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Dendritic Macromolecules at the Interface of Nanoscience and Nanotechnology

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Summary: As a result of their unique architecture and structural as well as functional versatility, dendrimers have generated considerable interest in numerous areas of the physical sciences, engineering, as well as the biological sciences. Both their size - in the 1-10 nm range – and their globular shape resemble those of many proteins suggesting a host of biomimetic and nanotechnological applications. This brief highlight describes some of our recent work with nascent applications of dendrimers as unimolecular nanoreactors, as nanoscale antennae for energy harvesting and transduction, and as nanosized carriers for diagnostic or therapeutic applications. While implementation of some of these applications may still be distant, the impatient critic might remember that new markets are not created overnight as demonstrated by the slow commercial acceptance of many promising molecules and technologies with development frequently extending decades after their initial discovery.

Keywords: catalysis, dendrimers, light-harvesting, nanotechnology, therapeutics

1 Shape, Flexibility and Molecular Ordering of Dendrimers

Dendrimers are size monodisperse, globular macromolecules in which all bonds emerge radially from a central focal point or core with a regular branching pattern and with repeat units that each constitute a branch point.[1-3] Numerous macromolecules possess some of the features of dendrimers, including high degree of branching and multiplicity of chain-ends and reactive sites. These include hyperbranched polymers – both natural (e.g. polysaccharides) and synthetic – as well as hybrid dendritic-linear polymers, dendronized polymers, comb-burst polymers, etc.[2,3] Despite unsupported claims to the contrary, none of these dendritic macromolecules match the ultimate properties of dendrimers. Even with true dendrimers, properties of the dendritic state,[3] such as core encapsulation[4] and unusual intrinsic viscosity behavior in solution,[5] are only reached when globularity is achieved at a certain generation or size

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threshold. The flexibility of dendrimers depend greatly on the generation or number of layers of repeat units, the choice of monomer repeat unit, including the number of branches, the number and type of bonds between branches, as well as the degrees of freedom available to interbranch bonds. In general, low generation dendrimers are quite flexible while high generation dendrimers may become rigid.[7]

A controversy concerning the shape of dendrimers and the placement of their chain ends either at the "periphery" of the globular macromolecule or back folded within its building blocks had arisen in recent years. This was fueled by a variety of calculations and measurements suggesting either back folding of the chain ends or supporting their peripheral arrangement[6]. In reality, free-energy rules and dendrimers react to their environment (i.e. adjust their shape and the placement of their functional groups) in order to minimize their free energy.[7] Thus a dendrimer might adopt a fully extended conformation and reach a volumetric maximum and an almost spherical shape in a good solvent while collapsing to a more compact volume in another or in the absence of a solvent with its final shape, chain-end placement, and other structural features determined by its intrinsic flexibility, the interactions of its various components (core, internal repeat units, chain-ends) and the interactions with its near neighbors or a surface. Similarly, the location of chain-ends (peripheral or back folded) in all but the most rigid structures is dictated by free energy.[7] If the chains ends possess favorable interactions, such as H-bonding or π-stacking, with the inner building blocks, back folding may be expected to occur, a phenomenon that may be exacerbated or mitigated by solvent. If the absence of favorable enthalpic contributions, entropy considerations usually disfavor the mixing of chain ends with dissimilar inner building blocks.[8] The existence of a "cavity" within dendrimers has been another topic of controversy. The structure of a dendrimer bears some analogy to that of a micelle although the dendrimer is a static unimolecular covalent assembly while the micelle is a dynamic supramolecular assembly. Like micelles, larger dendrimers possess a volume and structural features that enable them to accommodate guest molecules,[9-10] particularly when enlarged by solvation with a good solvent. However, with the possible exception of very specialized structures such as shell-crosslinked dendrimers, dendrimers do not possess a permanent and rigid cavity. Small guests that can penetrate the volume of a dendrimers as a result of favorable enthalpic interactions may remain encapsulated following collapse of a solvated structure.

Encapsulation may become permanent as in Meijer's "dendritic box"[11] if the peripheral density of the dendritic structure is increased to rigidify the macromolecule while guest molecules are located within the extended volume of a dendrimers.

