequently been used for modeling a number of intercalating molecules and derivatives in order to derive structure-activity relationships. Several examples of this approach will be given in the presentation, and its scope and limitations will be discussed.

Identification of a Bioactive Conformer of Glutamate at a Quisqualate-Sensitive Glutaminergic Receptor

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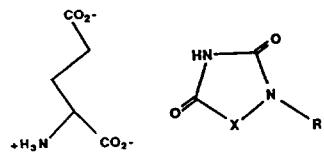
L-Glutamate **(1)** is now regarded as an important excitatory neurotransmitter in a number of vertebrate and invertebrate systems in which three principal receptor types have been identified: quisqualate (Q, N-methyl-D-aspartate (NMDA) and kainate (K). As part of our ongoing structure/activity program on glutamate agonists and antagonists, several glutamate analogues have been synthesized and evaluated in the well-defined, quisqualate-sensitive, glutaminergic synapse of the locust leg nerve-muscle preparation. In order to provide a molecular rationale for our experimental observations, a combination of X-ray crystallographic analyses, semiempirical MO calculations and molecular mechanics studies have been used to identify a bioactive conformer of glutamate at this Q receptor.



	R	X
(2)	$-CH_2CH(NH_2)CO_2H$	O
(3)	$-CH_2CH(NH_2)CO_2H$	CH_2
(4)	-Me	O
(5)	-Me	CH_2
(6)	-Me	NH

(1)

Semiempirical MNDO calculations on model systems (4–6), together with X-ray crystallographic data on quisqualic acid **(2)** and its carbon analogue **(3)**, have shown that the amide nitrogen attached to the alkyl side chain in systems **(2)**, **(4)** and **(6)** is in fact pyramidal. Those in the hydantoin type systems **(3)** and **(5)** are, as expected, planar. Data from the MNDO calculations along with observed crystal lattice structures suggest that the barriers to inversion in the pyramidal nitrogens (Scheme 1) are low, comparable to bond rotation ener-