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Polymeric Prodrug

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POLYMERIC PRODRUG

The presently-disclosed subject matter includes compounds that comprise an initiator and an active agent that is covalently bonded to the initiator through a ring-opening polymerization process, an atom-transfer radical polymerization process, a Michael addition reaction, or a ring-opening metathesis polymerization process. In some embodiments the active agent includes simvastatin. The presently-disclosed subject matter also includes methods for making the compositions and methods for using the compositions to treat tissue wounds.

Abstraction

The presently-disclosed subject matter includes compounds that comprise an initiator and an active agent that is covalently bonded to the initiator through a ring-opening polymerization process, an atom-transfer radical polymerization process, a Michael addition reaction, or a ring-opening metathesis polymerization process. In some embodiments the active agent includes simvastatin. The presently-disclosed subject matter also includes methods for making the compositions and methods for using the compositions to treat tissue wounds.

10 Claims, 9 Drawing Sheets
References Cited

OTHER PUBLICATIONS


* cited by examiner
Figure 1
Figure 2

Figure 3
Figure 6
Figure 7

Figure 8

Cumulative Drug Release (mg)

Days

- 1 M NaOH
- PBS
Figure 11

![Graph showing bone area and newly formed mature bone percentages.

Figure 12

Chemical structures and reactions:
- Tbd
- Simvastatin
- Molecule with a bond reaction indicated
- MPEG-simvastatin diblock

Chemical formulas and reactions:
- Molecule compositions and reactions indicated with chemical structures.
Figure 13A

Figure 13B
POLYMERIC PRODRUG

RELATED APPLICATIONS

This Application claims the benefit of U.S. Provisional Patent Application No. 61/885,915, filed Oct. 2, 2013, the entire disclosure of which is hereby incorporated by reference.

GOVERNMENT SUPPORT

This invention was made with government support under grant number DGE-0653710 awarded by National Science Foundation (NSF). The government has certain rights in the invention.

TECHNICAL FIELD

The present invention generally relates to polymeric prodrugs. In particular, certain embodiments of the present invention relate to polymer compounds that degrade to release an active agent.

BACKGROUND

The need to treat bone defects resulting from age-related degenerative disease, trauma, and reconstructive surgery continues to grow at a significant rate. An estimated 2.2 million bone grafting procedures are performed annually worldwide at a cost of $2.5 billion. Considering the "graying" of the U.S. population, in which the percentage of persons 55 years and older is expected to nearly double over the next 30 years, tremendous societal impact is inevitable. Consequently, the market for orthopedic biomaterials will continue to expand.

Autologous (i.e., autografts) and cadaveric (i.e., allografts) bone are the most commonly used grafting materials to treat bone defects. Each, however, has drawbacks related to morbidity associated with a second surgical procedure for autografts and the potential for disease transmission with allografts. These observations have led to development of synthetic materials and tissue engineering approaches for use in bone regeneration. Bone graft substitutes have used materials of natural and synthetic origin. Ongoing developments focus on enhancing biological activity, such as by incorporating stem cells (e.g., mesenchymal stem cells) and growth factors (e.g., bone morphogenetic protein (BMP) 2) into bone graft substitute materials.

Synthetic biodegradable polymers are also used for drug delivery and to aid in tissue regeneration. Candidate materials include polyanhydrides, polylactides, polycarbonates, and polyorthoesters. The majority of resorbable synthetic polymers utilized for drug delivery and tissue engineering belong to the polyester family, such as polylactic acid), poly(lactic acid), and poly(lactic-co-glycolic acid) (PLGA). These materials are relatively biocompatible, can degrade by the hydrolytic cleavage of ester bonds, and have degradation and mechanical properties that can be tailored by changing monomer ratio. Their degradation products of glycolic and lactic acid are metabolically removed from the body by conversion to carbon dioxide and water in the Krebs cycle. Other common polyesters include poly(e-caprolactone), polyvalerolactone, polydioxanone, and their blends and copolymers.

Furthermore, although certain drugs are intended for systemic therapy, many are most effective if targeted to or placed within a specific site. To this end, drugs are routinely encapsulated in polymers. Entrapment in a solid matrix protects the molecules from environmental effects, and controlled release can be achieved. Persistent challenges, however, include instability of encapsulated drugs, incomplete release, and initial burst.

