FEATURE ARTICLE

ChemComm
www.rsc.org/chemcomm

Dendrimers as guests in molecular recognition phenomena†

Winston Ong, Marielle Gómez-Kaifer and Angel E. Kaifer*
Center for Supramolecular Science and Department of Chemistry, University of Miami, Coral Gables, FL 33124-0431, USA. E-mail: akaifer@miami.edu; Fax: 1 305 444 1777; Tel: 1 305 284 3468

Received (in Cambridge, UK) 27th January 2004, Accepted 10th March 2004
First published as an Advance Article on the web 14th April 2004

Dendrimers are highly branched macromolecules which may engage in host–guest interactions, acting as either hosts or guests; this review is specifically concerned with the binding behavior of dendrimers containing single or multiple guest residues interacting with individual, freely diffusing hosts.

Introduction

Research interest on functionalized dendrimers has exploded in recent years and several comprehensive reviews have documented the large amount of research work in this area.[1] One of the key properties of dendrimers is that they usually exhibit inner cavities, which can be occupied by smaller molecules. This is a simple form of molecular recognition in which the dendrimer can be considered the host (receptor) and the trapped molecule would be the guest (substrate). Small molecule trapping was observed very early in the development of dendrimers, although the experiments of Meijer and co-workers exploited the trapping of small molecules in very elegant ways.[2] A number of groups have developed this idea further by incorporating *designed binding sites* within the dendrimer innards, thus imparting more specific recognition properties to the dendrimer hosts. An excellent example of such systems is illustrated by Diederich and Felber's 'dendrophanes', which contain a single cyclophane binding site at the core of dendritic compounds.[3] Very recently, the groups of Zimmerman and Suslick have pioneered the preparation of synthetic hosts by molecular imprinting inside dendrimers.[4] All these systems, in which dendrimers behave as high molecular weight, structurally complex hosts, resemble proteins as they interact with small molecules (ligands). While the biological inspiration of this type of research work is clear, dendrimers offer further research possibilities with strong biological overtones. Many molecular recognition processes in living systems take place at interfaces. For example, a substrate group, covalently attached to a large macromolecular structure or assembly (such as a membrane), is recognized and bound by a suitable molecular receptor of variable size (Fig. 1).

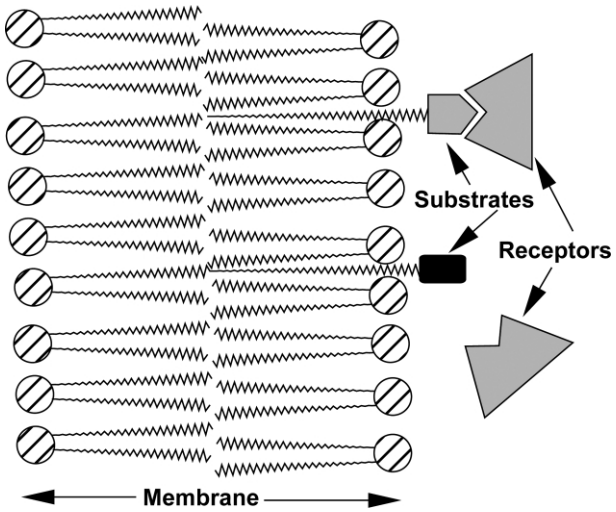


Fig. 1 Biological molecular recognition at a membrane interface. In biochemical terms, membrane-anchored substrate residues are often called 'receptors', which may be bound by antibodies, proteins or other freely diffusing biological macromolecules acting as the molecular receptors depicted in this schematic drawing.

More specifically, T cell recognition of cell surfaces involves a series of molecular recognition events taking place between residues attached to their respective cell membranes.[5] In this case, the molecular recognition partners come together and interact while connected to large, presumably inert, structures, which may, however, exert considerable influence on the binding events. Such considerations stimulated our interest in the possibility of using dendrimers as guests, rather than hosts, in molecular recognition phenomena and begs the obvious question: *How does the dendrimer mass and bulk affect the binding affinity between one or more dendrimer-attached guest residues and free hosts in the solution?* This article reports progress in this area with two types of systems: (a) dendrimers containing a single guest residue, and (b) dendrimers containing multiple guest residues on their peripheries. Owing to space limitations, complex systems involving recognition *via* ligand coordination to metal sites,[6] or mechanically linked

Winston Ong obtained a B.S. Degree in Chemistry (1996) from De La Salle University (Manila, Philippines), and a Ph.D. Degree in Chemistry (2002) from the University of Miami, the latter under the supervision of Angel E. Kaifer. He is currently pursuing further postdoctoral training at Louisiana State University in the laboratory of Robin McCarley. His overall research interests include the synthesis, molecular recognition, encapsulation and self-assembly of simple and dendritic property-active systems.

Marielle Gómez-Kaifer holds a B.F.A. in Photography and Painting, a B.S. in Chemistry (second major in Microbiology and Immunology) and a Ph.D. in Chemistry from the University of Miami. Her research interests focus on supramolecular electrochemistry and magnetic resonance spectroscopy.

Angel E. Kaifer holds an undergraduate degree in Chemistry (1977) from Universidad Autónoma de Madrid, Spain, and a Ph.D. (1984) from the University of Puerto Rico, Rio Piedras. He did postdoctoral work at the University of Texas, Austin, and joined the faculty at the University of Miami in 1985, where he is currently a Professor in the Department of Chemistry. His research interests focus on the electrochemistry of supramolecular systems and nanometer-sized systems, such as dendrimers, molecular capsules and metal nanoparticles.

† In memory of Dr Moisés Morán.

DOI: 10.1039/b401186d

systems such as those of Stoddart and co-workers[7] will not be covered in this review.

