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Harnessing the Chemical Diversity of the Natural Product Magnolol for the Synthesis of Renewable, Degradable Neolignan Thermosets with Tunable Thermomechanical Characteristics and Antioxidant Activity

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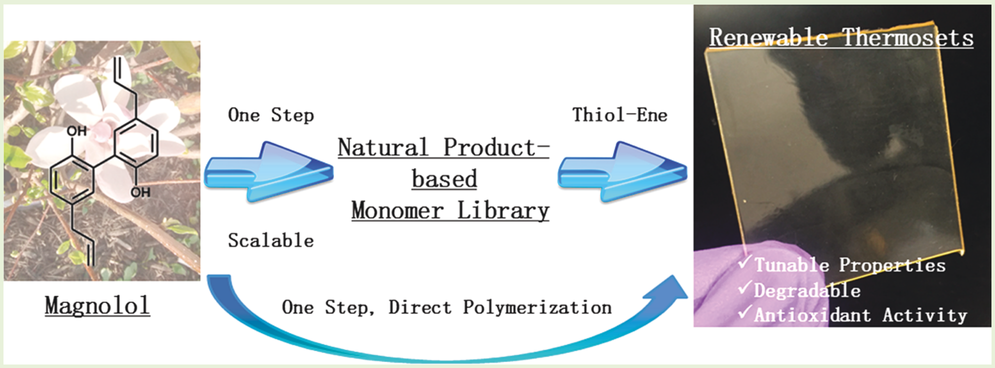
Supporting Information



ABSTRACT: Magnolol, a neolignan natural product with antioxidant properties, contains inherent, orthogonal, phenolic, and alkenyl reactive groups that were used in both direct thermoset synthesis, as well as the stepwise synthesis of a small library of monomers, followed by transformation into thermoset materials. Each monomer from the small library was prepared via a single step functionalization reaction of the phenolic groups of magnolol. Thermoset materials were realized through solvent-free, thiol−ene reactions, and the resulting cross-linked materials were each comprised of thioether and ester linkages, with one retaining the hydrophilic phenols from magnolol, another having the phenols protected as an acetonide, and two others incorporating the phenols into additional cross-linking sites via hydrolytically labile carbonates or stable ether linkages. With this diversity of chemical compositions and structures, the thermosets displayed a range of thermomechanical properties including glass transition temperatures, T_g, 29−52 °C, onset of thermal degradation, T_d, from about 290−360 °C, and ultimate strength up to 50 MPa. These tunable materials were studied in their degradation and biological properties with the aim of exploiting the antioxidant properties of the natural product. Hydrolytic degradation occurred under basic conditions (pH = 11) in all thermosets, but with kinetics that were dependent upon their chemical structures and mechanical properties: 20% mass loss was observed at 5, 7, 27, and 40 weeks for the thermosets produced from magnolol directly, acetonide-protected magnolol, bis(allyl carbonate)-functionalized magnolol, and bis(allyl ether)-functionalized magnolol, respectively. Isolated degradation products and model compounds displayed antioxidant properties similar to magnolol, as determined by both UV−vis and in vitro reactive oxygen species (ROS) assays. As these magnolol-based thermosets were found to also allow for extended cell culture, these materials may serve as promising degradable biomaterials.

■ INTRODUCTION

There is a great interest to incorporate renewable resources into monomers and polymers in order to reduce use of petroleum-based materials, comply with legal and societal standards, and to develop advanced polymeric materials with emergent properties, particularly for engineering or biomedical applications.[1] Concurrently, thiol−ene reactions have become pervasive methods for the development of polymeric materials on account of the inherent chemoselectivity, high yields, and often solvent-free reaction conditions associated with their chemistry.[2] The union of these two areas has been the subject of much attention recently and has resulted in the synthesis of

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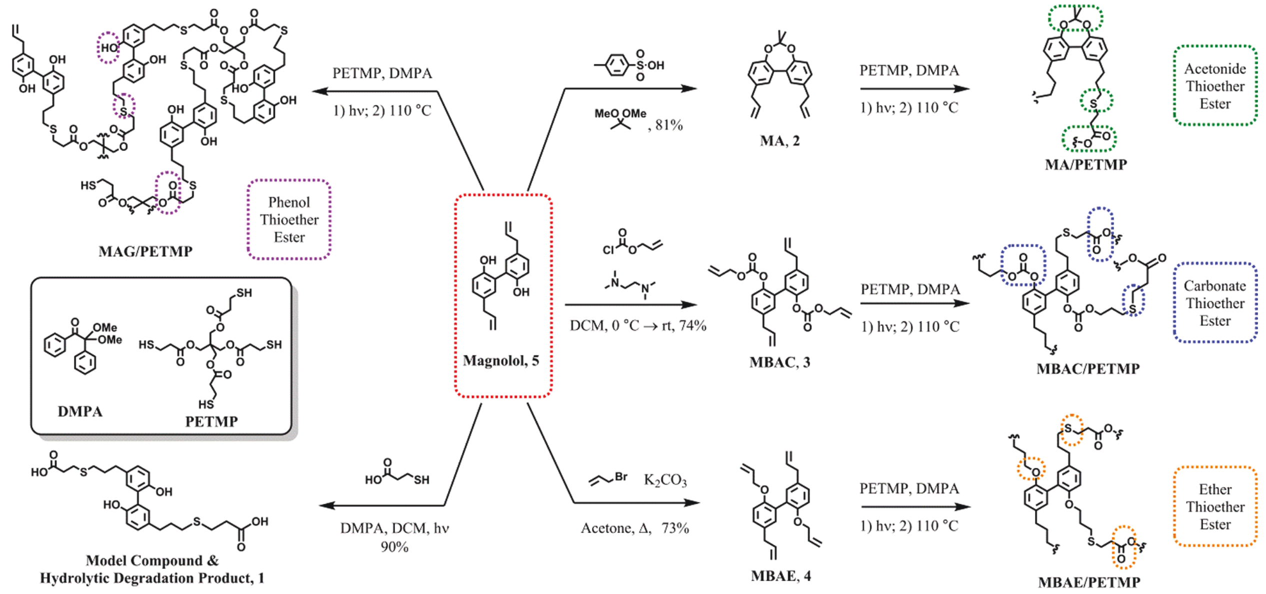
Scheme 1. Synthetic Approach to Magnolol Monomers, Thermosets, and Model Compound