Therefore, since cells and tissues require exposure to bioactive agents at particular concentrations and doses for certain durations, limited release kinetics is a shortcoming of many drug delivery systems in regenerative medicine. Consequently, some have attempted to develop drugs conjugated to polymers to extend release duration. Among others, water soluble polymers, such as poly(ethylene glycol), polyllysine, polyglutamic acid, and N-(2-hydroxypropyl)methacrylamide (HPMA) have been used for this purpose. With these systems, drugs are attached as pendants linked to the polymeric backbone via ester, amide, and hydrazone bonds. Depending on the spacer molecule chosen, drug release can be prolonged until cleavage in a desired environment, such as pH-sensitive release in a lysosome. Other applications include targeting to specific cells and prolonging circulation time by shielding the drug from degradative enzymes and preventing opsonization. The number of molecules (i.e., payload) that can be attached to the backbone, however, is limited by the number of functional groups required for conjugation.

Hence, there remains a need for degradable compositions and compounds for treating tissue wounds, including bone tissue wounds, that can release drugs at a wound site in a controlled manner. There also remains a need for such compositions and compounds whose payload is not limited by the number of functional groups present on a polymer backbone.

SUMMARY

The presently-disclosed subject matter meets some or all of the above-identified needs, as will become evident to those of ordinary skill in the art after a study of information provided in this document.

This summary describes several embodiments of the presently-disclosed subject matter, and in many cases lists variations and permutations of these embodiments. This summary is merely exemplary of the numerous and varied embodiments. Mention of one or more representative features of a given embodiment is likewise exemplary. Such an embodiment can typically exist with or without the feature(s) mentioned; likewise, those features can be applied to other embodiments of the presently-disclosed subject matter, whether listed in this summary or not. To avoid excessive repetition, this summary does not list or suggest all possible combinations of such features.

In some implementations of the presently-disclosed subject matter, a compound is provided that comprises an initiator and an active agent that is covalently bonded to the initiator through a ring-opening polymerization process, an atom-transfer radical polymerization process, a Michael addition reaction, or a ring-opening metathesis polymerization process. In some implementations the initiator comprises one or more hydroxyl groups configured to react and bond to the active agent, and exemplary initiators can include methoxy(polyethylene glycol) (mPEG). In some implementations the active agent includes a lactone group, and exemplary active agents include statins, such as simvastatin. In this regard, in some implementations the compounds include a molar ratio of the initiator to the active agent is about 1:1 to about 1:100.

Therefore, since cells and tissues require exposure to bioactive agents at particular concentrations and doses for certain durations, controlled release can be achieved. Persistent challenges, however, include instability of encapsulated drugs, incomplete release, and initial burst.

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In some implementations of the presently-disclosed subject matter, a compound is provided that comprises an initiator and an active agent that is covalently bonded to the initiator through a ring-opening polymerization process, an atom-transfer radical polymerization process, a Michael addition reaction, or a ring-opening metathesis polymerization process. In some implementations the initiator comprises one or more hydroxyl groups configured to react and bond to the active agent, and exemplary initiators can include methoxy(polyethylene glycol) (mPEG). In some implementations the active agent includes a lactone group, and exemplary active agents include statins, such as simvastatin. In this regard, in some implementations the compounds include a molar ratio of the initiator to the active agent is about 1:1 to about 1:100.
In some implementations the compounds are a polymer that includes a linear or a branched structure. In some instances the compounds can include a molecular weight of about 500 Da to about 80 kDa. Exemplary compounds can further comprise a targeting agent that is selective for a target substance.

In some implementations the compound is a copolymer, and in specific embodiments the compound is a diblock copolymer. For instance, certain embodiments of the present compounds include the formula:

\[
\begin{align*}
\text{H} - \text{O} - \text{O} - & \text{OH} \\
(\text{m}) & \text{OH} \\
\text{O} - & \text{OH} \\
\text{O} - & \text{OH} \\
\text{O} - & \text{OH} \\
\end{align*}
\]

wherein \( n \) is about 1 to about 44 and \( m \) is about 1 to about 258.

The presently-disclosed subject matter also provides methods for making a compound. In some implementations the methods for making a compound include providing an initiator, mixing an active agent with the initiator to form a mixture, adding a catalyst to the mixture, and then reacting the mixture via a ring-opening polymerization process to form a compound that includes the initiator and the active agent. In some implementations the catalyst is selected from a metallic catalyst, an enzymatic catalyst, an organic catalyst, and combinations thereof.

Further still, the presently-disclosed subject matter provides methods for treating a wound in a subject with the presently-disclosed compounds. In some implementations, the methods for treating a wound comprise administering a composition to the wound of a subject, the composition including an initiator and an active agent that is covalently bonded to the initiator through a ring-opening polymerization reaction to form poly(ethylene glycol)-block-poly(simvastatin) (loaded).