2 Designing Free-energy Driven Unimolecular Nanoreactors

Catalysis with highly branched macromolecules, primarily dendrimers, has been well documented in the literature.[12] Depending on the location of the catalytic site(s), dendritic catalysts with either multiple active sites located at chain-ends or constituting a catalytically active core have been described.[12] The first approach offers the advantage of relatively high catalyst loading, however, the dendritic backbone merely serves as a support and its favorable structural and functional features cannot be completely utilized. In contrast, the second approach enables encapsulation of the active site,[4] thereby offering protection from unfavorable deactivation mechanisms. Moreover, the placement of catalytic moieties at the core or the interior of a globular dendritic structure allows for the fine-tuning of overall molecular properties and catalytic activity by modification of the periphery and the interior environment, respectively. Early work by Moore and Suslick[13] explored dendritic Mn-porphyrins that exhibited improved stability in solution compared to the free metalloporphyrin as a result of encapsulation by the surrounding polymer. The dendritic catalyst also appeared to provide increased regioselectivity in simple epoxidation reactions using various dienes as substrates. The greater significance of this early study, however, was a clear confirmation of previous findings that small molecules could penetrate the congested exterior of the dendritic scaffold for reaction[4a] at the core. Since then, several groups have investigated the effect of the dendritic superstructure on the catalytic activity of encapsulated sites. In most cases few advantages could be associated with the dendritic character of the new catalytic moieties.[14,15] A critical reading of some recently published work suggests that mass transport within the dendritic shell may well be a significant factor and that product inhibition could be responsible for the relatively limited performance of several systems.[15]

In view of our reading of these previous findings, our approach to dendrimers catalysis has included a global design of not only the catalytic site itself, but also its environment to incorporate appropriate mass transport "molecular machinery". Thus the dendrimers should not only provide a reaction center, but it should also function, much like an enzyme, as a unimolecular nanometer size "reactor" transporting and concentrating substrate to the environment that promotes reaction, and removing product from the catalytic site as soon as it is produced. In recent work[16,17] we have demonstrated this concept and shown that by utilizing the chemical nature and structural features of the dendritic building blocks, tailored microenvironments can be generated that assist in transporting substrate and product, as well as in stabilizing crucial intermediates and transition states. The key concept makes use of contrasting polarity between the dendritic inner and outer environments coupled with a polarity difference occurring during the course of the catalyzed chemical transformation. The amphiphilic design leads to preferential accumulation of substrates and, in some instances, stabilization of transition states or intermediates in the interior, while the product is simultaneously expelled into the external medium thereby preventing inhibition. An added feature of this free energy-driven mass transport is the fast kinetics that result from the high local concentration of substrate that can be achieved at the site of catalysis within the dendrimer.

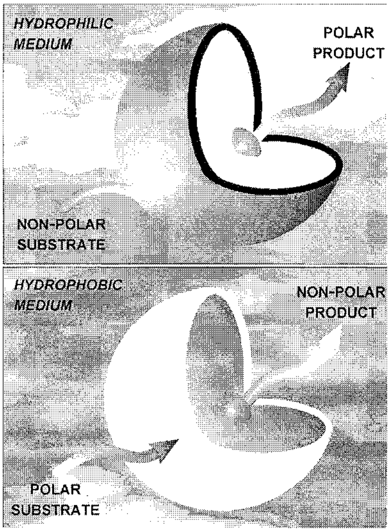

Fig. 1. Design of free-energy based catalytic nanoreactor.

With this key concept in mind, it is possible to delineate criteria pertinent to the general design of efficient polymeric catalysts. Irrespective of the catalytic site(s) to be incorporated into the polymer, the primary consideration for molecular design depends on the intrinsic polarity change associated with the specific transformation. Depending on the relative polarity of the product compared to the substrate, two types of catalysts can be envisioned (Figure 1). In the case of a polarity increase from substrate to product, the reaction will be performed in a more hydrophilic medium utilizing a regular micelle-like but unimolecular, shape-persistent dendrimer. This design is particularly attractive since many catalytic transformations, such as oxidations, involve polarity increases and catalysis in water is of great importance for the development of environmentally friendly chemical processes. In a similar manner, if polarity decreases during the course of the reaction, one would employ a reverse unimolecular micelle in a hydrophobic solvent.