natural product-based monomers from eugenol,[3] isosorbide,[4] limonene,[5] phenolic acids,[6] polyols,[7] and others[1a] en route to polymers via thiol—ene chemistry. There is particular interest in polymers that retain the biological properties of the natural products from which they are made, as is the case of antibacterial networks from linalool and isosorbide.[4c] Natural products and their derivatives that are amenable to thiol—ene polymerization may serve as starting points in the design of polymers that include interesting biological properties. Furthermore, the potential to produce new materials with emergent properties for which there may be no close petrochemical equivalent drives the exploration of new natural products for polymeric materials.

One class of natural products that has been underdeveloped for renewable monomers and polymers is neolignans. Neolignans are structurally diverse natural products biosynthesized through oxidative dimerization of a core phenylpropanoid unit[8] and have been well-studied for their anti-inflammatory and antioxidant properties, among others.[9] They are typically isolated from plant sources, such as the *Magnolia* genus, in which individual neolignans are present up to several weight percent.[9] A recent example of a neolignan-based polycarbonate developed in our lab from honokiol, a natural product containing both phenolic and alkenyl functionalities, showed excellent cell compatibility and cell culturing capabilities.[10] The regioisomeric neolignan, magnolol, was unable to undergo carbonylation of the phenolic groups to establish an analogous linear polycarbonate, likely due to steric constraints. However, the 2,2′-biphenolic structure of magnolol provides opportunities for chemical modification or protection via ring closure to afford tricyclic structures that retain the alkenyl moieties. Magnolol is of particular interest, therefore, not only for potential biological properties, but also in that it is highly functional, allowing for either direct polymerization, or the rapid and scalable syntheses of diverse libraries of both renewable monomers and polymers. Because the antioxidant properties[11] of magnolol involve intriguing redox chemistry of the phenols in the presence of peroxyl radicals,[12] without reliance on the allylic moieties, we designed a strategy whereby the alkenyl moieties could be used in forming a polymer network via thiol—ene chemistry, with variation in the chemistry conducted upon the phenolic groups to allow for monomer and polymer diversification and investigation of the physicochemical, thermomechanical, and biological properties. Based on the polymer design, the phenolic groups may be revealed during hydrolytic degradation and subsequently scavenge radicals in the presence of an oxidative media.

Herein, we present the scalable syntheses of thermoset materials based on magnolol by solvent-free polymerization with a commercially available, multifunctional thiol comonomer through photoinitiated thiol—ene chemistry. The partially biobased, tunable thermosets were thoroughly characterized and differences in the resulting key thermomechanical and hydrolytic degradation properties were rationalized by the structure—property relationships of the magnolol-based monomers. Complementary synthetic-biologic interactions were investigated by cell viability, cell adhesion and proliferation, and radical scavenging and antioxidant assays. These studies have shown promising results for use of magnolol-based thermosets in biomedical applications where tunable polymer properties, including degradation, are desired and where beneficial biological activity of the degrading thermoset might be leveraged without the need for additional small molecule therapeutics.

■ RESULTS AND DISCUSSION

Because the antioxidant characteristics of magnolol are due to the radical scavenging ability of its phenolic groups, our original aim to employ the alkenyl functionalities for thiol—ene copolymerization of magnolol with multifunctional comonomers necessitated a study of its reactivity in a model photoinitiated radical thiol—ene reaction (Scheme 1, lower left). A monofunctional thiol was used for these model studies to be able to monitor the reaction and isolate the products as small molecules for detailed analyses to confirm that the thiol—ene reaction would proceed in high conversion without significant radical scavenging by the phenols that could lead to cyclohexadienone formation and other side reactions. In addition, the monofunctional thiol was selected to be 3-mercaptopropionic acid, specifically, as the bis-

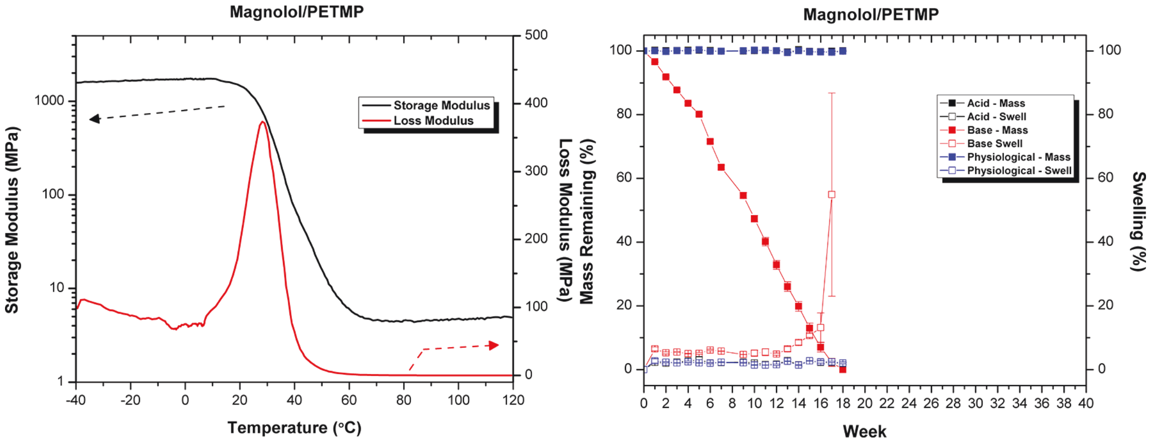


Figure 1. Characterization of thermosets synthesized directly from magnolol and PETMP. Dynamic mechanical analysis exhibiting loss in mechanical properties near room temperature and T_g, as measured by the peak in the loss modulus, E'', at 29 °C (left). Degradation of thermoset pellets ($n = 3$, about 110 mg each), in PBS media (pH = 3.0, 7.4, and 11.0), as measured by mass remaining and swelling extent (right).

