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A molecular dynamics study of the structure and inter-particle interactions of polyethylene glycol-conjugated PAMAM dendrimers

Hwankyu Lee^{1,*} and Ronald G. Larson²

¹Laboratory of Computational Biology, National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, MD 20892, USA

²Department of Chemical Engineering, Biomedical Engineering, Mechanical Engineering, and Macromolecular Science and Engineering Program, University of Michigan, Ann Arbor, MI 48109, USA

Abstract

We performed molecular dynamics (MD) simulations of one or two copies of polyethylene glycol of molecular weight 550 (PEG550) and 5000 (PEG5000) Daltons, conjugated to generation 3 (G3) to 5 (G5) polyamidoamine (PAMAM) dendrimers with explicit water using a coarse-grained model. We found the radii of gyration of these dendrimer-PEG molecules to be close to those measured in experiments by Hedden and Bauer (*Macromolecules* **2003**, 36, 1829). Densely grafted PEG ligands (>50% of the dendrimer surface) extend like brushes, with layer thickness in agreement with theory for starlike polymers. Two dendrimer-PEG complexes in the box drift away from each other, indicating that no aggregation is induced by either short or long PEG chains, conflicting with a recent view that the cytotoxicity of some PEGylated particles might be due to particle aggregation for long PEG lengths.

INTRODUCTION

Polyamidoamine (PAMAM) dendrimers, which are regular hyper-branched polymers with many surface terminal groups, have been widely studied for biomedical applications such as drug delivery and antitumor therapeutics because of their controlled mass, uniform structure, surface functionality, and good water solubility.^{1–6} Drugs and imaging molecules can be attached to dendrimers, and these complexes have been successfully targeted to specific cancer cells *in vitro* and *in vivo*. Also, many experiments using atomic force microscopy, neutron scattering, and NMR have shown that the mechanism and efficiency of targeting can be engineered through attachment of ligands to the surfaces of dendrimers.^{7,8} In particular, the group of Banaszak Holl, Orr, Baker, and co-workers have shown that cationic amine terminals of the dendrimer induce nonspecific binding and cause pore formation in membranes, which can lead to internalization of the dendrimers and increased cytotoxicity.^{9,10} To reduce the toxicity of the dendrimer, the dendrimer surface must be made electrically neutral. The above group also showed that after acetylation of the terminal groups, these dendrimers do not induce pore formation in membranes, and show much less toxicity.^{9–11} While acetylation can improve the solubility of dendrimer, attachment of polyethylene glycol (PEG) or polyethylene oxide (PEO), which is called PEGylation, can do so to a greater degree.¹² Also, PEGylation can

*Corresponding author leeh3@nhlbi.nih.gov.

sterically shield drug molecules, increasing their circulation lifetime in the bloodstream. Hence, PEGylation has been widely used for many drug molecules.^{13,14}

PEGylated dendrimers have shown increased water solubility, extended periods of release of drug molecules, and have been successfully used as nonviral gene vectors targeting brain, breast and prostate cancers.^{15–23} However, experiments showing these potential applications have used different lengths of PEG chains and dendrimer sizes, which can affect the encapsulation efficiency.²⁴ Kim et al. showed that ligating only 25% or less of the terminal groups on the dendrimer with PEG550 chains (here, “550” means the molecular weight of the PEG) may reduce cytotoxicity of the dendrimer, while retaining good water solubility.¹⁶ However, longer PEG2000, more densely conjugated to the dendrimer surface, increases cytotoxicity at high dendrimer concentration. It was suggested that this change in cytotoxic behavior at increased PEG length was caused by intermolecular aggregation, although no direct evidence for aggregation was provided. Yang et al. also suggested agglomeration of dendrimers conjugated with PEG5000 chains.²⁵ However, other experiments with different nanoparticles have shown that PEG attached to the nanoparticle surface can significantly reduce the extent of aggregation.^{26,27} These different experimental observations might be due to different properties of various nanoparticles, but the effect of PEG conjugation on interactions between nanoparticles has not been resolved.

