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(54) Benævnelse: **LÆGEMIDDEL TIL ORAL INDGIVELSE OMFATTENDE MGBG OG FREMGANGSMÅDER TIL BEHANDLING AF SYGDOM**

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## DESCRIPTION

**[0001]** The present invention relates to oral pharmaceutical compositions comprising methylglyoxal bis(guanylhydrazone) (MGBG; mitoguazone) for use in the treatment of pain when orally administered to a subject, as well as MGBG for use in the treatment of pain.

**[0002]** Disclosed herein are new oral pharmaceutical compositions of polyamine analogs, polyamine biosynthesis inhibitors, polyamine inhibitors of AMD-I and regulators of osteopontin, and their application for the treatment of conditions including those modulated by osteopontin or associated with elevated levels or activity of osteopontin, and those related to viral infections such as human immunodeficiency virus (HIV).

**[0003]** MGBG (methylglyoxal bis(guanylhydrazone); mitoguazone) is a competitive polyamine inhibitor of S-adenosyl methionine decarboxylase (AMD-I), which catalyzes the synthesis of spermidine, a polyamine. The amino-acid-derived polyamines have long been associated with cell growth and cancer, and specific oncogenes and tumor-suppressor genes regulate polyamine metabolism. Inhibition of polyamine synthesis has proven to be generally ineffective as an anticancer strategy in clinical trials, but it is a potent cancer chemoprevention strategy in preclinical studies. Despite its novel mechanism of action and promising preclinical data, initial clinical trials of MGBG were ceased in the middle of 1960s due to severe toxicity especially to self-renewing normal tissues such as the bone marrow and intestinal tract (particularly severe mucositis), which were both dose and schedule dependent.

**[0004]** Regardless, research has continued with MGBG. A number of studies have examined potential uses in combination with other chemotherapeutic agents and innovative dosing regimens, designed to minimize side effects and dose where possible. Others have focused on elucidating MGBG's modes of action in the body. Yet others have investigated MGBG's activity in diseases other than cancer.

**[0005]** Perhaps due to negative clinical findings in these early studies, to date, MGBG has been confined to intravenous use. As a practical matter, this presents a number of problems for treatment of many diseases, particularly of chronic or recurrent conditions. Administration via IV injection or infusion must be done by a medical professional in a hospital setting. This not only presents an inconvenience and increased cost to the subject, but it also exposes him or her to hospital-based infections and illnesses, this latter both from venipuncture and the hospital or clinic visit itself. In immunocompromised individuals such as, for example, those with HIV or AIDS, individuals undergoing treatment with immune system suppressors, and the elderly, this is a relevant concern. Thus, a subject with a long-term chronic condition such as an autoimmune or hyperproliferative disorder, or a doctor treating such a subject, might find the cost, inconvenience, and risks of such a treatment more important than any potential therapeutic benefits the drug might offer.

**[0006]** An oral formulation of MGBG, in contrast, would present several benefits. First, an oral

formulation, for example a simple pill or tablet, may be taken outside of a hospital setting, increasing the potential for subject ease of use and compliance. This permits a subject to avoid infection risks concomitant with IV administration and hospital visits. Where early treatment can prevent the development of disease complications, this is of particular benefit. Chronic low-dose administration of MGBG is practically impossible in an IV formulation. Additionally, oral delivery typically avoids the high concentration peak and rapid clearance associated with an IV bolus dose. Yet another advantage of an oral drug would be the ability to formulate MGBG as a combination composition with one or more other therapeutic agents.

**[0007]** In this context, White (White, F. R.; Cancer Chemotherapy Reports, 24; 1962; pp. 79-84) describes the compound MGBG, the pharmacological properties thereof, and its use in the treatment of cancer. Further, Oliverio *et al.* (Oliverio, V. T. et al.; Journal of Pharmacology and Experimental Therapeutics, 141; 1963; pp. 149-156) describes the pharmacokinetic properties of radioactively labeled MGBG. Moreover, WO 2009/018368 A1 describes methods for treating neuropsychiatric disorders by administration of D-amino acid oxidase inhibitors (DAAOIs). Finally, US 2007/0078187 A1 describes methods for treating viral infections using *inter alia* MGBG.

**[0008]** The present invention relates to the following items:

1. 1. An oral pharmaceutical composition, comprising MGBG together with at least one oral pharmaceutically acceptable excipient, for use in the treatment of pain when orally administered to a subject.
2. 2. The oral pharmaceutical composition for use as recited in item 1, comprising about 25 mg to about 350 mg of MGBG.
3. 3. The oral pharmaceutical composition for use as recited in item 1 or item 2, formulated as a tablet or capsule.
4. 4. The oral pharmaceutical composition for use according to any one of items 1 to 3, wherein the pain is selected from the group consisting of inflammatory pain, pain due to nerve injury, chronic pain, intractable cancer pain, complex regional pain syndrome, neuropathic pain, surgical or post-surgical pain, dental pain, pain resulting from dermal injury, lower back pain, headaches, migraine, tactile allodynia, and hyperalgesia.
5. 5. The oral pharmaceutical composition for use according to any one of items 1 to 4, wherein the subject is a human.
6. 6. The oral pharmaceutical composition for use according to any one of items 1 to 5, wherein the pain is chronic.
7. 7. The oral pharmaceutical composition for use according to any one of items 1 to 5, wherein the pain is acute.
8. 8. MGBG for use in the treatment of pain.
9. 9. MGBG for use according to item 8, wherein the MGBG is orally administered.
10. 10. MGBG for use according to item 8 or item 9, wherein the pain is selected from the group consisting of inflammatory pain, pain due to nerve injury, chronic pain, intractable cancer pain, complex regional pain syndrome, neuropathic pain, surgical or post-surgical pain, dental pain, pain resulting from dermal injury, lower back pain, headaches,

migraine, tactile allodynia, and hyperalgesia.

11. 11. MGBG for use according to any one of items 8 to 10, wherein the pain is chronic.
12. 12. MGBG for use according to any one of items 8 to 10, wherein the pain is acute.

**[0009]** Further, disclosed herein are oral pharmaceutical formulations of MGBG and other polyamine analogs, polyamine biosynthesis inhibitors, polyamine inhibitors of AMD-I and regulators of osteopontin. Also disclosed are methods for the treatment of diseases comprising the administration of MGBG and other polyamine analogs, polyamine biosynthesis inhibitors, polyamine inhibitors of AMD-I and regulators of osteopontin.

**[0010]** Additionally, methods for the treatment of pain comprising the administration of MGBG and other polyamine analogs, polyamine biosynthesis inhibitors, polyamine inhibitors of AMD-I and regulators of osteopontin are disclosed.

**[0011]** The figures show:

Figure 1 shows the results of a murine carrageenan-induced model of inflammation. Change in paw edema over time is plotted against time.

Figure 2 shows the results of a murine carrageenan-induced model of pain. Change in paw withdrawal latency, a measure of hyperalgesia, over time is plotted against time.

**[0012]** Accordingly, provided herein is a pharmaceutical composition for oral delivery, comprising a polyamine analog or polyamine biosynthesis inhibitor together with at least one pharmaceutically acceptable oral excipient.

**[0013]** In certain embodiments, the polyamine analog or polyamine biosynthesis inhibitor is a compound disclosed herein.

**[0014]** In certain embodiments, the polyamine analog or polyamine biosynthesis inhibitor is one known in the art.

**[0015]** Also provided herein is an oral pharmaceutical composition, comprising polyamine analog or polyamine biosynthesis inhibitor together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma polyamine analog or polyamine biosynthesis inhibitor level when orally administered to a subject.

**[0016]** Also provided herein is an oral pharmaceutical composition, comprising polyamine analog or polyamine biosynthesis inhibitor together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma polyamine analog or polyamine biosynthesis inhibitor level for the treatment of pain when orally

administered to a subject.

**[0017]** In certain embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma level of a polyamine analog or polyamine biosynthesis inhibitor for a period of at least 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 30, 36, or 48 hours. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least a 6-hour period. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least a 12-hour period. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least an 18-hour period. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least a 24-hour period.

**[0018]** In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 25, 50, 55, 60, 65, 75, 80, 85, 90, or 95 percent of the peak plasma concentration for at least 4 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 75% of the peak plasma concentration for at least 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, or 24 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 75% of the peak plasma concentration for at least 4 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 75% of the peak plasma concentration for at least 6 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 75% of the peak plasma concentration for at least 8 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 50% of the peak plasma concentration for at least 8 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 50% of the peak plasma concentration for at least 12 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 50% of the peak plasma concentration for at least 18 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of a polyamine analog or polyamine biosynthesis inhibitor of at least 25% of the peak plasma concentration for at least 18 hours. In further embodiments, the peak plasma concentration is a therapeutically effective concentration. In yet further embodiments, the percentage of peak plasma concentration is therapeutically effective over the given time period.

**[0019]** In certain embodiments, the pharmaceutical composition comprising the polyamine analog or polyamine biosynthesis inhibitor has an oral bioavailability of at least 10, 20, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 45, 50, 55, or 60 percent. In further

embodiments, the pharmaceutical composition has an oral bioavailability of at least 10%, 20%, 25%, 30%, 35%, 40%, or 45%. In further embodiments, the pharmaceutical composition has an oral bioavailability of at least 30%, at least 35%, at least 40% or at least 45%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 20%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 30%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 35%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 40%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 45%. In certain embodiments, the pharmaceutical composition has an oral bioavailability which yields a therapeutically effective plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least a 24 hour period in the subject with once-daily dosing. In certain embodiments, the pharmaceutical composition has an oral bioavailability which yields a therapeutically effective plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least a 24 hour period in the subject with twice-daily dosing. In certain embodiments, the pharmaceutical composition has an oral bioavailability which yields a therapeutically effective plasma level of a polyamine analog or polyamine biosynthesis inhibitor for at least a 24 hour period in the subject with thrice-daily dosing.

**[0020]** In certain embodiments, the pharmaceutical composition comprising the polyamine analog or polyamine biosynthesis inhibitor has a half life of at least 4, 6, 8, 10, 12, 14, 16, 18, 19, 20, 21, 22, 23, 24, 26, 28, 30, or 36 hours. In certain embodiments, the pharmaceutical composition has a half life of at least 12 hours. In further embodiments, the pharmaceutical composition has a half life of at least 18 hours. In further embodiments, the pharmaceutical composition has a half life of at least 20 hours. In further embodiments, the pharmaceutical composition has a half life of at least 24 hours. In certain embodiments, the pharmaceutical composition has a half life of at least 48, 72, 96, or 120 hours.

**[0021]** Additionally provided herein is a pharmaceutical composition for oral delivery, comprising MGBG together with at least one pharmaceutically acceptable oral excipient.

**[0022]** Also provided herein is an oral pharmaceutical composition, comprising MGBG together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma MGBG level when orally administered to a subject.

**[0023]** Also provided herein is an oral pharmaceutical composition, comprising MGBG together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma MGBG level for the treatment of pain when orally administered to a subject.

**[0024]** In certain embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma MGBG level for a period of at least 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 30, 36, or 48 hours. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma MGBG level for at least a 6-hour period. In further embodiments, the pharmaceutical composition yields a

therapeutically effective systemic plasma MGBG level for at least a 12-hour period. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma MGBG level for at least an 18-hour period. In further embodiments, the pharmaceutical composition yields a therapeutically effective systemic plasma MGBG level for at least a 24-hour period.

**[0025]** In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 25, 50, 55, 60, 65, 75, 80, 85, 90, or 95 percent of the peak plasma concentration for at least 4 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 75% of the peak plasma concentration for at least 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, or 24 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 75% of the peak plasma concentration for at least 4 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 75% of the peak plasma concentration for at least 6 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 75% of the peak plasma concentration for at least 8 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 50% of the peak plasma concentration for at least 8 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 50% of the peak plasma concentration for at least 12 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 50% of the peak plasma concentration for at least 18 hours. In certain embodiments, the pharmaceutical composition yields a plasma level of MGBG of at least 25% of the peak plasma concentration for at least 18 hours. In further embodiments, the peak plasma concentration is a therapeutically effective concentration. In yet further embodiments, the percentage of peak plasma concentration is therapeutically effective over the given time period.

**[0026]** In certain embodiments, the pharmaceutical composition comprising MGBG has an oral bioavailability of at least 10, 20, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 45, 50, 55, or 60 percent. In further embodiments, the pharmaceutical composition has an oral bioavailability of at least 10%, 20%, 25%, 30%, 35%, 40%, or 45%. In further embodiments, the pharmaceutical composition has an oral bioavailability of at least 30%, at least 35%, at least 40% or at least 45%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 20%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 30%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 35%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 40%. In certain embodiments, the pharmaceutical composition has an oral bioavailability of at least 45%. In certain embodiments, the pharmaceutical composition has an oral bioavailability which yields a therapeutically effective plasma level of MGBG for at least a 24 hour period in the subject with once-daily dosing. In certain embodiments, the pharmaceutical composition has an oral bioavailability which yields a therapeutically effective plasma level of MGBG for at least a 24 hour period in the subject with twice-daily dosing. In certain embodiments, the pharmaceutical composition has an oral bioavailability which yields a therapeutically effective plasma level of MGBG for at

least a 24 hour period in the subject with thrice-daily dosing.

**[0027]** In certain embodiments, the pharmaceutical composition comprising MGBG has a half life of at least 4, 6, 8, 10, 12, 14, 16, 18, 19, 20, 21, 22, 23, 24, 26, 28, 30, or 36 hours. In certain embodiments, the pharmaceutical composition has a half life of at least 12 hours. In further embodiments, the pharmaceutical composition has a half life of at least 18 hours. In further embodiments, the pharmaceutical composition has a half life of at least 20 hours. In further embodiments, the pharmaceutical composition has a half life of at least 24 hours. In certain embodiments, the pharmaceutical composition has a half life of at least 48, 72, 96, or 120 hours.

**[0028]** Also provided herein is a pharmaceutical composition comprising MGBG together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma MGBG level when orally administered to a subject, which does not have substantially dose-limiting side effects. In certain embodiments, said side effects are gastrointestinal. In further embodiments, said gastrointestinal side effects are chosen from nausea, vomiting, diarrhea, abdominal pain, oral mucositis, oral ulceration, pharyngitis, stomatitis, and gastrointestinal ulceration. In further embodiments, said gastrointestinal side effects are chosen from inhibition of gastrointestinal mucosal proliferation, inhibition of migration of developing epithelial lumen cells, and inhibition of differentiation of stem or progenitor cells into epithelial lumen cells. In certain embodiments, said side effects are chosen from thrombocytopenia, leukopenia, phlebitis, laryngitis, cellulitis, dermatitis, and hypoglycemia.

**[0029]** Also provided herein is a low-dose oral pharmaceutical composition for chronic delivery, comprising a therapeutically effective amount of MGBG and at least one pharmaceutically acceptable excipient, which does not have substantial gastrointestinal side effects. In certain embodiments, the low-dose oral pharmaceutical composition for chronic delivery, comprising a therapeutically effective amount of MGBG and at least one pharmaceutically acceptable excipient, which does not have substantial gastrointestinal side effects, yields a therapeutically effective plasma level of MGBG for at least a 24 hour period in the subject with once-daily dosing.

**[0030]** In certain embodiments, the pharmaceutical composition is formulated as a tablet or capsule. For example, in certain embodiments, the pharmaceutical composition comprises:

0.1-50% of a polyamine analog or a polyamine biosynthesis inhibitor;

0.1-99.9% of a filler;

0-10% of a disintegrant;

0-5% of a lubricant; and,

0-5% of a glidant.

**[0031]** In certain embodiments, the pharmaceutical composition comprises:

0.1-50% of MGBG;  
0.1-99.9% of a filler;  
0-10% of a disintegrant;  
0-5% of a lubricant; and,  
0-5% of a glidant.

**[0032]** In further embodiments,

said filler is chosen from a sugar, a starch, a cellulose, and a poloxamer;  
said disintegrant is chosen from povidone and crospovidone;  
said lubricant is magnesium stearate; and  
said glidant is silicon dioxide.

**[0033]** In further embodiments,

said filler is chosen from lactose and microcrystalline cellulose;  
said disintegrant is chosen from povidone and crospovidone;  
said lubricant is magnesium stearate; and  
said glidant is silicon dioxide.

**[0034]** In certain embodiments, the pharmaceutical composition comprises:

10-300 mg of a polyamine analog or a polyamine biosynthesis inhibitor, making  
up 2-50% of the tablet content or capsule fill content;  
0-10% of a disintegrant;  
0-5% of a lubricant;  
0-5% of a glidant; and  
30-98% of a filler.

**[0035]** In certain embodiments, the pharmaceutical composition comprises:

10-300 mg of MGBG, making up 2-50% of the tablet content or capsule fill content;  
0-10% of a disintegrant;

0-5% of a lubricant;

0-5% of a glidant; and

30-98% of a filler.

**[0036]** In further embodiments, the pharmaceutical composition comprises

0.1-10% of a binder;

0-5% of a surfactant;

0-10% of an intergranular disintegrant; and

0-10% of an extragranular disintegrant.

**[0037]** In further embodiments, the pharmaceutical composition may additionally comprise

0-10% of a binder;

0-5% of a surfactant;

0-10% of an intergranular disintegrant; and

0-10% of an extragranular disintegrant.

**[0038]** In further embodiments,

said binder is chosen from copolyvidone, hydroxypropyl-cellulose, hydroxypropylmethylcellulose, and povidone;

said surfactant is chosen from polyoxyethylene (20) sorbitan monooleate, a poloxamer, and sodium lauryl sulfate;

said intergranular disintegrant is chosen from croscarmellose sodium, sodium starch glyconate, and crospovidone; and

said extragranular disintegrant is chosen from croscarmellose sodium, sodium starch glyconate, and crospovidone.

**[0039]** Also provided herein is a method of treating or delaying the onset or development of a condition in a subject in need thereof comprising the administration of an oral pharmaceutical composition comprising MGBG and at least one pharmaceutically acceptable excipient. In certain embodiments, the oral pharmaceutical composition is delivered in a therapeutically effective amount. In certain embodiments, said oral pharmaceutical composition has an oral bioavailability of at least 30%. In certain embodiments, said oral pharmaceutical composition

does not have substantially dose-limiting side effects. In certain embodiments, the plasma level of MGBG is at least 75% of the peak plasma concentration for 4 or more hours. In further embodiments, said oral pharmaceutical composition yields a therapeutically effective systemic plasma MGBG level for at least a 12-hour period when orally administered to a subject. In certain embodiments, said condition is chosen from a proliferative disorder, an inflammatory disease, and an autoimmune disease, and neuropathy. In certain embodiments, said condition is chosen from rheumatoid arthritis, osteoarthritis, multiple sclerosis, HIV neuropathy, and HIV associated dementia.

