6-20-2006

Highly Lipophilic Camptothecin Intermediates and Prodrugs and Methods of Preparation Thereof

Thomas G. Burke
University of Kentucky

Dennis P. Curran

Wu Du

Click here to let us know how access to this document benefits you.

Follow this and additional works at: https://uknowledge.uky.edu/markey_patents

Part of the Oncology Commons

Recommended Citation
Burke, Thomas G.; Curran, Dennis P.; and Du, Wu, "Highly Lipophilic Camptothecin Intermediates and Prodrugs and Methods of Preparation Thereof" (2006). Markey Cancer Center Faculty Patents. 2.
https://uknowledge.uky.edu/markey_patents/2

This Patent is brought to you for free and open access by the Cancer at UKnowledge. It has been accepted for inclusion in Markey Cancer Center Faculty Patents by an authorized administrator of UKnowledge. For more information, please contact UKnowledge@lsv.uky.edu.
HIGHLY LIPOPHILIC CAMPTOTHECIN INTERMEDIATES AND PRODRUGS AND METHODS OF PREPARATION THEREOF

Inventors: Thomas G. Burke, deceased, late of Lexington, KY (US); Lori Latus, legal representative, Lexington, KY (US); Dennis P. Curran, Pittsburgh, PA (US); Wu Du, San Diego, CA (US)

Assignees: University of Kentucky Research Foundation, Lexington, KY (US); University of Pittsburgh, Pittsburgh, PA (US)

Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 0 days.

This patent is subject to a terminal disclaimer.

Appl. No.: 10/843,822
Filed: May 12, 2004

Related U.S. Application Data
Provisional application No. 60/469,805, filed on May 12, 2003.

Int. Cl.
C07F 7/10

U.S. Cl. .......................................................... 546/14

Field of Classification Search .................. 546/14
See application file for complete search history.

References Cited
U.S. PATENT DOCUMENTS
5,552,156 A 9/1996 Burke
5,736,156 A 4/1998 Burke
6,290,167 B1 9/2001 Burke et al.
6,376,676 B1 4/2002 Curran et al.

OTHER PUBLICATIONS


2 Claims, No Drawings

ABSTRACT
The present invention relates to novel, highly lipophilic silatecan intermediates and prodrugs of DB-67 and other silatecans.
OTHER PUBLICATIONS
* cited by examiner
This application claims the benefit of U.S. Provisional Patent Application Ser. No. 60/469,805 filed on May 12, 2003.

This invention was made with Government support under NIH Grant Number 1R01CA63653. The Government may have certain rights in this invention.

**TABLE 1**

Clinical candidates and FDA-approved analogs in the camptothecin family of antitumor agents

<table>
<thead>
<tr>
<th>Aqueous Solubility</th>
<th>Compound</th>
<th>R^1</th>
<th>R^2</th>
<th>R^3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soluble</td>
<td>Topotecan/TPT</td>
<td>H</td>
<td>OH</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>CDK602</td>
<td>H</td>
<td>C(CH_3)NH(CH_3)_2</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>Irinotecan/CPT-11</td>
<td>C_2H_5</td>
<td>H</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>GI-147211/C/GG-211</td>
<td>CH_3</td>
<td>H</td>
<td>H</td>
</tr>
<tr>
<td>Insoluble</td>
<td>Camptothecin</td>
<td>H</td>
<td>H</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>9-AC</td>
<td>H</td>
<td>NH_2</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>9-NC/Rubitecan</td>
<td>H</td>
<td>NO_2</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>SN-38</td>
<td>C_2H_5</td>
<td>H</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>DB-87</td>
<td>Si(CH_3)_2C(CH_3)_3</td>
<td>H</td>
<td>H</td>
</tr>
<tr>
<td></td>
<td>MDCPT</td>
<td>H</td>
<td>H</td>
<td>H</td>
</tr>
</tbody>
</table>

**TECHNICAL FIELD**

The present invention relates to novel intermediates and prodrugs of highly lipophilic 7-silylalkylcamptothecins (silicateins).