Additional features and advantages of the present invention will also become evident to those of ordinary skill in the art after a study of the description, figures, and non-limiting examples in this document.

**DESCRIPTION OF THE DRAWINGS**

Illustrative aspects of embodiments of the present invention will be described in detail with reference to the following figures wherein:

FIG. 1 includes a schematic diagram showing a ring-opening polymerization reaction between simvastatin and poly(ethylene glycol) methyl ether (mPEG) that yields a mPEG-poly(simvastatin) diblock copolymer.

FIG. 2 includes a graph showing molecular weights of mPEG-polysimvastatin copolymers analyzed by gel permeation chromatography (GPC).

FIG. 3 includes a graph showing the molecular weights of mPEG-polysimvastatin copolymers that were synthesized at different temperatures as a function of time.

FIG. 4 includes a graph showing the polydispersity (PDI) of mPEG-polysimvastatin copolymers that were synthesized at different temperatures as a function of time.

FIG. 5 includes a graph showing a \(^1\)H-NMR analysis of a mPEG-polysimvastatin copolymer.

FIG. 6 includes a graph showing fourier transform infrared (FTIR) spectra of (a) mPEG, (b) simvastatin, (c) a simvastatin and mPEG blend (100:1), and (d) a purified mPEG-polysimvastatin copolymer.

FIG. 7 includes a schematic diagram showing a hydrolytic degradation mechanism of a mPEG-poly(simvastatin) copolymer that results in the release of simvastatin molecules.

FIG. 8 includes a graph showing the change in absorbance of a degrading mPEG-simvastatin copolymer as a function of time in 1 M NaOH and in phosphate buffered saline (PBS).

FIG. 9 includes images showing histological differences four weeks post-implantation in woven bone layers that were been treated with either no drug (blank) or with an embodiment of the present simvastatin-loaded compounds (loaded).

FIG. 10 includes a graph showing the thickness of woven bone layers formed over calvaria that were treated with either no drug (blank) or with an embodiment of the present simvastatin-loaded compounds (loaded).

FIG. 11 includes a graph showing the new bone area and percentage of new formed mature bone formed over calvaria that were treated with either no drug (blank) or with an embodiment of the present simvastatin-loaded compounds (loaded).

FIG. 12 includes a schematic diagram showing an anionic ring-opening polymerization reaction to form poly(ethylene glycol)-block-poly(simvastatin).

FIG. 13A includes a gel permeation chromatogram showing the peaks of an embodied compound synthesized with triazabicyclodecene (TBD) as well as the peaks of the initial reactants simvastatin and 550 Da mPEG.

FIG. 13B includes a gel permeation chromatogram showing the peaks of an embodied compound synthesized with triazabicyclodecene (TBD) as well as the peaks of the initial reactants simvastatin and 2 kDa mPEG.

FIG. 13C includes a gel permeation chromatogram showing the peaks of an embodied compound synthesized with stannous octoate as well as the peaks of the initial reactants simvastatin and 5 kDa mPEG.

**DESCRIPTION OF EXEMPLARY EMBODIMENTS**

The details of one or more embodiments of the presently-disclosed subject matter are set forth in this document. Modifications to embodiments described in this document, and other embodiments, will be evident to those of ordinary skill in the art after a study of the information provided in this document. The information provided in this document, and particularly the specific details of the described exemplary embodiments, is provided primarily for clearness of understanding, and no unnecessary limitations are to be understood therefrom.

The presently-disclosed subject matter includes compounds that comprise polymeric prodrugs. More specifically, embodiments of the present compounds include an initiator and an active agent, wherein the active agent is covalently bonded to the initiator through a ring-opening polymerization process, an atom-transfer radical polymer-
methyl ether-block-simvastatin) copolymer. Within a physiological environment, the copolymer can degrade by hydrolysis of ester bonds into the osteogenic biomolecules of simvastatin for therapeutic treatment. Similar to the initiator, the active agent can comprise an active agent carrier.

In this regard, the mechanisms of polymer degradation and drug release of the present compounds can be the same since the chemical structure of the compound is comprised of the polymeric prodrug. In other words, since the active agent is directly incorporated into the present compounds, degradation of the compounds releases the active agent into the surrounding environment. Furthermore, polymerization of the prodrug (e.g., simvastatin) eliminates certain limitations on the number of prodrug molecules provided by the compounds relative to current polymer-prodrug systems. As a result, relatively increased concentrations of active agents can be provided in the present compounds, which can provide more efficient therapeutic effects and localized release of active agents.