**[0040]** In certain embodiments, said proliferative disorder is chosen from a cancer, psoriasis, psoriatic arthritis and atopic dermatitis. In certain embodiments, the neuropathy is chosen from peripheral neuropathy, diabetic neuropathy, entrapment neuropathy (carpel tunnel syndrome), postherpetic neuralgia (PHN), chemotherapy-induced neuropathy, and HIV neuropathy.

**[0041]** In certain embodiments, the condition is chosen from a proliferative disorder, rheumatoid arthritis, osteoarthritis, multiple sclerosis, HIV neuropathy, and HIV associated dementia. In certain embodiments, the proliferative disorder could, for example, be chosen from a cancer, psoriasis, psoriatic arthritis, and atopic dermatitis. In certain embodiments, the treatment results in decreased levels or activity of osteopontin. In certain embodiments, the method further comprises assaying for osteopontin level, and based on the level of osteopontin, administering an additional therapeutically effective amount of an oral pharmaceutical composition comprising MGBG and at least one pharmaceutically acceptable excipient.

**[0042]** Also provided is an oral pharmaceutical composition, comprising a polyamine analog or polyamine biosynthesis inhibitor together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma level of the polyamine analog or polyamine biosynthesis inhibitor for the treatment of pain when orally administered to a subject. Also provided is an oral pharmaceutical composition, comprising MGBG together with at least one oral pharmaceutically acceptable excipient, which yields a therapeutically effective systemic plasma level of MGBG for the treatment of pain when orally administered to a subject.

**[0043]** Also provided herein is a method of treatment of pain in a subject in need thereof comprising the administration of a polyamine analog or a polyamine biosynthesis inhibitor, or a salt or protected derivative thereof. Also provided herein is a method of treatment of pain in a subject in need thereof comprising the administration of MGBG. In certain embodiments, the MGBG is administered in a therapeutically effective amount. Further provided is a method of treatment of pain in a subject in need thereof comprising the administration of a therapeutically effective amount of a pharmaceutical composition comprising MGBG and at least one pharmaceutically acceptable excipient.

**[0044]** In certain embodiments, the pain is chosen from inflammatory pain, pain due to nerve injury, chronic pain, intractable cancer pain, complex regional pain syndrome, neuropathic

pain, surgical or post-surgical pain, dental pain, pain resulting from dermal injury, lower back pain, headaches, migraine, tactile allodynia, and hyperalgesia. In certain embodiments, the pain is chronic. In other embodiments, the pain is acute. In certain embodiments, the pain is inflammatory pain.

**[0045]** In certain embodiments, the administration of MGBG or its pharmaceutical composition is oral. In other embodiments, the administration is intravenous.

**[0046]** In certain embodiments, the administration is a combination of oral and intravenous. In certain embodiments, the first administration is oral and the second IV; in others the first is IV and the second oral; in either case, additional oral or IV dosing may follow. In certain embodiments, the pain is surgical or post-surgical pain. For example, in certain embodiments, the pre-surgical administration is oral and the peri-surgical administration is IV; in others the pre-surgical administration is IV, the pre-surgical administration is also IV, and the post-surgical administration is oral. In either case, additional oral or IV dosing may follow. In certain embodiments, the pre-, peri-, and post- surgical administration is IV.

**[0047]** Also provided herein is a method of treatment of HIV neuropathy in a subject in need thereof comprising the administration of a therapeutically effective amount of an oral pharmaceutical composition comprising MGBG and at least one pharmaceutically acceptable excipient. In certain embodiments, the plasma level of MGBG is at least 75% of the peak plasma concentration for 4 or more hours.

**[0048]** Also provided herein is a method of treatment of a condition in a subject in need thereof comprising the administration of:

an oral pharmaceutical composition comprising MGBG and at least one pharmaceutically acceptable excipient; and

another therapeutic agent.

**[0049]** In certain embodiments, the MGBG is delivered in a therapeutically effective amount. In other embodiments, the MGBG is delivered in a subtherapeutic amount. In certain embodiments, the other therapeutic agent is delivered in a therapeutically effective amount. In other embodiments, the other therapeutic agent is delivered in a subtherapeutic amount. In certain embodiments, the MGBG and the other therapeutic agent are delivered together in amounts which would individually be subtherapeutic but which together are therapeutically effective. In other embodiments, the MGBG and the other therapeutic agent are delivered together in amounts which are individually therapeutically effective.

**[0050]** Additionally provided herein are methods: of regulation of osteopontin activity or levels in a subject as well as for treating or preventing conditions associated with an increased level or activity of osteopontin in a subject; of decreasing levels or the activity of osteopontin in a

cell, comprising contacting a cell with an effective amount of an agent that inhibits S-adenosyl methionine decarboxylase ("AMD-I"), or inhibits polyamine biosynthesis; of decreasing the level or activity of osteopontin in a cell, comprising contacting the cell with an effective amount of MGBG, a salt of MGBG, or a protected derivative of MGBG; of treating or preventing a condition associated with an increased level or activity of osteopontin, comprising administering to a subject in need of such treatment an effective amount of an agent that inhibits S-adenosyl methionine decarboxylase, or inhibits polyamine biosynthesis in the subject.

**[0051]** Additionally provided herein is a method of treating a condition. The method comprises administering to a subject in need of such treatment an effective amount of MGBG, a salt of MGBG, a protected derivative of MGBG, or a polyamine analog or polyamine biosynthesis inhibitor or a salt, a protected derivative, or a stereoisomer thereof, wherein the condition is chosen from Crohn's disease, Parkinson's disease, inflammatory bowel disorder, multiple sclerosis (MS), amyotrophic lateral sclerosis (ALS), hepatitis, HBY, HCV, nephritis, cerebritis, glomerulonephritis, rheumatoid arthritis, type 2 diabetes, cardiac fibrosis and angiotensin type II associated hypertension, osteoporosis, a mast cell produced IgE mediated hypersensitivity immune reaction, peripheral sensory neuropathy associated with HIV infection or diabetes mellitus, asthma, autism, dermatomyositis, frailty, obesity, primary biliary cirrhosis, primary sclerosing cholangitis, post-radiation syndrome, psoriatic arthritis, sarcoidosis, scleroderma with or without pulmonary fibrosis, a kidney related autoimmune condition, diabetic nephropathy, a diabetic vascular complication, and a lymphoproliferation related autoimmune condition.

**[0052]** Additionally provided herein is a method of decreasing osteopontin secretion from monocytes or macrophages, comprising contacting a monocyte or macrophage with an effective amount of an agent that inhibits S-adenosyl methionine decarboxylase or inhibits polyamine biosynthesis in the monocyte or macrophage. Further provided herein is a method of decreasing osteopontin secretion from monocytes or macrophages, comprising contacting a monocyte or macrophage with an effective amount of MGBG or a salt or protected derivative thereof.

**[0053]** Additionally provided herein is a method of decreasing differentiation of macrophages from monocytes, comprising contacting a monocyte with an effective amount of an agent that inhibits S-adenosyl methionine decarboxylase or inhibits polyamine biosynthesis in the monocyte. In certain embodiments the agent is MGBG, or a salt or protected derivative thereof.

**[0054]** Described herein is a method of treating or preventing a condition associated with an increased level or activity of osteopontin. The method comprises administering to a subject in need of such treatment an effective amount of an agent that regulates the activity of osteopontin. The condition can be any condition now known, or later discovered, to be associated with an increased level or activity of osteopontin. Examples of conditions associated with an increased activity of osteopontin include, but are not limited to, autoimmune conditions, inflammatory conditions, neoplastic growth and tumor metastases. In one embodiment, the

condition associated with an increased level or activity of osteopontin is infiltration of immune cells to an affected area or increased level of CD14/CD16 macrophages in a subject.

**[0055]** In another embodiment, conditions associated with an increased activity of osteopontin include, but are not limited to, multiple sclerosis (MS), atherosclerosis and related coronary conditions, rheumatoid arthritis, lupus, nephritis, cerebritis, Crohn's disease, osteoporosis, inflammatory bowel disorder, breast cancer, ovarian cancer, pancreatic cancer, bladder cancer, lung cancer, colon cancer, gastric carcinomas, esophageal carcinomas, squamous cell carcinomas of the head or neck, prostate cancer, thyroid cancer, melanoma, kidney cancers, renal cell carcinomas, endometrial cancer, small intestine cancer, duodenal cancer, cholangiocarcinoma, astrocytoma, AIDS lymphoma, follicular lymphoma, T-cell lymphoma, B-cell lymphoma, proliferative retinopathy, vitreoretinopathy, diabetic retinopathy, macular degeneration, non-HIV associated dementia, HIV- and AIDS-associated dementia, focal segmental glomerulosclerosis, membrane proliferative glomerulonephropathy, psoriasis, herpes virus associated disease, Castleman's disease, Kaposi's sarcoma, Alzheimer's disease, type 2 diabetes, cardiac fibrosis and angiotensin type II associated hypertension, mast cell produced IgE mediated hypersensitivity immune reactions, prelymphomatous or lymphoproliferation related autoimmune conditions, angioimmunoblastic lymphadenopathy (AILD), glomerulonephritis and other glomerular diseases, immunoglobulin A (IgA) nephropathy, Amyotrophic Lateral Sclerosis (ALS), hepatitis including HBV and HCV, peripheral sensory neuropathy associated with HIV infection or diabetes mellitus, asthma, autism, dermatomyositis, frailty, obesity, Parkinson's disease, primary biliary cirrhosis, primary sclerosing cholangitis, post-radiation syndrome, psoriatic arthritis, sarcoidosis, scleroderma with or without pulmonary fibrosis, kidney related autoimmune conditions, diabetic nephropathy and other diabetic vascular complications.

**[0056]** According to yet another aspect, described herein is a method of decreasing osteopontin secretion from monocytes or macrophages. The method comprises contacting a monocyte or macrophage with an effective amount of an agent that regulates the activity of osteopontin.

**[0057]** According to yet another aspect, described herein is a method of decreasing differentiation of macrophages from monocytes. The method comprises contacting a monocyte with an effective amount of an agent that regulates the activity of osteopontin. In certain embodiments the agent that regulates the activity of osteopontin is MGBG, or a salt or protected derivative thereof.

**[0058]** Also provided herein is a method of reducing viral load of an infected subject comprising administering to a subject infected by an immunodeficiency virus a therapeutically effective amount of a polyamine analog or a polyamine biosynthesis inhibitor whereby reducing the viral load in the subject infected by the immunodeficiency virus. In certain embodiments the polyamine biosynthesis inhibitor is MGBG, or a salt or protected derivative thereof.

**[0059]** Also provided herein is a method for treating an immunodeficiency viral infection

comprising administering to a subject infected by an immunodeficiency virus a therapeutically effective amount of a polyamine analog or polyamine biosynthesis inhibitor and at least one an antiviral agent. Also provided herein is a method for preventing the onset of AIDS or conditions secondary to HIV infection comprising administering to a subject infected with human immunodeficiency virus a therapeutically effective amount of a polyamine analog. In certain embodiments the polyamine biosynthesis inhibitor is MGBG, or a salt or protected derivative thereof.

**[0060]** Also provided herein is a pharmaceutical formulation which comprises a polyamine analog or polyamine biosynthesis inhibitor, an antiviral agent, and a pharmaceutically acceptable carrier, which may optionally be provided as a kit. In certain embodiments, this pharmaceutical formulation is in an amount of a single dose suitable to be taken by a subject in need of such treatment. In certain embodiments, said treatment is for a viral infection.

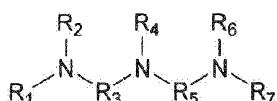
**[0061]** The agent useful in the methods described herein can be any agent that decreases the activity of osteopontin. In one embodiment, the agent is capable of inhibiting S-adenosyl methionine decarboxylase ("AMD-I") or any pathway containing AMD-I, e.g., any entity upstream or downstream of a pathway containing AMD-I, especially any pathway containing AMD-I and associated with adenosine production. In another embodiment the agent is capable of inhibiting polyamine biosynthesis or any pathway involved in polyamine biosynthesis. In general, a pathway containing AMD-I or adenosine is understood to refer to a pathway in which either AMD-I or adenosine is involved, including, for example, as a substrate, catalyst, product or by-product.

**[0062]** The agent can be any kind of known or later discovered agent that can inhibit the activity of the enzyme S-adenosyl methionine decarboxylase, can inhibit polyamine biosynthesis in, for example, a cell. In one embodiment, the agent is a chemical agent, including, but not limited to, organic molecules and salts, protected derivatives and stereoisomers thereof, inorganic molecules or various ionic or elemental entities.

**[0063]** Compounds for use in the methods and compositions disclosed herein include polyamine analogs and polyamine biosynthesis inhibitors, as well as salts, prodrugs, solvates, anhydrous forms, protected derivatives, structural isomers, stereoisomers, amino acid conjugates, and porphyrin conjugates thereof. Any polyamine analog is suitable for use in the methods described herein.

**[0064]** Exemplary polyamine analogs used in the methods described herein include compounds of the structural formulas 1, 2, 3, 4, 5, 6, and 7 and the corresponding stereoisomers, salts, and protected derivatives thereof.

Formula 1 has the structure

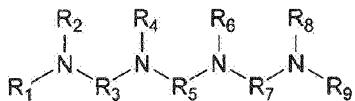


wherein

$R_1$ ,  $R_2$ ,  $R_4$ ,  $R_6$  and  $R_7$  are independently chosen from hydrogen, alkyl and aryl; and

$R_3$  and  $R_5$ , are alkyl groups.

Formula 2 has the structure

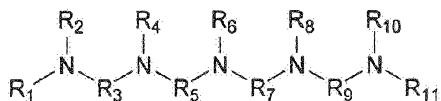


wherein

$R_1$ ,  $R_2$ ,  $R_4$ ,  $R_6$ ,  $R_8$ , and  $R_9$  are independently chosen from hydrogen, alkyl and aryl; and

$R_3$ ,  $R_5$  and  $R_7$  are alkyl groups.

Formula 3 has the structure

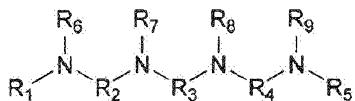


wherein

$R_1$ ,  $R_2$ ,  $R_4$ ,  $R_6$ ,  $R_{10}$  and  $R_{11}$  are independently chosen from hydrogen, alkyl and aryl; and

$R_3$ ,  $R_5$ ,  $R_7$  and  $R_9$  are alkyl groups.

Formula 4 has the structure



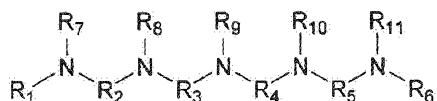
wherein

$R_1$  and  $R_5$  are independently chosen from methyl, ethyl, n-propyl, and isopropyl;

$R_2$ ,  $R_3$ , and  $R_4$  are independently chosen from C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>2</sub>-C<sub>6</sub> alkenyl, C<sub>3</sub>-C<sub>6</sub> cycloalkyl, C<sub>1</sub>-C<sub>6</sub> alkyl-C<sub>3</sub>-C<sub>6</sub> cycloalkyl- C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>3</sub>-C<sub>10</sub> aryl, and C<sub>1</sub>-C<sub>6</sub> alkyl-C<sub>3</sub>-C<sub>10</sub> aryl-C<sub>1</sub>-C<sub>6</sub> alkyl; and

$R_6$ ,  $R_7$ ,  $R_8$  and  $R_9$  are independently chosen from hydrogen, methyl, and ethyl;

Formula 5 has the structure



wherein

$R_1$  and  $R_6$  are independently chosen from methyl, ethyl, n-propyl, and isopropyl;

$R_2$ ,  $R_3$ ,  $R_4$  and  $R_5$  are independently chosen from C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>2</sub>-C<sub>6</sub> alkenyl, C<sub>3</sub>-C<sub>6</sub> cycloalkyl, C<sub>1</sub>-C<sub>6</sub> alkyl- C<sub>3</sub>-C<sub>6</sub> cycloalkyl- C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>3</sub>-C<sub>10</sub> aryl, and C<sub>3</sub>-C<sub>10</sub> aryl-C<sub>1</sub>-C<sub>6</sub> alkyl; and

$R_7$ ,  $R_8$ ,  $R_9$ ,  $R_{10}$  and  $R_{11}$  are independently chosen from hydrogen, methyl, and ethyl.

**[0065]** In another embodiment, the polyamine analogs are compounds of the structures 2 and 3, wherein

$R_3$ ,  $R_5$ ,  $R_7$  and  $R_9$  are independently  $(CH_2)_x$  groups;

$x$  is an integer from 2 to 6; and

$R_4$ ,  $R_6$  and  $R_8$  are hydrogen atoms.

**[0066]** In yet another embodiment, the polyamine analogs are compounds of the structures 2 and 3, wherein

$R_3$ ,  $R_5$ ,  $R_7$  and  $R_9$  are independently  $(CH_2)_x$  groups;

$x$  is an integer from 2 to 6;

$R_4$ ,  $R_6$  and  $R_8$  are hydrogen atoms;

$R_1$  and  $R_{10}$  are alkyl groups; and

$R_2$  and  $R_{11}$  are hydrogen atoms.

**[0067]** In yet another embodiment, the polyamine analogs are compounds of the structures 2 and 3, wherein

$R_3$ ,  $R_5$ ,  $R_7$  and  $R_9$  are independently  $(CH_2)_x$  groups;

$x$  is an integer from 2 to 6;

$R_4$ ,  $R_6$  and  $R_8$  are hydrogen atoms;

$R_1$  and  $R_{10}$  are alkyl groups;

$R_2$  and  $R_{11}$  are hydrogen atoms; and

the polyamine analogs have a molecular weight less than 500.

**[0068]** Further embodiments of compounds of the structure 4 include those wherein  $R_6$ ,  $R_7$ ,  $R_8$  and  $R_9$  are hydrogen.

**[0069]** In other embodiments,  $R_1$  and  $R_5$  are ethyl.

**[0070]** In yet further embodiments,

$R_6$ ,  $R_7$ ,  $R_8$  and  $R_9$  are hydrogen; and

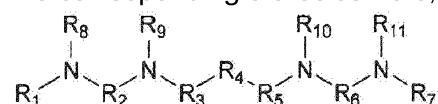
$R_1$  and  $R_5$  are ethyl.

**[0071]** In yet further embodiments,

$R_2$  and  $R_4$  are independently chosen from  $C_1$ - $C_6$  alkyl; and

$R_3$  is chosen from  $C_1$ - $C_6$  alkyl,  $C_2$ - $C_6$  alkenyl,  $C_3$ - $C_6$  cycloalkyl,  $C_1$ - $C_6$  alkyl- $C_3$ - $C_6$  cycloalkyl-  $C_1$ - $C_6$  alkyl,  $C_3$ - $C_{10}$  aryl, and  $C_1$ - $C_6$  alkyl- $C_3$ - $C_{10}$  aryl- $C_1$ - $C_6$  alkyl.