Atomistic and coarse-grained (CG) molecular dynamics (MD) simulations of PAMAM dendrimers have revealed the size, shape, internal structure, dynamics, and thermodynamic properties of dendrimers.^{28–33} More complex systems, including the interactions of dendrimers with lipid bilayers,^{34–38} with self-assembled monolayers (SAMs),^{39,40} and with polyelectrolytes, including DNA,^{41–46} have been studied using both atomistic and CG simulations. These studies have explored dendrimer-induced pore formation in lipid bilayers and multivalent binding of dendrimers with the surfaces of SAMs, among other topics. In particular, simulations of dendrimers with polyelectrolytes or DNA have shown that the linear chains grafted onto a charged dendrimer surface can wrap around the dendrimer if there are enough opposite charges on the linear chain. Although interactions between dendrimers and linear polyelectrolytes have been widely studied, the interactions between dendrimers and PEG chains have not been simulated and are less understood.

In this work, we perform MD simulations of PEG-conjugated dendrimers using the CG dendrimers^{36,47} and PEG⁴⁸ force fields parameterized in our previous work. To verify our CG models and understand the conformation of the PEG chains grafted onto the dendrimer surface, we simulate a dendrimer-PEG complex with different PEG lengths, grafting densities, and particle concentrations in water. Radii of gyration of the dendrimer-PEG are compared with experimental values measured for the same length and PEG conjugation density.⁴⁹ Also, thicknesses of the PEG layers are compared with a theory for the conformation of star-shaped polymers.⁵⁰ In addition to simulations of a single dendrimer-PEG, two molecules of dendrimer-PEG are also simulated in a single simulation box to understand the effects of the PEG length on the inter-particle interaction. Distances between two dendrimers, diffusion coefficients of the center of mass of each dendrimer-PEG, and radial distribution functions of PEG monomers on two different dendrimer-PEG complexes are computed.

METHODS

All simulations and analyses were performed with the GROMACS4.0.3 simulation package^{51,52} with the coarse-grained (CG) force field, “MARTINI”, developed by Marrink et al.^{53,54} We previously developed CG force fields for PAMAM dendrimers^{36,47} and polyethylene oxide (PEO)⁴⁸ within the framework of the MARTINI force field. For the CG dendrimer, chemical moieties were mapped onto four different CG bead types based on their

charges and hydrogen bonding properties. The CG dendrimer model showed radii of gyration of acetylated and un-acetylated G3 to G7 dendrimers that were in agreement with experimental values.^{36,47} For the CG PEO, the monomer unit (C-O-C) was modeled as a CG bead. For PEO-PEO and PEO-water interactions, a SNda type ($\sigma_{ij} = 4.3$ and $\epsilon_{ij} = 3.375$) was used, while, for interactions of PEO with other molecules, a SNa type was used, where the interactions of SNa with other bead types are already built into the MARTINI force field. PEO beads are connected by a weak harmonic potential $V_{\text{bond}}(R)$ with an equilibrium distance

$R_{\text{bond}} = 0.33 \text{ nm}$ ($V_{\text{bond}}(R) = \frac{1}{2} K_{\text{bond}} (R - R_{\text{bond}})^2$, where $K_{\text{bond}} = 17000 \text{ kJ mol}^{-1} \text{ nm}^{-2}$), an angle potential with an equilibrium angle

$\theta_0 = 130^\circ$ ($V_{\text{angle}}(\theta) = \frac{1}{2} K_{\text{angle}} \{\cos(\theta) - \cos(\theta_0)\}^2$, where $K_{\text{angle}} = 85 \text{ kJ mol}^{-1}$), and a dihedral

