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Synthesis and Characterization of Bola-Type Amphiphilic Dendritic Macromolecules

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ABSTRACT: The synthesis of fourth-generation bola-type linear−dendritic macromolecules is described using a high yielding divergent method. These systems are composed of a non-immunogenic linear poly(ethylene glycol) core and dendritic wedges of succinic acid and glycerol connected by ester linkages in a triblock architecture. Three molecular weight PEGs (3400, 10 000, and 20 000 g/mol) were used in the synthesis of low, medium, and high molecular weight macromolecules. The molecular weight data were determined by MALDI−TOF and SEC. The macromolecules were further characterized by modulated differential scanning calorimetry (MDSC) and critical aggregation concentration (CAC) as determined by fluorescence spectroscopy. Cell toxicity studies were performed with human umbilical vein endothelial cells (HUVEC). These macromolecules are of interest for a variety of biomedical applications including the repair of ocular wounds.

Introduction

Our continuing use of sutures, three-dimensional scaffolds, drug delivery vehicles, and polymer therapeutics demonstrates the ever-increasing interest in synthesizing polymers for the medical and biotechnological fields.[1,2] Within these applications, linear polymers are widely used as temporary scaffold materials for wound repair and tissue engineering applications.[3] For example, poly(lactic acid) (PLA) is a degradable water-insoluble polyester that has been successfully used in a number of applications; however, it has limited potential for functionalization and property optimization. Recently, other polymer architectures, like dendrimers, have been used for drug delivery, medical imaging, and wound repair.[4−9] Dendrimers[10,11] are composed of an interior core, internal branching regions, and a large number of external functional groups. Compared to linear polymers, dendrimers are monodisperse and possess a higher solubility, a lower viscosity, and the ability to adopt a globular shape in solution.[10−13] Today, dendrimers are synthesized using either the divergent[14−18] or convergent approach.[19−25] In the divergent strategy, synthesis is initiated from the core and proceeds outward in a series of coupling and activation steps. The convergent strategy initiates growth from the exterior dendrimer segments (dendrons) and proceeds inward by coupling the dendrons to a core.

Dendritic-based macromolecular structures were first reported in the late 1970s by Buhleier et al.[26] This report was later followed by the synthesis of a complete poly(amidoamine), PAMAM, dendrimer in the mid-1980s by both Tomalia and Newkome using a divergent strategy, wherein methyl acrylate was coupled to an ammonia core followed by an exhaustive amidation with ethylenediamine.[27−30] This approach was a significant advancement toward overcoming problems of purity and low yields in the preparation of dendritic macromolecules.[30,31] The ability to synthesize well-defined structures lead to an increase in the number and type of dendritic macromolecules synthesized and studied in the past two decades.[32−37] Of the various dendritic macromolecules studied, linear−dendritic block copolymers are finding every increasing uses because of the interesting assembles they can form in solution, on surfaces, or in the presence of other macromolecules (e.g., DNA).[38−50]

In this report, the synthesis of linear−dendritic amphiphilic macromolecules composed of succinic acid, glycerol, and poly(ethylene glycol) (PEG) is described using the divergent approach (Figure 1). These dendritic bola-type macromolecules possess an ABA triblock architecture where a linear B block, consisting of a hydrophilic PEG linker, is flanked by two dendritic A blocks of glycerol and succinic acid connected via ester linkages. The presence of hydrolyzable ester linkages allows for degradation of the copolymers to natural metabolites, such as succinic acid, which is present in the Krebs cycle, and glycerol, which is the precursor to triacylglycerols.[51,52] For these studies, PEG was selected because of its non-immunogenicity.[53] These copolymers belong to a family of dendritic macromolecules that have been used for ocular wound repair, drug delivery, and cartilage tissue engineering.[54−59] Herein, we report the synthesis and characterization of benzylidene protected and hydroxy-terminated bola-type dendritic−linear copolymers up to a fourth generation, consisting of succinic acid and glycerol with three different PEG cores (molecular weight of 3400, 10 000, and 20 000 g/mol).

Results and Discussion

The divergent synthesis of the triblock linear−dendritic macromolecules (Schemes 1 and 2) proceeds from the core and builds outward in an iterative fashion, wherein an esterification reaction with an activated monomer is followed by a hydrogenolysis reaction. The activated monomer was synthesized in two steps by the addition of *cis*-1,3-*O*-benzylidene glycerol to 1.2 equiv of succinic anhydride in pyridine to afford 2-(*cis*-1,3-*O*-benzylidene glycerol) succinic acid monoester in 75% yield. Subsequent esterification with DCC afforded the anhydride (**2**) of the succinic acid monoester in 64% yield.[54]