**BACKGROUND OF THE INVENTION**

Camptothecin and related analogs (Table 1) are emerging as a prominent class of agents useful in the treatment of cancer. The camptothecins display a unique mechanism of action: stabilization of the covalent binding of the enzyme topoisomerase I (topo I), an intranuclear enzyme that is overexpressed in a variety of tumor lines, to DNA. This drug/enzyme/DNA complex leads to reversible, single strand nicks. According to the fork collision model, the DNA nicks are converted to irreversible and lethal double strand DNA breaks during replication.

The camptothecin class of anticancer agents have exhibited unusual reactivity in vivo, both with respect to drug hydrolysis and blood protein interactions. These factors have hindered the pharmaceutical development and clinical implementation of camptothecins. In terms of hydrolysis, each of the camptothecins shown in Table 1 contains an α-hydroxy-δ-lactone pharmacophore.

At physiological pH of 7 and above this functionality is reactive, readily converting to the biologically inactive “ring opened” carboxylate form. Thus, as a result of the labile α-hydroxy-δ-lactone pharmacophore, an equilibrium is established between two distinct drug species: 1) the biologically active lactone form where the lactone ring remains closed; and 2) a biologically-inactive carboxylate form generated upon the hydrolysis of the lactone ring of the parent drug.

This hydrolysis problem with camptothecin and many analogs (e.g., 9-aminocamptothecin, 9-nitrocamptothecin) is exacerbated in human blood. In human blood and tissues, the camptothecin equilibrium of active lactone form vs. inactive carboxylate form can be strongly modulated by the presence of human serum albumin (HSA). The lactone form...
of camptothecin binds to HSA with moderate affinity yet the carboxylate form of camptothecin binds much more tightly than the carboxylate, displaying the 150-fold enhancement in its affinity. Thus, the preferential binding of the carboxylate form to HSA drives the equilibrium to the right in favor of the carboxylate, resulting in the lactone ring hydrolyzing more rapidly and completely (than when camptothecin is in an aqueous solution without HSA).

The development of 7-silylcamptothecins (or silatecans) has resulted in the identification of agents with improved human blood stability and activities. Recent rational design efforts have resulted in the identification of A,B-ring modified camptothecins displaying improved human blood stabilities combined with potent anti-topoisomerase I activities. Dual 7,10-substitution (where the 10-substituent is a hydroxy group) results in camptothecins displaying vastly improved human blood stabilities. SN-38 contains this dual 7-alkyl-10-hydroxy substitution pattern and in 1994 it was shown that these structural modifications block SN-38 from associating with the high affinity camptothecin carboxylate binding pocket on HSA.

More recently the design of another dual 7,10-modified camptothecin has been described. The new agent is 7-t-butylidimethylsilyl-10-hydroxycamptothecin (DB-67). DB-67 displays markedly improved human blood stability and potent anti-topoisomerase I anticancer activity. The design of DB-67 was based upon the following two considerations: 1) dual 7,10-substitution patterns eliminate the highly specific binding of carboxylate form over lactone form by HSA; and 2) lactone stabilization is further promoted by enhanced lipophilicity or lipid bilayer partitioning. Lipophilicity promotes camptothecin drug stability by favoring lactone partitioning into blood cells, thereby protecting the active lactone forms from hydrolysis. The key α-hydroxy-6-lactone pharmacophore in DB-67 displays superior stability in human blood when compared with FDA-approved topotecan, CPT-11, and several other clinically relevant camptothecin analogs. DB-67 displayed a t½ of 130 min, and a % lactone at equilibrium value of 30 in human blood; the t-butylidimethylsilyl group enhances lipophilicity and thereby promotes drug associations with blood cells. DB-67 is 25 times lipophilic than camptothecin and readily incorporates as its active lactone form into cellular and liposomal bilayers. Equally important, the dual 7-alkylsilyl and 10-hydroxy substitution in DB-67 blocks the associations of the carboxylate form of DB-67 with the high affinity carboxylate binding pocket on HSA. Together, the enhanced lipophilicity and altered HSA interactions provide DB-67 with the highest human blood stability when compared with clinically relevant camptothecins containing the conventional α-hydroxy-α-lactone pharmacophore.