Single site dendrimer guests

While the majority of the early work in the area of dendrimers as guests involved multi-site systems (*vide infra*), more recent work on single site structures has provided salient results for assessing binding affinity in dendrimer systems. We will discuss single site dendrimer guests first in order to explore the effects of intermolecular interactions in the host–guest pair, as well as the effects of dendrimer growth and shape on the binding process. Interest in dendrimer guest systems arose in the author's group due to the desire to study molecules that better approximate some of the directional reactivity and selectivity exhibited by some redox proteins in their electron transfer reactions. To that end, some of our earliest work in this area dealt with unsymmetric ferrocenyl and dansyl dendrimers.[8] Based on Newkome-type dendrimers, structures **1**–**3** and **4**–**6** (Fig. 2) show hydrophilic properties resulting from their terminal carboxylic acid residues. Ferrocenyl dendrimers **1**–**3** and their interaction with host **7**, β-cyclodextrin (β-CD), were investigated in aqueous solution by voltammetric techniques.[8] The effects of β-CD binding on the voltammetric response of ferrocene derivatives have been well documented.[9] Briefly, electron transfer does not take place directly from the inclusion complex, but rather the complex must first dissociate prior to the electrochemical oxidation step. This is an example of a CE (chemical-electrochemical) mechanism, in electrochemical jargon, in which the electron transfer is preceded by a chemical step (complex dissociation). The degree of binding affinity between the ferrocenyl compound and β-CD is typically evidenced by both a positive shift in the half-wave potential ($E_{1/2}$) and a decrease in the voltammetric current due to the slower diffusion of the β-CD complex compared to that of the free ferrocenyl derivative. Results for dendrimers **1**–**3** showed that increasing dendrimer growth inhibits binding of β-CD to the ferrocenyl dendrimer. This finding suggested that growth of the dendrimer interferes with the approach of the CD host and its inclusion of the ferrocene moiety, likely due to steric crowding (*vide infra*). These results were further borne out by ^{1}H-NMR spectroscopic and steady-state and time-resolved fluorescence measurements obtained with dansyl dendrimers **4**–**6**, which demonstrated that binding of β-CD to the second and third generation dendrimers **5** and **6** progressively weakened as compared to that observed with guest **4** or a model dansyl compound.[8] We should note, however, that the association constants for both ferrocenyl and dansyl groups with β-CD are relatively low, typically 10^3 M^{-1} or lower. In contrast, binding studies performed in the laboratory of our collaborator, Professor Frank Bright (SUNY-Buffalo), with a polyclonal anti-dansyl antibody (α-DA) and dendrimers **4**–**6** showed that dendrimer growth, while having an attenuating effect on the binding affinity, did not quell the binding of α-DA to the same extent as in the case of the β-CD host. Studies by steady-state fluorescence anisotropy showed that the strong binding obtained for α-DA to dansylamine (K_a of the order of 10^7 M^{-1}) was only moderately attenuated in the dansyl dendrimers **4**–**6** (10^6 M^{-1} for all three dendrimers), suggesting that sufficiently strong intermolecular interactions between a host–guest pair will not be greatly attenuated by dendrimer growth.[8] Under such conditions the flexible character of the dendrimer will allow for its rearrangement or folding away in order to accommodate binding and the corresponding host–guest complex will be formed due to its considerable thermodynamic stability.

More recent work with viologen-containing dendrimers and cucurbit[7]uril has further elucidated this point. The interaction of viologen derivatives with the host cucurbit[7]uril (CB7), **8**, has been the subject of interest of both our group and that of Kimoon Kim and co-workers (Fig. 3).[10,11] Methyl viologen (MV^{2+}), **9**, shows a moderately high association constant with cucurbit[7]uril,

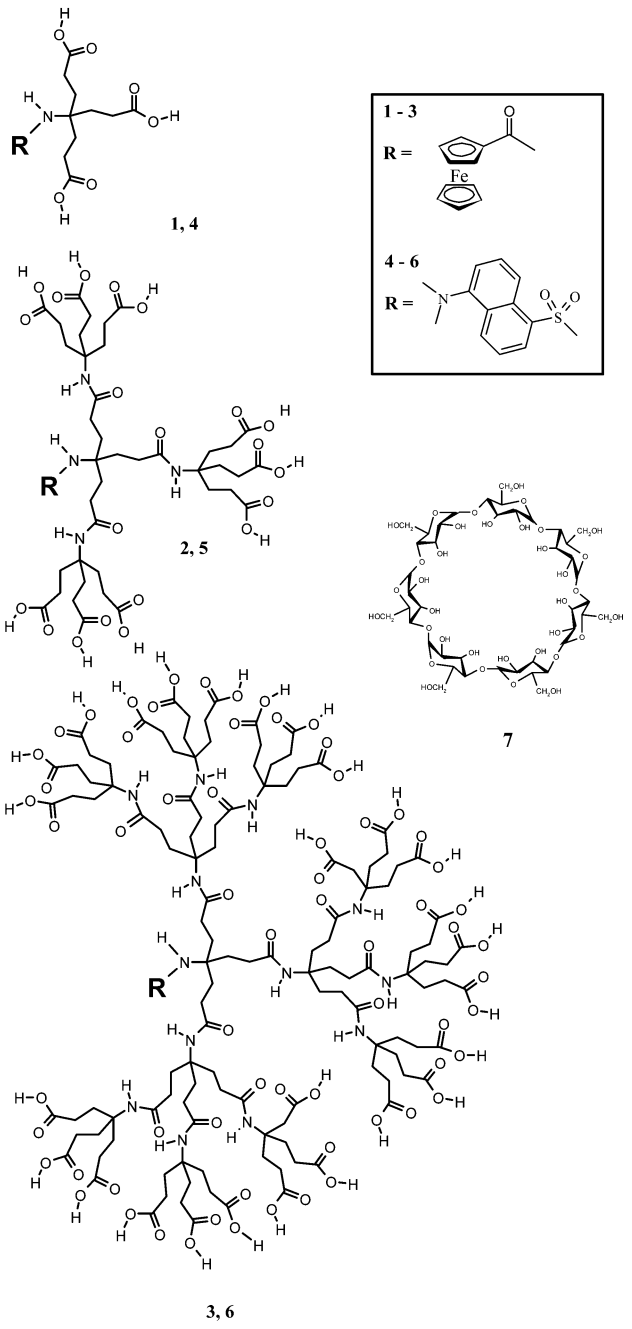
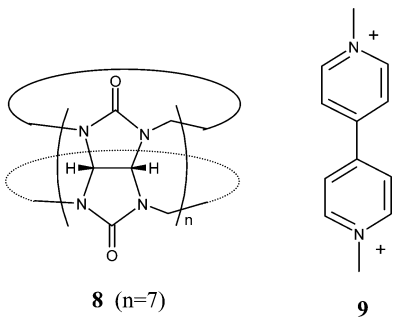