(thioetherpropionic acid) adduct **1** and its monothioether analog have the chemical structures of the hydrolytic degradation products from the thiol−ene copolymerization of magnolol with pentaerythritol tetrakis(3-mercaptoproprionate) (PETMP). In the presence of about 0.05 mol % photoinitiator (2,2-dimethoxy-2-phenylacetophenone, DMPA, mp = 70 °C), 2 equiv of 3-mercaptopropionic acid were allowed to undergo reaction with magnolol under ultraviolet irradiation ($\lambda = 352$ nm, 40 W) in deuterated dichloromethane at room temperature. The reaction was monitored by ^{1}H NMR spectroscopy and found to reach 100% conversion at 10 min, with no undesirable byproduct formation noticed (Figure S1). The model reaction was then scaled up from about 100 mg magnolol to several grams in a round-bottom flask with degassed dichloromethane to model the scale at which polymerizations would occur and to yield desirable quantities of this model compound for future studies. At this larger scale but under identical conditions, full alkene conversion required 2 h. The viscous oil was purified by column chromatography (15 → 100% ethyl acetate in hexanes) and concentrated to yield a clear, colorless oil that was then allowed to solidify under atmospheric conditions to afford **1** in 90% yield.

Although the phenols of magnolol did not significantly impede the model thiol−ene reaction in solution, direct thermoset synthesis in the bulk utilizing magnolol as an A_2 monomer and widely available pentaerythritol tetrakis(3-mercaptoproprionate) (PETMP) as a tetra-functional B_4 comonomer encountered challenges. At room temperature, the solid magnolol monomer and photoinitiator did not dissolve or mix well into the viscous thiol comonomer (Figure S2). Attempts to realize bulk polymerization through photoinitiation at room temperature were unsuccessful. However, with manageably low melting points for DMPA and magnolol (mp = 102 °C), the mixture of comonomers and photoiniator were melted at 110 °C in an oven to produce a clear oil that was transferred hot into preshaped molds. The resins were then polymerized in a UV oven (352 nm, 40 W) for 2 h followed by a post-cure at 110 °C for an additional 2 h. Similar to the observed differences in the model reactions performed on small and large scales in solution, thermoset synthesis in bulk performed on scales up to several grams in molds (ca. 0.5 cm thick) necessitated a longer UV exposure for gelation. The resulting clear, colorless magnolol/PETMP (MAG/PETMP) thermosets were stiff but pliable after handling. Notably, attempts to thermally initiate DMPA and yield polymer in the absence of UV irradiation were unsuccessful, even after heating for 10 h at 110 °C; no gelation was observed and solid precipitated from the mixture upon cooling to room temperature.

Initial characterization was performed to determine the thermomechanical properties and chemical resistance under basic, acidic, and physiologically relevant pH at physiological temperature (Figure 1). The MAG/PETMP thermoset was thermally stable (onset of thermal degradation, $T_d = 350$ °C) and showed a modest storage modulus (E' @ 25 °C = 1.10 ± 0.14 GPa). The glass transition temperature, T_g, was slightly above room temperature, as measured by dynamic mechanical analysis (DMA; $T_g = 29 \pm 1$ °C). The thermomechanical properties also appeared to have an effect on the hydrolytic stability in that, although hydrophobic, at physiological temperature, the MAG/PETMP thermoset immediately swelled and began hydrolytic degradation in basic PBS media (pH = 11.0; Figure 1b). Full degradation occurred by 18 weeks under basic conditions, while samples exposed to acidic and pH = 7.4 conditions displayed little swelling and no degradation over the course of the study. It is expected that degradation occurred through hydrolysis of the ester moieties of the repeat units derived from PETMP.

To expand upon the performance of this initial thermoset, a small library of magnolol-based monomers was desired, wherein cross-linked polymer networks would be synthesized via thiol−ene polymerization and the resulting thermomechanical and degradation properties could be controlled through the initial monomer design and further exploited in material applications without requiring extensive synthetic procedures or processing. With this in mind, magnolol was functionalized in three separate reactions to yield monomers containing di- or tetra-alkenyl groups on multigram (up to decagram) scales and >70% yields. Magnolol acetonide, MA (**2**), a tricyclic, difunctional A_2 monomer containing an acid-labile phenolic protecting group was synthesized by organic acid-catalyzed acetonide formation with 2,2-dimethoxypropane in 81% yield through a procedure adapted from Schinazi and co-workers.[13] Of the tetra-functional A_4 monomers, magnolol bis(allyl carbonate), MBAC (**3**), intended to serve as a base-labile monomer, was synthesized in 74% yield by addition of allyl