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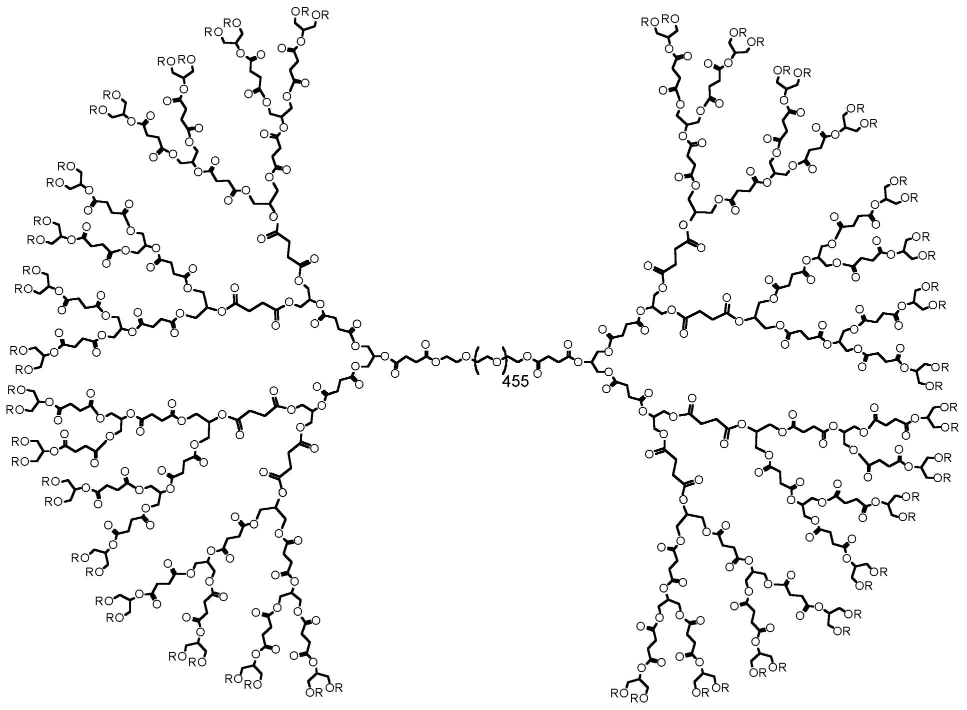


Figure 1. Chemical structure of ([G4]-PGLSA-OH)$_2$-PEG$_{20000}$ bola-type dendritic−linear copolymer. R = H or benzylidene acetal.

The zeroth (G0) through fourth (G4) generation dendritic macromolecules were synthesized according to a report by Carnahan et al.,[54] which described the preparation of the 3400 g/mol M_w PEG-based dendritic macromolecules. The zeroth generation *b*enzy*lid*ene protected *p*oly(*gl*ycerol *s*uccinic *a*cid) PEG macromolecule, ([G0]-PGLSA-bzld)$_2$-PEG, was obtained by coupling 8 equiv of **2** with each of the three PEG polymers (3400, 10 000, and 20 000 g/mol, corresponding to **3a,b,c**, respectively) in the presence of 1.6 equiv of DMAP in DCM. The esterification reactions were stirred for 1, 3, and 7 days, corresponding to the 3400, 10 000, and 20 000 g/mol PEG copolymers, and quenched with 1-butanol before isolation in cold ether. The tetrahydroxy-terminated ([G0]-PGLSA-OH)$_2$-PEG, **4a,b,c**, macromolecules were obtained by cleaving the benzylidene acetal group via a hydrogenolysis reaction using 10% w/w Pd/C and 60 psi of H_2 in THF followed by precipitation in cold ether. The first generation benzylidene-protected macromolecule (**(**[G1]-PGLSA-bzld)$_2$-PEG, **5a,b,c**) was prepared by reacting 16 equiv of **2** with **4** and 3.2 equiv of DMAP in DCM. The benzylidene acetal groups were cleaved via a hydrogenolysis reaction with 10% w/w Pd/C, 60 psi H_2 in THF, to afford **(**[G1]-PGLSA-OH)$_2$-PEG, **6a,b,c** after isolation in cold ether.