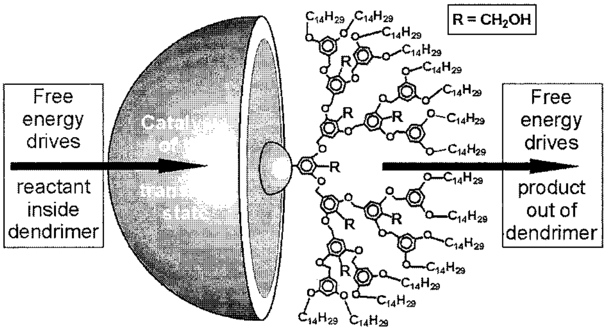

Fig. 2. Transition state catalyst for unimolecular elimination.

As proof of concept, a rather unsophisticated, reverse micelle-like dendrimer (Figure 2) was designed to catalyze an E1-type elimination process.[16] The catalyst consists of a hydrophilic interior that favors a low energy transition state for the carbenium intermediate while also providing for preferential entry of the substrate. Peripheral alkyl chains provide a hydrophobic exterior that offers solubility in non-polar solvents, such as hexanes. In E1-type elimination reactions involving tertiary alkyl halides, high turnover numbers (17400) were observed leading to almost quantitative conversions with very low catalyst loading (<0.01 mol %). Presumably, the slightly polar alkyl halide is drawn and concentrated into the polar core, where formation of the cationic intermediate and subsequent elimination occur to yield the non-polar alkene that is driven from the core to the non-polar corona and then to surrounding solvent. Solid $NaHCO_3$ present in the hexane phase serves as an acid acceptor. In this manner, the free energy of the system can minimized at each event and high efficiencies can be obtained.

The generality of the amphiphilic design concept has also been demonstrated and applied by Hecht[17] to excited state catalysis. Using an amphiphilic dendritic photocatalyst (Figure 3), Hecht was able to affect a [4+2]cycloaddition between singlet oxygen ($^1O_2$) and cyclopentadiene (CP). The cycloadduct is further reduced *in situ* to the allylic diol in the presence of thiourea. The large polarity increase associated with the overall transformation dictated our choice for the relative polarities of the inner and outer compartments. By encapsulating a $^1O_2$-sensitizing benzophenone core into a globular dendrimer having a hydrophobic interior and hydrophilic surface, it was possible to demonstrate the effect of dendrimer size on the performance of this nanoscale photoreactor. Since singlet oxygen lifetimes are greatest in nonpolar environments, increasing the generation of the dendrimer, led to the expected increase in CP conversion. A noteworthy feature of this system is that the bimolecular reaction benefits from both the high internal substrate concentration and the enhanced lifetime of singlet oxygen in the hydrophobic core environment.

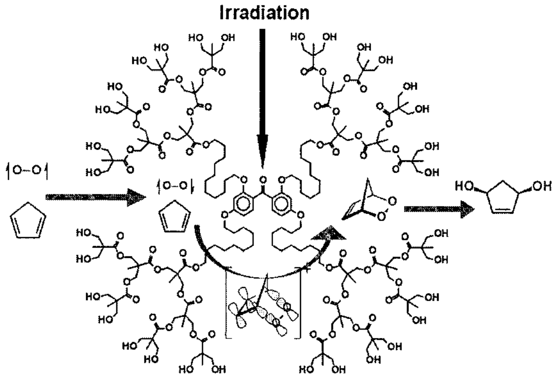

Fig. 3. Photocatalyst for bimolecular cycloaddition.

Although this area of research is still in its infancy, these initial findings are encouraging and demonstrate the importance of the amphiphilic motif for rational catalyst design. We are currently extending this theme exploring a variety of reactions with different polymer architectures to gain further insight into important aspects regarding the possible roles of the backbone and of the cooperativity of multiple catalytic sites. Furthermore, a strategy that combines light harvesting with photocatalysis is also under active investigation as a step towards more evolved systems that begin to mimic the complexity and combination of function found in photosynthesis.