In vitro cytotoxicity assays have shown that DB-67 is of comparable potency relative to camptothecin and 10-hydroxycamptothecin, as well as the FDA approved analogs topotecan and CPT-11. In addition, cell-free cleavage assays revealed that DB-67 forms more stable topoisomerase I cleavage complexes than camptothecin or SN-38. In terms of in vitro potency, DB-67 has been shown to display activity against human glioma in a murine model. Overall, these stability and activity profiles of DB-67 indicate how rational drug design can result in new, highly lipophilic agents displaying improved pharmacological properties.

In this invention we describe novel, highly lipophilic intermediates and prodrugs of DB-67 and other silatecans.

Detailed Description of the Invention

Our previous rational synthetic efforts described above yielded a series of silatecan agents. 7-t-Butylidimethylsilyl-10-hydroxycamptothecin, which we have termed DB-67, is an example of a silatecan that displays improved biological properties. In the current invention we describe novel intermediates and prodrugs of DB-67 and related 7-silylcamptothecins or silatecans.

The invention includes compounds with the following structures A and B:

R1 and R2 are independently the same or different and are hydrogen, an alkyl group, an alkenyl group, an alkynyl group, an alkoxy group, an aryloxy group, an acyloxy group, OCH2COR12 wherein R12 is an alkyl group, an alkoxycarbonyloxy group, a halogen, a hydroxy group, a nitro group, a cyano group, an azido group, a formyl group, a hydrazino group, COHR13 wherein R13 is an alkyl group, an alkoxy group, an amino group, or a hydroxy group, SR14, wherein R14 is hydrogen, an OCH2COR13, an alkyl group, or an aryl group; or R1 and R2 together form a group of the formula O(CH2)nO wherein n represents an integer 1 through 6; R3 is H, a nitro group, a halogen atom, an amino group, a hydroxy group, or a cyano group, or R3 and R4 together form a group of the formula O(CH2)nO wherein n represents an integer 1 through 6; R3 is H, F, an alkyl group, an alkenyl group, an alkynyl group, a trialkylsilyl group or an aryl group; R3 is C1-C12 alkyl group, an alkenyl group, a benzyl group or a propargyl group; R6, R7 and R8 are independently a C1-C15 alkyl group, a C2-C15 alkynyl group, a C2-C15 alkenyl group, an aryl group or a CH2R13 group, wherein R13 is an integer between 1 and 15 and R13 is a hydroxy group, alkyl group, amino group, an alkylamino group, an alkenylamino group, a dialkylamino group, a halogen atom, a cyano group or a nitro group; R9 is a hydrogen, an alkyl group, an alkenyl group, an alkynyl group, an aryl group, an aryloxy group, or an acyloxy group; R10 is an alkenylene group, an alkylene group or an alkenylene group.

The invention also includes compounds with the following structures C and D:
wherein R₁ and R₂ are independently the same or different and are hydrogen, an alkyl group, an alkenyl group, an alkynyl group, an alkoxy group, an aryl group, an acyl group, a carbamoyloxy group, a halogen, a hydroxy group, a nitro group, a cyano group, an azido group, a formyl group, a hydrazino group, C(O)R₁, wherein R₁ is an alkyl group, an alkoxy group, an amino group or a hydroxy group, SR₁, wherein R₁ is hydrogen, C(O)R₁, an alkyl group, or an aryl group; or R₁ and R₂ together form a group of the formula O(CH₂)ₚO wherein p represents an integer 1 through 6; R₃ is H, a nitro group, a halogen atom, an amino group, a hydroxy group, or a cyano group, or R₂ and R₃ together form a group of the formula O(CH₂)ₚO wherein p represents an integer 1 through 6; R₄ is H, F, an alkyl group, an alkenyl group, an alkynyl group, a trialkylsilyl group or an alkoxy group; R₅ is a C₁₋₁₅ alkyl group, an alkyl group, a benzyl group or a propargyl group; R₆, R₇ and R₈ are independently a C₁₋₁₅ alkyl group, a C₂₋₁₅ alkenyl group, a C₂₋₁₅ alkynyl group, an aryl group or a (CH₂)₂R₈ group, wherein q is an integer between 1 and 15 and R₁₋₅ is a hydroxy group, alkoxyl group, an amino group, an alkenylamino group, an alkenylamido group, a halogen group, a cyano group or a nitro group; R₉ is a hydrogen, an alkyl group, an alkenyl group, an alkynyl group, an alkoxy group, an aryl group, or an acyloxy group; R₁₀ is an alkenyl group, an alkene group or an alkenylene group; R₁₁ is (CH₂)₂NR₂NR₂ wherein L₁ may be an integer ranging from 1–30 and R₁₀ and R₁₁ are independently the same or different and are hydrogen, a C₁₋₁₅ alkyl group, a C₂₋₁₅ alkenyl group, a C₂₋₁₅ alkynyl group, an aryl group, a (CH₂)₂R₈ group, a (CH₂)₂C(O)R₈ group or a (CH₂)₂CO₂R₈ wherein Y may be an integer ranging from 1 to 15 and R₉ is a hydroxy group, a thiol group, an alkylthiol, a silyl group, an alkoxy group, an amino group, an alkenylamino group, a diazenylamino group, a cyano group, a nitro group. R₁¹ may also be a hydrogen, an alkyl group, an alkenyl group.