potential $V_{\text{dihedral}}(\phi) = \sum_{i=1}^4 K_{\phi,i} (1 + \cos(n_i \phi - \phi_i))$, where $\phi_1 = 180^\circ$ and $K_{\phi,1} = 1.96 \text{ kJ mol}^{-1}$ for $n_1 = 1$; $\phi_0 = 0^\circ$ and $K_{\phi,2} = 0.18$ for $n_2 = 2$; $\phi_0 = 0^\circ$ and $K_{\phi,3} = 0.33$ for $n_3 = 3$; $\phi_0 = 0^\circ$ and $K_{\phi,4} = 0.12$ for $n_4 = 4$. The CG PEO model predicted radii of gyration, diffusivities, hydrodynamic radii, and the distributions of bond lengths, angles, dihedrals and end-to-end distances that were in excellent agreement with those from all-atom simulations and experiments.⁴⁸ In particular, the experimentally observed transition from ideal to swollen-coil behavior at $1600 < M_w < 2000$ was successfully captured.⁴⁸ The CG PEO model can be applied to both PEO and PEG with different numbers of CG beads, as described in our previous work. Details of the force field parameters and verification of the CG dendrimer and PEG models are given in our previous work.^{36,47,48}

A cutoff of $12 \text{ \AA}$ was used for Lennard-Jones (LJ) and electrostatic interactions. The LJ potential was smoothly shifted to zero between 9 and $12 \text{ \AA}$, and the Coulomb potential was smoothly shifted to zero between 0 and $12 \text{ \AA}$. A temperature of 298 K was maintained by applying a Berendsen thermostat in the NPT ensemble.⁵⁵ A time step of 8 fs was used, and the coordinates were saved every 20 ps for analysis.

Equilibration of dendrimers

Single CG G3, G4, and G5 dendrimers were solvated in boxes of size of 10 nm /side. The final system includes a dendrimer, ~ 7500 CG waters (representing ~ 30000 real waters), and enough counterions to achieve electroneutrality (32 , 64 , and 128 Cl^- for G3, G4, and G5 dendrimers, respectively). Simulations were performed for 200 ns , and the last 150 ns were used for analysis.

Equilibration of a dendrimer-PEG conjugate

The equilibrated CG PEG550 (12-mers, $M_w \approx 550$) and PEG5000 (113-mers, $M_w \approx 5000$) from our previous work⁴⁸ are used to obtain initial configurations of PEG molecules. 30 , 60 , and 88 copies of PEG5000 were attached to the surfaces of G3, G4, and G5 dendrimers, respectively. For example, 88 copies of PEG5000 were equally distributed by attaching them randomly to 88 of the 128 surface terminals on a G5 dendrimer. PEGylation neutralizes the dendrimer terminals, and hence PEG-attached G3, G4, and G5 respectively have 2 , 4 , and 40 cationic charges remaining after the attachment of 30 , 60 , and 88 PEGs, respectively. Each dendrimer-PEG complex was solvated in a box of size of 29 nm /side, and the final system includes a dendrimer-PEG, ~ 200000 CG waters (~ 800000 real waters), and 2 – 40 counterions (Cl^-) to make the system neutral. For G4 dendrimers, 32 and 60 copies of PEG550, and 32 copies of PEG5000 were also attached, and the same procedure was performed except that the dendrimer-PEG550 system includes ~ 65000 CG waters in a box of size of 20 nm /side. Table

1 describes the numbers of molecules and surface charges for each system. Simulations were performed for 350 ns, and the last 150 ns were used for analysis.

Equilibration of two dendrimer-PEG complexes

The final configurations of dendrimer-PEG molecules simulated in water were used as the initial configurations for studies with multiple molecules. Two complexes of PEG550- or PEG5000-attached G4 dendrimers were initially clustered together in a box of size 25 or 40 nm/side, respectively. The lengths of the PEG chains and the numbers of PEG chains per dendrimer are given in Table 1. Initial distances between center of mass of the dendrimer-PEG complexes were set closely to two times of radius of gyration for each complex (second column of Table 2). The final system with PEG550 includes two dendrimer-PEG complexes, ~126000 CG waters, and 8 counterions for electroneutrality. For the system with PEG5000, ~522000 CG waters were included. Simulations were performed for 400 ns, and the last 200 ns were used for the analysis.

RESULTS AND DISCUSSION

Dendrimers and the dendrimer-PEG complexes were simulated in water with different sizes and numbers of dendrimers and PEG ligands. These systems are named “G3”, “G3P5000-30”, “G4”, “G4P550-32”, “G4P550-60”, “G4P5000-32”, “G4P5000-60”, “G5”, “G5P5000-88”, “TG4P550-60”, and “TG4P5000-60”, where the first, second, and third number describe the dendrimer generation, molecular weight of the PEG, and the number of PEG molecules, respectively. The first initial “T” specifies the systems with two copies of dendrimer-PEG molecules; see Table 1.