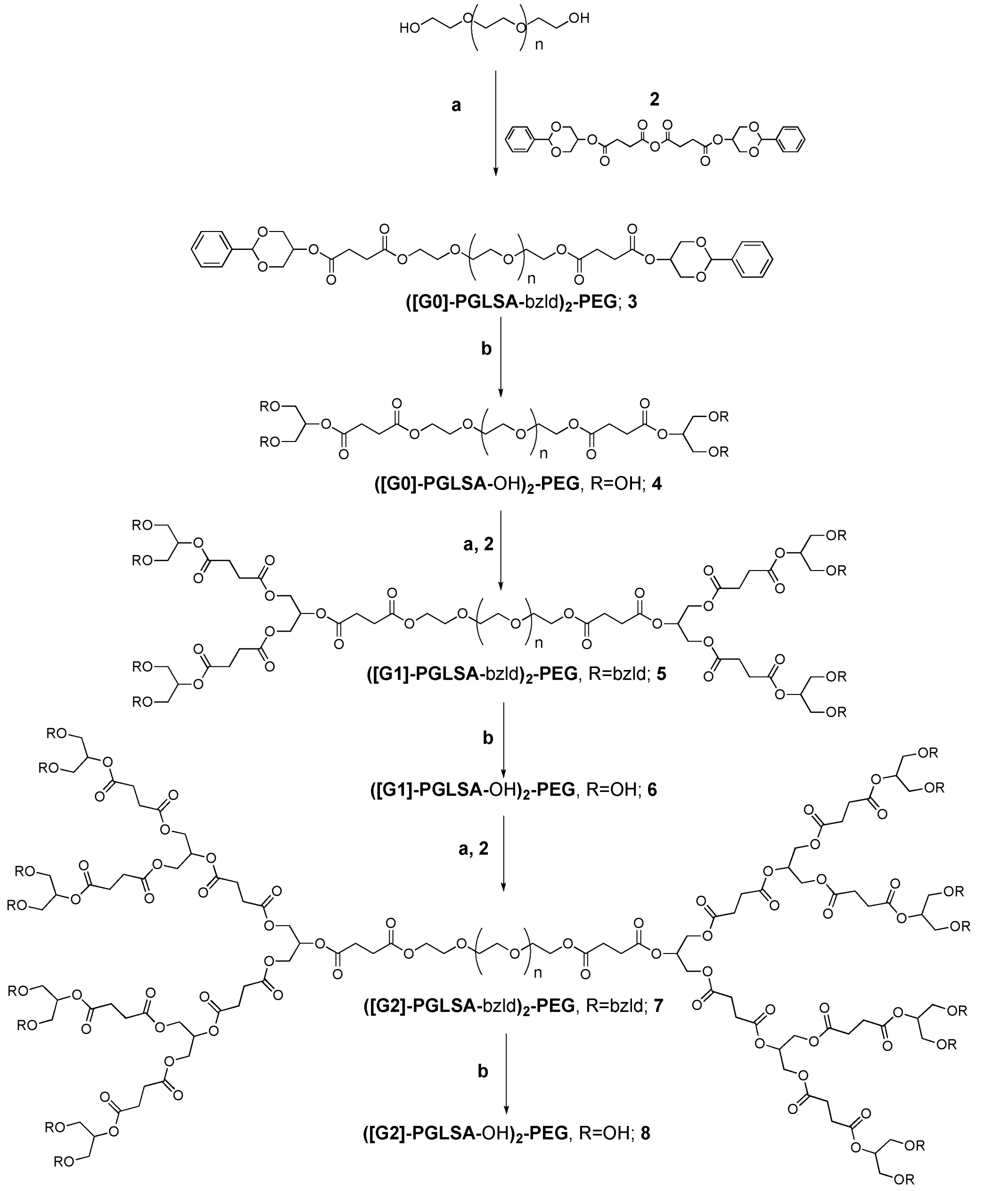
The second generation deprotected dendritic macromolecules **(**[G2]-PGLSA-OH)$_2$-PEG, **8a,b,c** were synthesized in a similar manner using 32 equiv of **2** and coupling with **6a,b,c** in the presence of 6.4 equiv of DMAP. Copolymers **8a,b,c** were obtained via a hydrogenolysis reaction with H_2 and a Pd/C catalyst in THF and isolated after precipitation in ether. The third generation deprotected dendritic macromolecules **(**[G3]-PGLSA-OH)$_2$-PEG, **10a,b,c** were obtained by treating **8a,b,c** with 64 equiv of **2** and 12.8 equiv of DMAP in DCM. A hydrogenolysis reaction cleaved the benzylidene acetal protecting group and afforded copolymers **10a,b,c** after precipitation in ether. The fourth generation of the hybrid dendritic macromolecules, **(**[G4]-PGLSA-OH)$_2$-PEG, **12a,b,c**, were synthesized using the same iteration of esterification (128 equiv of **2** and 25.6 equiv of DMAP) and deprotection (Pd/C, 60 psi H_2 in THF) reactions. Copolymers **11a,b,c** and **12a,b,c** were obtained after precipitation in cold ether. The yields for all the esterification reactions ranged from 86 to 97%, while the yields for the hydrogenolysis reactions ranged from 87 to 98%. The **(**[Gn]-

Table 1. MALDI-TOF and SEC Data for ([Gn]-PGLSA)$_2$-PEG$_{3400,\ 10000,\ \text{and}\ 20000}$ Macromolecules

	3400 g/mol PEG				10 000 g/mol PEG				20 000 g/mol PEG			
		calcd	MALDI	SEC[a]		calcd	MALDI	SEC		calcd	MALDI	SEC
dendritic macromolecule	no.	M_n	M_n[b]	M_n[b]	no.	M_n	M_n	M_n[c]	no.	M_n	M_n	M_n[d]
OH-PEG-OH		3366	3351	3550		10000	12923	11216		20000	22142	20645
([G0]-PGLSA-bzld)$_2$-PEG	**3a**	3891	3875	3750	**3b**	13448	13427	12016	**3c**	22668	22672	32755
([G0]-PGLSA-OH)$_2$-PEG	**4a**	3714	3696	3500	**4b**	13274	13193	11373	**4c**	22493	22467	20645
([G1]-PGLSA-bzld)$_2$-PEG	**5a**	4763	4749	3790	**5b**	14323	14259	12602	**5c**	23542	23564	14997
([G1]-PGLSA-OH)$_2$-PEG	**6a**	4411	4394	4440	**6b**	13970	13961	11064	**6c**	23189	22999	20645
([G2]-PGLSA-bzld)$_2$-PEG	**7a**	6509	6492	4680	**7b**	16068	16003	13515	**7c**	25287	26031	28418
([G2]-PGLSA-OH)$_2$-PEG	**8a**	5804	5788	5210	**8b**	15364	15275	12710	**8c**	24583	25455	19924
([G3]-PGLSA-bzld)$_2$-PEG	**9a**	10000	9961	6900	**9b**	19559	19506	14315	**9c**	28779	29481	14474
([G3]-PGLSA-OH)$_2$-PEG	**10a**	8591	8575	7820	**10b**	18150	17998	13451	**10c**	27667	27958	25546
([G4]-PGLSA-bzld)$_2$-PEG	**11a**	16983	16968	7950	**11b**	24508[e]	*f*	16352	**11c**	35759	36029	8528
([G4]-PGLSA-OH)$_2$-PEG	**12a**	14163	14146	8990	**12b**	21688[e]	21956	15680	**12c**	32939	*f*	13920