All compounds of the present invention including the β-hydroxylactone group can exist in racemic form, enantiomerically enriched from, and enantiomerically pure form.
Some of the camptothecin analogs of the present invention can be prepared for pharmaceutical use as salts with inorganic acids such as, but not limited to, hydrochloride, hydrobromide, sulfate, phosphate, and nitrate. The camptothecin analogs can also be prepared as salts with organic acids such as, but not limited to, acetate, tartrate, fumarate, succinate, citrate, methanesulfonate, p-toluenesulfonate, and stearate. Other acids can be used as intermediates in the preparation of the compounds of the present invention and their pharmaceutically acceptable salts.

For purification, administration or other purposes, the E-ring (the lactone ring) may be opened with alkali metal such as, but not limited to, sodium hydroxide or calcium hydroxide, to form opened E-ring analogs of compounds of the present invention. The intermediates thus obtained are more soluble in water and may be purified to produce, after treatment with an acid, a purified form of the camptothecin analogs of the present invention.

All the novel compounds of the present invention whether in racemic, enantiomerically enriched or enantiomerically pure form display good biological activity while also possessing favorable characteristics for active loading into liposomal particle drug delivery systems of the micelle type disclosed and described in U.S. Pat. Nos. 5,316,771; 5,552,156 and 5,736,156. Pre-made liposomes can be efficiently loaded with any of the novel compounds of this invention using pH gradients.

In brief, in the active core loading process an amine-containing agent is loaded into the particle. For example, a gradient created by ammonia gas diffusing out of the liposome can result in diffusion or active loading of the compound/agent of the present invention inward to the core of the particle. The chemical gradient across the membrane creates a driving force for the compound/agent to replace the lost NH₃ from the interior of the liposome. Once inside the acidic confines of the core, the compound/agent becomes protonated and remains within the core, as its positive charge impedes retro-diffusion across the liposome bilayer. The protonated amine also prevents the occurrence of nucleophilic attack of the amine on the lactone carbonyl. As liposomes can be actively and/or passively targeted to the tumor, the liposome encapsulated compound/agent can be effectively concentrated at the tumor site, thereby reducing exposure of the healthy host tissues to the cytotoxic agent yet enhancing exposure at the tumor target.

The tumor targeted approach involving liposomal delivery of core loaded compound/agent addresses multiple clinical issues. For example, reduced systemic toxicity can be achieved. Enhanced exposure at the tumor site in terms of relative amounts of drug reaching the tumor can also be achieved. Furthermore, enhanced exposure at the tumor site can be achieved in terms of prolonging the exposure of drug there.