**[0072]** Additional polyamine analogs useful herein include compounds of the formula 6, and the corresponding stereoisomers, salts, and protected derivatives thereof:



wherein

$R_4$  is chosen from  $C_2$ - $C_6$  n-alkenyl,  $C_3$ - $C_6$  cycloalkyl,  $C_3$ - $C_6$  cycloalkenyl, and  $C_3$ - $C_6$  aryl;

$R_3$  and  $R_5$  are independently chosen from a single bond,  $C_1$ - $C_6$  alkyl, and  $C_1$ - $C_6$  alkenyl;

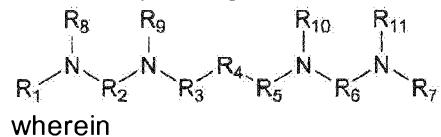
$R_2$  and  $R_6$  are independently chosen from  $C_1$ - $C_6$  alkyl,  $C_1$ - $C_6$  alkenyl,  $C_3$ - $C_6$  cycloalkyl,  $C_3$ - $C_6$  cycloalkenyl, and  $C_3$ - $C_6$  aryl;

$R_1$  and  $R_7$  are independently chosen from hydrogen,  $C_1$ - $C_6$  alkyl, and  $C_2$ - $C_6$  alkenyl; and

$R_8$ ,  $R_9$ ,  $R_{10}$ , and  $R_{11}$  are hydrogen.

**[0073]** In certain embodiments of the compounds of formula 6,  $R_1$  and  $R_7$  are independently chosen from  $C_1$ - $C_6$  alkyl and  $C_2$ - $C_6$  alkenyl.

**[0074]** Additional polyamine analogs useful herein include compounds of the formula 7, and the corresponding stereoisomers, salts, and protected derivatives thereof:



$R_4$  is chosen from  $C_1$ - $C_6$  n-alkyl and  $C_1$ - $C_6$  branched alkyl;

$R_3$  and  $R_5$  are independently chosen from a single bond or  $C_1$ - $C_6$  alkyl;

$R_2$  and  $R_6$  are independently chosen from  $C_1$ - $C_6$  alkyl,  $C_1$ - $C_6$  alkenyl,  $C_3$ - $C_6$  cycloalkyl,  $C_3$ - $C_6$  cycloalkenyl, or  $C_3$ - $C_6$  aryl;

$R_1$  and  $R_7$  are independently chosen from H,  $C_1$ - $C_6$  alkyl, or  $C_2$ - $C_6$  alkenyl; and

$R_8$ ,  $R_9$ ,  $R_{10}$ , and  $R_{11}$  are hydrogen.

**[0075]** In certain embodiments of the compounds of formula 7

$R_2$  and  $R_7$  are independently chosen from  $C_1$ - $C_6$  alkyl or  $C_2$ - $C_6$  alkenyl;

$R_4$  is chosen from  $C_1$ - $C_6$  saturated n-alkyl and  $C_1$ - $C_6$  saturated branched alkyl; and

$R_3$  and  $R_5$  are independently chosen from a single bond and  $C_1$ - $C_6$  saturated n-alkyl.

**[0076]** According to another embodiment described herein, the agent is a chemical moiety that inhibits the activity of S-adenosyl methionine decarboxylase, inhibits polyamine biosynthesis, and/or increases the activity of adenosine.

**[0077]** Examples of such moieties include, but are not limited to, those listed in Table 1. Irrespective of the form of the moiety listed in Table 1, it is understood that it includes, as applicable, a salt, protected derivative, and stereoisomer thereof.

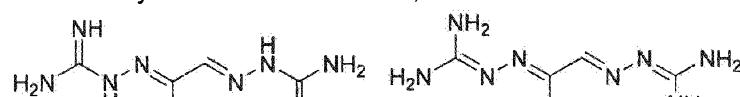
Table 1.

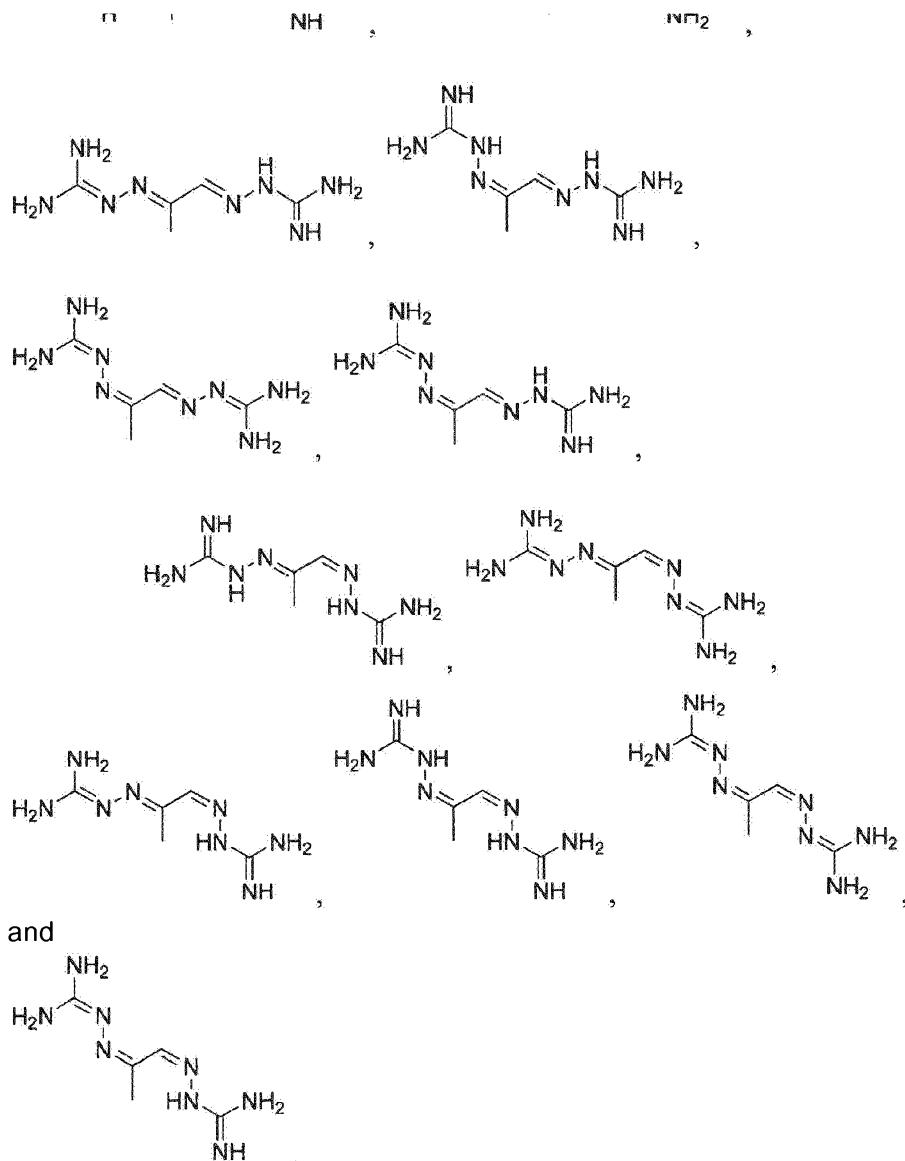
Compound	Official Name (Not IUPAC)	Pub Chem ID
Decarboxylated SAM	s-adenosyl-3-methylthiopropylamine	5351154
Mitoguazone or "MGBG"	Methylglyoxal bis(guanylhydrazone)	9561662
EGBG	Ethylglyoxal bis(guanylhydrazone)	2354
Berenil	Diminazene or Diminazene aceturate	4735
Pentamidine	4- [5-(4-carbamimidoylphenoxy)penoxy] benzenecarboximidamide 5'-(Dimethylsulphino)-5'-deoxyadenosine S-adneosyl-4-methylthiobutyrate S-adenosyl-S-methyl-L-cysteine	
AMA	S-(5'-Deoxy-5'-adenosyl) methylthioethylhydroxylamine	
EMGBG	Ethylmethylglyoxal bis(guanylhydrazone)	
DEGBG	Diethylglyoxal bis(guanylhydrazone)	9574151
CGP-33'829	6-((2-carbamimidoylhydrazone)methyl) picolinimidamide	5479208
CGP-36'958		
CGP-39'937	2,2'-bipyridine-6,6'-bis(carboximidamide)	
CGP-48664 or CGP48664A or SAM 364A	4-amidinoindan-1-one 2'-amidinohydrazone	5486811
AbeAdo or MDL-73 811	5'-[[(Z)-4-amino-2-butenyl] methylamino]-5'-deoxyadenosine	6436013
MAOEA	5'-deoxy-5'-[N-methyl-N-[2-(aminoxy)ethyl] amino] adenosine	3081018
MHZPA	5'-deoxy-5'-[N-methyl-N-(3-hydrazinopropyl)amino]adenosine	122092
MHZEA	5'-deoxy-5'-[(2-hydrazinoethyl)-methylamino] adenosine	
AdoMac	S-(5'-deoxy-5'-adenosyl)-1-ammonio-4-(methylsulfonio)-2cyclopentene	3083364
AdoMao	S-(5'-deoxy-5'-adenosyl)-1-aminoxy-4-(methylsulfonio)-2-cyclopentene	
APA	1-Aminoxy-3-aminopropane	65020
AOE-PU	N-[2-aminoxyethyl]-1,4-diaminobutane	

Compound	Official Name (Not IUPAC)	Pub Chem ID
AP-APA	1-aminoxy-3-N-[3-aminopropyl]-aminopropane 1,11-bis(ethyl)norspermine	
BES	1,8-bis(ethyl)spermidine	
BES	1,12-bis(ethyl)spermine	
DESPM	N1,N12-diethylspermine	
BE-3-3-3	1,11-bis(ethylamino)-4,8-diazaundecan	
BE-4-4-4	1,14-bis(ethylamino)-5,10-diazatetradecane	
DEHOP or DEHSPM	Diethylhomospermine, N1,N14-diethylhomospermine	
DENOP	diethyl-norspermine	
BE-4-4-4-4	1,19-bis(ethylamino)-5,10,15-traza-nonadecane	
SL1103 7	N-ethyl-N'-(2-(3'-ethylamino-propylamino methyl)-cis-cyclopropylmethyl)-propane 1,3-diamine tetrahydrochloride	
SL1103 8	N-ethyl-N'-(2-(3'-ethylamino-propylamino methyl)-trans-cyclobutylmethyl)-propane 1,3-diamine tetrahydrochloride	
SL11044	N-ethyl-N'-(2-(3'-ethylamino-propylamino methyl)-transcyclopropylmethyl)-propane 1,3-diamine tetrahydrochloride	
SL11047 or SL47	N,N'-bis(3-ethylaminopropyl)-cis-but-2-ene-1,4-diaminetetrahydrochloride	
SL11093 or SL93	N,N'-(cyclopropane-1,2-diylbis(methylene))bis(N4-ethylbutane-1,4-diamine)	

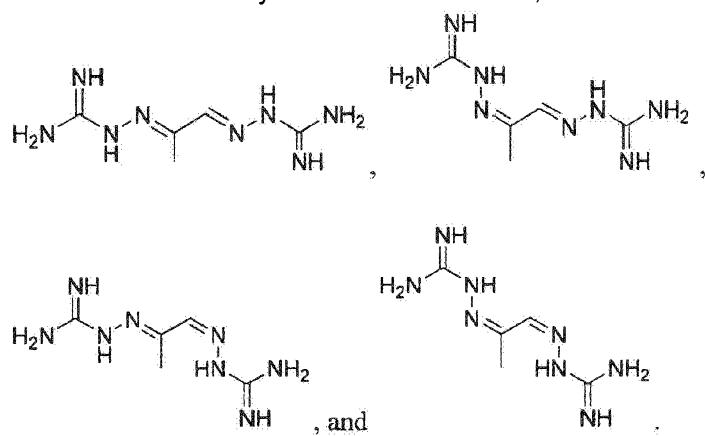
**[0078]** In yet another embodiment, the agent is a compound chosen from MGBG, MDL73811, CGP48664, Berenil, Pentamidine, SL47, and SL93, or a combination of two or more thereof. In yet another embodiment, the agent is MGBG, SL47 or SL93. In still another embodiment, two or more agents are used in the methods described herein to regulate the activity of osteopontin. The two or more agents can be used either sequentially or simultaneously.

**[0079]** MGBG is 1,1'[(methylethanediylidene)dinitrilodiguanidine and is also known as methylglyoxal bis(guanylhydrazone), methyl-GAG, Me-G, and mitoguazone. As used herein, MGBG includes the free base and salts thereof. It is commonly, but not necessarily, used as a dihydrochloride. MGBG may be present as any one of the following isomers, or a tautomer and/or a syn/anti isomer thereof, mixture of one or more thereof:

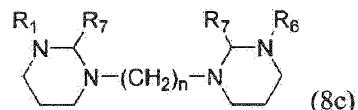
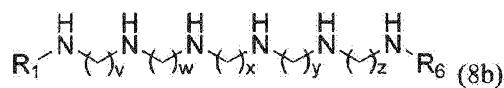
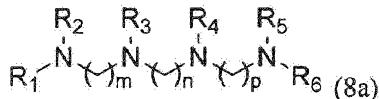




**[0080]** In certain embodiments, MGBG may be present one of the following isomers, or a tautomer and/or a syn/anti isomer thereof, mixture of one or more thereof:



**[0081]** In certain embodiments, compounds have a structure chosen from Formulas 8a-8c:



$\text{R}_1$ - $\text{R}_6$  are chosen from hydrogen, alkyl and aralkyl having from 1 to 12 carbon atoms, provided that, in formula (8a),  $\text{R}_1$ , and  $\text{R}_6$  are not hydrogen;

$\text{R}_7$  chosen from hydrogen, alkyl, aryl and aralkyl having from 1 to 12 carbon atoms;

$\text{m}$ ,  $\text{n}$ , are each independently an integer from 3 to 6, inclusive; and

$\text{v}$ ,  $\text{w}$ ,  $\text{x}$ ,  $\text{y}$ , and  $\text{z}$  are each independently an integer from 3 to 10, inclusive.

**[0082]** Additional disclosure may be found in WO98/10766, for example on pp. 3-4.

**[0083]** In certain embodiments, compounds have a structure of Formula 9a:



wherein

$\text{A}$  is independently selected from the group consisting of  $\text{C}_1$ - $\text{C}_6$  alkyl,  $\text{C}_2$ - $\text{C}_6$  alkenyl,  $\text{C}_2$ - $\text{C}_6$  alkynyl,  $\text{C}_3$ - $\text{C}_6$  cycloalkyl,  $\text{C}_3$ - $\text{C}_6$  aryl, and  $\text{C}_3$ - $\text{C}_6$  cycloalkenyl;

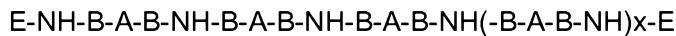
$\text{B}$  is independently selected from the group consisting of: a single bond,  $\text{C}_1$ - $\text{C}_6$  alkyl, and  $\text{C}_2$ - $\text{C}_6$  alkenyl; and

$\text{E}$  is independently selected from the group consisting of hydrogen,  $\text{C}_1$ - $\text{C}_6$  alkyl,  $\text{C}_2$ - $\text{C}_6$  alkenyl,  $\text{C}_2$ - $\text{C}_6$  alkynyl,  $\text{C}_3$ - $\text{C}_6$  cycloalkyl,  $\text{C}_3$ - $\text{C}_6$  aryl, and  $\text{C}_3$ - $\text{C}_6$  cycloalkenyl;

with the proviso that either at least one  $\text{A}$  moiety is selected from the group consisting of  $\text{C}_2$ - $\text{C}_6$  alkenyl,  $\text{C}_2$ - $\text{C}_6$  alkynyl,  $\text{C}_3$ - $\text{C}_6$  cycloalkyl,  $\text{C}_3$ - $\text{C}_6$  aryl, and  $\text{C}_3$ - $\text{C}_6$  cycloalkenyl, or at least one  $\text{B}$  moiety is selected from the group consisting of  $\text{C}_2$ - $\text{C}_6$  alkenyl; and all salts, hydrates, solvates, and stereoisomers thereof.

**[0084]** In another embodiment, the conformationally restricted polyamine analog is selected

from among the group of compounds of the formula 9b:



wherein:

A is independently selected from the group consisting of C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>2</sub>-C<sub>6</sub> alkenyl, C<sub>2</sub>-C<sub>6</sub> alkynyl, C<sub>3</sub>-C<sub>6</sub> cycloalkyl, C<sub>3</sub>-C<sub>6</sub> aryl, and C<sub>3</sub>-C<sub>6</sub> cycloalkenyl;

B is independently selected from the group consisting of: a single bond, C<sub>1</sub>-C<sub>6</sub> alkyl, and C<sub>2</sub>-C<sub>6</sub> alkenyl; and

E is independently selected from the group consisting of hydrogen, C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>2</sub>-C<sub>6</sub> alkenyl, C<sub>2</sub>-C<sub>6</sub> alkynyl, C<sub>3</sub>-C<sub>6</sub> cycloalkyl, C<sub>3</sub>-C<sub>6</sub> aryl, and C<sub>3</sub>-C<sub>6</sub> cycloalkenyl;

x is an integer from 2 to 16;

with the proviso that either at least one A moiety is selected from the group consisting of C<sub>2</sub>-C<sub>6</sub> alkenyl, C<sub>2</sub>-C<sub>6</sub> alkynyl, C<sub>3</sub>-C<sub>6</sub> cycloalkyl, C<sub>3</sub>-C<sub>6</sub> aryl, and C<sub>3</sub>-C<sub>6</sub> cycloalkenyl, or at least one B moiety is selected from the group consisting of C<sub>2</sub>-C<sub>6</sub> alkenyl;

and all salts, hydrates, solvates, and stereoisomers thereof.