Fig. 2 Structures of ferrocene (**1**–**3**) and dansyl (**4**–**6**) dendrimer guests and the β-cyclodextrin host (**7**).



8 (n=7) **9**

Fig. 3 Structures of the cucurbit[7]uril host (**8**) and its dicationic guest methyl viologen (**9**).

of the order of 10^5 M^{-1} as determined by electronic absorption spectroscopy, in aqueous media of low ionic strength.[11] Based on

this finding and our interest in dendrimer guests, we prepared hydrophilic dendrimers **10a**–**12a** (Fig. 4), in which the viologen

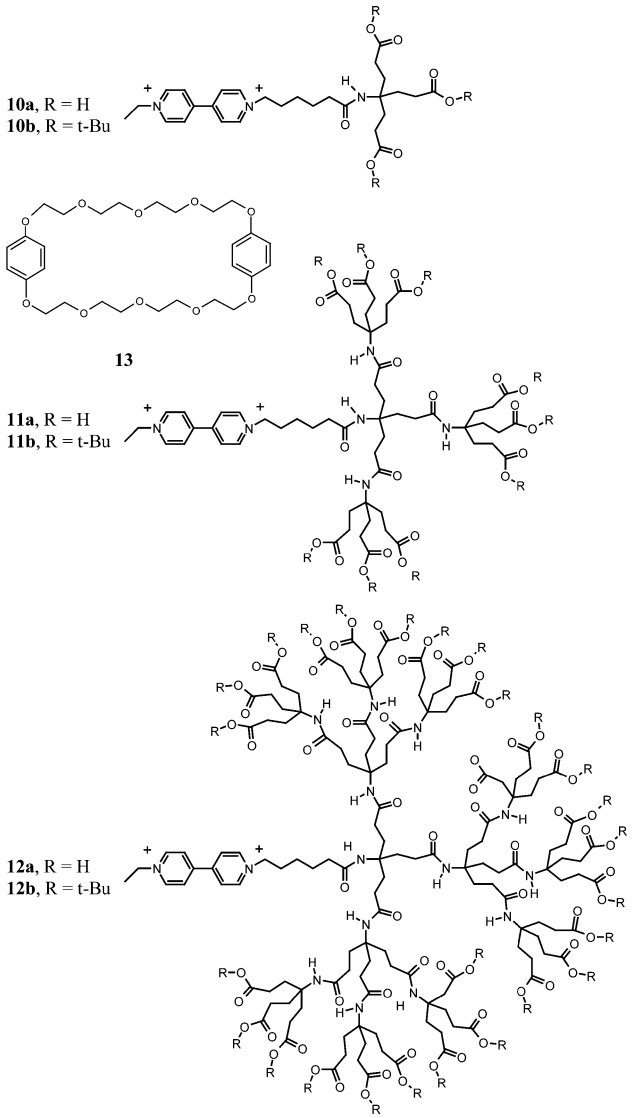


Fig. 4 Structures of Newkome-type viologen dendrimer guests and the host BPP34C10 (**13**).

guest is attached to a Newkome-type dendrimer *via* a tether. Notice that this tether is longer than the direct amide connections used with the ferrocene and dansyl dendrimers. We did attempt to prepare viologen dendrimers with shorter tethers in order to increase the structural similarities with dendrimers **1**–**6**. Unfortunately, shorter tethers led to rather unstable dendrimers. Although the binding affinity of the CB7–MV^{2+} complex depends on the composition of the medium, in acidic solutions of relatively low ionic strength dendrimer growth in **10a**–**12a** shows negligible effects on the stability of the complexes formed with CB7, that is the association constants remain in the range of 10^5 M^{-1}.[12] Several factors may influence these findings, which differ from those seen with the ferrocenyl dendrimers (**1**–**3**) and dansyl dendrimers (**4**–**6**) and their complexes with β-CD. The five-methylene tether, which permits CB7 to approach the dendrimer with fewer steric limitations, may play a role, but other experimental results suggest that this role is rather limited. Furthermore, from a statistical standpoint there is an advantage in that either opening of the CB7 can bind the guest, while in the case of β-CD, only one of the host openings provides ideal docking for optimal inclusion of the ferrocenyl or dansyl guest. The most relevant factor, however, is likely the overall higher binding association strength between the viologen moiety and CB7. These conclusions are further supported by recent work in which the binding association of structurally related dendrimers **10b**–**12b** and crown *bis-para*-phenylene-34-crown-10 (BPP34C10), **13**, was examined.[13–14] The crown host exhibits a relatively low association constant for the viologen moiety, *e.g.* of the order of 10^2–10^3 M^{-1} with MV^{2+}. Hydrophobic viologen dendrimers **10b**–**12b** were studied in acetone and acetonitrile solutions and were found to have significantly reduced association with the crown host as the dendrimer growth increased. Thus, the role of the five carbon tether or any statistical benefit from the host's symmetry appears to be far less significant than the overall strength of the host–guest binding interaction.