The present invention also provides a method of treating a patient, which comprises administering a pharmacologically effective amount of a compound of the present invention. The compound may, for example, be administered to a patient afflicted with cancer and/or leukemia. The compounds of the present invention may also act as antiviral (for example, anti-HIV) agents and antiparasitic agents. The pharmacologically effective amount or dosage is preferably between 0.01 to 80 mg of one of the compounds of structures A, B and/or C per kg of body weight. More preferably, the pharmacologically effective amount or dosage is preferably between 0.1 to 40 mg of one or more of the compounds per kg of body weight. In general, a pharmacologically effective amount or dosage contains an amount of one of the compounds effective to display antileukemic and/or antitumor (anticancer) behavior. Pharmaceutical compositions containing as an active ingredient of one of the compounds of the present invention including a pharmaceutically acceptable salt thereof in association with a pharmaceutically acceptable carrier or diluent are also within the scope of the present invention.

The novel compounds of the present invention display good biological activity while also possessing favorable characteristics for active loading into liposomal particle drug delivery systems of the micelle type disclosed and described in U.S. Pat. Nos. 5,316,771; 5,552,156 and 5,736,156. Pre-made liposomes can be efficiently loaded with the novel compounds of this invention using pH gradients.

In brief, in the active core loading process an amine-containing agent is loaded into the particle. For example, a gradient created by ammonia gas diffusing out of the liposome can result in diffusion or active loading of the compound/agent of the present invention inward to the core of the particle. The chemical gradient across the membrane creates a driving force for the compound/agent to replace the lost NH₃ from the interior of the liposome. Once inside the acidic confines of the core, the compound/agent becomes protonated and remains within the core, as its positive charge impedes retro-diffusion across the liposome bilayer. The protonated amine also prevents the occurrence of nucleophilic attack of the amine on the lactone carbonyl. As liposomes can be actively and/or passively targeted to the tumor, the liposome encapsulated compound/agent can be effectively concentrated at the tumor site, thereby reducing exposure of the healthy host tissues to the cytotoxic agent yet enhancing exposure at the tumor target.

The tumor targeted approach involving liposomal delivery of core loaded compound/agent addresses multiple clinical issues. For example, reduced systemic toxicity can be achieved. Enhanced exposure at the tumor site in terms of relative amounts of drug reaching the tumor can also be achieved. Furthermore, enhanced exposure at the tumor site can be achieved in terms of prolonging the exposure of drug there.

The compounds of the present invention may be administered in a dose which is effective at inhibiting the enzyme topoisomerase I. These amounts are generally from about 1–80 mg/kg of body weight per week, preferably about 2–40 mg/kg per week.

The compounds of the present invention may be administered as a pharmaceutical composition containing the compounds and a pharmaceutically acceptable carrier or diluent. The compounds can be administered as their ring open salt forms, since relactonization to their active forms can occur in the body (especially at sites of reduced pH). The active material can also be mixed with other active materials which do not impair the desired action and/or supplement the desired action. The compounds/active materials according to the present invention can be administered by any route, for example, orally, nasally, parenterally, intravenously, intradermally, subcutaneously, or topically, in liquid or solid form.

For the purposes of parenteral therapeutic administration, the active ingredient may be incorporated into a solution or suspension. The solutions or suspensions may also include the following components: a sterile diluent such as water; for injection, suspensions of liposomal particles whereby the particles contain stable, active drug within the core of the particle in a pH controlled and protected environment or associated to the outside of the particle or any of the bilayers of the particle, saline solution, fixed oils, polyethylene
glycols, glycerine, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl parabens; antioxidants such as ascorbic acid or sodium bisulfite; chelating agents such as ethylenediaminetetraacetic acid; buffers such as acetates, citrates or phosphates and agents for the adjustment of tonicity such as sodium chloride or dextrose. The parenteral preparation can be enclosed in ampoules, disposable syringes or multiple dose vials made of glass or plastic.

Another mode of administration of the compounds of this invention is oral. Oral compositions will generally include an inert diluent or an edible carrier. They may be enclosed in gelatin capsules or compressed into tablet. For the purpose of oral therapeutic administration, the aforesaid compounds may be incorporated with excipients and used in the form of tablets, troches, capsules, elixirs, suspensions, syrups, wafers, chewing gums and the like.