Table 1. Magnolol Thermosets Showing Tunable Thermomechanical Properties by TGA, DSC, DMA, and Tensile Testing

thermoset	T_d (°C)	T_g DSC (°C)	T_g DMA[a] (°C)	E'[b] (GPa)	E_r[c] (MPa)	Young's modulus (MPa)	ultimate Strength (MPa)	strain (%)
MAG/PETMP	350	23	29	1.10 ± 0.14	4	386 ± 65	28 ± 5	68 ± 25
MA/PETMP	359	30	33	1.25 ± 0.09	7	384 ± 37	26 ± 3	131 ± 29
MBAC/PETMP	293	46	52	1.33 ± 0.02	38	735 ± 48	48 ± 2	18 ± 6
MBAE/PETMP	361	47	52	1.21 ± 0.13	41	794 ± 91	50 ± 2	20 ± 7

[a] As measured by the peak of the loss modulus, E''. [b] Storage modulus measured at 25 °C. [c] Rubbery modulus measured at 100 °C. DMA measurements: T_g, E', and E_r; Tensile testing: Young's modulus, ultimate strength, and strain.

chloroformate to a solution of magnolol in dichloromethane and tetramethylethylenediamine at 0 °C following a similar procedure from previously reported work in our group,[7] while magnolol bis(allyl ether), MBAE (**4**), a hydrolytically stable monomer, was prepared in 73% yield using slight excess of allyl bromide with potassium carbonate in acetone under reflux (Scheme 1). Although we performed these syntheses in batches to yield about 15 g monomer, the straightforward synthetic procedures to access the various chemically functionalized monomers from magnolol are readily scalable.

Similar to the direct synthesis of thermosets from magnolol, cross-linked thermosets were achieved through reaction of **2**, **3**, or **4** with PETMP using stoichiometric equivalences of functional group and in the presence of 1 wt % DMPA/thiol under UV irradiation followed by a post-cure at 110 °C. Although magnolol is a beige solid in its commercial form (>98% purity), the purified magnolol-based monomers **2**−**4** were transparent, colorless oils, and mixed well with comonomer and photoinitiator. No heating was required to mix or transfer the resin to the molds and the solvent-free polymerizations ultimately yielded transparent films. Comparison of FTIR spectra of the films to the spectra of the monomers did not indicate the presence of free thiol (S−H stretch, ∼2600 cm^{-1}; Figure S3) and showed consumption of alkene (str. C═C stretch, ∼1650 cm^{-1}, C−H bend, ∼900 cm^{-1}; Figure S4) in the thermosets.

As was expected, the thermoset compositions containing monomers with higher functionality (and a greater expected degree of cross-linking), MBAC/PETMP and MBAE/PETMP, displayed higher T_gs by DSC (46 and 47 °C) as compared to the difunctional magnolol-based thermosets, MA/PETMP (30 °C) and MAG/PETMP (23 °C; Table 1). Interestingly, the same trend was not observed thermogravimetrically, as MAG/PETMP, MA/PETMP, and MBAE/PETMP showed significantly higher thermal stabilities−onsets of thermal degradation at 350, 359, and 361 °C, respectively, compared to MBAC/PETMP, which began degrading at about 295 °C (Figure S5). The MBAC/PETMP thermoset did display a second thermal degradation event near about 360 °C and a final thermal degradation event near about 425 °C, as did the thermosets synthesized from monomers **2**, **4**, and **5**. All other variables equivalent and given the high conversion of monomers to thioether, we hypothesize this difference in thermal stability among the polymers is a result of a more thermally labile carbonate linkage in MBAC/PETMP. Similar degradation temperatures have been observed for other polycarbonate cross-linked networks synthesized via thiol−ene chemistry.[4a,7]

With these results, we decided to further investigate the fundamental materials properties and potential performance of the thermosets relevant to biomedical materials by DMA (dry and submersion) and tensile testing analyses. The same T_g trend was observed by DMA as for DSC, whereby the MA thermoset showed a transition at 33 °C (as measured by the peak of the loss modulus, E''), just below physiological temperature, while MBAC and MBAE thermosets were about 20 °C higher (Table 1 and Figure 2). All T_gs were higher than the thermoset synthesized directly from magnolol (MAG/PETMP, 29 °C). Although containing the same degree of cross-linking, assuming a complete thiol−ene reaction, the slightly higher glass transition of MA/PETMP may be attributed to the rigidity of this monomer with its tricylic structure compared to the thermoset from the magnolol monomer. Given the differences in T_g, there is a greater than 1000 MPa difference in the storage moduli, E', between MBAE/PETMP (1250 MPa) or MBAC/PETMP (1210 MPa) and MA/PETMP (100 MPa) or MAG/PETMP (110 MPa) at 37 °C. At physiological temperature, the latter two thermosets are at the midpoints of their glass transition regions. However, these results are in contrast with the material properties at room temperature, which show little change in E' for all four thermosets as they are still well in the glassy region. Additionally, the degree of cross-linking was noticeably higher in A_4B_4 MBAC and MBAE thermosets compared to the A_2B_4 MA/PETMP and MAG/PETMP thermosets, as measured by the rubbery modulus, E_r, in accordance with the theory of rubber elasticity.[14] These differences, although seemingly modest at room temperature, may have large effects under physiological conditions.

The effects of compositional and structural differences of the thermosets on mechanical properties were also revealed by tensile testing. Each thermoset displayed a modest ductile behavior at a strain rate of 5 mm/min; however, MA/PETMP showed a drastically higher strain at failure and lower modulus and ultimate strength than the more highly cross-linked counterparts. The mechanical behavior of MAG/PETMP was similar to MA/PETMP, with the exception of having early strain at break. In addition to the chemical compositional differences across this series of thermosets, the relatively lower cross-linking density for the MA- and MAG-derived A_2B_4 materials and their associated lower T_g values (vs the A_4B_4-based structures) may have contributed to the mechanical properties effects due to the testing conditions (room temperature ≈27 °C) being near their glass transitions, but still 25 °C lower than thermosets from MBAC and MBAE (Figure 2c).