A further, less explored, point of interest in these systems is the effect that dendrimer shape and host orientation may have on the strength of host–guest association. In collaboration with the group of Prof. Carlos Peinador (Universidade da Coruña, Spain), we have recently begun to investigate viologen dendrimers based on Fréchet-type building blocks (Fig. 5).[15] Dendrimers **14**–**16** and

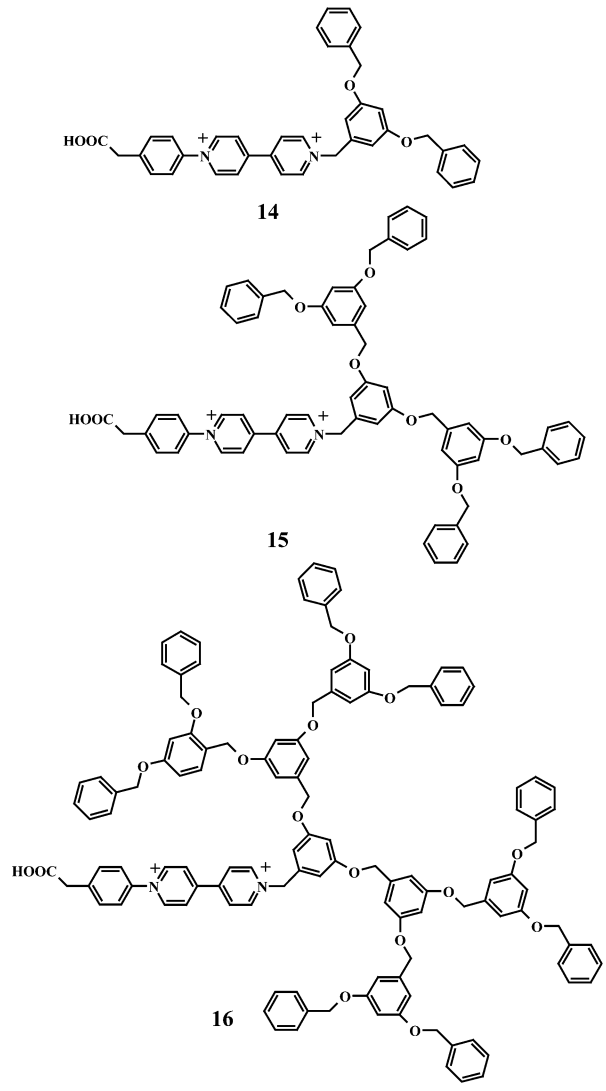


Fig. 5 Fréchet-type dendrimer guests surveyed for binding with crown host **13**.

their interaction with host **13** were studied. In contrast to the results observed for dendrimers **10a**–**12a**, little if any change is observed in the association constants of **14**–**16** with crown host **13** from the corresponding value observed for **13** and methyl viologen (**9**). We have postulated that the lower flexibility and the more two-dimensional character of the Fréchet-type dendrimers (based on an AB_2-type building block) may foster greater ease of binding, even in more branched structures, such as a third generation dendrimer.

In constrast to this, Newkome-type dendrimers (based on an AB_3-type building block) are more flexible and may hinder the encapsulation of the apical guest residue by the host. In any instance, these results afford an interesting comparison of the properties of two widely used dendrimer architectures.

Fig. 6 illustrates and defines some of the relevant parameters that may control the extent of hindrance that the host experiences as it approaches the guest residue, which is covalently attached to the dendrimer. Parameter *X* represents the length of the tether linking the guest residue to the amine functional group on the apical or focal position of the Newkome-type dendron framework. Parameter ϕ represents the angle at which the host approaches the guest moiety for optimal binding interaction. The host can approach the guest with its main axis in a range of orientations *vs.* the main axis of the dendrimer, which is basically considered to be aligned with the tether connecting the guest residue to the dendron's focal point. For instance, $\phi = 0°$ describes the sliding motion of the host CB7 over the viologen moiety to form the optimum inclusion complex. The other extreme corresponds to an angle of *ca.* 90°. This situation is anticipated for the binding of β-CD to ferrocenyl dendrimers, as the wider opening of this host will include the unsubstituted cyclopentadienyl ring of the ferrocene group. A large ϕ angle requires the host to make a closer approach to the dendrimer structure, even if the guest functional group is connected to the dendron through a tether of significant length, and steric considerations may become more important than in binding situations characterized by $\phi \approx 0°$. We can consider, for instance, the binding of β-CD to dendrimer **3**. Based on many studies of β-CD binding to simple ferrocene derivatives, we can assume that β-CD will approach the ferrocenyl dendrimer at a ϕ angle of *ca.* 90°, thereby interacting closely with the third generation dendrimer's lower periphery. In contrast, with the equivalent dansyl dendrimer **6**, β-CD approaches the guest moiety at an angle of $\phi \approx 0°$. Thus, the overall weak association in dansyl-β-CD systems (of the order of 10^2 M^{-1}) is most likely the determining factor for the rapid decrease in binding observed in these systems, regardless of the orientation of the host–guest binding, which would appear to favour easier binding in the system.