The tablets, pills, capsules and the like may contain the following ingredients: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a disintegrating agent such as alginic acid, Primogel, corn starch and the like; a lubricant such as magnesium stearate or Sterolets; a glidant such as a colloidal silicon dioxide; and a sweetening agent such as sucrose or saccharin or flavoring agent such as peppermint, methyl salicylate, or orange flavoring may be added. When the dosage unit is in the form of a capsule, it may contain, in addition to material of the above type, a liquid carrier such as a fatty oil. Other dosage unit forms may contain other various materials which modify the physical form of the dosage unit, for example, as coatings. Thus tablets or pills may be coated with sugar, shellac, or other enteric coating agents. A syrup may contain, in addition to the active compounds, sucrose as a sweetening agent and certain preservatives, dyes and coloring and flavors. Materials used in preparing these various compositions should be pharmaceutically or veterinarily pure and non-toxic in the amount used.

The following examples are presented to further illustrate the invention, but it is not to be considered as limited thereto.

**EXAMPLE 1**

Preparation of DB-67 20(S) α-aminoalkanoic esters

DB-67 α-aminoalkanoic esters were synthesized as following:
DB-67 (1 equiv) was dissolved in anhydrous dichloromethane, and DIPEA (5 equiv) was added under nitrogen atmosphere. The reaction mixture was stirred for 10 min at room temperature followed by the addition of MOMCl (5 equiv). The resulting solution was left stirring overnight. The reaction was terminated. Then the reaction mixture was diluted by dichloromethane and washed several times with water, dried (MgSO₄), filtered, and concentrated. Purification by chromatography on silica gel gave 10-MOM protected compound.

1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDCI, 3 equiv) was slowly added to a solution of 10-MOM protected compound (1 equiv), N-tert-butoxycarbonylaminino acid (3 equiv) and 4-(Dimethylamino)pyridine (DMAP, 0.6 equiv) and anhydrous dichloromethane, which had been pre-cooled to 0°C. The resulting solution was allowed to warm to room temperature and stirred overnight. The organic fraction was washed with HCI (0.1 N) and water and then extracted several times with CH₂Cl₂. Organic layers were combined and dried with MgSO₄, followed by filtration and concentration. Purification of this residue by flash chromatography on silica gel gave chloromethyl methyl ether (MOM) and N-BOC protected prodrug. Deprotection of MOM and N-BOC groups was achieved simultaneously by stirring MOM and N-BOC protected prodrug in dichloromethane and trifluoroacetic acid for 30 minutes at room temperature, followed by concentration under high vacuum and recrystallization in methanol and ethyl ether. Purity of final products was determined by HPLC and prodrug purity was found to be greater than 98% pure.

**EXAMPLE 2**

Synthesis of DB-67 20-O-ester of N-methylglycine, mono (trifluoroacetate)
EXAMPLE 3

Conversion of DB-67 20(S)-ω-aminoalkanoic esters to lactam, ortho lactone and acetic acid prodrugs

At physiological pH of 7 or above, the nucleophilicity of the amine group of DB-67 20(S)-glycinate ester manifests itself and cyclization to the C-21 carbonyl carbon occurs. This cyclization triggers a rapid and convenient non-enzymatic decomposition process that releases active DB-67. DB-67 20(S)-glycinate 1 decomposed to produce several products: the closed-ring lactone form of DB-67 5, the ring-opened carboxylate form of DB-67 6, and two novel decomposition products 3 and 4 generated following the formation of an unusual six membered morpholine 2,5-dione ring 2 (or lactam intermediate). The lactam intermediate arose by intramolecular nucleophilic attack of the amino group on the lactone E-ring carbonyl carbon of DB-67 and is in fast equilibrium with structures 3 (which we refer to as the ortho lactone) and 4. Ortho lactone 3 arose by a second intramolecular reaction within the lactam intermediate and 3 exhibited the same mass as DB-67 20(S)-glycinate ester but with a strikingly different fragmentation pattern. 4 arose by a competitive intermolecular reaction to the lactam intermediate and reacted to release both 5 and 6. ESIMS/MS showed that DB-67 glycinate ester fragmentation pattern [m/z(%)] was: 536 (100%), 461 (50%), 433 (20%); DB-67 ortho lactone fragmentation pattern was: 536 (100%), 518 (10%).