**[0085]** In another embodiment, x is 4, 6, 8, or 10.

**[0086]** In another embodiment, x is 4. In another embodiment, x is 6.

**[0087]** In another embodiment, x is 8.

**[0088]** In another embodiment, x is 10.

**[0089]** In another embodiment, the conformationally restricted polyamine analog is selected from among the group of compounds of the formula 9c:



wherein:

A is independently selected from the group consisting of C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>2</sub>-C<sub>6</sub> alkenyl, C<sub>2</sub>-C<sub>6</sub> alkynyl, C<sub>3</sub>-C<sub>6</sub> cycloalkyl, C<sub>3</sub>-C<sub>6</sub> aryl, and C<sub>3</sub>-C<sub>6</sub> cycloalkenyl;

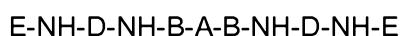
B is independently selected from the group consisting of: a single bond, C<sub>1</sub>-C<sub>6</sub> alkyl, and C<sub>2</sub>-C<sub>6</sub> alkenyl; and

E is independently selected from the group consisting of C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>1</sub>-C<sub>6</sub> alkanol, C<sub>3</sub>-C<sub>6</sub> cycloalkanol, and C<sub>3</sub>-C<sub>6</sub> hydroxyaryl, with the proviso that at least one E moiety be selected from the group consisting of C<sub>1</sub>-C<sub>6</sub> alkanol, C<sub>3</sub>-C<sub>6</sub> cycloalkanol, and C<sub>3</sub>-C<sub>6</sub> hydroxyaryl; and

x is an integer from 0 to 16;

and all salts, hydrates, solvates, and stereoisomers thereof.

**[0090]** In another embodiment, the conformationally restricted polyamine analog is selected from among the group of compounds of the formula 9d:



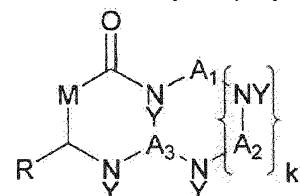
wherein A is selected from the group consisting of C<sub>2</sub>-C<sub>6</sub> alkene and C<sub>3</sub>-C<sub>6</sub> cycloalkyl, cycloalkenyl, and aryl;

B is independently selected from the group consisting of a single bond and C<sub>1</sub>-C<sub>6</sub> alkyl and alkenyl;

D is independently selected from the group consisting of C<sub>1</sub>-C<sub>6</sub> alkyl and alkenyl, and C<sub>3</sub>-C<sub>6</sub> cycloalkyl, cycloalkenyl, and aryl;

E is independently selected from the group consisting of hydrogen, C<sub>1</sub>-C<sub>6</sub> alkyl and alkenyl; and all salts, hydrates, solvates, and stereoisomers thereof.

**[0091]** In another embodiment, the conformationally restricted polyamine analog is selected from macrocyclic polyamines of the formula 9e:



wherein

A<sub>1</sub>, each A<sub>2</sub> (if present), and A<sub>3</sub> are independently selected from C<sub>1</sub>-C<sub>8</sub> alkyl;

each Y is independently selected from hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl;

M is selected from C<sub>1</sub>-C<sub>4</sub> alkyl;

k is 0, 1, 2, or 3; and

R is selected from C<sub>1</sub>-C<sub>32</sub> alkyl;

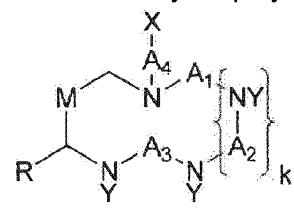
and all salts, hydrates, solvates, and stereoisomers thereof.

**[0092]** In additional embodiments, the Y group is hydrogen or -CH<sub>3</sub>.

**[0093]** In another embodiment, A<sub>1</sub>, each A<sub>2</sub> (if present), and A<sub>3</sub> are independently selected from C<sub>2</sub>-C<sub>4</sub> alkyl.

**[0094]** In yet another embodiment, M is -CH<sub>2</sub>-.

**[0095]** In another embodiment, the conformationally restricted polyamine analog is selected from macrocyclic polyamine analogs of the formula 9f:



wherein

A<sub>1</sub>, each A<sub>2</sub> (if present), and A<sub>3</sub> are independently selected from C<sub>1</sub>-C<sub>8</sub> alkyl;

A<sub>4</sub> is selected from C<sub>1</sub>-C<sub>8</sub> alkyl or null;

X is selected from - hydrogen, -Z, -CN, -NH<sub>2</sub>, -C(=O)- C<sub>1</sub>-C<sub>8</sub>-alkyl, or-NHZ, with the proviso that when A<sub>4</sub> is null, X is hydrogen, -C(=O)-C<sub>1</sub>-C<sub>8</sub>-alkyl, or -Z;

Z is selected from the group consisting of an amino protecting group, an amino capping group, an amino acid, and a peptide;

each Y is independently selected from hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl;

M is selected from C<sub>1</sub>-C<sub>4</sub> alkyl;

k is 0, 1, 2, or 3; and

R is selected from C<sub>1</sub>-C<sub>32</sub> alkyl;

and all salts, hydrates, solvates, and stereoisomers thereof.

**[0096]** In certain embodiments, A<sub>4</sub> is null.

**[0097]** In other embodiments, X is -Z, and -Z is hydrogen.

**[0098]** In other embodiments, X is -Z, and -Z is 4-morpholinocarbonyl.

**[0099]** In other embodiments, X is -Z and -Z is acetyl.

**[0100]** In other embodiments, X is -Z and -Z is t-Boc or Fmoc.

**[0101]** In other embodiments, Y is -CH<sub>3</sub>.

**[0102]** In other embodiments, M is -CH<sub>2</sub>-.

**[0103]** In still further embodiments, k is 1.

**[0104]** In further embodiments, A, and A<sub>3</sub> are -CH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>-.

**[0105]** In still further embodiments, - CH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>-.

**[0106]** In still further embodiments, R is C<sub>13</sub>H<sub>27</sub>.

**[0107]** In yet further embodiments, one or more of the specific limitations on A<sub>4</sub>, X, Z, Y, M, k, A<sub>1</sub>, A<sub>3</sub>, and R are combined.

**[0108]** In further embodiments of macrocyclic polyamine analog compounds, A<sub>4</sub> is C<sub>1</sub>-C<sub>8</sub> alkyl;

X is -NHZ; and

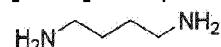
Z is selected from one of the 20 genetically encoded amino acids (alanine, cysteine, aspartic acid, glutamic acid, phenylalanine, glycine, histidine, isoleucine, lysine, methionine, asparagine, proline, glutamine, arginine, serine, threonine, valine, tryptophan, tyrosine), a peptide of the formula acetyl-SKLQL-, a peptide of the formula acetyl-SKLQ-I3-alanine-, or a peptide of the formula acetyl-SKLQ-.

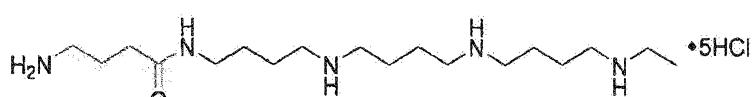
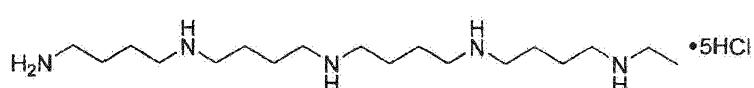
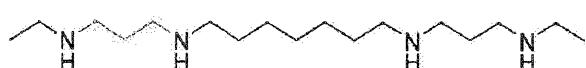
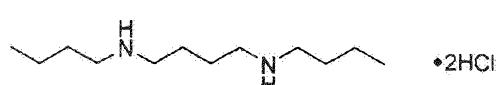
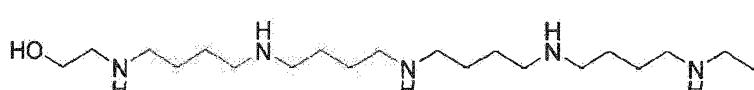
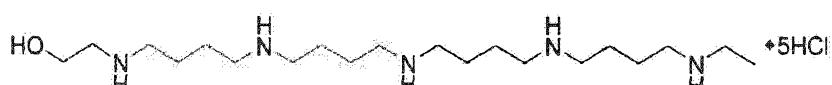
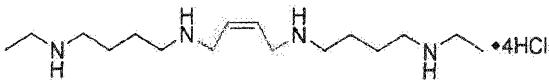
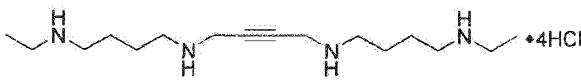
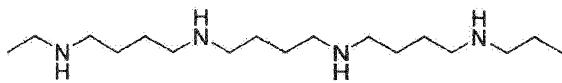
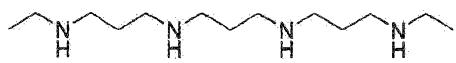
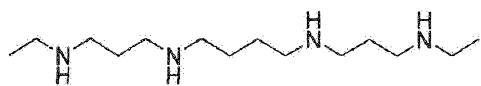
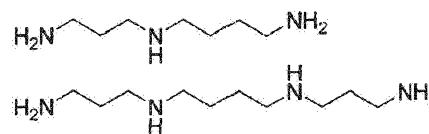
**[0109]** In these cases, where Z is an amino acid or peptide, the therapeutic agent to be used is a polyamine-amino acid conjugate or polyamine-peptide conjugate.

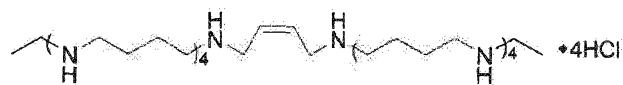
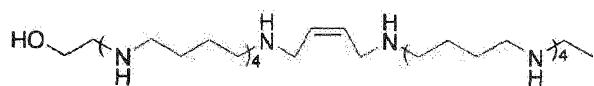
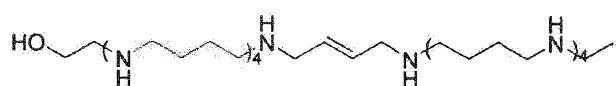
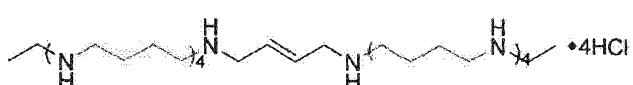
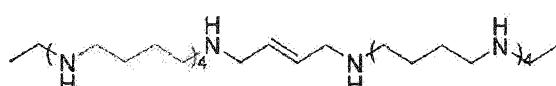
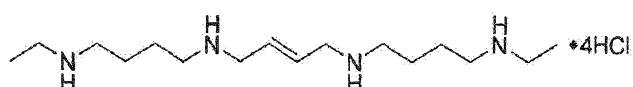
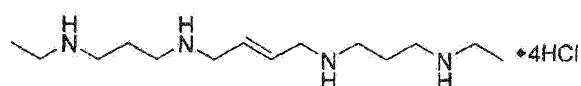
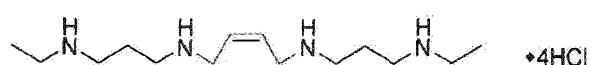
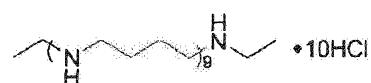
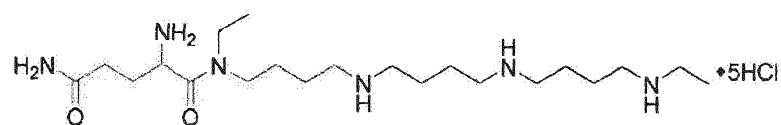
**[0110]** In one embodiment, the only conformational restriction of the polyamine analog is due to a carbon-carbon double bond (an ethenyl group, C=C) in the molecule.

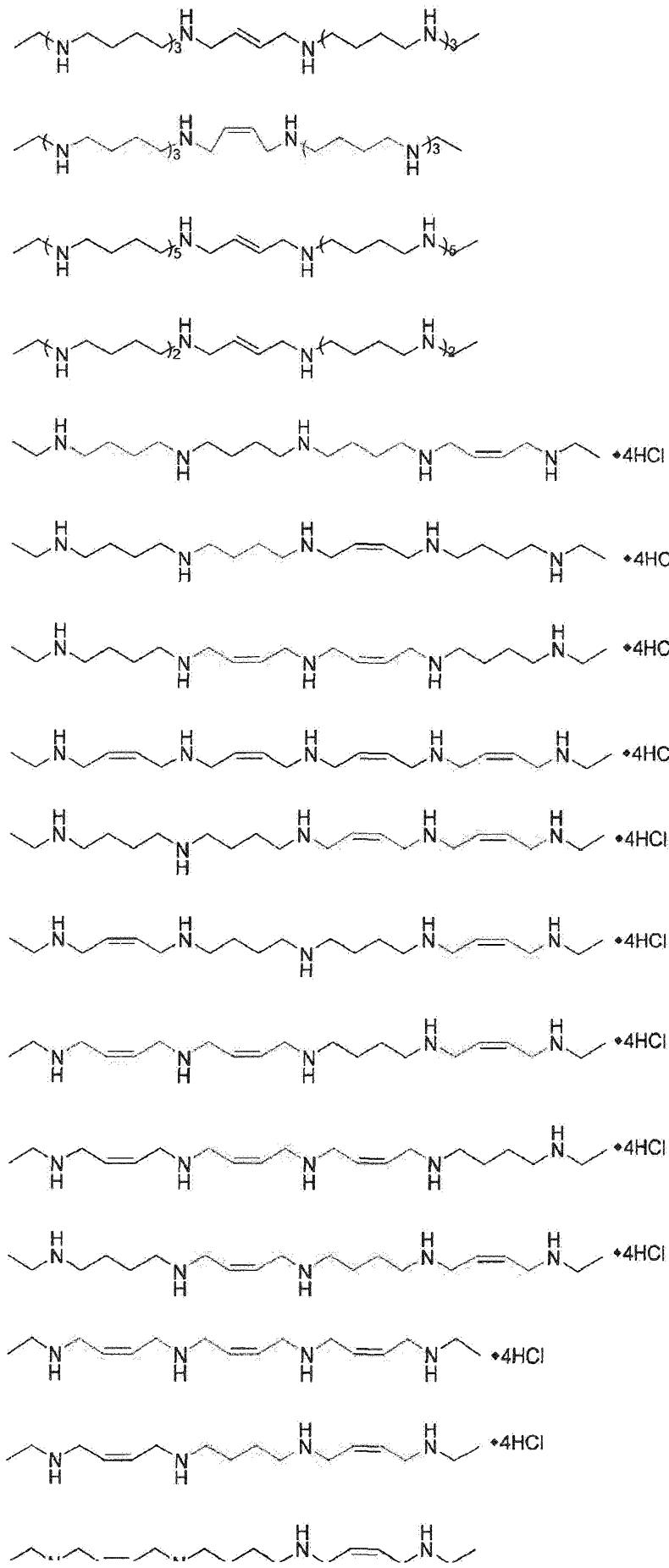
**[0111]** In another embodiment, the only conformational restriction of the polyamine analog is due to a cycloalkyl group, such as a cyclopropyl group, in the molecule.

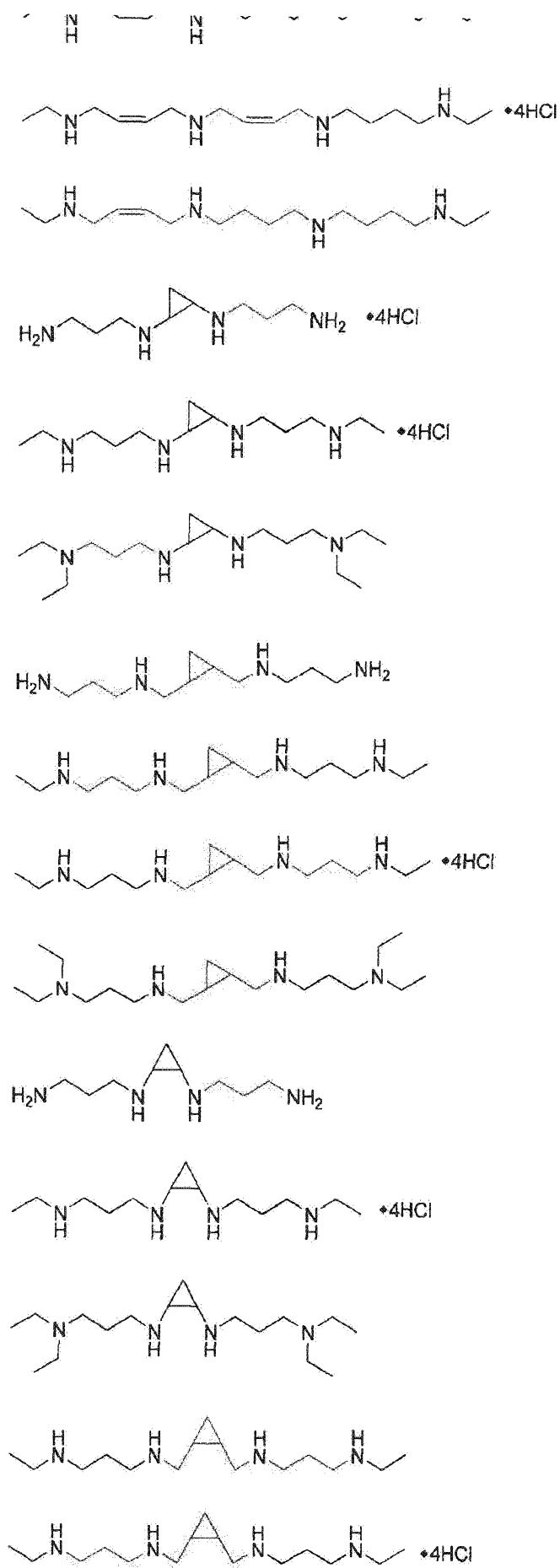
**[0112]** Compounds include, but are not limited to:

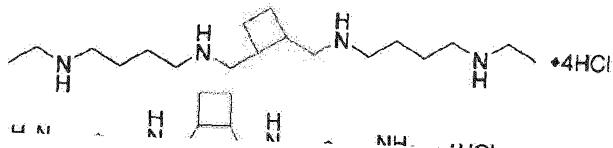
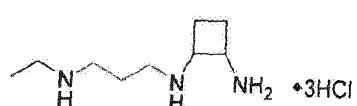
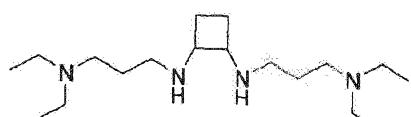
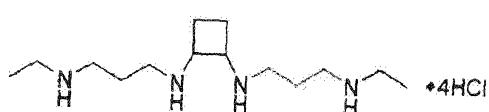
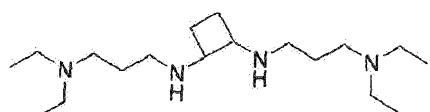
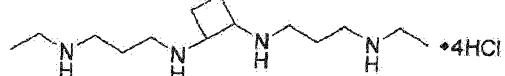
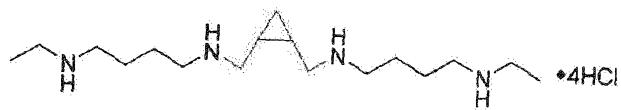