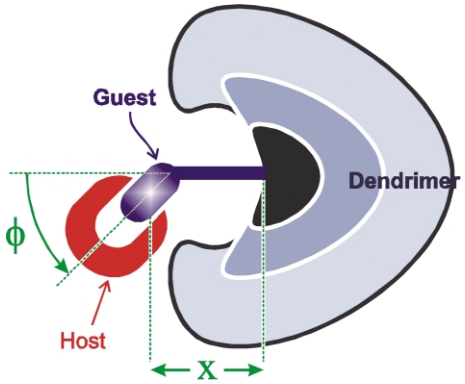


Fig. 6 Relevant parameters (*X*, ϕ) that may exert partial control on the extent of steric hindrance experienced by the host as it approaches the dendrimer-attached guest residue in order to form the host–guest complex.

Table 1 collects all of the binding constants measured with Newkome-type dendrimers containing a single guest residue at their apical positions. The table also shows the free energy changes (ΔG) for the corresponding complexation equilibria and their incremental changes ($\Delta\Delta G$) between dendrimer generations. The $\Delta\Delta G_{3-1}$ values provide an estimate of the overall change in free energy resulting from the growth of the dendron from 1st to 3rd generation. One would expect to see similar $\Delta\Delta G_{3-1}$ values in all cases if this parameter reflected only the energy of the dendritic component folding away from the guest residue. Clearly, the recorded values vary considerably from case to case and, remarkably, the largest $\Delta\Delta G_{3-1}$ values are observed for the cases in which β-CD is the host. This finding suggests that perhaps the hydrophobic interactions responsible for binding of ferrocenyl and dansyl residues by the β-CD host are more strongly affected by dendron growth than other interactions (ion–dipole, *etc.*) prevalent in binding by the CB7 host, as well as by the BPP34C10 crown in non-aqueous solution. It is noteworthy to mention here that the lowest $\Delta\Delta G_{3-1}$ value recorded in our experiments corresponds to the binding between the α-DA polyclonal antibody and the dansyl dendrimers. Unfortunately, the specific values of the parameters *X* and ϕ are not known in this case, because the detailed structure of the anti-dansyl antibody has not been elucidated. However, the binding affinity between this antibody and simple dansyl derivatives is clearly the highest one among all these host–guest pairs investigated in our group.

Based on these studies of single site guest dendrimers we can draw several conclusions about host–guest association in these systems. Dendrimer shape, flexibility and generation (size) are indeed important to determine the overall strength of the binding interactions. Tethering of the guest moiety to the dendrimer nucleus may improve the ease of binding, especially if the host must approach the guest from a relatively large angle ($\phi \gg 0°$) that could promote steric hindrance with the dendrimer periphery. However, while all of the foregoing points are salient, *the original strength of the association between the host–guest pair is the single most important factor in determining the overall level of molecular recognition that will remain, as the dendrimer grows from the first*

Table 1 Structural and thermodynamic parameters (measured at 25 °C) for the complexation in aqueous solution of Newkome-type, dendritic guests by various hosts

Guest	Generation	Host	ϕ^a	X^a/no. of C atoms	K/L mol^{-1}	ΔG/kcal mol^{-1}	$\Delta\Delta G_{n-(n-1)}$/ kcal mol^{-1}	$\Delta\Delta G_{3-1}$/ kcal mol^{-1}
1	1st	β-CD	*ca.* 90	0	950	−3.99		
2	2nd	β-CD	*ca.* 90	0	250	−3.21	0.78	
3	3rd	β-CD	*ca.* 90	0	50	−2.28	0.93	1.71
4	1st	β-CD	*ca.* 0	0	136	−2.86		
5	2nd	β-CD	*ca.* 0	0	< 1	−0.40	2.46	
6	3rd	β-CD	*ca.* 0	0	NB[c]	−0.0	0.40	2.86
4	1st	α-DA	?	0	4×10^6	−8.85		
5	2nd	α-DA	?	0	2×10^6	−8.45	0.40	
6	3rd	α-DA	?	0	1.5×10^6	−8.28	0.17	0.57
10a	1st	CB7	*ca.* 0	5	5.5×10^4	−6.35		
11a	2nd	CB7	*ca.* 0	5	5.7×10^4	−6.38	−0.03	
12a	3rd	CB7	*ca.* 0	5	1.3×10^4	−5.52	0.86	0.83
10b[b]	1st	**13**	*ca.* 0	5	374	−3.45		
11b[b]	2nd	**13**	*ca.* 0	5	210	−3.11	0.35	
12b[b]	3rd	**13**	*ca.* 0	5	92	−2.63	0.48	0.83

[a] Parameters defined in Fig. 6. [b] Values obtained in acetonitrile. [c] No binding detected.

to the third generation. While this is a seemingly obvious conclusion, the rationalization of the actual generation-to-generation changes in binding affinity, as observed in the various cases surveyed in this work, appears to be much more complicated. A factor that may also be important is the main type of intermolecular force holding the host–guest complex together and the effect that dendrimer growth may exert on the microenvironment polarity around the guest and on the magnitude of the relevant host–guest intermolecular interaction forces.