[a] All the SEC data are based on PEG standards in THF using Polymer Laboratories PLgel MIXED-E 3 μm column at a flow rate of 1 mL/min. [b] PDIs ranged from 1.02 to 1.04. [c] PDIs ranged from 1.06 to 1.19. [d] PDIs ranged from 1.12 to 1.24. [e] Calculated molecular weight based on the use of a different batch of PEG 10 000 M_w (MALDI M_n10 891). [f] Unable to identify the peak from baseline.

Scheme 1. Divergent Synthesis of ([G2]-PGLSA-OH)$_2$-PEG Hybrid Dendritic Macromolecule[a]



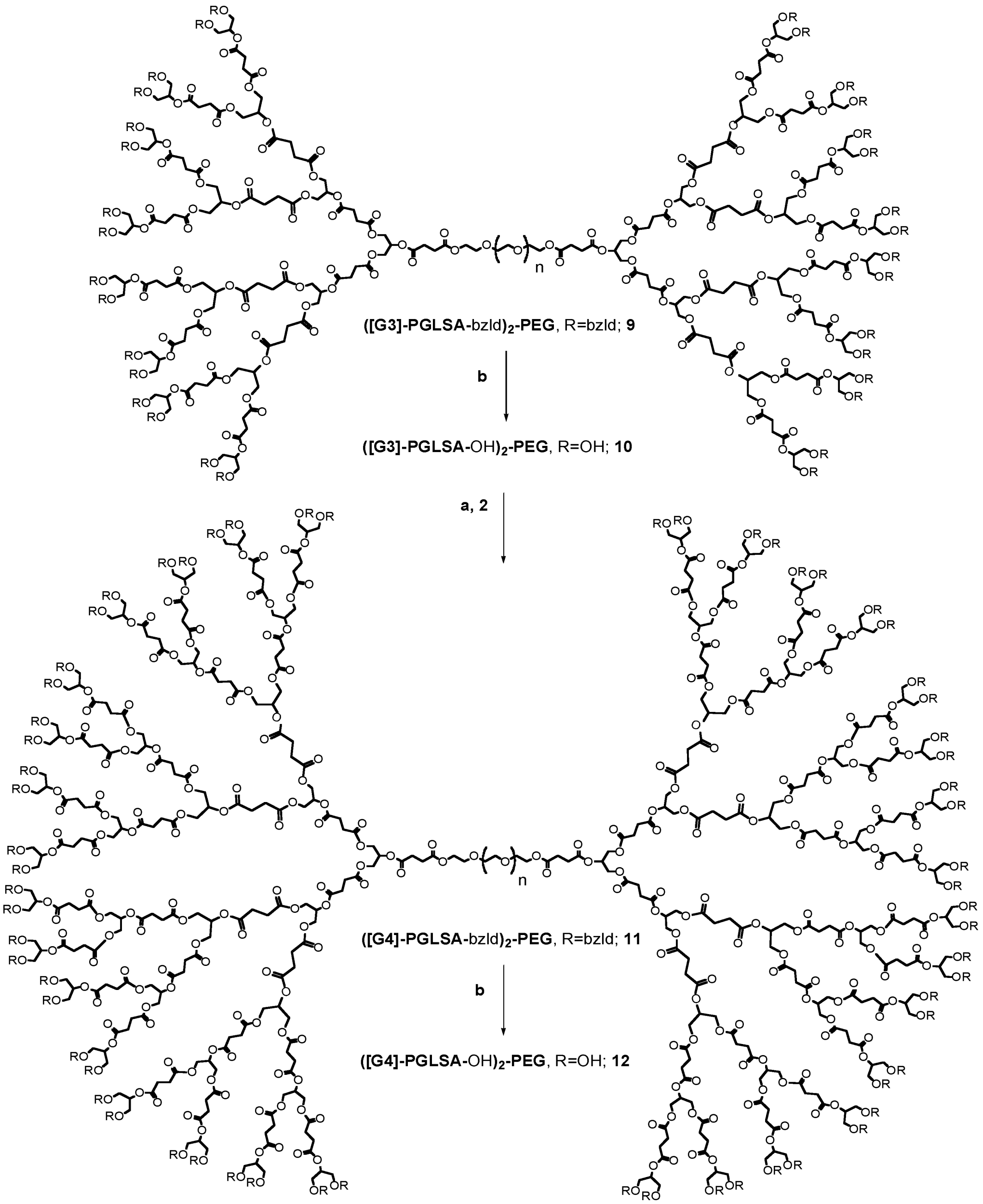
[a] The scheme depicts the general synthetic route using the 3400, 10 000, and 20 000 g/mol PEG polymers, where *n* is 77, 227, and 455, respectively.