In certain embodiments the presently-disclosed subject matter includes novel compounds and methods for making the same that include a biodegradable poly(ethylene glycol methyl ether-block-simvastatin) copolymer. Within a physiological environment, the copolymer can degrade by hydrolysis of ester bonds into the osteogenic biomolecules of simvastatin for therapeutic treatment.

The terms “biodegradable,” “degradable,” or the like are used interchangeably herein to refer to compounds and compositions that degrade under physiological conditions and/or are metabolized or excreted with little to no adverse effects to a subject. In certain embodiments, the compounds are metabolized or excreted without permanent damage to the subject. Biodegradable compounds can be hydrolytically degradable, can require cellular and/or enzymatic action to fully degrade, or both. Biodegradable compounds also include compounds that are broken down within cells. Degradation may occur by hydrolysis, oxidation, enzymatic processes, phagocytosis, or other processes. Degradation rates for compounds can vary, and may be on the order of hours, days, weeks, months, or years, depending on the embodiment of the compound. In some embodiments the compounds and compositions include prodrugs that can degrade to form active agents.

Additionally or alternatively, embodiments of the presently-disclosed compounds can be biocompatible. The term “biocompatible” as used herein describes a characteristic of compounds that do not typically induce undesirable or adverse side effects when administered in vivo. For example, biocompatible compounds may not induce side effects such as significant inflammation and/or acute rejection. It will be recognized that “biocompatibility” is a relative term, and some side effects can be expected even for some compounds that are biocompatible. In some embodiments, a biocompatible compound does not induce irreversible side effects, and in some embodiments a compound is biocompatible if it does not induce long term side effects.

As stated above, embodiments of the present compounds can comprise an initiator and an active agent. The initiator is not particularly limited except that it must be capable of forming a polymeric material with the active agent. In some embodiments the initiator is a monomer compound that can undergo a ring-opening polymerization process with the active agent. For instance, in some embodiments the initiator comprises one or more hydroxyl groups, and can be referred to herein as a hydroxylated monomer. In some embodiments it is preferable that the initiator have one group (e.g., hydroxyl group) that can initiate a ring-opening polymerization process, such as methoxypoly(ethylene glycol) (mPEG).

The type and size of the initiator can also be varied to tune the characteristics of the resulting compound. In some embodiments the initiator and/or the active agent has a hydrophobic character or a hydrophilic character. In a specific embodiment comprising a mPEG initiator, the initiator has a hydrophilic character. Thus, by tuning the relative concentration and/or size of the mPEG initiator, the resulting compound can be imparted with a relatively more hydrophilic or hydrophobic character.

The hydrophilic and hydrophobic characteristics of a compound can be tuned to, among other things, adjust the degradation rates of a compound. As described herein, some embodiments of compounds degrade via hydrolysis. Thus, for such embodiments, the degradation of the compound can increase as the hydrophilic character of the compound increases. In other words, hydrophilic compounds that more easily attract and absorb water permit can permit a greater degree of hydrolysis relative to more hydrophobic compounds that repel or at least to not permit water to contact the compound as easily. For instance, in some embodiments the relative concentration and/or molecular weight of a hydrophilic mPEG initiator can be increased in a compound to increase the hydrophilicity and relative degradation rate of the compound via hydrolysis.

The active agent to be used in embodiments of the presently-disclosed subject matter is also not particularly limited so long as it is capable of forming a polymeric material with the initiator. In this regard, the term “active agent” is used herein to refer to compounds or entities that alter, promote, speed, prolong, inhibit, activate, or otherwise affect biological or chemical events in a subject. Exemplary active agents include, but are not limited to, osteogenic agents, osteoinductive agents, and osteoconductive agents, anti-cancer agents, antibiotics, immunosuppressants, anti-viral agents, inhibitors, anti-histamines, anti-parasite agents, anti-protozoal agents, anti-fungal agents, analgesics, anti-inflammatory agents, anti-angiogenic factors, angiogenic factors, targeting agents, polypeptides, cells, polynucleotides, viruses, and vaccines. In some embodiments, the bioactive agent is a drug and/or a small molecule, such as a statin. Similar to the initiator, the active agent can comprise one or more groups that are suitable for undergoing a ring-opening polymerization process with the initiator.