Multiple site dendrimer guests

The earliest work in the field of dendrimer guests involved multi-site systems, in which the guest moieties reside on the dendrimer surface. The group of Morán and Cuadrado first reported ferrocenyl-terminated diaminobutane (DAB) or poly(propyleneimine) (PPI) dendrimers with up to 64 terminal residues in 1994.[16] Dendrimers **17**–**19** (Fig. 7), with 4, 8 and 16 ferrocene peripheral

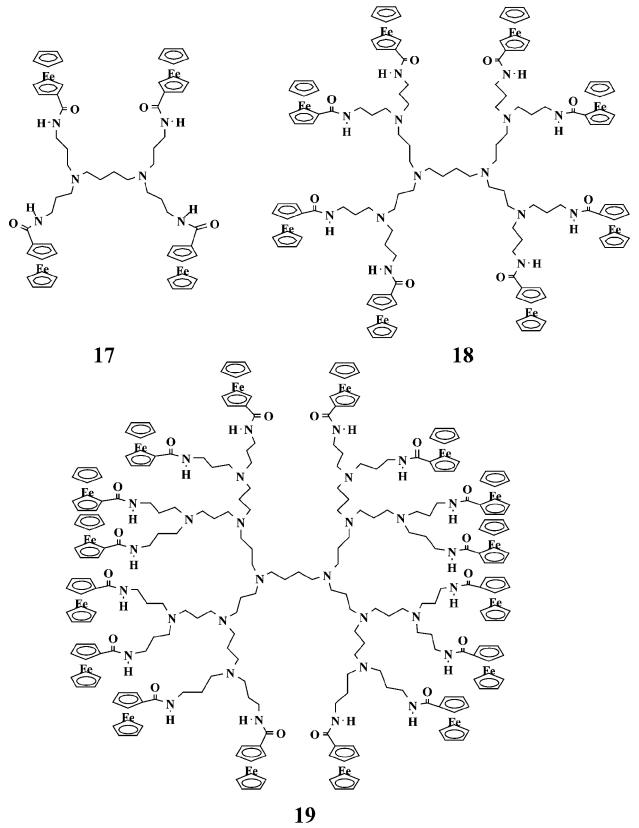


Fig. 7 DAB dendrimers containing multiple ferrocenyl sites on their peripheries.

residues, respectively, and their binding interactions with β-CD were studied in a fruitful collaboration with Morán, Cuadrado and co-workers.[17] In these systems, the limited solubility of dendrimers **17**–**19** required a less than ideal method of prolonged mixing of dendrimer-loaded CH_2Cl_2 and β-CD-containing aqueous solutions in order to effect the phase transfer of the dendrimers into the aqueous solution. Compound **19** showed the lowest solubility in β-CD-containing aqueous solution, suggesting that steric hindrance due to the increased proximity of ferrocene subunits prevents full complexation by 16 β-CD hosts. The voltammetric behaviour of these dendrimers in the presence of β-CD was considerably more complex than that observed in the single site case.

Dendrimers **17** and **18**, which can be fully bound with β-CDs, display a single voltammetric wave in the presence of excess host, indicating that all the ferrocene units are equivalent and undergo independent monoelectronic oxidations.[17] This would indicate that, in the presence of β-CD, all the ferrocene units are bound. In contrast, dendrimer **19** exhibited two waves in the presence of excess β-CD, the first at a less positive potential (+0.38 V *vs.* SCE) corresponding to uncomplexed ferrocene residues, while the second, at a more positive potential (+0.51 V *vs.* SCE), corresponds to the β-CD-complexed ferrocenes. Addition of β-CD to the solution did not drive further binding, once again suggesting that steric congestion limits the number of ferrocene residues which can be complexed by the bulky CD hosts. Conceptually, the dendrimer framework provided a three-dimensional template for organising the β-CD receptors, while the reversible electrochemical oxidation of the ferrocene units affords a mechanism for tempering the binding affinity with the CDs. A similar concept was employed the following year with a similar series of DAB dendrimers containing 4, 8 and 16 cobaltocenium units. This organometallic subunit is bound by β-CD upon one-electron reduction from cobaltocenium to cobaltocene.[18] In this scenario, the dendrimers undergo peripheral complexation by β-CD hosts upon 'electrochemical activation' of the cobaltocenium units. Furthermore, clear electrochemical detection of the extent of β-CD association is evidenced by the dramatic changes in the shape of the cyclic voltammograms observed for these dendrimers.[19]

Reinhoudt and co-workers have reported a related investigation with adamantyl-terminated DAB dendrimers, whose structures are shown in Fig. 8.[20] The authors prepared generations 1 to 5, and the

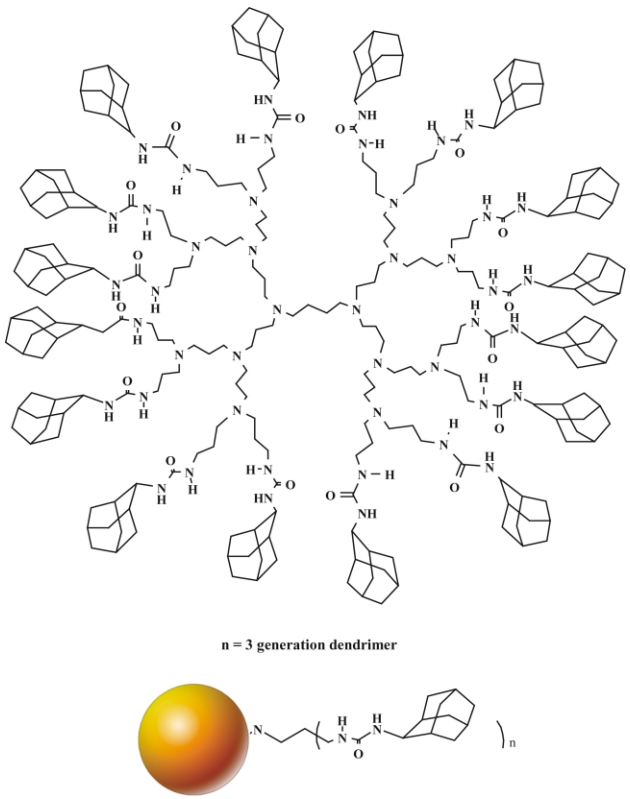


Fig. 8 Reinhoudt's adamantyl dendrimers. In the scheme, *n* is the generation number and the number of peripheral adamantyl residues is given by the expression 2^{n+1}.