PGLSA)$_2$-PEG$_{10000 \text{ and } 20000}$ macromolecules were course, non-adhesive powders which were easier to handle than the ([Gn]-PGLSA)$_2$-PEG$_{3400}$ macromolecules. The protected and deprotected G0 through G2 3400 PEG-based macromolecules are soluble in common organic solvents. The protected G3 and G4 3400 PEG-based macromolecules are soluble in DCM, THF, and ethyl acetate. The deprotected G3 and G4 3400 PEG-based macromolecules are soluble in DMF, EtOH, and H_2O. The protected and deprotected G0 through G4 10 000 and 20 000 PEG-based macromolecules are soluble in common organic solvents. The protected G0 and G1 and deprotected G0 through G4 10 000 and 20 000 PEG-based macromolecules are soluble in H_2O.

The benzylidene acetal group (bzld) was selected as the protecting group for the primary glycerol hyroxyls since it can be readily cleaved under mild conditions by hydrogenolysis. The success of the esterification and deprotection reactions was monitored by ^{1}H NMR spectroscopy, where the PEG served as a tag to monitor the change in proton integrations with each successive generation. For example, Figure 2 shows the ^{1}H NMR spectrum of a protected and deprotected third generation copolymer. The characteristic aromatic protons from the benzylidene protecting group appeared at 7.32 and 7.46 ppm (a), the benzylidene acetal proton appeared at 5.45 ppm (b), and the internal and external glycerol methine protons appeared at 5.19 (c) and 4.71 ppm (d) (Figure 2A). The methylene glycerol protons are shifted upfield at 4.12−4.27 ppm (e), while the PEG protons appeared as a broad multiplet at 3.47−3.78 ppm (f). The succinic acid protons are shifted upfield at 2.65−2.76 ppm (g). As shown in Figure 2B, upon removal of the benzylidene protecting groups by a hydrogenolysis reaction, the aromatic and acetal protons at 7.32, 7.46 (a), and 5.45 ppm disappear (b).

Molecular weights and PDIs were determined by MALDI−TOF and SEC. The MALDI−TOF data (Table 1) showed a bell-shaped distribution, which shifted to higher molecular weights with the attachment of more monomer units, corresponding to the next higher generation. A decrease in the number-average molecular weight (M_n) was observed with the removal of the benzyliene acetal protecting group in each generation. With a low molecular weight PEG (3400 g/mol), the adjacent peaks in the bell-shaped curve were present with a 44 g/mol mass difference, corresponding to the individual

Scheme 2. Divergent Synthesis of ([G4]-PGLSA-OH)$_2$-PEG Macromolecule from ([G2]-PGLSA-OH)$_2$-PEG[a]



[a] The scheme depicts the general synthetic route using the 3400, 10 000, and 20 000 g/mol PEG polymers where *n* is 77, 227, and 445, respectively.

oxyethylene units of PEG. As the molecular weight of PEG increased from 3400 to 10 000 and 20 000 g/mol, the oxyethylene peaks become less apparent, and the bell-shaped curve shifted to a broad peak. The MALDI spectra for the protected and deprotected third generation copolymer, for which the NMR is shown in Figure 2, is found in the SI for comparison. With SEC, an accurate measure of the molecular weight of hybrid dendritic−linear copolymers was more difficult to obtain with a calibration based on linear PEG standards. The data from Table 1 show a decrease in M_n as the dendritic generation number increased. This is likely a result of the hybrid copolymers adopting a more globular structure in solution with increasing molecular weight.

The melting (T_m) and glass transition (T_g) temperatures of hybrid copolymers were determined using MDSC to further characterize these polymers. The T_m decreased for the **(**[Gn]-PGLSA-OH)$_2$-PEG$_{10000\text{ and }20000}$ macromolecules from a value of 62 and 64 °C for simple 10 000 and 20 000 g/mol PEG-diol polymers to 42.6 to 49.2 °C as the generation number increased to a G4. The T_m decreased from 49 °C for 3400 g/mol PEG-diol to 36.5 °C as the generation increased to the **(**[G2]-PGLSA-OH)$_2$-PEG$_{3400}$ macromolecule. A T_g was observed at −13.6 °C for the ([G3]-PGLSA-bzld)$_2$-PEG$_{3400}$, which reduced to −18 °C for the **(**[G4]-PGLSA-OH)$_2$-PEG$_{3400}$ macromolecules (Table 2). As the molecular weight of the dendritic portion exceeded the PEG molecular weight, the copolymer exhibited a glass transition, which was observed with the **(**[Gn]-PGLSA)$_2$-PEG$_{3400}$ macromolecule. For both the **(**[Gn]-PGLSA)$_2$-PEG$_{10000\text{ and }20000}$ macromolecules, the PEG molecular weight portion exceeded the dendritic molecular weight portion for the zeroth through fourth generations, and a T_m was observed.