In some embodiments the active agent and/or the initiator is selected to provide the compounds with one or more osteoconductive, osteoinductive, osteogenic, and osteointe-
The present compounds can form polymers having various configurations. In some embodiments the polymers are branched. In other embodiments the polymers are unbranched. Furthermore, in some embodiments the compounds are a copolymer comprising any suitable orientation of the initiator and the active agent. The term “copolymer” as used herein refers to a polymer formed of two or more different types of monomer units. Furthermore, in some embodiments the copolymer is an amphipathic block copolymer, wherein a “block” copolymer refers to a structure comprising one or more sub-combinations of constitutional or monomeric units. Thus, the term copolymer is inclusive of block copolymers. In some embodiments the copolymer is a diblock copolymer comprising two blocks. Exemplary diblock copolymers can include a block comprised of initiator monomers and a block comprised of active agent monomers. For example, in embodiments comprising an mPEG initiator and a simvastatin active agent, the compounds can include the following formula:

![Chemical structure diagram](image)

wherein \( n \) is about 1 to about 44, and \( m \) is about 1 to about 248. The compounds are not limited to diblock copolymers, and can include alternating copolymers and the like.

By virtue of directly binding the active agent to the initiator, the present compounds can form prodrugs of an active agent. The term “prodrug” is used herein to refer to an inactive or relatively less active form of an active agent that becomes active through one or more metabolic processes in a subject. For example, the present compounds can be administered to a subject as a prodrug that includes an initiator bound to an active agent, and, by virtue of being degraded by a metabolic process, the active agent is released from the compound in its active form. In some instances hydrolytic degradation of the present compounds in vivo converts the prodrug compounds from an inactive or relatively inactive form to an active form. In specific embodiments hydrolytic degradation releases biologically active simvastatin from a relatively inactive or less active mPEG-simvastatin copolymer.

The presently disclosed subject matter also includes pharmaceutical compositions comprising the compounds described herein as well as a pharmaceutically acceptable carrier. The term “pharmaceutically acceptable carrier” refers to sterile aqueous or nonaqueous solutions, suspensions, emulsions, as well as sterile powders for reconstitution into sterile solutions or dispersions just prior to use. The compositions can thus be in the form of a cream, paste, lotion, liquid, or the like.

Proper fluidity can be maintained, for example, by the use of coating materials such as lecithin, by the maintenance of the required particle size in the case of dispersions, and by the use of surfactants. These compositions can also contain adjuvants such as preservatives, wetting agents, emulsifying agents and dispersing agents. It can also be desirable to include isotonic agents such as sugars, sodium chloride and the like. The compositions can be sterilized, for example, by filtration through a bacterial-retaining filter or by incorporating sterilizing agents in the form of sterile solid compositions which can be dissolved or dispersed in sterile water.
or other sterile injectable media just prior to use. Suitable inert carriers can include sugars such as lactose.

Suitable formulations include aqueous and non-aqueous sterile solutions that can contain antioxidants, buffers, bacteriostats, bactericidal antibiotics, and solutes that render the compositions isotonic with the bodily fluids of the intended recipient; and aqueous and non-aqueous sterile suspensions, which can include suspending agents and thickening agents.

The compositions can take such forms as suspensions, solutions, or emulsions in oily or aqueous vehicles, and can contain formulation agents such as suspending, stabilizing and/or dispersing agents. Alternatively, the compositions can be in powder form for constitution with a suitable vehicle, e.g., sterile pyrogen-free water, before use.

The compositions can be presented in unit-dose or multidose containers, for example sealed ampoules and vials, and can be stored in a frozen or freeze-dried (lyophilized) condition requiring only the addition of sterile liquid carrier immediately prior to use.

The presently-disclosed subject matter further includes methods for making a compound. In some embodiments the methods comprise providing an initiator, mixing an active agent with the initiator to form a mixture, adding a catalyst to the mixture, and reacting the mixture to form a compound that includes the initiator and the active agent. The step of reacting the mixture to form the compound can comprise reacting the mixture via a ring-opening polymerization process.

With respect to the catalyst, various known catalysts may be utilized. Such catalysts include, but are not limited to, metallic catalysts, such as stannous octoate, aluminum isopropoxide, and yttrium isopropoxide, enzymatic catalysts, such as porcine pancreatic and Candida Antartica lipases, and organic systems (organic catalysts), such as trizazacyclodecane and carboxylic acids with an alcohol. In some embodiments the mixture is provided with about 0.5 to 5 wt % of catalyst. In some embodiments the mixture for preparing the compositions includes about 0.1 to 5 wt % of the catalyst.