Dependence of the PEG conformation on grafting density

Figure 1 shows snapshots of dendrimer-PEG complexes at the end of the simulations. Since PEG chains are attached to 94% of the dendrimer surface, PEG are very crowded and extended towards water. This is observed more significantly in the longer PEG-attached dendrimer, G4P5000-60, where a dendrimer is almost completely covered by PEG. Figure 2 shows that radii of gyration (R_g) reach apparent steady-state values at ~100 ns, indicating that systems are well equilibrated. In Table 2, the R_g of dendrimer-PEG's calculated from the simulations agree quantitatively with those obtained from the neutron scattering experiments with the same lengths and numbers of PEG's,⁴⁹ indicating that the CG model can capture the experimentally measured conformations of dendrimer-PEG. Table 3 shows the aspect ratios, I_z/I_x and I_z/I_y , where I_z , I_y , and I_x are principal moments of inertia, ordered such that $I_z > I_y > I_x$. The relative shape anisotropy, κ^2 , is given by $\kappa^2 = 1 - 3I_2/I_1^2$, where I_1 and I_2 are the first and second invariants of the radius of gyration tensor ($I_1 = I_x + I_y + I_z$, $I_2 = I_x I_y + I_y I_z + I_x I_z$). A linear array of skeletal atoms is characterized by $\kappa^2 = 1$, while a molecule with tetrahedral or higher symmetry is characterized by $\kappa^2 = 0$.⁵⁶ Simulations of PEG-free dendrimers in water show that G3 to G5 dendrimers form ellipsoidal shapes, in agreement with our previous simulations.³⁰ After attaching PEG onto the dendrimer surfaces, aspect ratios and anisotropies of the dendrimer-PEG complexes significantly decrease, indicating that attachment of PEG chains makes the dendrimer-PEG almost spherical.

To understand the dependence of the PEG conformation on the PEG grafting density on the dendrimer surface, the outer radii of the dendrimer (R_d), the outer radii of the dendrimer-PEG (R), and thickness of the PEG layer ($L = R - R_d$) were calculated (Figure 3). R_d was calculated from the root-mean-squared (RMS) distance from the dendrimer terminals to the dendrimer center of mass (COM), and R was calculated from the RMS distance from the ending monomers of PEG to the dendrimer-PEG COM. Table 2 shows that the layer thicknesses of both PEG550

and PEG5000 with many PEG attachments are greater than for fewer attachments, indicating the conformation of PEG on the dendrimer surface is affected by numbers of the grafted chains.

Daoud and Cotton⁵⁷ used polymer brush theory to show that polymer chains attached to small spheres are less stretched than those on planar surfaces. Vagberg et al.⁵⁰ modified the model and derived a simple analytical formula to predict the thickness of the polymer layer. Applying their formula to the dendrimer-PEG complex, the outer radius (R) of the dendrimer-PEG, given

in Table 2, is calculated using $R = \left[N_{EO} l^{1/v} \frac{8f^{(1-v)/2v}}{3v4^{1/v}} + R_d^{1/v} \right]^v$, where N_{EO} is the number of EO monomers per chain (12 and 113 for PEG550 and PEG5000, respectively), l is the statistical length of the monomer (which is equivalent to the bond length of the CG bead, 3.3 Å), v is the Flory exponent (3/5 for a good solvent), and f is the number of grafted chains. The R_d values were obtained from simulations. Table 2 shows that the L values obtained from the simulations are close to those calculated from the analytical theory, suggesting that the CG model behaves similarly to the brush theory for a starlike polymer.

Figure 4 shows density profiles, normalized so that they integrate to unity, for PEG as a function of distance from the COM of the dendrimer. For both PEG550 and PEG5000 with lower PEG attachment density, the mass of PEG is concentrated close to the COM of the dendrimer. Figure 5 shows average distances between the n^{th} -monomer of the PEG (numbering starting at the point of attachment to the surface) and the dendrimer-PEG COM. As n for PEG5000 increases, its distance from the dendrimer-PEG COM increases almost linearly, implying that PEG does not strongly fold back towards the dendrimer surface.