Next, the amphiphilicity of the bola-type macromolecules was determined by measuring their critical aggregation concentration (CAC) using a pyrene-based fluorescence technique. In this method, the increase in pyrene fluorescence as a function of copolymer concentration was monitored.[60] The CAC was

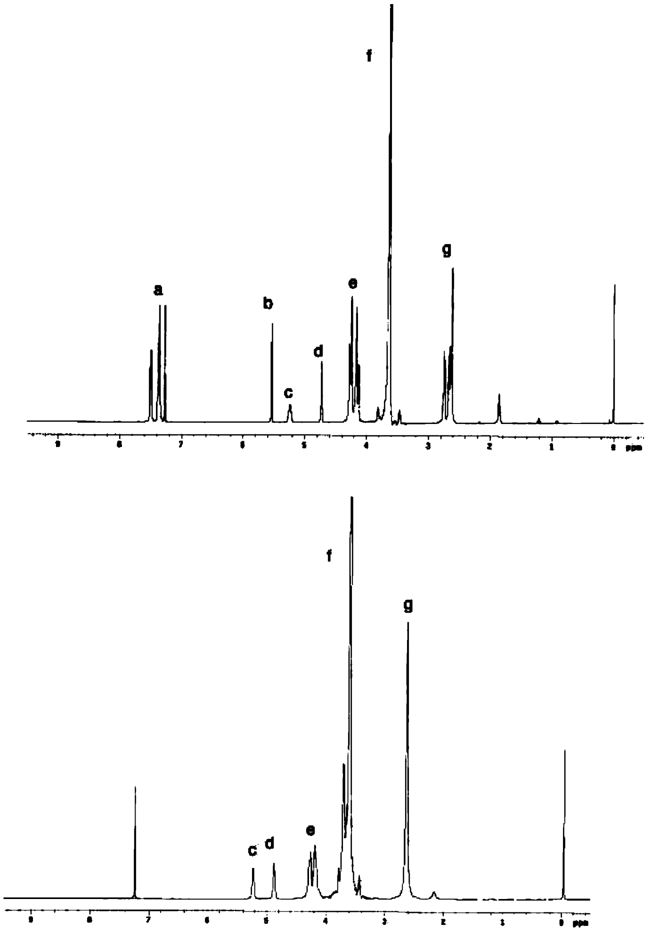


Figure 2. ^{1}H NMR spectra of (A) ([G3]-PGLSA-bzld)$_2$-PEG$_{10000}$ and (B) ([G3]-PGLSA-OH)$_2$-PEG$_{10000}$.

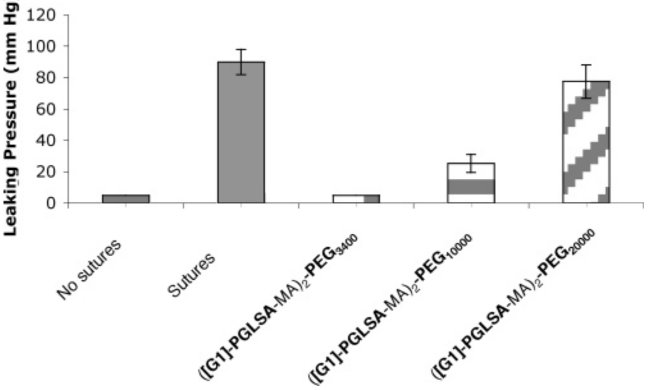


Figure 3. Leaking pressures for the 4.1 mm central lacerations with three different hydrogel adhesives at 5% w/v. All adhesives were evaluated on enucleated porcine eyes ($n = 3$).

determined for each generation of the ([Gn]-PGLSA)$_2$-PEG$_{3400,\ 10000,\ \text{and}\ 20000}$ dendritic macromolecules, both protected and deprotected, up to a first generation. As the copolymers increased in size from a protected second generation to a protected fourth generation, the materials were insoluble in aqueous solutions even at very low concentrations (i.e., 10^{-7} M). Therefore, only deprotected copolymers were evaluated from the second to the fourth generations. As the concentration of the copolymers was varied from 10^{-6} to 10^{-2} M, the intensity of pyrene emission at 373 nm increased, and the CAC was determined as the concentration where a sharp increase in fluorescence was observed (Table 3).