The reaction conditions as well as the reaction duration can be modified to tune the properties of the resulting compound. In some embodiments the step of reacting the mixture is performed at a predetermined temperature. The predetermined temperature can be, but is not limited to, temperatures of about 20° C., 30° C., 40° C., 50° C., 60° C., 70° C., 80° C., 90° C., 100° C., 110° C., 120° C., 130° C., 140° C., 150° C., 160° C., 170° C., 180° C., 190° C., 200° C., 210° C., 220° C., 230° C., 240° C., 250° C., or any temperatures therebetween. In some implementations, the molecular weight of the resulting compound increases as the reaction temperature increases.

Similarly, the reaction can be allowed to proceed for a predetermined time period. The time period can be about 1 to about 25 hours, including about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, or 25 hours. In some embodiments the polydispersity and/or the molecular weight of the resulting compounds increase as the reaction time is increased.

Further still, the presently-disclosed subject matter includes methods for treating a wound in a subject. In some embodiments the method comprises administering a composition to the wound, the composition including a compound that comprises an initiator and an active agent that is covalently bonded to the initiator through a ring-opening polymerization process. The various methods of treatment that are described herein can utilize any of the compounds that are described in this paper.
is released into the surrounding environment in its active form. The delivery of active agent that results from the degradation of a composition can be beneficial when treating a localized wound on a subject. Degradation of the composition can also provide a sustained and controllable release of active agent at a wound site.

Furthermore, the composition may be administered in several different forms. In some embodiments the composition is administered as a film. The film can be contacted with a wound on a subject. In other embodiments the composition is administered as nano- or microparticles, an oil-in-water emulsion, or as a three-dimensional scaffold. The scaffold itself can be porous or non-porous. Accordingly, the step of administering the composition, depending on the type of composition, the wound to be treated, and the like, can include injecting the composition on to a wound, contacting the composition on to a wound site, applying the composition as a paste or liquid on to a wound, or the like.

The present compositions and compounds can also be used as coating materials to coat medical devices, implants, or other devices configured to be implanted in or on a subject. The device can therefore include medical devices, biomaterials, or both. Devices that have been coated with the present compounds and compositions can provide a sustained and controlled delivery of active agent. The release of active agent from a coating can help ameliorate or accelerate the treatment of an existing wound. The release of active agent from a coating can also help mitigate adverse effects that may be caused by the implanted device.

EXAMPLES

The presently-disclosed subject matter is further illustrated by the following specific but non-limiting examples. The examples may include compilations of data that are representative of data gathered at various times during the course of development and experimentation related to the presently-disclosed subject matter. Furthermore, some of the examples described herein may be prophetic examples.

Example 1

This Example describes a process for making exemplary compounds comprising mPEG and a simvastatin active agent. This Example also describes procedures conducted to characterize the synthesized copolymers.

Stannous octoate was used for lactone polymerization given its relatively low cost, low toxicity, and high efficiency. With this reaction, the metal catalyst first forms a complex with the hydroxyl group of the initiator (i.e., mPEG) to form an alkoxide. The more reactive alkoxide then begins chain propagation by coordinating with the lactone ring of the monomer, followed by insertion of the ring into the alkoxide’s metal-oxygen bond. Throughout the process, the alkoxide acts as a nucleophile by attacking the carbon of the ring’s carbonyl group leading to cleavage of the acyl bond and extended chain formation. FIG. 1 shows a simplified representation of the exemplary simvastatin ROP reaction.

Methoxypoly(ethylene glycol) (mPEG) served as initiator in the ring-opening polymerization (ROP) reactions. Advantages of mPEG include that it comprises a single hydroxyl functionality and a boiling point suitable for the higher temperature reactions that can obtain higher molecular weight polymer.

The initial synthesis began with heating a 1:100 molar ratio of 5 kDa mPEG to simvastatin at 120°C for 2 hours and then at 150°C for 1 hour in a sand bath. After adding stannous octoate catalyst, reactions were allowed to proceed at different temperatures and for different times, with the reaction vessel continuously purged with nitrogen gas. The reaction product was precipitated in methylene chloride and diethyl ether followed by vacuum filtration. Molecular weights of the resulting polymers were analyzed by gel permeation chromatography (GPC) (FIG. 2). A distinct leftward shift can be seen as simvastatin “monomers” were consumed in the ROP reaction. FIGS. 3 and 4 show quantification of the molecular weight and polydispersity of the polymers as a function of reaction temperature at 24 hours. In these procedures, polymers reaching nearly 80 kDa were synthesized.