As the dry thermoset properties may be altered in response to plasticization under physiological conditions, in situ matrix relaxation DMA was also performed in PBS at 37 °C. The wet material properties may yield additional insight into the ultimate material performance in a biological application. The change in E' over time followed a similar trend to the T_g of the dry thermosets. The results were fitted to dual-exponential decay models which yielded average storage moduli lifetimes, τ, of 8 min (MAG/PETMP), 10 min (MA/PETMP), 27 min (MBAC/PETMP), and 41 min (MBAE/PETMP) (Figure S6). The dual-exponential fit accounts for the response of the

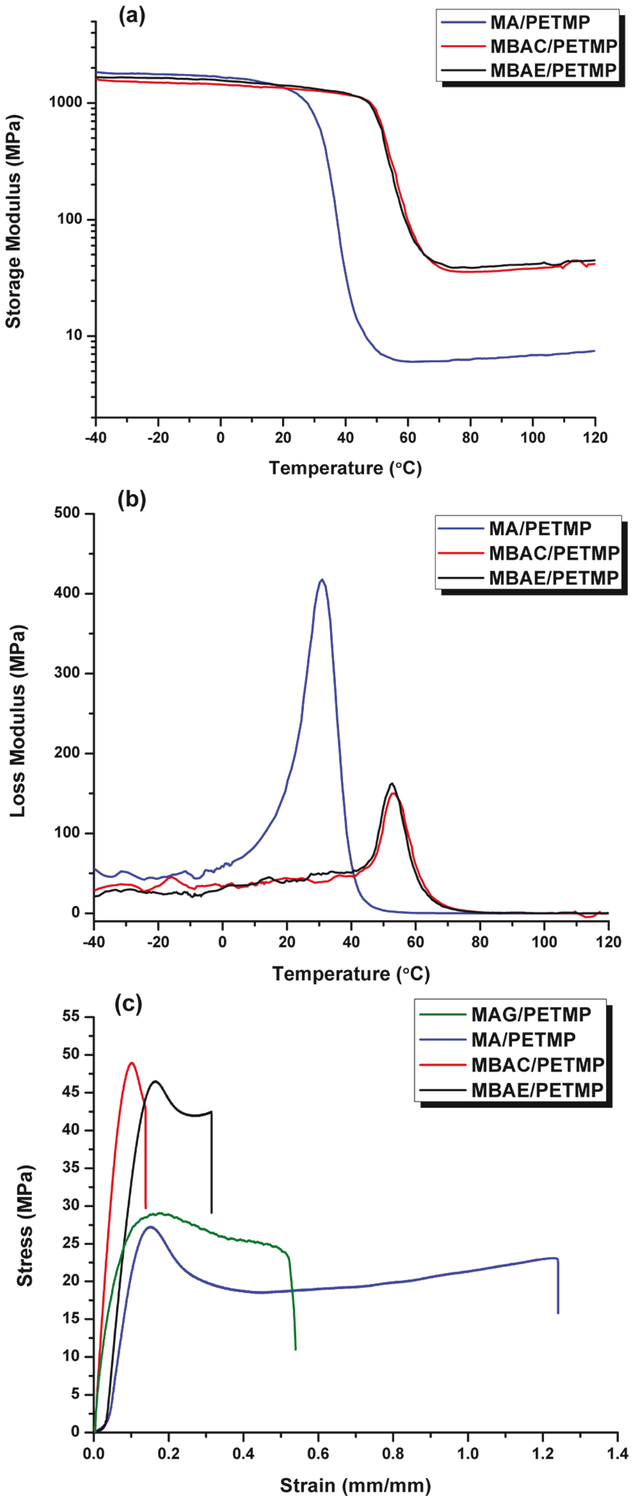


Figure 2. Thermomechanical results comparing MAG/PETMP, MA/PETMP, MBAC/PETMP, and MBAE/PETMP. (a) Storage moduli, E', showing distinct differences above physiological temperature, particularly in rubbery modulus, E_r, measured at 100 °C. (b) T_gs as measured by the peak in the loss modulus, E''. (c) Tensile testing performed at room temperature, 5 mm/min strain rate.

thermosets to both temperature change (room temperature to physiological temperature, Δ12 °C) followed by solvent plasticization of the thermosets. The overall loss in mechanical properties, as measured by the change in the storage moduli, $\Delta E'$, followed an inverse relationship, whereby MAG/PETMP had the largest loss over time ($\Delta E'$ = 1200 MPa), while MBAE/PETMP showed the least significant change ($\Delta E'$ = 400 MPa). MA/PETMP, similar to MAG/PETMP, displayed a large and quick change in mechanical properties when placed under physiological conditions and we anticipated that swelling and subsequent hydrolysis might occur most readily in this thermoset (compared to MBAC and MBAE thermosets) at a rate comparable to the degradation observed for MAG/PETMP. Interestingly, the surface chemistry did not appear to have a substantial effect on the plasticization and loss in mechanical properties upon submersion in PBS. Furthermore, there was not a significant effect of the chemical composition on the water contact angles, other than the phenol-containing MAG/PETMP was slightly more hydrophilic (θ_{water} = 75°) than the thermosets having the phenolic groups consumed (θ_{water} = 88−94°; Figure S7).