As the generation was increased form ([G0]-PGLSA-OH)$_2$-PEG$_{3400}$ to a ([G4]-PGLSA-OH)$_2$-PEG$_{3400}$, the CAC values decreased from 57 to 5 mM. Likewise, as the copolymers increased in molecular weight from ([G0]-PGLSA-OH)$_2$-PEG$_{10000\ \text{and}\ 20000}$ to the final ([G4]-PGLSA-OH)$_2$-PEG$_{10000\ \text{and}\ 20000}$, the CAC decreased from 44 and 50 mM to 4 and 7 mM, respectively. As shown in Table 3, the CAC values were larger for a ([Gn]-PGLSA-OH)$_2$-PEG compared to a ([Gn]-PGLSA-bzld)$_2$-PEG macromolecule within the same generation. Within one generation, as the PEG was increased from a 3400 to a 20 000 molecular weight, the CAC remained relatively constant. This suggests that the dendritic portion of the copolymer, not the PEG molecular weight, significantly influences the value of the CAC. Upon going from a protected ([G1]-PGLSA-bzld)$_2$-PEG to a deprotected ([G1]-PGLSA-OH)$_2$-PEG, the alcohol-terminated macromolecules have a larger CAC value. It was also observed that the ([G0]-PGLSA-bzld)$_2$-PEG$_{3400,\ 10000,\ \text{and}\ 20000}$ through ([G2]-PGLSA-OH)$_2$-PEG$_{3400,\ 10000,\ \text{and}\ 20000}$ macromolecules had similar CAC values, while the greatest CAC change occurred between the second and third generations. The CAC values decreased from 40, 45, and 30 mM for the ([G2]-PGLSA-OH)$_2$-PEG$_{3400}$, ([G2]-PGLSA-OH)$_2$-PEG$_{10000}$, and ([G2]-PGLSA-OH)$_2$-PEG$_{20000}$ macromolecules to 11, 18, and 14 mM for the ([G3]-PGLSA-OH)$_2$-PEG$_{3400}$, ([G3]-PGLSA-OH)$_2$-PEG$_{10000}$, and ([G3]-PGLSA-OH)$_2$-PEG$_{20000}$ macromolecules, respectively. At the fourth generation, the CAC values further decreased to 5, 4, and 7 mM, respectively, indicative of the strong propensity to aggregate at low concentrations.

Next, preliminary cytotoxicity studies were performed using human umbilical vein endothelial cells (HUVEC), and the IC$_{50}$ values were determined for a protected and deprotected zeroth generation and a deprotected third generation copolymer of one PEG molecular weight. The ([G0]-PGLSA-bzld)$_2$-PEG$_{10000}$, ([G0]-PGLSA-OH)$_2$-PEG$_{10000}$, and ([G3]-PGLSA-OH)$_2$-PEG$_{10000}$ macromolecules were chosen to evaluate cell viability as a function of end groups and generations. From the cell toxicity study, the IC$_{50}$ values were determined to be 3.44×10^{-4}, $>1 \times 10^{-2}$, and 7.15×10^{-3} M for the ([G0]-PGLSA-bzld)$_2$-PEG$_{10000}$, ([G0]-PGLSA-OH)$_2$-PEG$_{10000}$, and ([G3]-PGLSA-

Table 2. MDSC Data for ([Gn]-PGLSA)$_2$-PEG$_{3400,\ 10000,\ \text{and}\ 20000}$ Macromolecules

	3400 g/mol PEG			10 000 g/mol PEG		20 000 g/mol PEG	
dendritic macromolecule	no.	T_m (°C)	T_g (°C)	no.	T_m (°C)	no.	T_m (°C)
OH-PEG-OH		49			62		64
([G0]-PGLSA-bzld)$_2$-PEG	**3a**	44.7		**3b**	54.7	**3c**	57.9
([G0]-PGLSA-OH)$_2$-PEG	**4a**	46.6		**4b**	55.8	**4c**	56.8
([G1]-PGLSA-bzld)$_2$-PEG	**5a**	38.8		**5b**	53.9	**5c**	56.3
([G1]-PGLSA-OH)$_2$-PEG	**6a**	41.9		**6b**	53.4	**6c**	57.2
([G2]-PGLSA-bzld)$_2$-PEG	**7a**	31.4		**7b**	52.1	**7c**	56.1
([G2]-PGLSA-OH)$_2$-PEG	**8a**	36.5		**8b**	52.3	**8c**	53.9
([G3]-PGLSA-bzld)$_2$-PEG	**9a**		−13.6	**9b**	49.9	**9c**	55.3
([G3]-PGLSA-OH)$_2$-PEG	**10a**		−38.2	**10b**	48.9	**10c**	52.3
([G4]-PGLSA-bzld)$_2$-PEG	**11a**		5.3	**11b**	42.5	**11c**	45.3
([G4]-PGLSA-OH)$_2$-PEG	**12a**		−18	**12b**	42.6	**12c**	49.2

Table 3. Critical Aggregation Concentration (CAC) Values for the Macromolecules[a]

	3400 g/mol PEG		10 000 g/mol PEG		20 000 g/mol PEG	
dendritic macromolecule	no.		no.		no.	
([G0]-PGLSA-bzld)$_2$-PEG	**3a**	44	**3b**	47	**3c**	47
([G0]-PGLSA-OH)$_2$-PEG	**4a**	57	**4b**	44	**4c**	50
([G1]-PGLSA-bzld)$_2$-PEG	**5a**	40	**5b**	37	**5c**	42
([G1]-PGLSA-OH)$_2$-PEG	**6a**	45	**6b**	50	**6c**	60
([G2]-PGLSA-OH)$_2$-PEG	**8a**	40	**8b**	45	**8c**	30
([G3]-PGLSA-OH)$_2$-PEG	**10a**	11	**10b**	18	**10c**	14
([G4]-PGLSA-OH)$_2$-PEG	**12a**	5	**12b**	4	**12c**	7

[a] All CAC values are reported as mM concentrations.