Copolymer molecular weight, composition, and polydispersity were characterized by GPC, by preparing 5 to 10 mg/ml samples in tetrahydrofuran. Select samples were prepared in deuterated chloroform for supplementary structural analysis by 1H-NMR (University of Kentucky NMR Facility; Lexington, Ky.) (FIG. 5). Fourier transform infrared (FTIR) spectroscopy was also be used to evaluate the ratio of functional groups unique to individual block components of the copolymer and carbonyl peak shift analysis (FIG. 6).

Degradation kinetics with associated measurement of released simvastatin-containing molecules were assessed for films cast from the synthesized copolymers. Following incubation in a physiological buffer for increasing periods, mass loss was quantified. FIG. 7 shows a schematic of the hypothesized degradation mechanism. Degradation studies were conducted at both physiological (neutral) and alkaline (up to pH 12) conditions. Over a six week period, slow polymer degradation with associated slow release of simvastatin was observed (FIG. 8). For 16-18 mg samples, the cumulative drug amounts released were 108 and 266 μg in neutral (phosphate-buffered saline, PBS) and alkaline (NaOH) solutions, respectively. After an initial burst of 59 μg in 24 hr, a zero-order release rate was observed in NaOH with a constant of 7.4 μg/hr-1 between 1 and 10 days. A first-order release rate followed with a constant of 21 d-1 for the remainder of the degradation period. In PBS, after an initial burst of 37 μg in 24 hr, 2.5 μg was released during the following 8 days. A zero-order release constant of 2.1 μg/d-1 was determined for the remainder of the degradation period. These degradation/release rates would yield formulations that can deliver the active agent for periods exceeding one year.

Example 2

This Example describes procedures to evaluate the efficacy of simvastatin-containing materials for treating bone wounds.

Simvastatin-based films are studied in a supracalvarial implantation model to assess osteogenic effects of simvastatin released from an erodable polymer system. This model enables testing of a material’s ability to enhance formation of bone from existing bone surfaces outward (i.e., appositional bone formation). For this purpose, blank (no drug) and simvastatin-releasing implants were placed on the exposed calvarium, from which the periosteum containing osteoprogenitor cells had been displaced.

Intimately opposed to the pre-existing lamellar bone of the calvarium was a layer of newly formed woven bone. The thickness of this layer depended on type of device. In the blank (no-drug control) animals, a low level of bone activity was observed, whereas simvastatin-loaded release devices
stimulated formation of much more woven bone over the mature, lamellar calvarial bone (FIG. 9). Quantitatively, whereas disturbing the periosteum stimulated an average of 100 μm of woven bone formation, controlled release enhanced bone formation by over 110% (p<0.05) (FIG. 10). FIG. 11 shows the total area of new bone and the percentage of mature matrix relative to total bone. Simvastatin-loaded implants elicited 163% larger new bone area compared to controls, and the percentage of mature bone matrix (26%) was also higher than that of the controls (21%) (p<0.05).

Example 3

This Example describes another exemplary process for synthesizing an embodiment of the present compounds using triazabicyclodecene (TBD) as a catalyst. Unless stated otherwise, the same synthesis procedure described in Example 1 was followed.

Triazabicyclodecene was selected as was selected as the organic system to be used as a catalyst because of its efficient performance at ambient temperatures, ability to work without the need of a co-catalyst, and accessibility. The ring-opening polymerization mechanism of TBD is anionic (FIG. 12). Without being bound by theory, the amidine imine 25 of TBD was selected as the co-catalyst, and accessibility. The ring-opening polymerization mechanism of TBD is anionic (FIG. 12). Without being bound by theory, the amidine imine 25 of TBD was selected as the co-catalyst, and this addition to the reaction mixture (i.e., both on mPEG and the propagating polysimvastatin block) to form the mPEG-poly(simvastatin) diblock copolymer through hydrogen bonding.

The results showed that TBD was able to form poly(ethylene glycol)-block-poly(simvastatin) with efficiency in 35 polymerization using 550 Da and 2 kDa mPEG. A leftward shift of the polymer peak or shoulder is seen relative to the mPEG peaks, indicating an increase in overall molecular weight of the chains, and hence, growth of the poly(simvastatin) block. Table 1 summarizes molecular weight and polydispersity results for the components and copolymers synthesized.