3 Dendrimers as Nanoscale Light Harvesting Antennae

The very structure of dendrimers, with a radially emanating layered arrangement of functionalities around a central core, makes them uniquely suited for applications involving energy harvesting and conversion.[18] Thus, a molecule may be designed that incorporates multiple peripheral dyes used to harvest energy and capable of electronic communication with a functional core where the concentrated energy is "reprocessed". For example, harvested broadband radiation may be up- or down-converted into mono-chromatic light, or transformed into electrical or chemical energy. Thus, dipole dipole interactions between chromophore may be used to effect Förster energy transfer between an array of terminal donor chromophores and a core acceptor dye, which may also be excited independently of the periphery. Since emission is observed from the core only, the system serves as a spatial and spectral energy concentrator or "molecular lens" (Figure 4).

Essentially, this mimics the primary events in photosynthesis, where the light-harvesting complex funnels its excitation energy to the special pair leading to subsequent charge separation. Two types of systems, which either use the dendritic architecture solely as a scaffold,[19,20] or involve the dendrimer backbone in the energy transfer event,[21-23] have been explored. Our group has shown that an amplification of the core acceptor emission may be achieved in high generation dendrons labeled with multiple peripheral donor chromophores.[20] The amplification effect, misunderstood by some, has its origin in the enhanced donor absorption cross-section and the extremely fast rate of through-space energy transfer from the peripheral chromophores to the core, therefore giving rise to efficient light harvesting.[20] As for many other common forms of "amplification", this amplification is simply a "reprocessing" of energy that, in this case, enables the core chromophore to emit more light (including also energy transferred from the multiple peripheral dyes) that it possibly could solely via its direct excitation. In a key finding, Moore et al. demonstrated that a significant acceleration of energy transfer could be achieved within dendrimers having an internal energy gradient, resulting from a stepwise decrease of the HOMO-LUMO gaps of the branching units when progressing toward the acceptor.[22] Balzani et al. have constructed bipyridine-based polynuclear metal complexes capable of controlling the direction of energy transfer via alteration of the excited state energies by introducing appropriate metals.[23] This strategy impressively demonstrated how supramolecular chemistry could be used to assemble multiple chromophores while controlling their relative orientation. The performance benefit of the dendritic architecture over that of a linear polymer has also been demonstrated recently.[24]

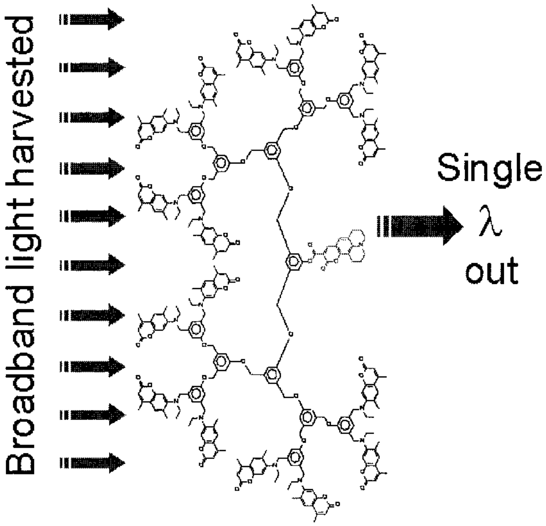

Fig. 4. Dendritic light harvesting antenna.

Recent work from Müllen et al., and from our own laboratory, has shown that cascade energy transfer involving a multiplicity of chromophores is possible.[25] Similarly, we have used two photon absorbing chromophores to effect up-conversion of energy using dendritic antennae.[26]