EXAMPLE 4

Procedure for the isolation of the ortho lactone from N-methyl DB-67 glycinate ester

N-methyl DB-67 glycinate trifluoroacetic acid salt was dissolved in DMF, 1.2 eq. triethylamine was then added and reaction stirred overnight at room temperature. Organic solvent was removed and a yellow precipitate was rinsed with water, filtered, and dried under vacuum. Yield: 98%.

EXAMPLE 5

Loading of Compounds of Present Invention into Liposomes

Remote “active” loading of prodrug into premade small unilamellar vesicles, with diameters of 100 nm, was carried out by using transmembrane ammonium sulfate gradients. Prodrug was added to a liposomal suspension where initially [(NH₄)₂SO₄]CORE: [(NH₄)₂SO₄]EXTERNAL; loading of the prodrug occurred as a result of base exchange (initiated by NH₃ gas molecules departing the liposome). Whereas undervatized camptothecin and DB-67 localize predominantly in the bilayer compartment of the liposome, their
20-OR prodrugs, where R) CO(CH₂)ₙNH₂ and n) 1–3, loaded with high efficiency (60 to 90%) into the core of liposomes at clinically relevant drug-to-lipid ratios (between 1:4 to 1:20). More importantly, these core-loaded liposomal formulations of camptothecin 4-aminobutanoate ester and DIB-67 4-amino butanoate ester exhibited markedly improved stabilities in whole blood relative to their free forms. Whereas the decomposition of free prodrug in both cases was extensive, liposomal entrapment prevented the degradation process from occurring, providing indirect evidence that the prodrug was effectively retained within the liposome for periods up to 40 h. These time periods are known to be sufficient for successful tumor-targeting to be achieved.

What is claimed is:

1. A compound having the formula:

```
R₁
OH
```

wherein R₁ and R₂ are independently the same or different and are hydrogen, an alkyl group, an alkenyl group, an alkynyl group, an alkylamino group, a dialkylamino group, a halogen atom, a nitro group, a cyano group, an alkoxy group, an amino group, an alkylamino group, an nitro group.

2. A compound having the formula:

```
R₁
OH
```

wherein R¹ and R² are independently the same or different and are hydrogen, an alkyl group, an alkenyl group, an alkynyl group, a phenyl group, a naphthoxy group, an acyloxy group, OC(O)OR¹² wherein R¹² is an alkyl group, an alkylamino group, a dialkylamino group, an alkenyl group, an alkenyloxy group, an alkynyl group, a phenyl group, a naphthyl group, a phenyl group, a naphthyl group, a (CH₂)ₙNR¹⁶R¹⁷ wherein n may be an integer ranging from 1 to 15 and R¹⁶ and R¹⁷ are independently the same or different and are hydrogen, a C₁₋₅ alkyl group, a C₂₋₅ alkenyl group, a phenyl group, a naphthyl group, an acyloxy group; R¹¹ is hydrogen, an alkyl group, an alkylamino group, an alkylamino group, a phenyl group, a naphthoxy group, or an acyloxy group; R¹⁰ is an alkylene group, an alkenylene group or an alkynylene group; R¹¹ is hydrogen, an alkyl group, an alkylamino group and (CH₂)ₙNR¹⁶R¹⁷ wherein n may be an integer ranging from 1 to 15 and R¹⁶ and R¹⁷ are independently the same or different and are hydrogen, a C₁₋₅ alkyl group, a C₂₋₅ alkenyl group, a phenyl group, a naphthyl group, a phenyl group, a naphthyl group, a (CH₂)ₙR¹⁸ group, a (CH₂)ₙC(O)R¹⁸ group or a (CH₂)ₙCO₂R¹⁸ wherein Y may be an integer ranging from 1 to 15 and R¹⁸ is a hydroxy group, a thiol group, an alkylthio group, a silyl group, an alkoxyl group, an amino group, an alkylamino group, a dialkylamino group, a halogen atom, a cyano group, a nitro group.