16-residue, third generation dendrimer is shown as an example. They achieved full complexation with β-CD, up to the $n = 4$ (32 residue) level, after protonating the DAB dendrimer framework in order to achieve a more rigid and extended framework that permits better β-CD association. (We should note that the ferrocenyl dendrimers **17**–**19** cannot be dissolved in strongly acidic solutions, since they would likely be oxidised under such conditions.) Reinhoudt's adamantyl dendrimers were quite insoluble even in acidic aqueous solution; however, subsequent complexation with β-CD vastly improved the solubility of the systems, allowing the solutions to be adjusted to neutral pH. This finding clearly

illustrates the issue of dendrimer shape on host binding. The more collapsed state of the dendrimer (*i.e.* unprotonated) does not permit full binding of the CD hosts. The authors' finding of *sterically induced stoichiometry* is clearly limited by the growth of the dendrimer and not the result of weakened cooperative association.[20] The uncomplexed adamantyl groups in the fifth generation (n = 64 residue) dendrimer were not found to form a hydrophobic surface on the dendrimer, *i.e.* the cyclodextrins fully cover the periphery of the dendrimer even if all adamantyl groups are not bound The authors observed aggregation due to partial collapse of the dendrimer structure at neutral pH and above that pH precipitation ensued. Kimoon Kim and co-workers have examined a closely related dendrimer system with protonated terminal diaminobutyl groups and cucurbit[6]uril, obtaining results similar to those of Reinhoudt, through the fourth generation DAB dendrimer.[21]

Several groups have explored the idea of biological applications of complex supramolecular systems based on dendrimers. Haensler, Szoka, and Tomalia were among the first[22] to employ dendrimers to form a compact association with DNA, utilising poly(amidoamine) (PAMAM) dendrimers.[22] In the arena of guest dendrimers, the group of Kimoon Kim has employed a ternary system based on the cucurbit[6]uril/DAB dendrimer complex mentioned above, and plasmid DNA molecule coding for firefly luciferase.[23] The system forms what can be termed a 'self-assembled gene carrier' based on the electrostatic association of the negatively charged DNA and the positively charged CB6-dendrimer complex, a method likened to the self-assembly of virus particles.[24] The authors found that although the DNA binding capacity of the dendrimers decreased as the generation increased, with the poly(propyleneimine) (PPI) peripheral moiety showing higher binding than either a diaminobutane (DAB) outer layer or a DAB-CB6 outer layer, the transfection efficiency of the gene carrier improved with generation increase. Rigidification and formation of a more compact shell at the exterior were postulated as reasons for the lower efficacy of binding in the ternary CB6-DAB system, though fortuitously, the transfection with the ternary system employing a fifth generation DAB dendrimer achieved better efficiency than did the same generation PPI dendrimer lacking the DAB termini and CB6 complex. The authors were planning to functionalize the CB6 in order to ligate it to peptides thereby making a three-component gene delivery system with directed function, a clear step in the direction of selective delivery of gene therapy.

Guest dendrimers have also found uses recently in the group of Hak-Sung Kim, who has created self-assembled monolayers (SAMs) of ferrocenyl/biotin-terminated PAMAM dendrimers.[25] After capping approximately 30% of the dendrimer termini with ferrocenyl groups, the dendrimers were attached to a SAM of mercaptoundecanoic acid on a gold electrode surface *via* amide linkages. The remaining free amino termini were then capped with biotin, creating a double functionalized dendritic monolayer. The dendrimer's ferrocene residues were then employed as mediators in the electron transfer reaction of glucose oxidase, which bears an FAD residue. When avidin or anti-biotin antibodies are bound to the dendritic monolayer the mediation response of the ferrocene is blocked, thereby indicating the presence of affinity binding to the biotin residues on the dendrimer monolayer. A complex of either type can be reversibly dissociated by competitive introduction of free solution biotin, which can then be washed away from the monolayer system. Problems due to the adsorption of glucose oxidase were encountered, requiring the addition of ferrocenyl-capped dendrimers to the electrolyte solution. Thus, this system, while intriguing, still poses some challenges but is another firm step in the direction of practical application of guest dendrimers.

In conclusion, we have presented a brief summary of the host–guest chemistry of guest dendrimers. The binding interactions of dendrimers containing a single guest residue with free hosts, ranging from cyclodextrins to antibodies, are generally attenuated by the growth of the guest's dendritic component. A detailed quantitative understanding of the generational changes observed is not available yet. However, the geometry of the host–guest interaction, as well as the strength and nature of the intermolecular forces responsible for binding, have been identified as relevant parameters affecting the host–guest phenomena. More work is necessary to improve our quantitative understanding of these effects. Dendrimers with multiple guests residues in their peripheries are also of great interest although the quantitation of binding affinities is more complicated due to the multiple binding sites per dendrimer and the possible effects (negative or positive) between proximate guest residues. The biological applications of complex supramolecular systems including dendrimers are very promising and provide additional support to continued research work in this area.

The authors are grateful to the National Science Foundation for the generous support of this research work. Fruitful collaborations with the groups of Frank Bright (SUNY-Buffalo) and Carlos Peinador (Universidade da Coruña, Spain) are also gratefully acknowledged. Special thanks to Beatriz Alonso, Carmen María Casado, Isabel Cuadrado, Blanca González and everyone else in the group of the late Professor Moisés Morán at Universidad Autónoma de Madrid, Spain.