The effects of the PEG length on the inter-particle interaction

Experimentally, Kim et al. showed that when PEG2000 fully saturated the dendrimer surface, cell viability was reduced at high concentration of the dendrimer-PEG complex, probably because of formation of intermolecular aggregates.¹⁶ Also, Yang et al. suggested that long PEG5000 chains likely penetrate into other complexes, leading to less efficient encapsulation of a hydrophobic compound.²⁵ To understand the effects of the PEG length on the inter-particle interaction, two copies of G4 dendrimer-PEG were simulated with different PEG lengths and dendrimer-PEG concentrations (Table 1).

Figure 6 shows snapshots from the beginning and the end of a simulation of TG4P550-60. Two complexes were initially clustered, but they drift apart by the end of the simulation. A similar trend was also observed for the nanoparticle with the longer PEG, TG4P5000-60. Figure 7 shows distances between centers of mass of two dendrimer-PEG complexes for TG4P550-60 and TG4P5000-60. The distance between two complexes of TG4P550-60 significantly increases over the simulation period, indicating that there is no aggregation of these dendrimer-PEG's. For TG4P5000-60, the same trend is observed, although an increase in the distance between two complexes is smaller than for TG4P550-60. To check if larger complex size and lower diffusivity for TG4P5000-60 cause less movement for insufficient simulation time, R_g and diffusion coefficients for TG4P550-60 and TG4P5000-60 were calculated. The radii of gyration of each dendrimer-PEG in TG4P550-60 and TG4P5000-60 are respectively $R_g = 2.70 \pm 0.02$ and 6.67 ± 0.18 nm, very close to those for single-dendrimer systems (respectively, G4P550-60 and G4P5000-60). Since R_g of a dendrimer-PEG is larger for TG4P5000-60 than for TG4P550-60, they may have different diffusivities. Diffusion coefficients of each dendrimer-PEG, D_{PBC} , were calculated from the slopes of the mean square displacements (MSD) of the centers of mass (COM) versus time. D_{PBC} were then corrected for finite size

effects using the formula developed by Yeh and Hummer,⁵⁸ $D = D_{PBC} + \frac{K_B T \xi}{6\pi\eta L}$, where K_B is Boltzmann's constant, L is the cubic box length, $\xi = 2.837297$, and η is the viscosity of the

medium (here, $\eta = 0.865$ cP at 298 K, calculated from the Stokes-Einstein relationship⁵⁹ with the translational friction constant of CG water obtained by Stoke's law with slip boundary conditions, as given in detail by the previous work⁴⁸). Note that the viscosity of the CG water is 0.865 cP at 298 K, close to the experimental value of 0.933 cP. A diffusion coefficient of CG water has previously been calculated from its MSD and corrected for finite size effects, yielding $D = 1.63 \times 10^{-5}$ cm²/s, which is close to the experimental value of 2.3×10^{-5} cm²/s.⁴⁸ Since the water diffusivity and viscosity are in reasonable semi-quantitative agreement with the experimental values, it is likely that the diffusivities of globular particles, such as the nanoparticles simulated here, are also semi-quantitatively accurate. Table 4 shows that diffusion coefficients for TG4P550-60 are higher than those for TG4P5000-60, which can be explained by the larger size of the dendrimer-PEG's in TG4P5000-60. For TG4P550-60, over a period of 400 ns, these diffusivities of around 0.11 nm²/ns ought to produce a mean-square drift in position of each of the two dendrimer-PEG's of around 44 nm², or a root-mean-square drift of 6.6 nm. This ought to lead to changes in separation between the two dendrimer-PEG's of around $\sqrt{2}$ times this magnitude (or around 9.4 nm over a 400 ns period), with large fluctuations about this value due to the stochastic nature of diffusion. This magnitude, along with large fluctuations in the separation, is consistent with what is observed for TG4P550-60 in Figure 7. For TG4P5000-60, diffusivity is around 0.06 nm²/ns, leading to changes in separation between the complexes of around 7 nm over a 400 ns period. These results show that an increase of ~6.5 nm in the inter-particle distance is lower than that for TG4P550-60, but evidently indicates separation between particles and no inter-particle aggregation. The two dendrimer-PEG's retain a center-of-mass separation of around 18 nm after 250 ns. The 18 nm separation is 5 nm greater than twice the radius of gyration of a single dendrimer-PEG (6.67 ± 0.18 nm), thus showing no indication of a dimer aggregate of the two dendrimer-PEG's. In Figure 8, radial distribution functions (RDF) of PEG chains between two dendrimer-PEG complexes for both TG4P550-60 and TG4P5000-60 show that although initially the PEG beads on the two dendrimer-PEG's are in close contact, the peak in the RDF significantly decreases at later times (200–400 ns). These results indicate that both short PEG550 and long PEG5000 chains do not induce aggregation of dendrimer-PEG, which does not support the long PEG2000/5000 chains-induced intermolecular aggregation suggested experimentally from the observations of the increased cytotoxicity and less efficient encapsulation.