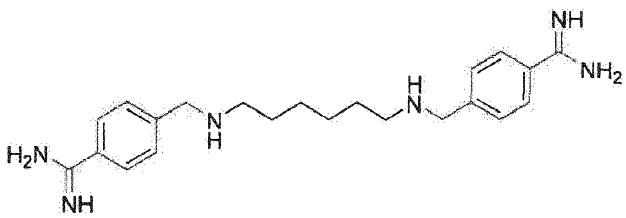
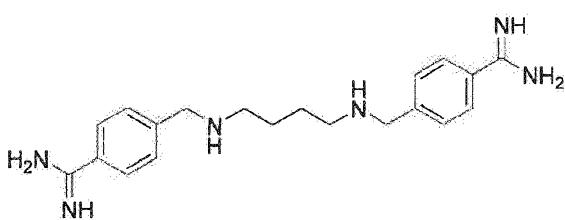
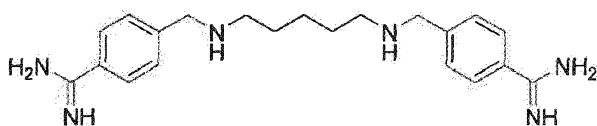
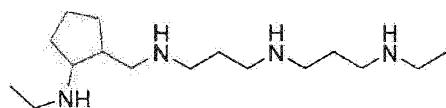
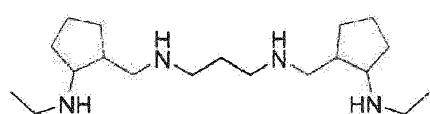
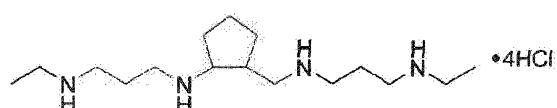
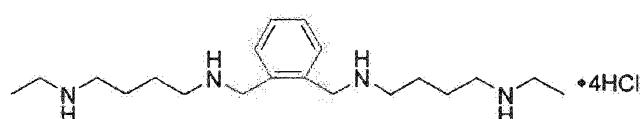
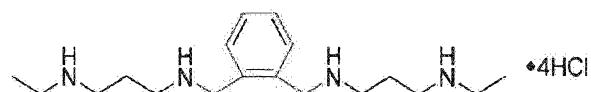
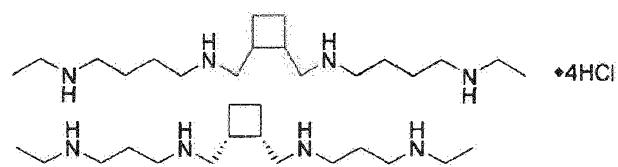


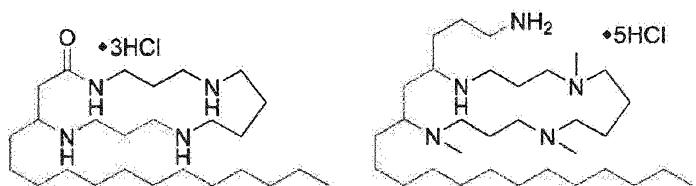
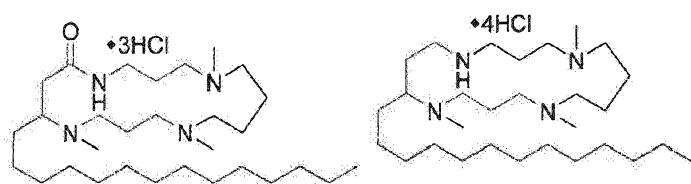
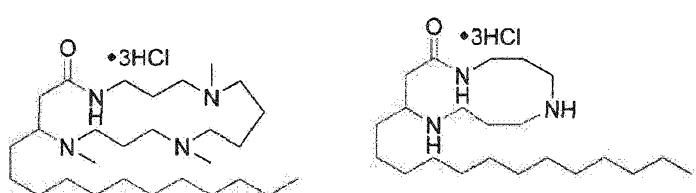
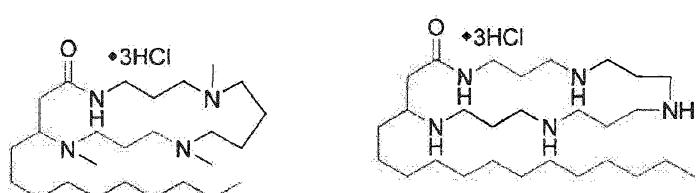
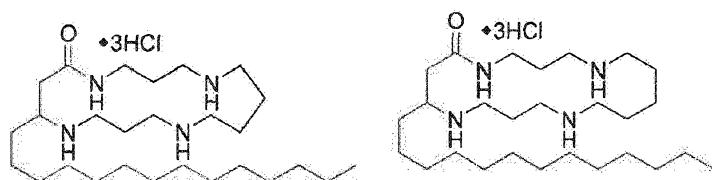
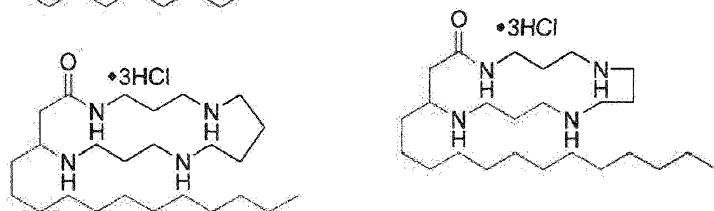
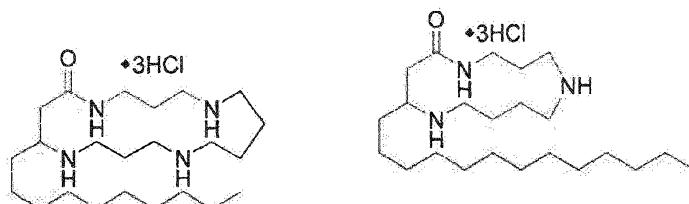
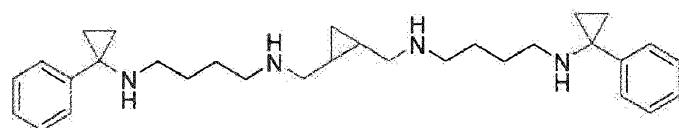




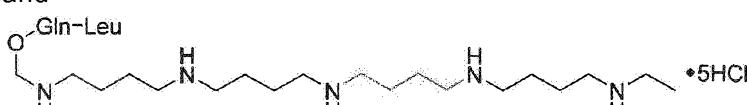








and



[0113] Additional disclosure may be found in WO2007/040535.

[0114] Additional analogs and derivatives include those encompassed by the following formula 10a:



wherein

R is selected from H or from the group of a straight or branched C1-50 saturated or unsaturated aliphatic, carboxyalkyl, carbalkoxyalkyl, or alkoxy; a C1-8 alicyclic; a single or multiring aryl substituted aliphatic; an aliphatic-substituted single or multiring aromatic; a single or multiring heterocyclic; a single or multiring heterocyclic 25 aliphatic; a C1-10 alkyl; an aryl sulfonyl; or cyano;

X may be -CO-, -SO<sub>2</sub>, or -CH<sub>2</sub>- , and

"polyamine" may be any naturally occurring, such as putrescine, spermine or spermidine, or synthetically produced polyamine.

[0115] Preferably, R is at least about C5, at least about C10, at least about C11, at least about C12, at least about C13, at least about C14, at least about C15, at least about C16, at least about C17, at least about C18, at least about C19, at least about C20, or at least about C22.

[0116] The linkage between X and the polyamine may be direct, wherein there are no atoms between X and the nitrogen of the amine group of the polyamine, or indirect, where there may be one or more atoms between X and the nitrogen of the amine group of the polyamine. The linkage between X and the polyamine may occur via any amino group within the polyamine, although a primary amino group is used in preferred embodiments described herein.

[0117] In preferred embodiments described herein where the linkage between X and the polyamine is indirect, the intervening one or more atoms are preferably those of an amino acid or a derivative thereof. In particularly preferred embodiments of this type, the intervening one or more atoms are those of lysine, aspartic acid, glutamic acid, ornithine, or 2,4-diaminobutyric acid. Preferred compounds of this type may be represented as in Formula 10b:



wherein

R is a straight or branched C10-50 saturated or unsaturated aliphatic, carboxyalkyl,

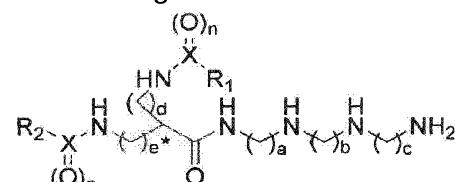
carbalkoxyalkyl, or alkoxy; a C1-8 alicyclic; a single or multiring aryl substituted or unsubstituted aliphatic; an aliphatic-substituted or unsubstituted single or multiring aromatic; a single or multiring heterocyclic; a single or multiring heterocyclic aliphatic; an aryl sulfonyl;

X is -CO-, -SO<sub>2</sub>-, or -CH<sub>2</sub>-; and

L is a covalent bond or a naturally occurring amino acid, ornithine, 2,4-diaminobutyric acid, or derivatives thereof.

**[0118]** The analogs and derivatives described herein may be optionally further substituted at one or more other positions of the polyamine. These include, but are not limited to, internal nitrogen and/or internal carbon atoms. In one aspect described herein, preferred substituents are structures that increase polyamine transport inhibition, binding affinity or otherwise enhance the irreversibility of binding of the compound to a polyamine binding molecule, such as the polyamine transporter, an enzyme or DNA. Such additional substituents include the aziridine group and various other aliphatic, aromatic, mixed aliphatic-aromatic, or heterocyclic multi-ring structures. Reactive moieties which, like aziridine, bind covalently to a polyamine transporter or another polyamine binding molecule, are also within the scope of this disclosure. Examples of reactive groups that react with nucleophiles to form covalent bonds include chloro-, bromo- and iodo-acetamides, sulfonylfluorides, esters, nitrogen mustards, etc. Such reactive moieties are used for affinity labeling in a diagnostic or research context, and may contribute to pharmacological activity in inhibiting polyamine transport or polyamine synthesis. The reactive group can be a reactive photoaffinity group such as an azido or benzophenone group. Chemical agents for photoaffinity labeling are well-known in the art (Flemming, S.A., *Tetrahedron* 1995, 51, 12479-12520).

**[0119]** A preferred aspect described herein relates to a polyamine analog or derivative that is a highly specific polyamine transport inhibitor with pharmaceutical utility as an anticancer chemotherapeutic. One class of a polyamine analog or derivative described herein that binds to a polyamine-binding site of a molecule and/or inhibits polyamine transport, is described by the following formula 10c:



wherein

a, b, and c independently range from 1 to 10;

d and e independently range from 0 to 30;

each X is independently either a carbon (C) or sulfur (S) atom, and

R<sub>1</sub> and R<sub>2</sub> are as described below, or each of R<sub>1</sub>X(O)<sub>n</sub>- and of R<sub>2</sub>X(O)<sub>n</sub>- are independently

replaced by H; and

\* denotes a chiral carbon position;

and with the provisos that

if X is C, then n is 1;

if X is S, then n is 2; and

if X is C, then the X(O) group may be CH<sub>2</sub> such that n is 0.

**[0120]** In the above formula, R<sub>1</sub> and R<sub>2</sub> are independently selected from H or from the group of a straight or branched C1-50 saturated or unsaturated aliphatic, carboxyalkyl, carbalkoxyalkyl, or alkoxy; a C1-8 alicyclic; a single or multiring aryl substituted aliphatic; an aliphatic-substituted single or multiring aromatic; a single or multiring aromatic or saturated heterocyclic; a single or multiring heterocyclic aliphatic; a C1-10 alkyl; an aryl sulfonyl; or cyano.

**[0121]** Examples of heterocyclic rings as used herein include, but are not limited to, pyrrole, furan, thiophene, imidazole, oxazole, thiazole, pyrazole, 3-pyrroline, pyrrolidine, pyridine, pyrimidine, purine, quinoline, isoquinoline, and carbazole.

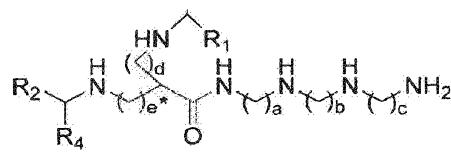
**[0122]** All of the above described aliphatic, carboxyalkyl, carbalkoxyalkyl, alkoxy, 30 alicyclic, aryl, aromatic, and heterocyclic moieties may, of course, also be optionally substituted with 1-3 substituents independently selected from halo (fluoro, chloro, bromo or iodo), lower alkyl (1-6C) and lower alkoxy (1-6C).

**[0123]** As used herein, carboxyalkyl refers to the substituent -R'-COOH wherein R' is alkylene; and carbalkoxyalkyl refers to -R'-COOR wherein R' and R are alkylene and alkyl respectively. In preferred embodiments, alkyl refers to a saturated straight- or branched-chain hydrocarbyl radical of 1-6 carbon atoms such as methyl, ethyl, n-propyl, isopropyl, n-butyl, t-butyl, n-pentyl, 2-methylpentyl, n-hexyl, and so forth. Alkylene is the same as alkyl except that the group is divalent. Aryl or alkyl sulfonyl moieties have the formula SO<sub>2</sub>R and alkoxy moieties have the formula -O-R, wherein R is alkyl, as defined above, or is aryl wherein aryl is phenyl, optionally substituted with 1-3 substituents independently selected from halo (fluoro, chloro, bromo or iodo), lower alkyl (1-6C) and lower alkoxy (1-6C).

**[0124]** A preferred group of compounds encompassed by the above is where d is 4 and e is 0.

**[0125]** An additional class of a polyamine analog or derivative described herein that binds to a polyamine-binding site of a molecule and/or inhibits polyamine transport, is described by the following formula 10d:

R<sub>3</sub>  
i



wherein

a, b, and c independently range from 1 to 10;

d and e independently range from 0 to 30;

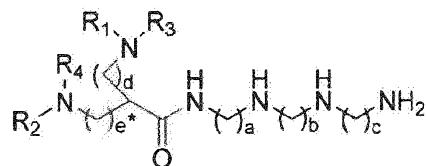
R<sub>1</sub> and R<sub>2</sub> are defined as above for formula 8c and

R<sub>3</sub> and R<sub>4</sub> are independently selected from organic substituents including-CH<sub>3</sub> and as defined above for R<sub>1</sub> and R<sub>2</sub> in formula 8c above. This grouping of analogs is produced by reductive amination of the free amino precursor with a ketone.

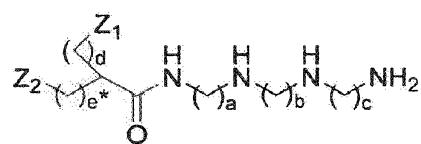
**[0126]** In one preferred embodiment described herein, R<sub>1</sub> and R<sub>2</sub> are identical and as described for formula 8c. Positions R<sub>3</sub> and R<sub>4</sub> may also be identical, and all of R<sub>1</sub> through R<sub>4</sub> may also be identical. Additionally, each of positions R<sub>1</sub>, R<sub>2</sub>, R<sub>3</sub>, and R<sub>4</sub> in formula 8d may also be independently H.

**[0127]** In an additional aspect described herein the proximal and/or the distal amino group relative to the polyamine (such as spermine) can be di-alkylated to form tertiary amines. These materials can be synthesized by reductive amination with a large excess of the carbonyl component. Additionally, these materials may be produced by a conjugate addition of the amine precursor to an α,β-unsaturated carbonyl or α,β-unsaturated nitrile.

**[0128]** Each of R<sub>1</sub>, R<sub>2</sub>, R<sub>3</sub>, and R<sub>4</sub> can be independently varied and are as defined as above for formula III. Each of R<sub>1</sub>, R<sub>2</sub>, R<sub>3</sub>, and R<sub>4</sub> may also be independently H. The values of a, b, c, d and e are as described above for formula 8d. This aspect is depicted in the following formula 10e:



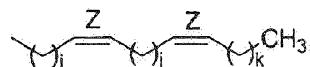
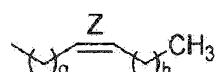
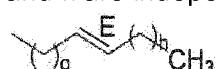
**[0129]** In a further aspect described herein, compounds which lack the proximal or distal amino group on the acyl portion of the molecule are also provided. These are represented by formula 10f:



wherein

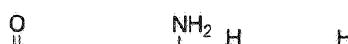
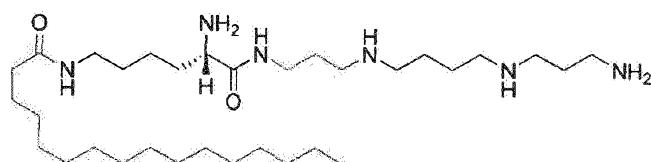
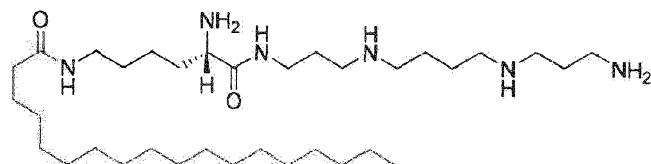
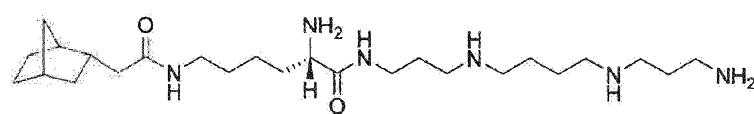
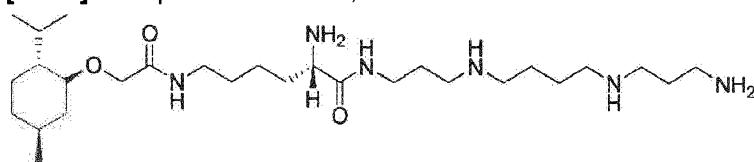
$Z_1$  is  $NR_1R_3$  and  $Z_2$  is selected from -  $R_1$ , - $CHR_1R_2$  or - $CR_1R_2R_3$  (wherein  $R_1$ ,  $R_2$ , and  $R_3$  are as defined above for formula 8c); or  $Z_2$  is  $NR_2R_4$  and  $Z_1$  is selected from - $R_1$ , - $CHR_1R_2$  or - $CR_1R_2R_3$  (wherein  $R_1$ ,  $R_2$ , and  $R_3$  are as defined above for formula 8d). Values for  $a$ ,  $b$ , and  $c$  independently range from 1 to 10;  $d$  and  $e$  independently range from 0 to 30. Compounds encompassed by formula V may be prepared by first coupling amino acid derivatives (modified to contain the non-amine containing  $Z$  group) to a polyamine followed by appropriate derivatization of the amine containing  $Z$  group. Chemistries for such reactions are known in the art and disclosed herein.