Notes and references

1 P. J. Gittins and L. J. Twyman, *Supramol. Chem.*, 2003, **15**, 5; J. Fréchet, *Proc. Natl. Acad. Sci. USA*, 2002, **99**, 4782; A. H. Bosman, H. M. Janssen and E. W. Meijer, *Chem. Rev.*, 1999, **99**, 1665; G. R. Newkome, E. He and C. N. Moorefield, *Chem. Rev.*, 1999, **99**, 1689; M. Fischer and F. Vögtle, *Angew. Chem., Int. Ed.*, 1999, **38**, 884.
2 J. F. G. A. Jansen, E. M. M. De Brabander-van den Berg and E. W. Meijer, *Science*, 1994, **266**, 1226.
3 F. Diederich and B. Felber, *Proc. Natl. Acad. Sci. USA*, 2002, **99**, 4778.
4 S. C. Zimmerman, M. S. Wendland, N. A. Rakow, I. Zharov and K. S. Suslick, *Nature*, 2002, **418**, 399.
5 M. M. Davis, M. Krogsgaard, J. B. Huppa, C. Sumen, M. A. Purbhoo, D. J. Irvine, L. C. Wu and L. Ehrlich, *Annu. Rev. Biochem.*, 2003, **72**, 717.
6 See, for example: Y. Tomoyose, D.-L. Jiang, R.-H. Jin, T. Aida, T. Yamashita and K. Horie, *Macromolecules*, 1996, **29**, 5236; G. R. Newkome, R. Guther, C. N. Moorefield, F. Cardullo, L. Echegoyen, E. Perez-Cordero and H. Luftman, *Angew. Chem., Int. Ed. Engl.*, 1995, **34**, 2023.
7 See, for example: A. M. Elizarov, S.-H. Chiu, P. T. Glink and J. F. Stoddart, *Org. Lett.*, 2002, **4**, 679.
8 C. M. Cardona, T. D. McCarley and A. E. Kaifer, *J. Org. Chem.*, 2000, **65**, 1857; C. M. Cardona, J. Alvarez, A. E. Kaifer, T. D. McCarley, S. Pandey, G. A. Baker, N. J. Bonzagni and F. V. Bright, *J. Am. Chem. Soc.*, 2000, **122**, 6139.
9 Similar experimental results have been reported for a first generation ferrocenyl glycodendrimer by: P. R. Ashton, V. Balzani, M. Clemente-León, B. Colonna, A. Credi, N. Jayaraman, F. Raymo, J. F. Stoddart and M. Venturi, *Chem. Eur. J.*, 2002, **8**, 673.
10 H.-J. Kim, W. S. Jeon, Y. H. Ko and K. Kim, *Proc. Natl. Acad. Sci. USA*, 2002, **99**, 5007.
11 W. Ong and A. E. Kaifer, *Org. Lett.*, 2002, **4**, 1791.
12 W. Ong and A. E. Kaifer, *Angew. Chem., Int. Ed.*, 2003, **42**, 2164.
13 J. C. Grindstaff, *Peripheral Modification and Binding Studies of Unsymmetric Viologen Dendimers*, M.S. Thesis, University of Miami, Coral Gables, FL, 2003.
14 J. Grindstaff, W. Ong, R. Toba, J. M. Quintela, C Peinador and A. E. Kaifer, *J. Am. Chem. Soc.*, submitted for publication.
15 R. Toba, J. M. Quintela, C. Peinador, E. Román and A. E. Kaifer, *Chem. Commun.*, 2001, 857; R. Toba, J. M. Quintela, C. Peinador, E. Román and A. E. Kaifer, *Chem. Commun.*, 2002, 1768.
16 B. Alonso, I. Cuadrado, M. Morán and J. Losada, *J. Chem. Soc., Chem. Commun.*, 1994, 2575.
17 R. Castro, I. Cuadrado, B. Alonso , C. M. Casado, M. Morán and A. E. Kaifer, *J. Am. Chem. Soc.*, 1997, **119**, 5760.
18 Y. Wang, S. Mendoza and A. E. Kaifer, *Inorg. Chem.*, 1998, **37**, 317.

19 B. Gonzalez, I. Cuadrado, B. Alonso, C. M. Casado, M. Morán, Y. Wang and A. E. Kaifer, *Chem. Commun.*, 1998, 2569.
20 J. J. Michaels, M. W. P. L. Baars, E. W. Meijer, J. Huskens and D. N. Reinhoudt, *J. Chem. Soc., Perkin Trans. 2*, 2000, 1914.
21 J. W. Lee, Y. H. Ko, S.-H. Park, K. Yamaguchi and K. Kim, *Angew. Chem., Int. Ed.*, 2000, **40**, 746.
22 J. F. Kukowska-Latallo, A. U. Bielinska, J. Johnson, R. Spindler, D. A. Tomalia and J. R. Baker, *Proc. Natl. Acad. Sci. USA*, 1996, **93**, 4897; M. X. Tang, C. T. Redemann and F. C. Szoka, *Bioconjugate Chem.*, 1996, **7**, 703; J. Haensler and F. C. Szoka, *Bioconjugate Chem.*, 1993, **4**, 372.
23 Y.-b. Lim, T. Kim, J. W. Lee, S.-M. Kim, H.-J. Kim and K. Kim, *Bioconjugate Chem.*, 2002, **13**, 1181.
24 For a thorough review of the use of dendrimers as synthetic gene vectors, see: J. Dennig and E. Duncan, *Rev. Mol. Biotechnol.*, 2002, **90**, 339.
25 H. C. Yoon, D. Lee and H.-S. Kim, *Anal. Chim. Acta*, 2002, **456**, 209.