The effect of long-range electrostatic interactions

The MARTINI force field was parameterized with the cutoff method for electrostatics.⁵³ Also, most cationic surfaces of the simulated dendrimers are covered by neutral PEG molecules and become neutral, leading to no electrostatic interactions. Therefore, the cutoff method was used for electrostatic interactions, assuming that the few remaining charges on the dendrimer surface and counterions do not strongly influence observations discussed above. To confirm this assumption, G5P5000-88 and TG4P550-60 were simulated with particle mesh Ewald summation (PME). Although G5P5000-88 has the highest numbers of surface charge and ions, its R_g with PME is 7.37 ± 0.03 nm, which is very close to that calculated from G5P5000-88 with the cutoff method. For TG4P550-60 with PME, the distance between centers of mass of two dendrimer-PEG complexes increases as a function of time, as observed in the same system with the cutoff method. The distance between the complexes reaches 12 nm at ~70 ns, and this increase in the distance occurs even faster than in the same system with cutoff, clearly showing no aggregation. These results indicate that long-range electrostatics do not strongly affect the conformation and interactions of highly neutralized dendrimers, although highly charged dendrimers may be affected by the treatment of electrostatics, as observed in our previous simulations of dendrimers with lipid bilayers.^{36,37}

CONCLUSIONS

Coarse-grained molecular dynamics (CG-MD) simulations of PAMAM dendrimer-PEG complexes with water were performed with different numbers and sizes of dendrimers and PEG's to investigate the effects of the PEG length and conjugate number on the conformation and aggregation of dendrimer-PEG. The CG dendrimer and PEG models were previously developed within the framework of the MARTINI force field,^{53,54} and their parameterization and verification are well described in our previous work.^{47,48} Radii of gyration of the dendrimer-PEG complexes agree quantitatively with those measured by neutron scattering experiments with the same lengths and numbers of attached PEG's. Also, the thickness of the PEG layer compares favorably with that calculated from the analytical formula from brush theory for a starlike polymer. At high PEG grafting density, PEG chains are crowded and more extended towards water than those with lower grafting density. Our results indicate that our CG dendrimer and PEG models successfully represent experimentally measured and theoretically predicted conformations of PEG grafted onto the dendrimer surface with different lengths and numbers of conjugated PEG's.

Two molecules of the dendrimer-PEG complex were simulated in the same simulation box, showing no aggregation of dendrimer-PEG for either length of PEG chain simulated. These findings thus do not support some suggestions in the literature that inter-particle aggregation can occur for PEG-coated nanoparticles, but instead support the view that PEGylation acts to stabilize nanoparticles against aggregation.

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G4P550-60

G4P5000-60

Figure 1. Snapshots at the end (350 ns) of simulations. Black dots represent dendrimers and red dots represent PEG molecules. Explicit water and ions are omitted for clarity. The images were generated with VMD.⁶⁰

Figure 2.
Radii of gyration of the dendrimer-PEG complexes as a function of simulation time.