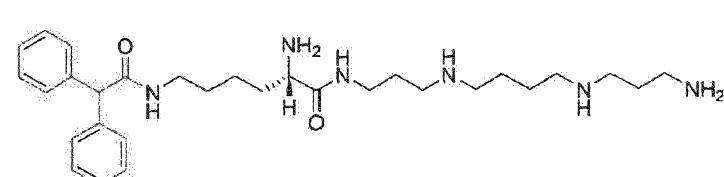
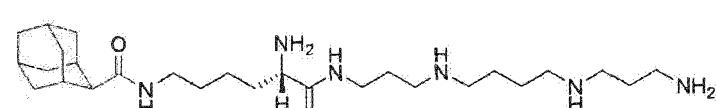
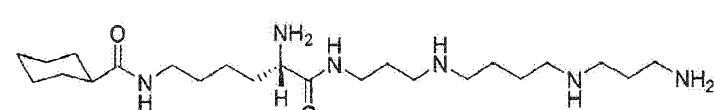
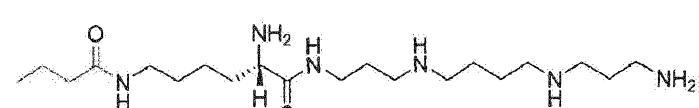
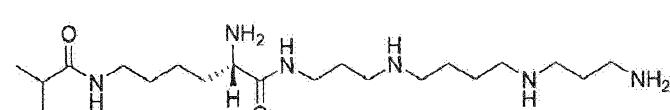
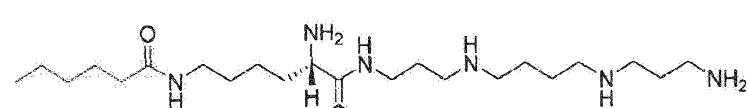
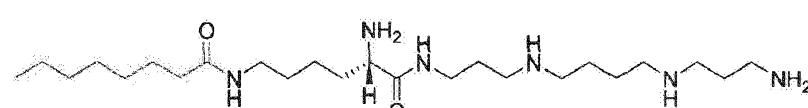
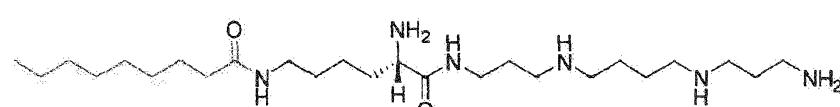
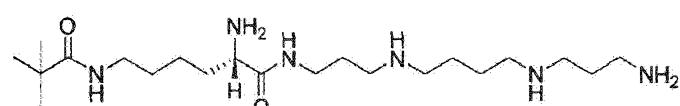
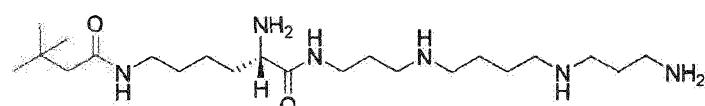
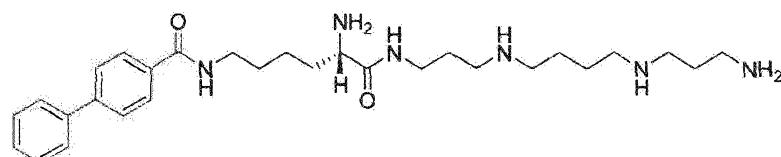
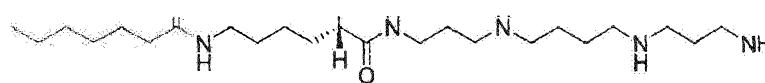
**[0130]** In preferred embodiments described herein, positions  $R_1$ ,  $R_2$ ,  $R_3$ , and  $R_4$ , of all the formulas set forth above are independently selected from the following, where each of  $g$ ,  $h$ ,  $i$ ,  $j$ , and  $k$  are independently selected from 0 to 15:



wherein  $E$  refers to "entgegen" and  $Z$  refers to "zusammen".

**[0131]** Compounds include, but are not limited to:

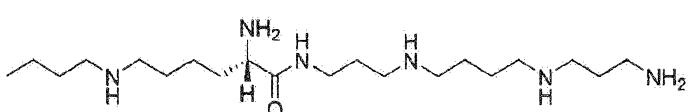
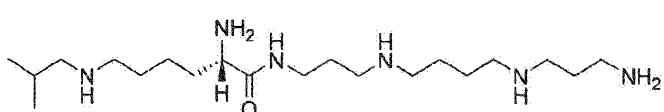
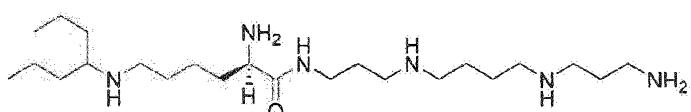
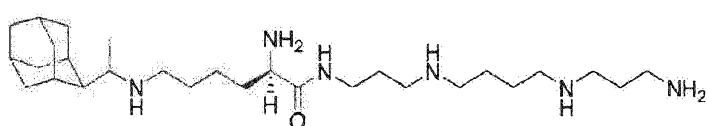
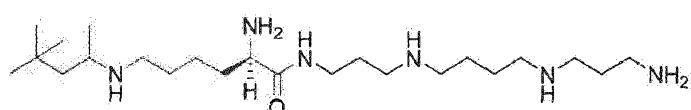
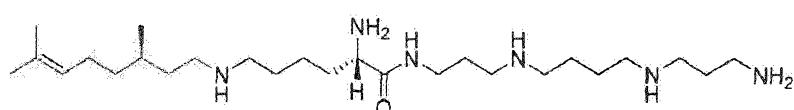
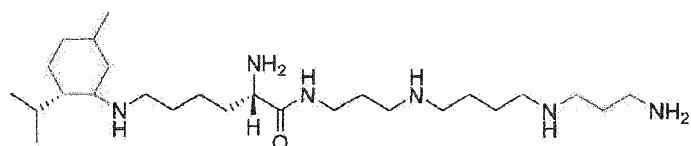
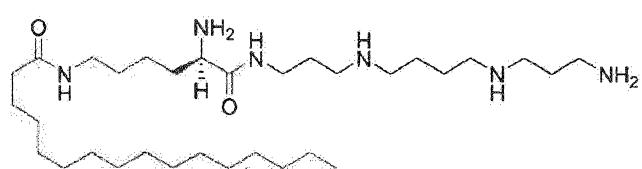
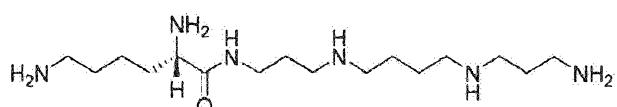
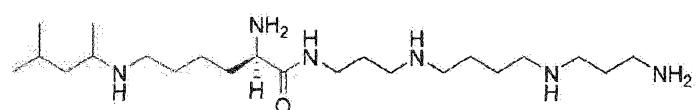
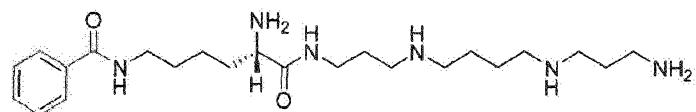
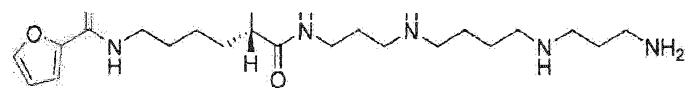




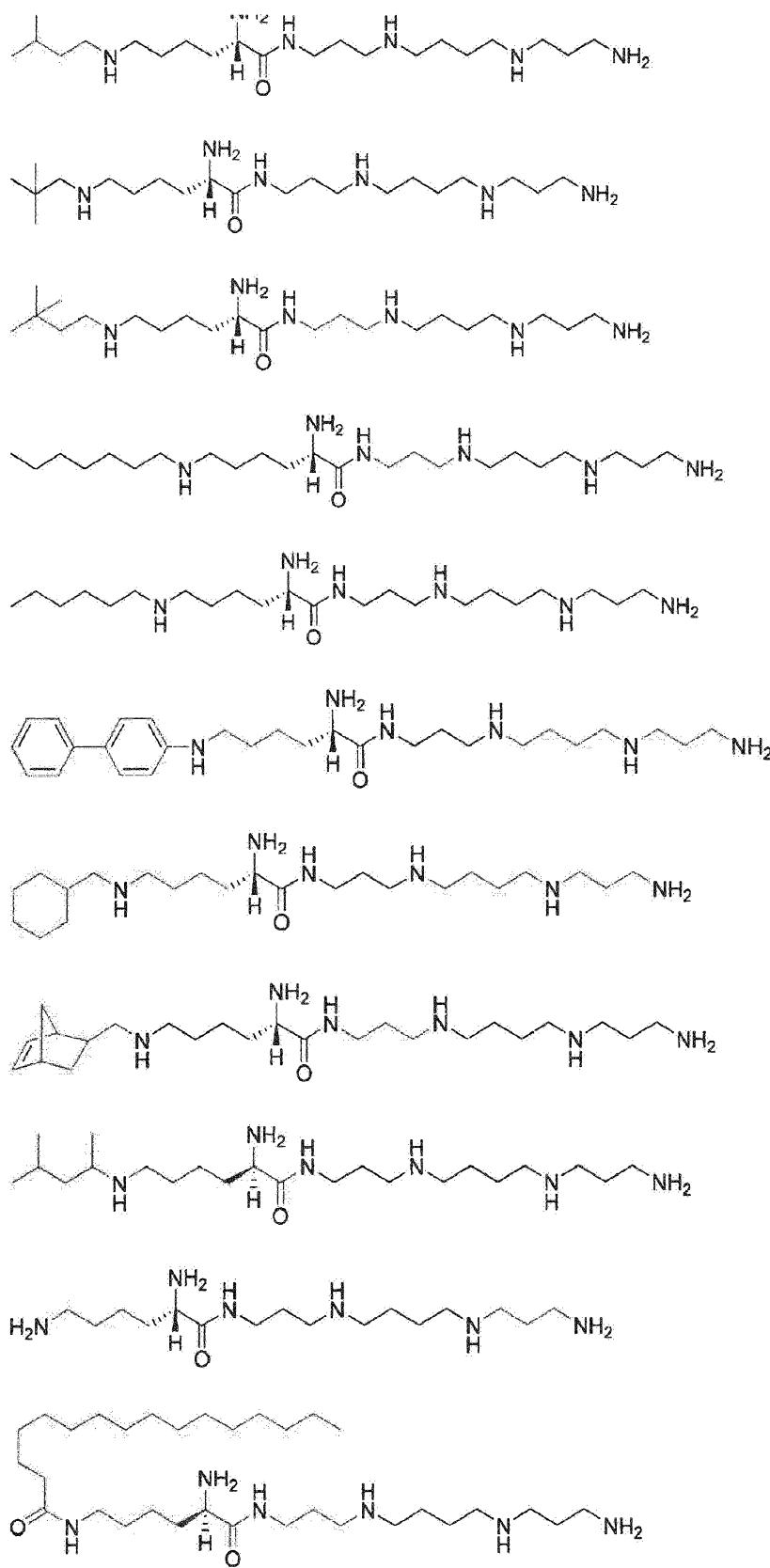
O

NH<sub>2</sub>

H



NH<sub>2</sub>



[0132] Additional disclosure may be found in WO2002/053519.

[0133] Additional analogs and derivatives include synthetic derivatives of original polyamines,

wherein a carbon atom of said original polyamine comprises an amide group, said synthetic derivative inhibiting the cellular uptake of a natural polyamine by specifically binding a cellular transporter for said natural polyamine.

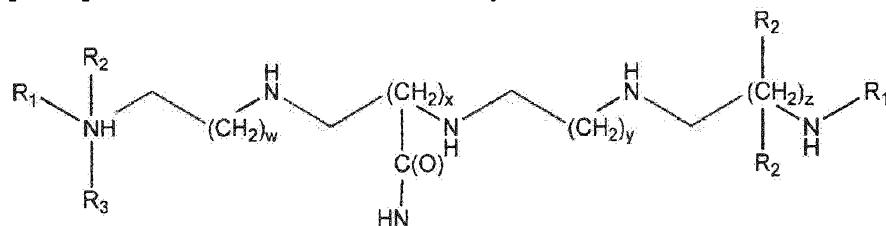
**[0134]** In certain embodiments, the carbon to which said amido group is located is between two internal nitrogen atoms of said original polyamine.

**[0135]** In certain embodiments, the synthetic derivative comprises a dimer of said original polyamine, the monomers of said dimer being linked together by a spacer side chain anchored to the amido group of each monomer.

**[0136]** In certain embodiments, the original polyamine is selected from the group consisting of putrescine, spermidine and spermine.

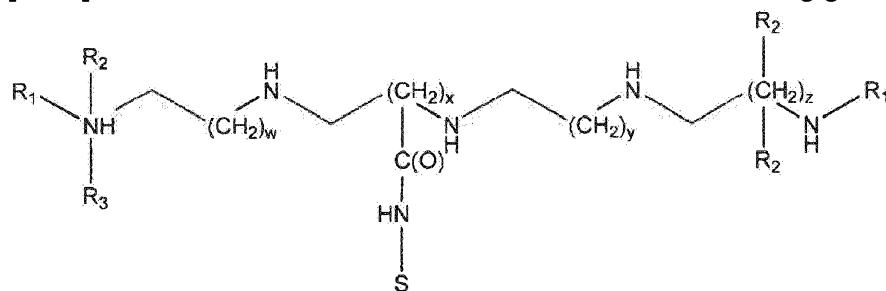
**[0137]** In certain embodiments, the original polyamine is spermine.

**[0138]** In certain embodiments, said synthetic derivative has the following general formula 11a:



in which R<sub>1</sub> and R<sup>1</sup><sub>1</sub> independently represent a hydrogen atom or an alkyl group having 1 to 2 carbon atoms, R<sub>2</sub>, R<sup>1</sup><sub>2</sub>, or R<sub>3</sub> and R<sup>1</sup><sub>3</sub> independently represent a hydrogen atom or a methyl group, w and z independently represent an integer of 2 or 3, x represents an integer from 0 to n, n represents an integer from 3 to 6, the sum of x and y equals n, and S represents a hydrogen atom or a molecule which cannot be captured by said natural polyamine transporter.

**[0139]** In certain embodiments, said monomer has the following general formula 11b:



in which R<sub>1</sub> and R<sup>1</sup><sub>1</sub> independently represent a hydrogen atom or an alkyl group having 1 to 2 carbon atoms, R<sub>2</sub>, R<sup>1</sup><sub>2</sub>, or R<sub>3</sub> and R<sup>1</sup><sub>3</sub> independently represent a hydrogen atom or a methyl group, w and z independently represent an integer of 2 or 3, x represents an integer from 0 to n, n represents an integer from 3 to 6, the sum of x and y equals n, and wherein the spacer side chain comprises a linear hydrocarbon-containing backbone of 3 to 8 atoms.

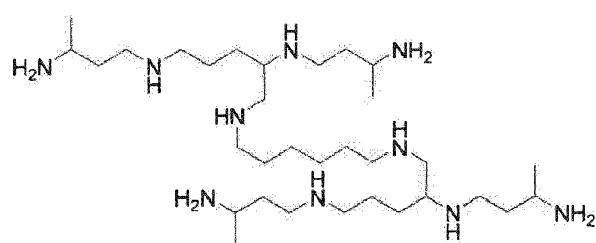
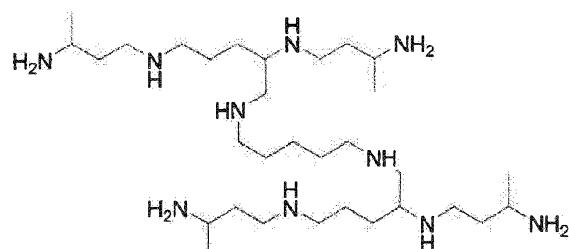
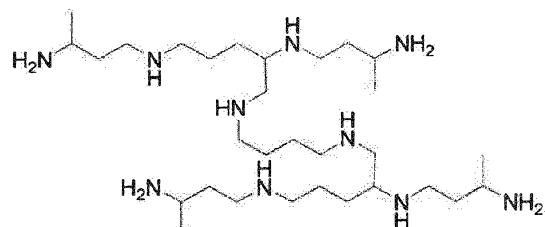
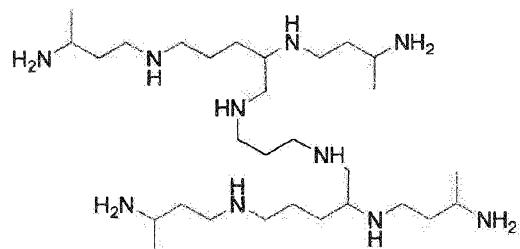
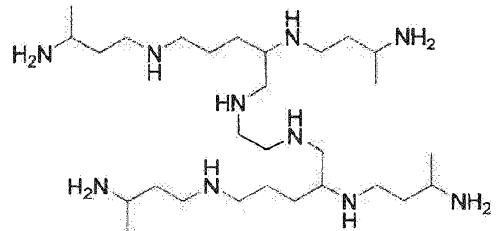
[0140] In certain embodiments, said backbone comprises sulfur, oxygen or nitrogen.

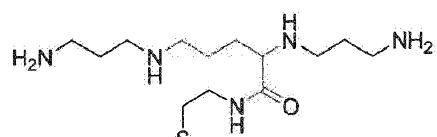
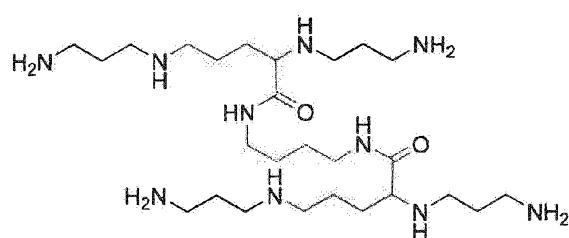
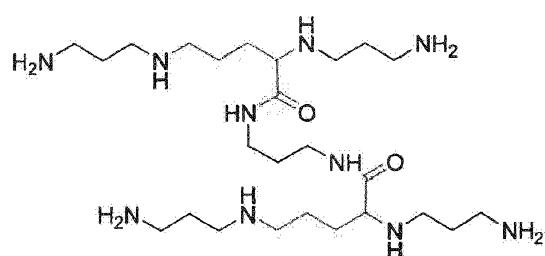
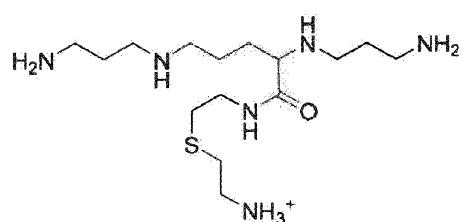
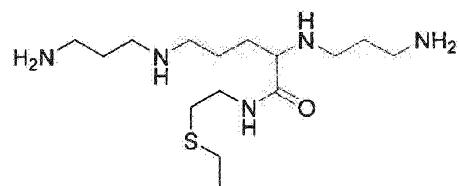
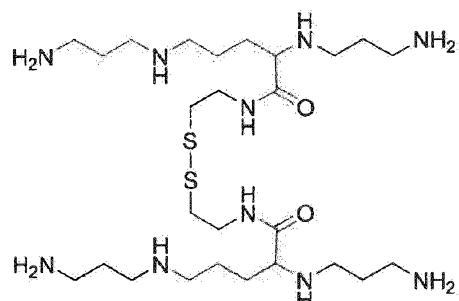
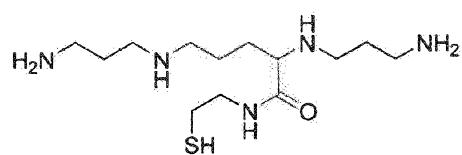
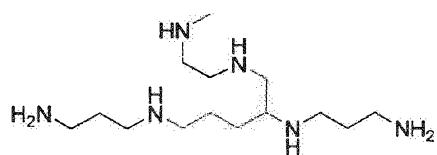
[0141] In certain embodiments, w=2, z=2 x=o and y=3.