Previous reports have examined renewable resources in degradable polymers to generate a biological response as the polymer degrades. Salicylic acid-based poly(anhydride ester)-s[15] and polyesters,[16] as well as ferulic acid-based poly-(carbonate amide)s[17] and poly(anhydride ester)s[18] have been studied for the potential antimicrobial or antioxidant properties of their degradation products. Salicylic acid-based polymers have similarly been examined as an antioxidant coating in drug eluting stents,[19] while materials based on naturally occurring biopolymers, such as modified dextran polymers[20] and melanin nanoparticles,[21] have been utilized to efficiently scavenge hydrogen peroxide and other reactive oxygen or nitrogen species. Polymer peroxide scavenging may also be leveraged as a cue for polymer degradation and release of therapeutic.[20,22] As these polymers and others[23] often show antioxidant properties as a result of the inherent radical scavenging ability of phenol moieties, functionalized magnolol thermosets which hydrolytically degrade to yield a phenol may similarly scavenge radicals and protect cells from oxidative stress. Radical scavenging may be particularly advantageous as protection in the local cellular environment from the response of the host immune system to a biomaterial. The cross-linking and modest hydrophobicity inherent to these magnolol-derived polymer networks should also result in a slow, controllable degradation and a sustained release of the magnolol degradation products.

Degradation studies were again performed in acidic, basic, and physiological pH PBS media at physiological temperatures but with samples from the phenolic-functionalized thermosets, MA/PETMP, MBAC/PETMP, and MBAE/PETMP (Figure 3). MA/PETMP began degrading within 2 weeks under basic conditions, had less than 50% mass remaining by week 12 and was fully degraded by week 30. The MBAC/PETMP thermoset showed only 5% degradation at week 15, while MBAE/PETMP did not exhibit significant mass loss until after week 17 under identical conditions. Visually, the pellets display distinct differences both in size and surfaces even by week 10 (Figure S8). There was no significant degradation observed for thermosets under acidic or neutral conditions during this time period; MA/PETMP had about 96% mass remaining under acidic and neutral conditions at 27 weeks, while MBAC and MBAE thermosets still had about 99% mass remaining. The <4% mass loss from MA/PETMP under acidic or neutral conditions may result, in part, from hydrolysis of the acetonide moiety that releases acetone but preserves the cross-linked polymer network. The stark difference in degradation behavior is likely a result of differences in the thermal response and

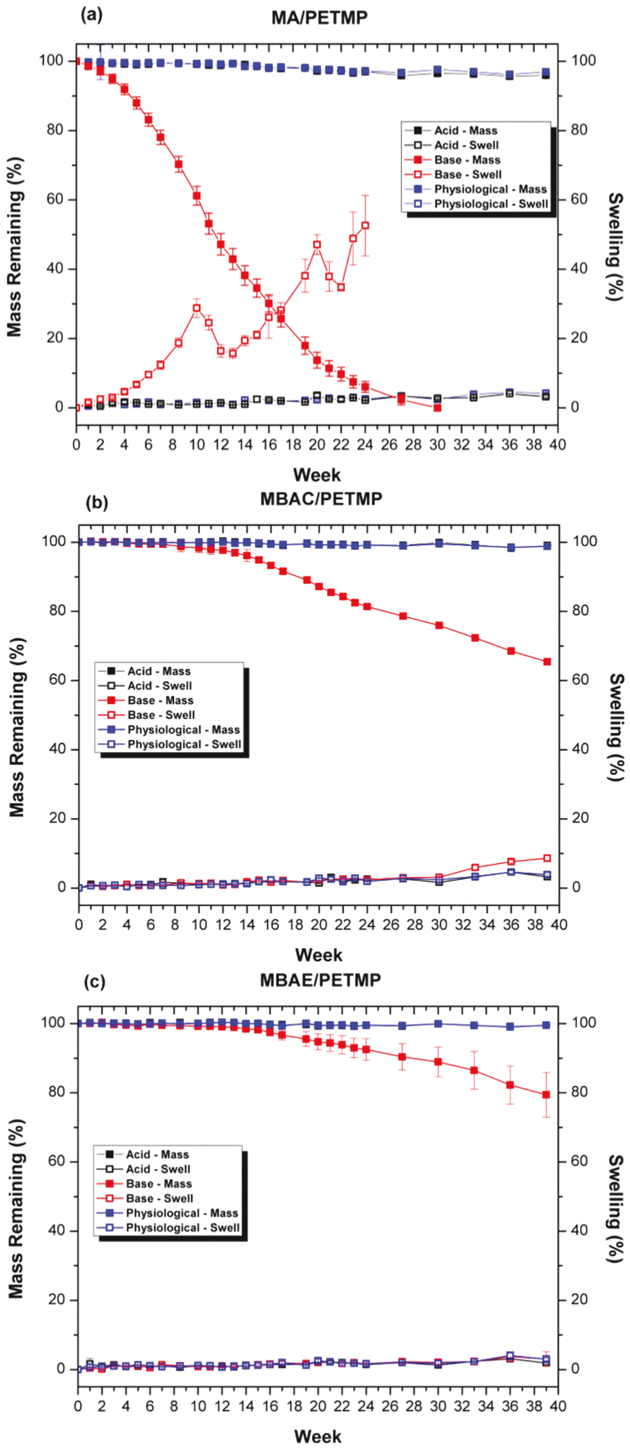


Figure 3. Hydrolytic degradation and swelling in PBS (a) MA/PETMP, (b) MBAC/PETMP, and (c) MBAE/PETMP under basic, acidic, and physiological pH at 37 °C.

plasticization of the polymers at physiological temperatures that initially leads to basic hydrolytic degradation of the pentaerythritol ester backbone. This hypothesis is supported by the results obtained from submersion DMA experiments and the observed T_gs of the dry materials.