Figure 3. Schematic representations of the brush configuration. R_d is the outer radius of the dendrimer, R is the outer radius of the dendrimer-PEG, and L is thickness of the PEG layer.

Figure 4. Normalized mass density profiles of PEG as a function of distance from center of mass (COM) of the dendrimer, averaged over the last 150 ns.

Figure 5. Average distances between n^{th} -monomer of PEG and center of mass of dendrimer-PEG. 1st monomer is next to the dendrimer surface, and 113th monomer is farthest from the dendrimer.

Figure 6. Snapshots at the beginning (0 ns, *left* image) and end (400 ns, *right* image) of a simulation of TG4P550-60. Black, red, and blue dots respectively represent dendrimer, PEG, and water molecules.

Figure 7.
Distances between centers of mass of two dendrimer-PEG complexes as a function of time.

Figure 8. Radial distribution functions of PEG monomers on two different dendrimer-PEG complexes (thin lines for 0–1 ns; thick dotted lines for 200–400 ns).

Table 1

List of simulations.

	No. of dendrimers or dendrimer-PEG's	Dendrimer			PEG		Simulation time (ns)
		Generation	No. of surface groups	No. of charges	No. of monomers in a PEG	No. of PEG molecules	
G3	1	3	32	32	-	-	200
G3P5000-30	1	3	32	2	113	30	350
G4	1	4	64	64	-	-	200
G4P550-32	1	4	64	32	12	32	350
G4P550-60	1	4	64	4	12	60	350
G4P5000-32	1	4	64	32	113	32	350
G4P5000-60	1	4	64	4	113	60	350
G5	1	5	128	128	-	-	200
G5P5000-88	1	5	128	40	113	88	350
TG4P550-60	2	4	64	4	12	60	400
TG4P5000-60	2	4	64	4	113	60	400

Table 2

The radius of gyration (R_g) and the outer radius (R) of the dendrimer-PEG, outer radius (R_d) of the dendrimer, and thickness of the PEG layer (L). All quantities are in nm.

	R_g		Exp ⁴⁹	R_d		R		$L (=R-R_d)$	
	Sim			Sim		Sim	Anal	Sim	Anal
G3P5000-30	5.98 ± 0.22		5.67 ± 0.26	2.42 ± 0.01		8.06 ± 0.16	7.50	5.64	5.08
G4P550-32	2.37 ± 0.06			2.31 ± 0.02		3.02 ± 0.04	3.13	0.71	0.82
G4P550-60	2.75 ± 0.02			2.53 ± 0.01		3.38 ± 0.03	3.47	0.85	0.94
G4P5000-32	5.77 ± 0.11			2.76 ± 0.08		7.81 ± 0.14	7.73	5.05	4.97
G4P5000-60	6.65 ± 0.10		7.06 ± 0.28	2.74 ± 0.01		8.87 ± 0.08	8.60	6.13	5.86
G5P5000-88	7.40 ± 0.04		7.67 ± 0.22	3.22 ± 0.02		9.72 ± 0.19	9.41	6.50	6.19

Table 3

Average values of aspect ratios and relative anisotropies of each dendrimer or dendrimer-PEG complex over the last 150 ns.

	Aspect ratio		Relative shape anisotropy
	I_z/I_x	I_z/I_y	
G3	1.91	1.13	0.032
G4	1.72	1.29	0.024
G5	1.64	1.44	0.024
G3P5000-30	1.25	1.10	0.004
G4P5000-60	1.15	1.08	0.002
G5P5000-88	1.13	1.09	0.001

Table 4

Diffusion coefficients ($10^{-7} \text{ cm}^2\text{s}^{-1}$) for dendrimer-PEG's. Each system includes two dendrimer-PEG's, leading to two values of the diffusion coefficient for each system. D_{PBC} was calculated directly from the trajectories using MSD, and then the final D was corrected for size artifacts from periodic boundary conditions.

	D_{PBC}	D
TG4P550-60	7.91 ± 0.20	10.8
	7.99 ± 0.24	10.9
TG4P5000-60	4.78 ± 1.00	6.58
	3.96 ± 0.47	5.76