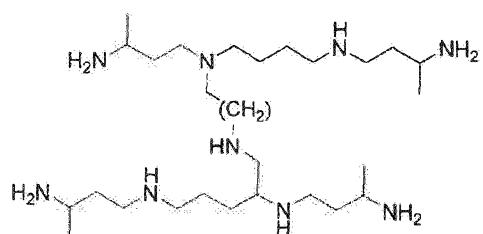
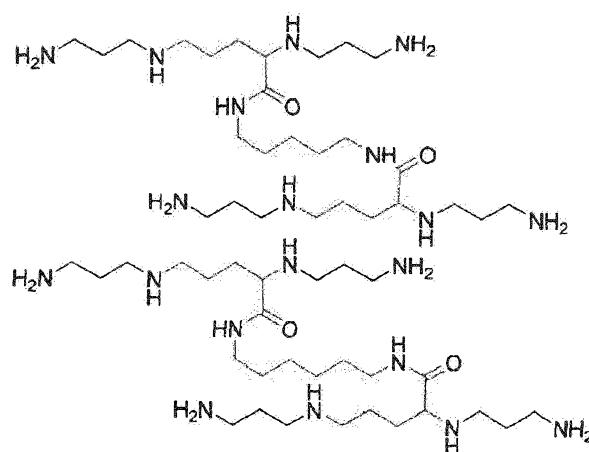
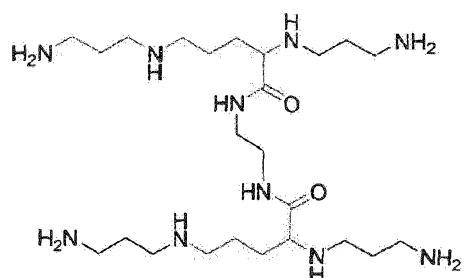
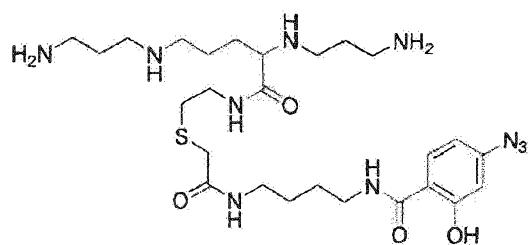
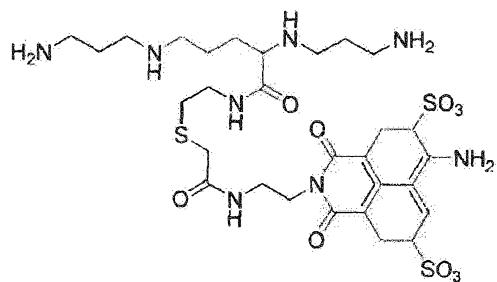
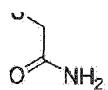
[0142] In certain embodiments, w=2, z=2, x=o and y=3.

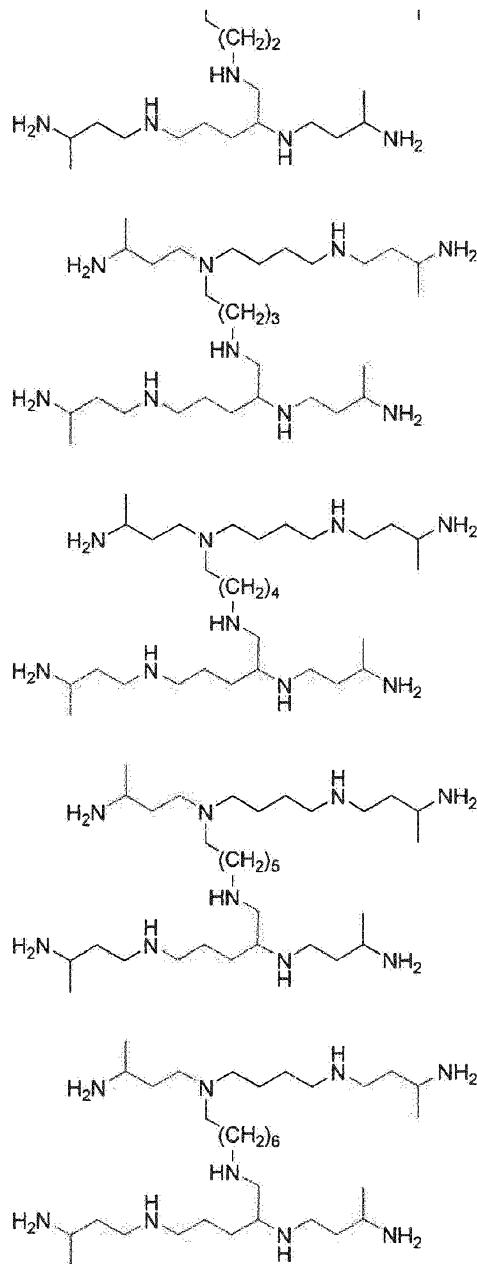
[0143] In certain embodiments, w=2, z=2, x=o and y=4.

[0144] Compounds include, but are not limited to:



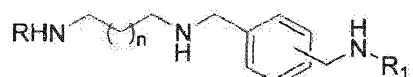






**[0145]** Additional disclosure may be found in WO98/17632.

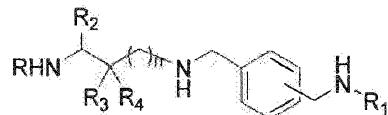
**[0146]** Additional analogs and derivatives include those encompassed by the following formula 12a:



wherein, n can be 0 to 8 and the aminomethyl functionality can be ortho, meta or para substituted, R is hydrogen, -CH<sub>3</sub>, -CH<sub>2</sub>CH<sub>3</sub>, 2-aminoethyl, 3-aminopropyl, 4-aminobutyl, 5-aminopentyl, 6-aminohexyl, 7-aminohethyl, 8-aminooctyl, N-methyl-2-aminooethyl, N-methyl-3-aminopropyl, N-methyl-4-aminobutyl, N-methyl-5-aminopentyl, N-methyl-6-aminohexyl, N-methyl-7-aminohethyl, N-methyl-8-aminooctyl, N-ethyl-2-aminooethyl, N-ethyl-3-minopropyl, N-ethyl-4-aminobutyl, N-ethyl-5-aminopentyl, N-ethyl-6-aminohexyl, N-ethyl-7-aminohethyl or N-

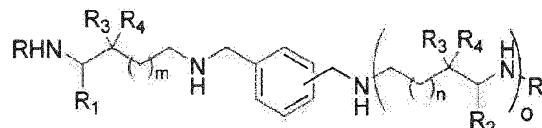
ethyl-8-aminoctyl and R, is a moiety selected from the group' consisting of a hydrogen or a straight or branched C1-20 saturated or unsaturated aliphatic; aliphatic amine but not propylamine when R =H, n=1 and the aminomethyl functionality is para substituted; an alicyclic; single or multi-ring aromatic; single or multi-ring aryl substituted aliphatic; aliphatic-substituted single or multi-ring aromatic; a single or multi-ring heterocyclic, a single or multi-ring heterocyclic-substituted aliphatic; an aliphatic-substituted aromatic; and halogenated forms thereof.

**[0147]** In certain embodiments, the analogs and derivatives that can be used according to this disclosure can be further modified as described in formula 12b:



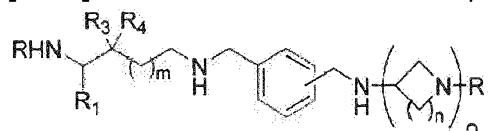
wherein n can be 0 to 8, R and R<sub>1</sub> are described as above, R<sub>2</sub> can be independently selected from hydrogen, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, and R<sub>3</sub> and R<sub>4</sub> may be the same or different and are independently selected from hydrogen, or fluorine.

**[0148]** In certain embodiments, compounds that can be used according to this disclosure are described in formula 12c:



wherein, m and n can be 0 to 7 independently, but m cannot equal n when R<sub>1</sub> equals R<sub>2</sub> and R<sub>3</sub> equals R<sub>4</sub>, o can be 2 to 4, R can be independently selected from H, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, R<sub>1</sub> and R<sub>2</sub> can be independently selected from hydrogen, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, and R<sub>3</sub> and R<sub>4</sub> may be the same or different and are independently selected from hydrogen or fluorine.

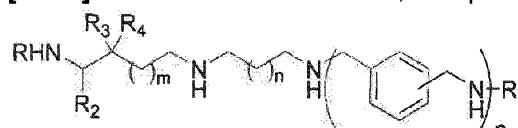
**[0149]** In certain embodiments, compounds have formula 12d:



wherein, R is hydrogen, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, m and n can be 0 to 7 independently and o

**[0150]** can be 2 to 4, R<sub>2</sub> can be independently selected from hydrogen, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, and R<sub>3</sub> and R<sub>4</sub> may be the same or different and are independently selected from hydrogen or fluorine.

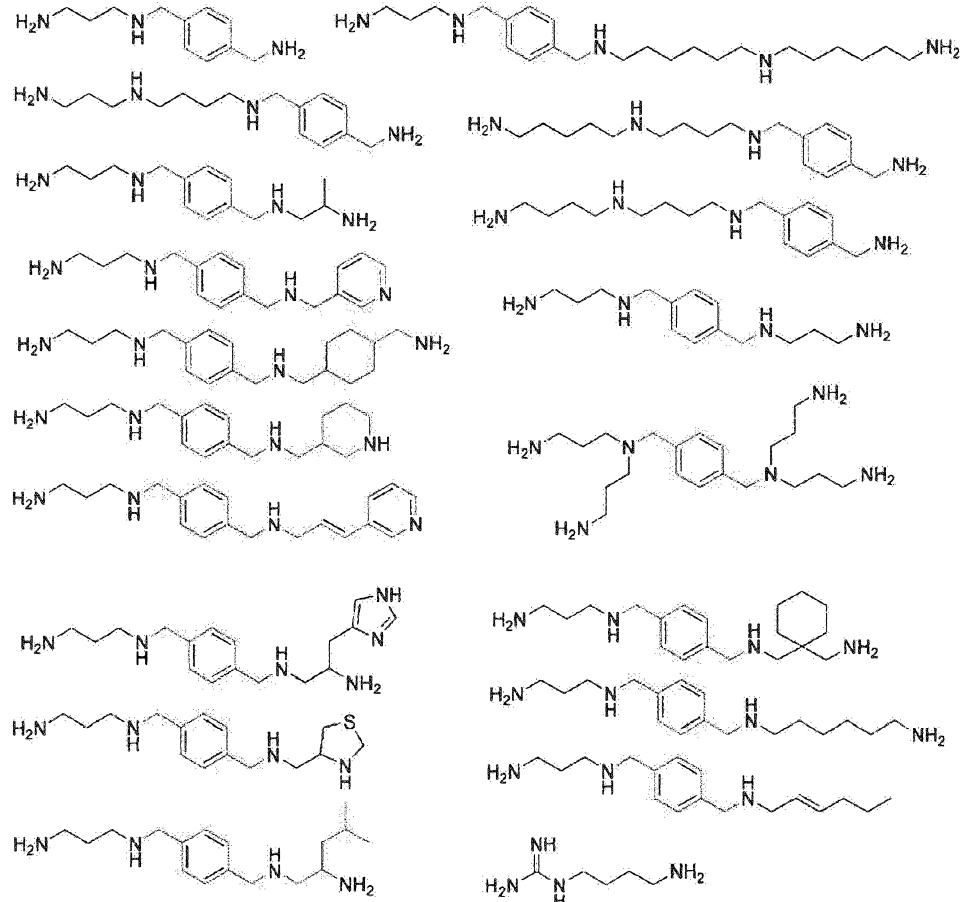
**[0151]** In certain embodiments, compounds described herein are represented by formula 12e:



wherein, R is hydrogen, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, m can be 0 to 7, n can be 0 to 8 and o can be 2 to 4, R<sub>2</sub> can be independently selected from hydrogen, -CH<sub>3</sub>,-CH<sub>2</sub>CH<sub>3</sub>, and R<sub>3</sub> and R<sub>4</sub> may be the

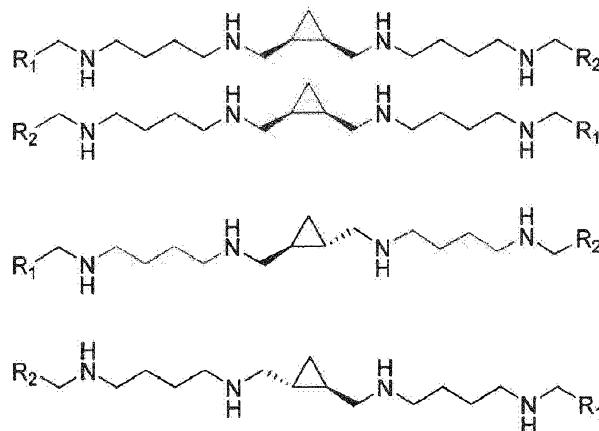
same or different and are independently selected from hydrogen or fluorine.

**[0152]** Compounds include, but are not limited to:



**[0153]** Additional disclosure may be found in WO05/105729.

**[0154]** Additional analogs and derivatives include compounds of the formulas 13a-d:



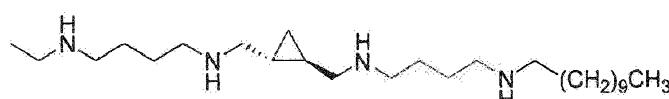
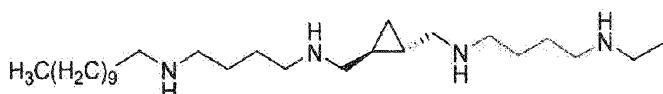
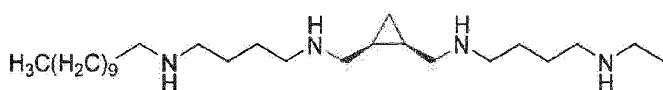
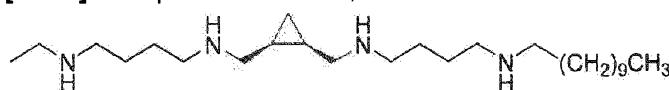
wherein R<sub>1</sub> and R<sub>2</sub> are independently selected from the group consisting of -C<sub>1</sub>-C<sub>10</sub> alkyl, -C<sub>3</sub>-C<sub>10</sub> cycloalkyl, -C<sub>1</sub>-C<sub>10</sub> alkylene-cycloalkyl, -C<sub>6</sub>-C<sub>10</sub> aryl, and -C<sub>1</sub>-C<sub>10</sub> alkylene-aryl, wherein when both R<sub>1</sub> and R<sub>2</sub> are alkyl, at least one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>2</sub>-C<sub>10</sub> alkyl and wherein both R<sub>1</sub>

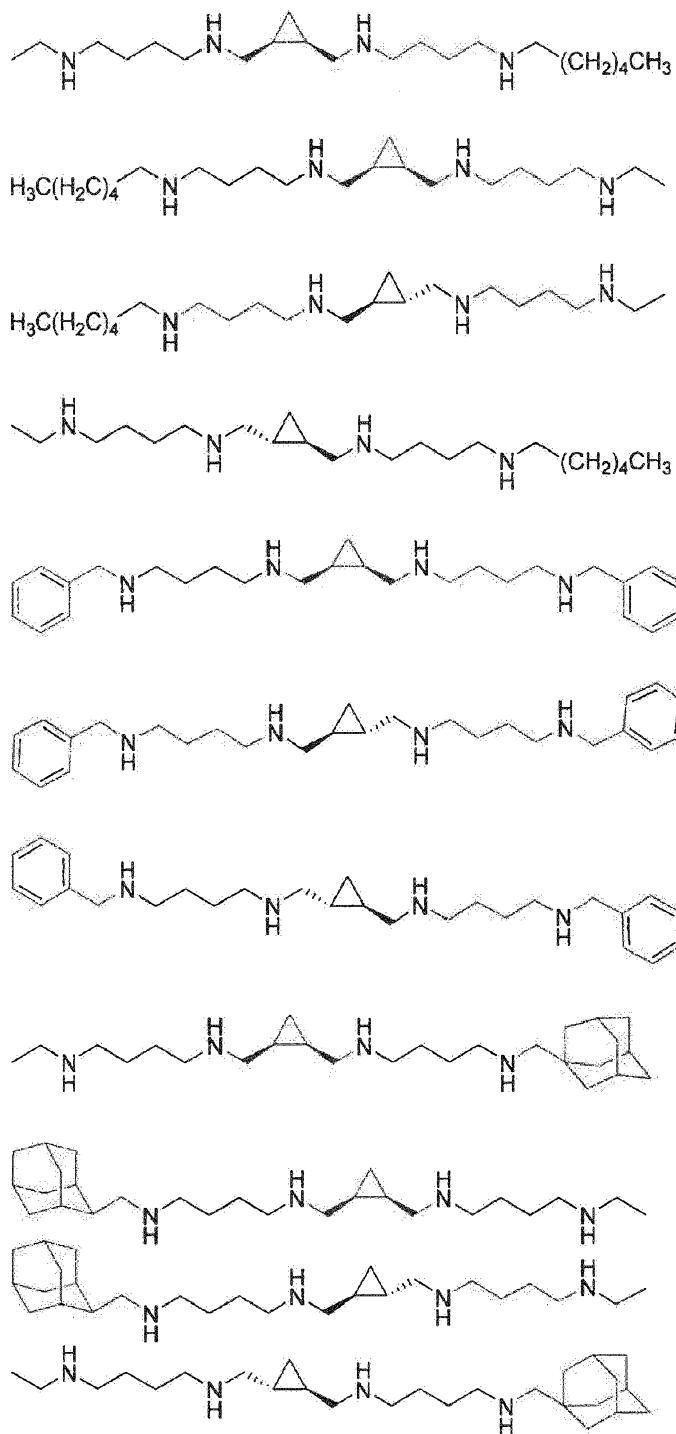
and R<sub>2</sub> are not *tert*-butyl; and all salts, hydrates, solvates, and stereoisomers thereof; and all mixtures of stereo isomers thereof, including racemic mixtures. In one embodiment, the substituents on the cyclopropyl ring are *trans* to each other. In another embodiment, the substituents on the cyclopropyl ring are *cis* to each other.