OH)$_2$-PEG$_{10000}$, respectively. These results suggest the materials are worthy of additional testing as would be required for in-vivo evaluation.

Although these macromolecules maybe of use for a number of applications, we are particularly interested in cross-linkable macromers as adhesives for the repair of ocular wounds. In a previous report, we described a methacrylated ([G1]-PGLSA-OH)$_2$-PEG$_{3400}$, ([G1]-PGLSA-MA)$_2$-PEG$_{3400}$, for securing a corneal laceration in vitro.[54] This photoactive adhesive was able to seal a 4.1 mm full-thickness corneal laceration when used at polymer weight percents of more than 30% by maintaining pressures well in excess of normal intraocular pressure of ≈15 mmHg. However, at lower polymer weight percents in solution, the adhesive did not perform as well. To determine whether by using larger molecular weight cross-linkable macromers we could reduce the wt % required to secure the wound, we prepared methacrylated G1 derivatives of the PEG 3400, 10 000, and 20 000 g/mol polymers and then performed in-vitro leaking pressure experiments. The methacrylated G1 derivates of the PEG 3400, 10 000, and 20 000 g/mol M_w were prepared by reacting **6a**,**b**,**c** with methacryloyl chloride, as we have reported.[54] For the leaking pressure studies, an enucleated eye with the cornea facing upward was held under a surgical microscope, and a 4.1 mm laceration was made with a keratome blade. This wound was then closed using either three interrupted 10−0 nylon sutures in a standard 3−1−1 suturing configuration or by application of 20 μL of a 5 wt % aqueous solution of the methacrylated G1 macromers and subsequent photo-cross-linking (argon ion laser; 200 mW, 1 s exposures; 50 s total irradiation time; the polymer solution contained ethyl eosin in 1-vinylpyrrolidinone and TEA as photoinitiator and cocatalyst; $N = 3$; for each copolymer tested). Next, saline was injected in the anterior chamber using a syringe inserted through the scleral wall until the repaired laceration leaked and the pressure was recorded. As shown in Figure 3, the mean leaking pressure for the ([G1]-PGLSA-MA)$_2$-PEG$_{3400}$, ([G1]-PGLSA-MA)$_2$-PEG$_{10000}$, and ([G1]-PGLSA-MA)$_2$-PEG$_{20000}$, formulations were <5, 25 ± 6, and 77 ± 28 mmHg, respectively. The control without any sutures leaked at less than 5 mmHg, and the leaking pressure for the control with three interrupted 10−0 nylon sutures was 90 ± 8 mmHg. The data show that by increasing the PEG molecular weight of this triblock linear−dendritic macromolecule, the strength of the adhesive improved when used at 5 wt %.

Conclusion

We have described the high yielding divergent synthesis of hybrid dendritic−linear copolymers based on biocompatible starting materials. The zeroth through fourth generations were obtained in an iterative process of esterification, by an activated monomer, followed by hydrogenolysis reactions in high yields. The success of each reaction was monitored by MALDI−TOF and SEC, where an increase in molecular weight was observed with each successive generation after coupling and a decrease in molecular weight was obtained within the same generation after deprotection. The melting and glass transition temperatures were determined by MDSC, where a range of T_m and T_g was observed for different PEG copolymers at varying generations. The ([Gn]-PGLSA)$_2$-PEG$_{3400}$ macromolecules displayed a T_m up to a deprotected second generation copolymer and a T_g up to a deprotected fourth generation copolymer. The ([Gn]-PGLSA)$_2$-PEG$_{10000\text{ and }20000}$ macromolecules showed only a T_m for all four generations. The CACs values were measured for the ([Gn]-PGLSA)$_2$-PEG$_{3400,\ 10000,\text{ and }20000}$ macromolecules. Generally, the CAC values decreased as the generation number increased. The deprotected copolymers showed a slight increase in the CAC values when compared to the protected zeroth and first generation copolymer for all three PEG cores. Overall, the fluorescence studies indicated that the dendritic portion influenced the CAC values more than the linear portion. HUVECs were used to determine the IC$_{50}$ values of ([G0]-PGLSA-bzld)$_2$-PEG$_{10000}$, ([G0]-PGLSA-OH)$_2$-PEG$_{10000}$, and ([G3]-PGLSA-OH)$_2$-PEG$_{10000}$, and relatively high concentrations of compound are required to observe cell toxicity. Photo-cross-linkable derivatives of the three different molecular weight macromolecules show varying abilities to secure a corneal laceration with the 5 wt % ([G1]-PGLSA-MA)$_2$-PEG$_{20000}$ formulation performing the best. In summary, dendritic macromolecules offer a number of potential advantages as well-defined materials for the preparation of biotechnological reagents, medical devices, and drug delivery vehicles.

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Supporting Information Available: Experimental details. This material is available free of charge via the Internet at http://pubs.acs.org.

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