We turned our attention toward translation of magnolol biological properties to the cross-linked networks. As a proof of principle, the radical scavenging ability of magnolol and a model degradation compound **1**, which would result from complete hydrolysis of MA/PETMP, MAG/PETMP, and MBAC/PETMP, were assessed using the DPPH$^{\bullet}$ assay. Results in Figure 4 show that the model degradation compound contained similar radical scavenging ability to the unmodified natural product. Additionally, as the thermosets degraded, the solubilized degradation fractions were expected to contain analogous degradation products with phenol moieties that scavenge radicals. Samples from the basic degradation of thermosets were, therefore, prepared in the same manner as magnolol and model degradation compound **1** after lyophilization. DPPH$^{\bullet}$ reduction exhibited a linear relationship to percent mass loss from both MAG/PETMP and MA/PETMP thermosets, while control samples from thermosets in which no degradation occurred showed no radical scavenging ability. In light of the controllable degradation of these samples, it is hypothesized that the release of radical scavenging degradation products could be used for the steady delivery of antioxidant supplementary to, or in replacement of, any additional therapeutic. It should be noted that for samples in which hydrolytic degradation would not lead to a phenol, such as the thermosets from **4**, there

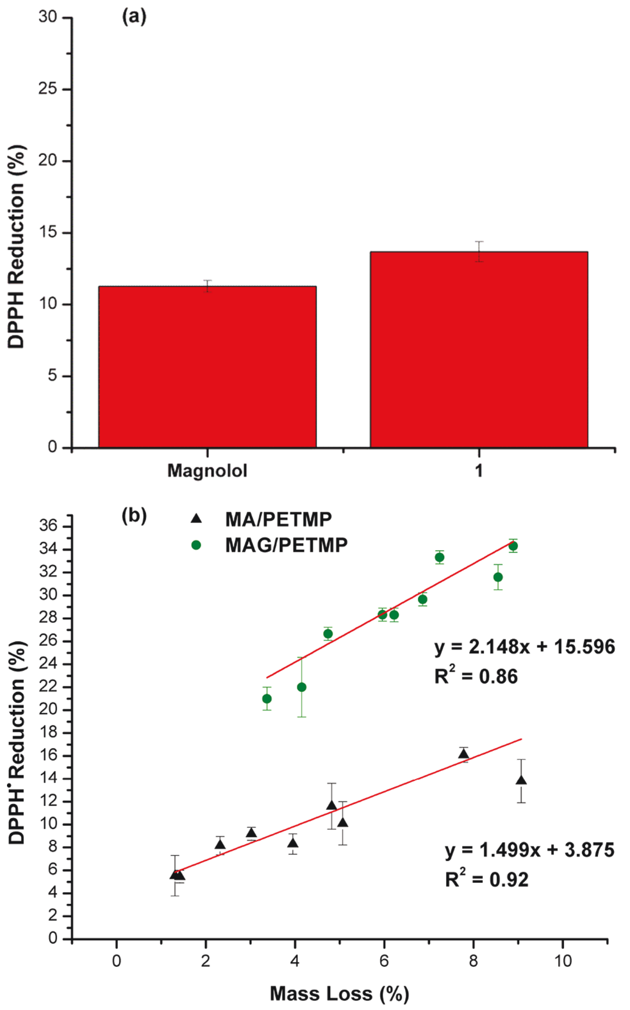


Figure 4. Radical scavenging by DPPH$^{\bullet}$ reduction. (a) Model degradation compound, **1**, compared to known natural product antioxidant, magnolol at working concentrations of 0.12×10^{-4} M. (b) Radical scavenging of MAG/PETMP and MA/PETMP degradation products compared to mass loss from the thermosets.

should be no significant antioxidant activity when comparing to MAG/PETMP or MA/PETMP for the same mass loss from the respective thermosets. DPPH$^\bullet$ reduction from MBAE/PETMP samples does not rise above background levels, however, both MBAE/PETMP and MBAC/PETMP degraded at sufficiently slower rates under basic conditions than either MA/PETMP or MAG/PETMP (vide supra) and, thus, samples from each thermoset could not be compared on an equivalent mass loss basis.

The effects of the materials on cells were evaluated in a series of studies in vitro. To demonstrate antioxidant activity, cytotoxicity assays were first performed on the polymer films and model degradation product, **1** (Figure S10). All thermosets showed high cell viability, and the cells also experienced good viability in the presence of **1** at mM concentrations for MC3T3 fibroblasts and coronary venular endothelial cells (CVECs), which we have previously used to investigate poly(honokiol carbonate) as a culture surface for biomedical applications.[10] In order to determine cell adaptability to the thermosets over longer periods of time, CVECs were plated on thin films of the polymers and media was replaced every 2−3 days to allow for culturing up to one month. After cell fixation, actin fibers and focal adhesions were stained with phalloidin and appropriate antibody-fluorophore conjugates, followed by imaging with confocal microscopy. The CVECs maintained a monolayer, showed good proliferation and adhesion to the thermosets, and were similar to the cell on the control surface at both short and long time periods (Figure 5).

Antioxidant activity of **1** was then assessed using a reactive oxygen species (ROS) assay performed with *tert*-butyl hydroperoxide (tBHP) in the presence of CVECs. Increasing concentrations of the model compound resulted in a general reduction of the ROS (Figure 6), however, the reduction was not as strong as that of magnolol (positive control) at a similar concentration: 10% reduction versus 20% reduction in ROS. Although not essential for radical scavenging, the allylic moieties of magnolol may also participate in scavenging radical species. Accounting for all radical scavenging moieties in the substrates may explain this difference between **1** and magnolol, however, the specific roles of the alkene and carboxylic acid moieties in the test compounds were not further investigated for these in vitro studies.