**TABLE 1**

<table>
<thead>
<tr>
<th>Sample</th>
<th>M₄ (Da)</th>
<th>M₅ (Da)</th>
<th>M₄/M₅</th>
<th>% of crude product</th>
</tr>
</thead>
<tbody>
<tr>
<td>simvastatin</td>
<td>300</td>
<td>320</td>
<td>1.0</td>
<td>0</td>
</tr>
<tr>
<td>550 Da mPEG</td>
<td>470</td>
<td>560</td>
<td>1.2</td>
<td>0</td>
</tr>
<tr>
<td>2 kDa mPEG</td>
<td>3200</td>
<td>3400</td>
<td>1.2</td>
<td>0</td>
</tr>
<tr>
<td>5 kDa mPEG</td>
<td>8500</td>
<td>8800*</td>
<td>1.1</td>
<td>0</td>
</tr>
<tr>
<td>550 Da mPEG-poly(simvastatin) 50:2 via TBD</td>
<td>6500</td>
<td>10400</td>
<td>1.6</td>
<td>21</td>
</tr>
<tr>
<td>2 kDa mPEG-poly(simvastatin) 50:2 via TBD</td>
<td>12700</td>
<td>15200</td>
<td>1.2</td>
<td>9.2</td>
</tr>
<tr>
<td>5 kDa mPEG-poly(simvastatin) 50:2 via TBD</td>
<td>13100</td>
<td>29500</td>
<td>2.3</td>
<td>62</td>
</tr>
</tbody>
</table>

*The GPC molecular weight of mPEG registered higher than the expected value because its chemistry differs from the polystyrene standards used for calibration.

While the terms used herein are believed to be well understood by one of ordinary skill in the art, definitions are set forth to facilitate explanation of the presently-disclosed subject matter.

Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which the presently-disclosed subject matter belongs. Although many methods, devices, and materials similar or equivalent to those described herein can be used in the practice or testing of the presently-disclosed subject matter, representative methods, devices, and materials are now described.

Following long-standing patent law convention, the terms “a,” “an,” and “the” refer to “one or more” when used in this application, including the claims. Thus, for example, reference to “a prodrug” includes a plurality of such prodrugs, and so forth.

Unless otherwise indicated, all numbers expressing quantities of ingredients, properties such as reaction conditions, and so forth used in the specification and claims are to be understood as being modified in all instances by the terms “about.” Accordingly, unless indicated to the contrary, the numerical parameters set forth in this specification and claims are approximations that can vary depending upon the desired properties sought to be obtained by the presently-disclosed subject matter.

As used herein, the term “about,” when referring to a value or to an amount of mass, weight, time, volume, concentration or percentage is meant to encompass variations in some embodiments of ±20%, in some embodiments of ±10%, in some embodiments of ±5%, in some embodiments of ±1%, in some embodiments of ±0.5%, and in some embodiments of ±0.1% from the specified amount, as such variations are appropriate to perform the disclosed method. It is also understood that there are a number of values disclosed herein, and that each value is also herein disclosed as “about” that particular number in addition to the value itself. For example, if the value “10” is disclosed, then “about 10” is also disclosed. It is also understood that each unit between two particular units are also disclosed. For example, if 10 and 15 are disclosed, then 11, 12, 13, and 14 are also disclosed.

Throughout this application, various publications are referenced. All such references, including the following references, are incorporated herein by reference.

REFERENCES


What is claimed is:

1. The compound having the formula:

$$\text{H}_2\text{C} \quad \text{O} \quad \text{O} \quad \text{OH} \quad \text{OH}$$

wherein:

n is about 1 to about 44; and

m is about 1 to about 258.

2. The compound of claim 1, further comprising a targeting agent.

3. The compound of claim 1, wherein the compound includes a linear or branched structure with a molecular weight of about 500 Da to about 80 KDa.

4. The compound of claim 1, wherein a molar ratio of the initiator to the active agent is about 1:1 to about 1:100.

5. A method for making the compound of claim 1, comprising:

- providing methoxypoly(ethylene glycol); mixing simvastatin with the methoxypoly(ethylene glycol) to form a mixture;
- adding a catalyst to the mixture; and
- reacting the mixture via a ring-opening polymerization process to form the compound of claim 1.

6. The method of claim 5, wherein a molar ratio of the methoxypoly(ethylene glycol) to simvastatin is about 1:1 to about 1:100.

7. The method of claim 5, wherein the catalyst is selected from a metallic catalyst, an enzymatic catalyst, an organic catalyst, and combinations thereof.

8. A method for treating a wound in a subject, comprising:

- administering the composition of claim 1 to the wound.

9. The method of claim 8, wherein the step of administering a composition includes contacting the wound with the composition.

10. The method of claim 8, wherein the wound is a bone wound, a skin wound, or a combination thereof.

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