4 The Potential of Dendrimers in Macromolecular Therapeutics

The efficient and targeted delivery of therapeutic agents is one of the great challenges of today's medicine. In particular, macromolecules may be used to modify the bioavailability and pharmacokinetics of known drugs as exemplified by Schering-Plough's PEG-Intron™, in which a short (MW = 12,000 D) strand of monomethoxy poly(ethylene glycol) [PEG] is attached to Interferon α-2b, a water-soluble protein (MW = 19,271D) produced by recombinant DNA techniques. The conjugate, a potent drug for the treatment of hepatitis C, vastly outperforms the free Interferon α-2b also used to treat the same disease. The conjugation of drugs to PEG ("PEGylation") is now studied extensively as a means of improving the performance of known as well as new drugs. However, with low molecular weight drugs, the conjugation of one or even two molecule of drug to one molecule of monomethoxy-PEG or PEG to form a prodrug leads to issues of low molar concentrations of active or high viscosity of the conjugate solution.[27]

Linear polymers may be used for the conjugation of multiple copies of a drug onto a single polymer chain as exemplified by Kopečeck and Duncan's polymeric drug conjugates based on poly(N-2-hydroxypropyl-methacrylamide) [HPMA].[28] While linear polymers such as HPMA show great promise, dendritic structures offer several advantages including lower polydispersity, better accessibility of reactive sites, better control over their number, better defined nanometer size, more compact shape, lower solution viscosity, and better ability to interact with receptor sites in multivalent fashion, etc.[29] Our program in the development of delivery vehicles for therapeutic agents has included both micro- and nanoparticulates based on pH-degradable polymers[30] for the delivery of vaccines and genes, and dendritic macromolecules for the conjugation and delivery of drugs.[31] General design concepts include structural features affording low toxicity water-soluble carriers with high drug loading capacities as exemplified by our linear-dendritic hybrids based on aliphatic polyester dendrons.[31] In early studies, targeting to tumor cells was based on Maeda's Enhanced Permeation and Retention[32] effect, which mandates relatively long plasma residence times, and thus requires optimization of the size of the dendritic-drug conjugate. Release of the free drug from the multivalent conjugates is based on the low pH that prevails in tumor tissue and the use of acid-labile linkages for attachment of the drug molecules to the dendritic carrier. In early work,[31,33] we have evaluated a number of different dendritic architectures based on aliphatic polyester dendrons for their suitability as drug carriers both *in vitro* and *in vivo*. These water soluble and non-toxic systems, can be used to conjugate potent anticancer drugs such as doxorubicin via acid-labile hydrazone linkages (Figure 5). Attachment of the drug to the dendritic carrier reduced its cytotoxicity and biodistribution experiments showed little accumulation of the DOX-polymer conjugate in vital organs while the serum half-life of the doxorubicin conjugate was significantly higher than that of the free drug. Thus, this new nanoscale drug carrier system exhibits promising characteristics for the development of new polymeric drugs.[31]

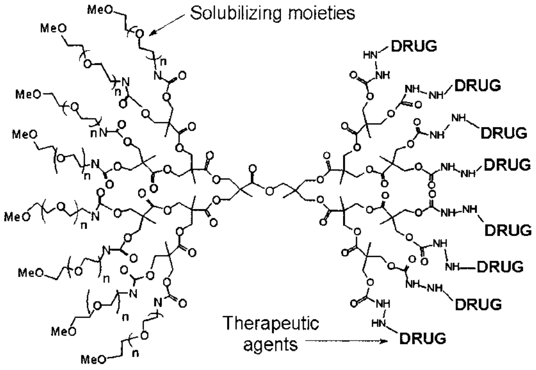

Fig. 5. Dendritic drug delivery system (from ref. 33).

5 Conclusion

It is clear that dendrimers have much to offer in numerous areas of the broad fields of nanoscience and nanotechnology. With a size typically ranging from 2-10nm, quite comparable to that of many proteins, dendrimers are large enough to allow significant tailoring of their structural features, physical properties, as well as surface and inner chemistry, yet they are small enough to allow their easy characterization and their use as mimics of many important biological macromolecules. There is little doubt that molecules possessing dendritic features and polyvalent character will find use in high added value applications ranging from nanotechnology to medicine. Whether these will be true dendrimers, dendritic hybrid, or even hyperbranched structures, really does not matter but the body of knowledge acquired through the fundamental study of structure-property-function relationships of dendrimers with precise and well-characterized structures will surely be of great value in the development of numerous classes of highly branched functional macromolecules with designs optimized for specific applications.

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