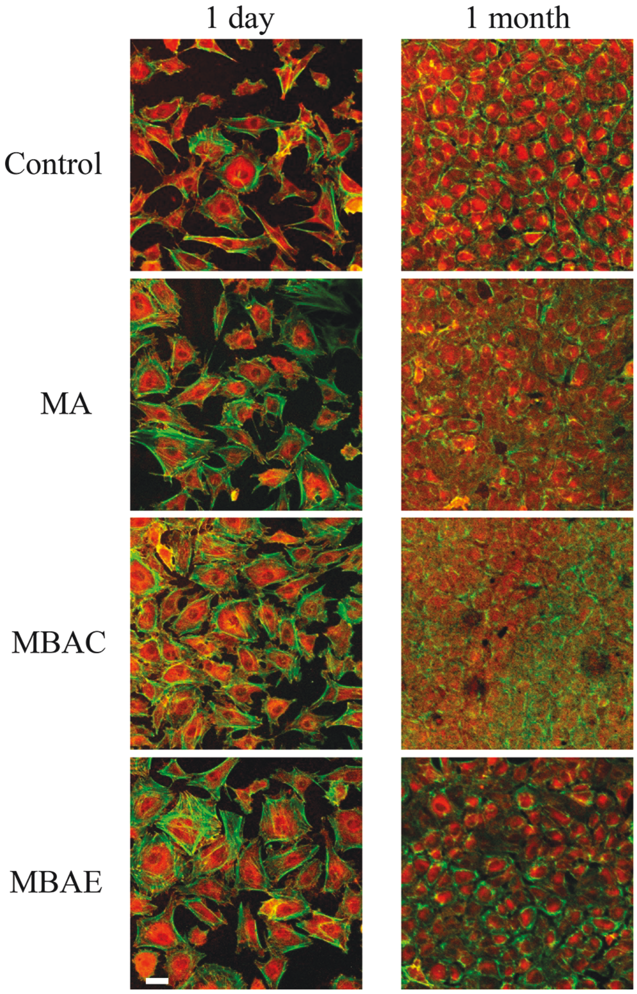


Figure 5. Laser scanning confocal microscopy of bovine coronary venular endothelial cells (CVECs) on magnolol-based thermosets after 1 day (left) and 1 month (right). Cells were cultured on the polymer or gelatin (control) coated glass surface and fixed with 4% paraformaldehyde. Cytoskeleton and focal adhesions were visualized by costaining with phalloidin-Alexa488 (green) and vinculin antibody with Alexa647-conjugated secondary antibody (red). Scale bar 30 μm.

■ CONCLUSIONS

A scalable, straightforward, and generally applicable route was developed for the production of renewable materials having a breadth of thermomechanical properties, hydrolytic degradation profiles and biological responses, each deriving from the neolignan magnolol—a previously unexplored natural product for polymer syntheses. The judicious choice of the highly functional starting material allowed for both direct thermoset synthesis and the development of a small library of monomers for solvent-free thiol−ene chemistry. The thermosets showed tunable thermomechanical properties and hydrolytic degradation rates that were influenced by the monomer design. Knowledge of the structure property relationships led to leveraging of the tunable thermomechanical properties in order to take advantage of the well-known, inherent radical scavenging and antioxidant properties of the natural product from which the polymer networks were synthesized. While no thermosets showed degradation under acidic and physiological pH, at basic pH, hydrolytic degradation occurred with drastically different rates on account of the thermomechanical properties and relative degree of cross-linking between the thermosets. Degradation products isolated from the thermosets synthesized from magnolol acetonide and magnolol displayed radical scavenging by the DPPH$^\bullet$ assay. These results were further corroborated by in vitro antioxidant assays with endothelial cells and a model degradation compound. This work lays a foundation for neolignan-based polymers wherein inherent biological properties of the natural product may be leveraged in degradable biomaterials and where straightforward, scalable chemical transformations are used to control the structure−property relationships for novel, biobased polymers. Although our initial interest in exploring these materials was directed toward degradable materials for use in biomedical applications, the relative hydrolytic stabilities highlight their potential in long-term implantable device technologies and also the possibility to pursue broader utility toward bioplastics that have promise to reduce environmental impact. Ongoing work is investigating the ability to conduct 3D printing for fabrication of intricate objects and also expanding the

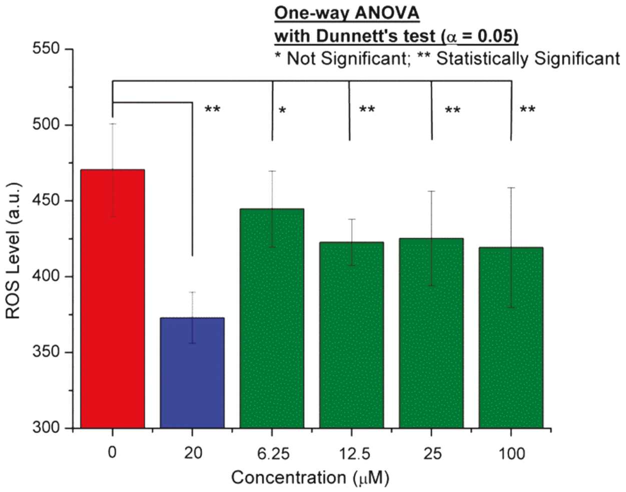


Figure 6. ROS reduction assay with CVECs in 1% FBS and 5 μM tBHP, comparing negative control (red), magnolol (positive control, blue), and model degradation compound, (**1**) (green). Data are presented as mean ± standard deviation. Significance respect to negative control was evaluated using both one-way ANOVA with Dunnett's post hoc test ($\alpha = 0.05$) and with Welch's t-test ($p < 0.05$).

degradation conditions to include enzymatic, microbial and other biodegradation studies.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.biomac.8b00771.

> Experimental methods, spectroscopic and thermomechanical characterization data, as well as hydrolytic degradation images and cell viability assays (PDF).

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Notes

The authors declare no competing financial interest.

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