**[0155]** In one embodiment, when both R<sub>1</sub> and R<sub>2</sub> are alkyl, at least one of R<sub>1</sub> and R<sub>2</sub> is straight-chain alkyl. In another embodiment, when both R<sub>1</sub> and R<sub>2</sub> are alkyl, both R<sub>1</sub> and R<sub>2</sub> are straight-chain alkyl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>2</sub>-C<sub>10</sub> alkyl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>4</sub>-C<sub>10</sub> alkyl. In one embodiment, both R<sub>1</sub> and R<sub>2</sub> are -C<sub>4</sub>-C<sub>10</sub> alkyl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>6</sub>-C<sub>10</sub> alkyl. In one embodiment, both R<sub>1</sub> and R<sub>2</sub> are -C<sub>6</sub>-C<sub>10</sub> alkyl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is selected from the group consisting of -C<sub>2</sub>-C<sub>4</sub> straight-chain alkyl and -C<sub>4</sub>-C<sub>10</sub> alkyl. In another embodiment, R<sub>1</sub> and R<sub>2</sub> are independently selected from the group consisting of -CH<sub>3</sub>, -(CH<sub>2</sub>)<sub>3</sub>CH<sub>3</sub>, and -(CH<sub>2</sub>)<sub>5</sub>CH<sub>3</sub>, provided that both R<sub>1</sub> and R<sub>2</sub> are not -CH<sub>3</sub>.

**[0156]** In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>3</sub>-C<sub>10</sub> cycloalkyl, -C<sub>1</sub>-C<sub>10</sub> alkylene-cycloalkyl, -C<sub>6</sub>-C<sub>10</sub> aryl, or -C<sub>1</sub>-C<sub>10</sub> alkylene-aryl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>3</sub>-C<sub>10</sub> cycloalkyl or -C<sub>1</sub>-C<sub>10</sub> alkylene-cycloalkyl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>3</sub>-C<sub>10</sub> cycloalkyl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>6</sub>-C<sub>10</sub> aryl or -C<sub>1</sub>-C<sub>10</sub> alkylene-aryl. In one embodiment, one of R<sub>1</sub> and R<sub>2</sub> is -C<sub>1</sub>-C<sub>10</sub> alkyl and the other is -C<sub>6</sub>-C<sub>10</sub> aryl. In another embodiment, both R<sub>1</sub> and R<sub>2</sub> are -C<sub>6</sub>-C<sub>10</sub> aryl. In another embodiment, both R<sub>1</sub> and R<sub>2</sub> are -C<sub>3</sub>-C<sub>10</sub> cycloalkyl. In one embodiment, the aryl group is benzene. In one embodiment, the cycloalkyl group is adamantyl. In one embodiment, the adamantyl group is 1-adamantyl. In another embodiment, the adamantyl group is 2-adamantyl. In another embodiment, R<sub>1</sub> and R<sub>2</sub> are independently selected from the group consisting of -CH<sub>3</sub>, phenyl, and adamantyl, provided that both R<sub>1</sub> and R<sub>2</sub> are not -CH<sub>3</sub>.

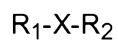
**[0157]** Compounds include, but are not limited to:





[0158] Additional disclosure may be found in WO2008/112251.

[0159] Additional analogs and derivatives include compounds of the formula 14a:



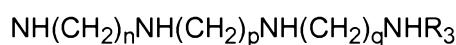
wherein

$R_1$  is H, or is a head group selected from the group consisting of a straight or branched C<sub>1-10</sub> aliphatic, alicyclic, single or multiring aromatic, single or multiring aryl substituted aliphatic, aliphatic-substituted single or multiring aromatic, a single or multiring heterocyclic, a single or multiring heterocyclic-substituted aliphatic and an aliphatic-substituted aromatic;

$R_2$  is a polyamine; and

X is CO, NHCO, NHCS, or SO<sub>2</sub>

**[0160]** In another embodiment of the above composition,  $R_2$  has the formula



wherein

n, p and q vary independently and n=p=q=1 to 12; and

$R_3$  is H; C<sub>1-10</sub> alkyl; C<sub>1-10</sub> alkenyl; C<sub>1-10</sub> alkynyl; alicyclic; aryl; aryl-substituted alkyl, alkenyl or alkynyl; alkyl-, alkenyl-, or alkynyl-substituted aryl; gauanidino; heterocyclic; heterocyclic-substituted alkyl, alkenyl or alkynyl; and alkyl-, alkenyl-, or alkynyl-substituted heterocyclic.

**[0161]** The above composition may further comprise, linked between X and  $R_2$ , a linker L and an additional group Y, such that said composition has the formula 14b:



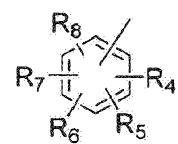
wherein

L is a C<sub>1-10</sub> alkyl; C<sub>1-10</sub> alkenyl; C<sub>1-10</sub> alkynyl, alicyclic, or heterocyclic;

X is CO, SO<sub>2</sub>, NHCO or NHCS; and

Y is CONH, SO<sub>2</sub>NH, NHCO, NHCONH, NHCSNH, NHSO<sub>2</sub>, SO<sub>2</sub>, O, or S.

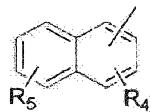
**[0162]** In the foregoing compositions  $R_1$  can have the formula:



wherein

$R_4$ ,  $R_5$ ,  $R_6$ ,  $R_7$ , and  $R_8$  are, independently, H, OH, halogen,  $NO_2$ ,  $NH_2NH(CH)_nCH_3$ ,  $N((CH)_nCH_3)_2$ , CN,  $(CH)_nCH_3$ ,  $O(CH)_nCH_3$ ,  $S(CH_2)_nCH_3$ ,  $NCO(CH_2)_nCH_3$ ,  $O(CF_2)_nCF_3$ , or  $CO-O(CH)_nCH_3$  where  $n=0$  to 10.

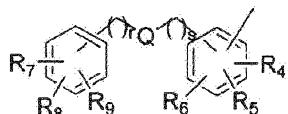
**[0163]** Alternatively,  $R_1$  has the formula:



wherein

$R_4$  and  $R_5$  are, independently, H, OH, halogen,  $NO_2$ ,  $NH_2NH(CH)_nCH_3$ ,  $N((CH)_nCH_3)_2$ , CN,  $(CH)_nCH_3$ ,  $O(CH)_nCH_3$ ,  $S(CH_2)_nCH_3$ ,  $NCO(CH_2)_nCH_3$ ,  $O(CF_2)_nCF_3$ , or  $CO-O(CH)_nCH_3$  where  $n=0$  to 10.

**[0164]** In yet another embodiment,  $R_1$  has the formula:



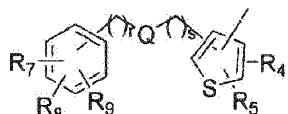
wherein

$r$  and  $s$  vary independently and  $r=s=0$  to 6;

$R_4$ ,  $R_5$ ,  $R_6$ ,  $R_7$ ,  $R_8$ , and  $R_9$  are, independently, H, OH, halogen,  $NO_2$ ,  $NH_2NH(CH)_nCH_3$ ,  $N((CH)_nCH_3)_2$ , CN,  $(CH)_nCH_3$ ,  $O(CH)_nCH_3$ ,  $S(CH_2)_nCH_3$ ,  $NCO(CH_2)_nCH_3$ ,  $O(CF_2)_nCF_3$ , or  $CO-O(CH)_nCH_3$  where  $n=0$  to 10;

$Q$  is CONH,  $SO_2NH$ , NHCO, NHCONH, NHCSNH,  $NHSO_2$ ,  $SO_2$ , O, or S.

**[0165]** Furthermore,  $R_1$  may have the formula:



wherein

$r$  and  $s$  vary independently and are 0 to 6;

$R_4$ ,  $R_5$ ,  $R_6$ , and  $R_7$  are, independently, H, OH,  $NO_2$ ,  $NH_2NH(CH)_nCH_3$ ,  $N((CH)_nCH_3)_2$ , CN,  $(CH)_nCH_3$ ,  $O(CH)_nCH_3$ ,  $S(CH_2)_nCH_3$ ,  $NCO(CH_2)_nCH_3$ ,  $O(CF_2)_nCF_3$ , or  $CO-O(CH)_nCH_3$  where  $n=0$  to 10; and

$Q$  is CONH,  $SO_2NH$ , NHCO, NHCONH, NHCSNH,  $NHSO_2$ ,  $SO_2$ , O, or S.

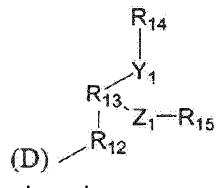
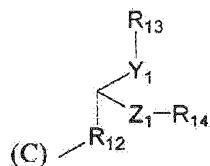
**[0166]** In the foregoing compositions, R<sub>1</sub> may be selected from the group consisting of naphthalene, phenanthrene, anthracene, pyrene, dibenzofuran, acridine, 2,1,3-benzothiodiazole, quinoline, isoquinoline, benzofuran, indole, carbazole, fluorene, 1,3-benzodiazine, phenazine, phenoxazine, phenothiazine, adamantane, camphor, pipiridine, alkylpiperazine, morpholine, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl, thiophene, furan, pyrrole, alkyl-1,2-diazole, alkylimidazole, alkyl-1H-1,2,3-triazol, alkyl-1H-1,2,3,4-tetrazole, thiazole, oxazole, 1,3,4-thiadiazole, pyridinyl, pyrimidine, 1,2-diazine, 1,4-diazine and 1,3,5-triazine, 4-dimethylaminoazobenzene, 3-phenyl-5-methylisooxazole, 3-(2-chlorophenyl)-5-methylisooxazole, 2-(4-chlorophenyl)-6-methyl-7-chloroquinoline, 6-chloroimidazo[2,1-β]thiazole, α-methylcinnamic acid, and 2-[1,2-dihydro-2H-1,4-benzodioxepinyl]thiazole.

**[0167]** R1 may also be a D- or L-amino acid.

**[0168]** Also provided is the above composition where R1 has a formula selected from the group consisting of

(A) R<sub>12</sub>-R<sub>13</sub>-Y<sub>1</sub>-R<sub>14</sub>

(B) R<sub>12</sub>-Y<sub>1</sub>-R<sub>13</sub>-Z<sub>1</sub>-R<sub>14</sub>



wherein

R<sub>12</sub> and R<sub>13</sub>, independently, are H, naphthalene, phenanthrene, anthracene, pyrene, dibenzofuran, acridine, 2,1,3-benzothiodiazole, quinoline, isoquinoline, benzofuran, indole, carbazole, fluorene, 1,3-benzodiazine, phenazine, phenoxazine, phenothiazine, adamantane, camphor, pipiridine, alkylpiperazine, morpholine, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl, thiophene, furan, pyrrole, alkyl-1,2-diazole, alkylimidazole, alkyl-1H-1,2,3-triazol, alkyl-1H-1,2,3,4-tetrazole, thiazole, oxazole, 1,3,4-thiadiazole, pyridinyl, pyrimidine, 1,2-diazine, 1,4-diazine and 1,3,5-triazine, 4-dimethylaminoazobenzene, 3-phenyl-5-methylisooxazole, 3-(2-chlorophenyl)-5-methylisooxazole, 2-(4-chlorophenyl)-6-methyl-7-chloroquinoline, 6-chloroimidazo[2,1-β]thiazole, α-methylcinnamic acid, or 2-[1,2-dihydro-2H-1,4-benzodioxepinyl]thiazole;

and further,

wherein a ring of  $R_{12}$ ,  $R_{13}$  or both in formulas (A), (B) and (D), is optionally substituted with one or more of OH, halogen,  $NO_2$ ,  $NH_2$ ,  $NH(CH)_nCH_3$ ,  $N((CH)_nCH_3)_2$ , CN,  $(CH)_nCH_3$ ,  $O(CH)_nCH_3$ ,  $S(CH_2)_nCH_3$ ,  $NCO(CH_2)_nCH_3$ ,  $O(CF_2)_nCF_3$ , or  $CO-O(CH)_nCH_3$  where  $n=0$  to 10

$R_{14}$  and  $R_{15}$  and, in formula (C),  $R_{13}$ , independently, are  $(CH_2)_n$ ,  $(CH_2)_nCH=CH$ ,  $(CH_2)_n(CH=CH)_mCO$ , or  $(CH_2)_nCO$  where  $n=0$  to 5 and  $m=1$  to 3;

$Y_1$ , and  $Z_1$ , independently, are  $CONH$ ,  $SO_2NH$ ,  $NHCO$ ,  $NHCONH$ ,  $NHCSNH$ ,  $NHSO_2$ ,  $SO_2^-$ ,  $NHSO_2$ ,  $SO_2$ , O, S, or  $COO$ ;

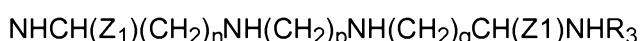
or

when  $R_1$  is of formula (A) or (B),  $Y_1$  represents a bond between a C or N atom of  $R_{12}$ , and a C or N atom of  $R_{13}$ , and  $Z_1$  represents a bond between a C or N atom of  $R_{13}$ , and a C or N atom of  $R_{14}$ ; or

when  $R_1$  is of formula (C) or  $Y_1$  represents a bond between the C and a C or N atom of  $R_{13}$  and  $Z_1$  represents a bond between the C and a C or N atom of  $R_{14}$ ; or

when  $R_1$  is of formula (D)  $Y_1$  represents a bond between a C or N atom of  $R_{12}$  and a C or N atom of  $R_{14}$  and  $Z_1$  represents a bond between a C or N atom of  $R_{13}$  and a C or N atom of  $R_{15}$ .

**[0169]** In the above compositions,  $R_2$  preferably has the formula



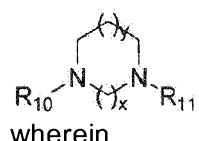
wherein

$n$ ,  $p$  and  $q$  vary independently and  $n=p=q=1$  to 12; and

$R_3$  is H;  $C_{1-10}$  alkyl;  $C_{1-10}$  alkenyl;  $C_{1-10}$  alkynyl; alicyclic; aryl; aryl-substituted alkyl, alkenyl or alkynyl; alkyl-, alkenyl-, or alkynyl-substituted aryl; guanidine or heterocyclic; and

$Z_1$  is  $CH_3$ ,  $CH_2CH_3$  or cyclopropyl.

**[0170]** In another embodiment,  $R_2$  has the formula:



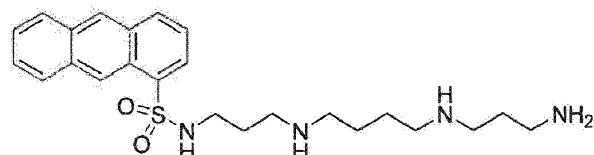
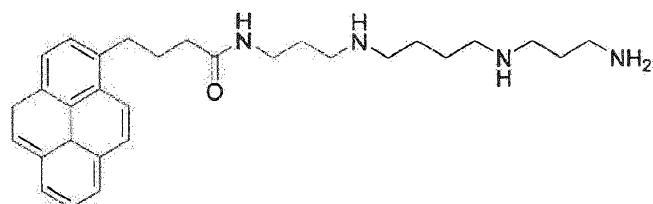
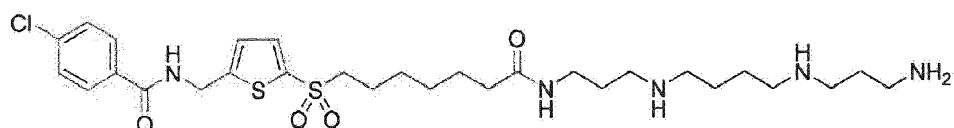
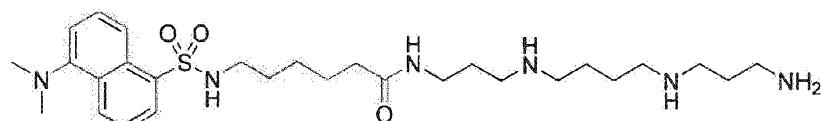
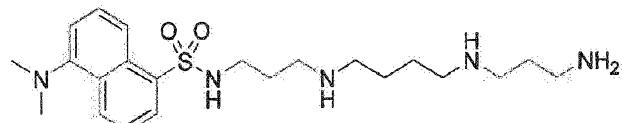
$x=1$  to 4;  $y=1$  to 3,

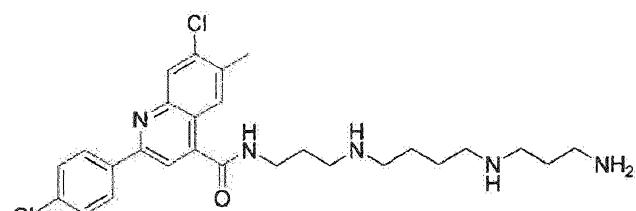
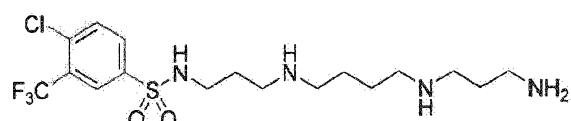
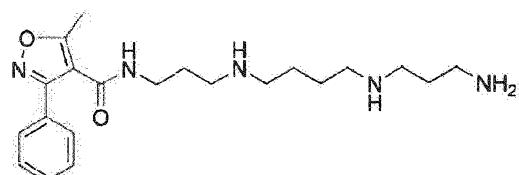
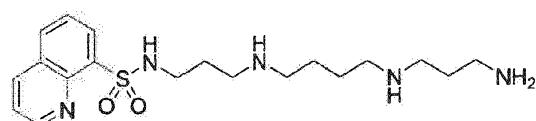
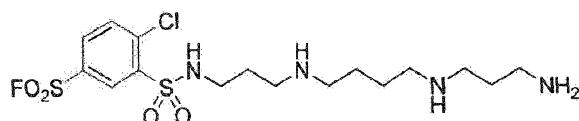
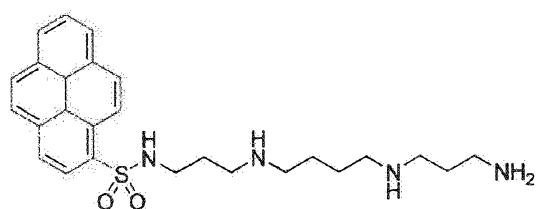
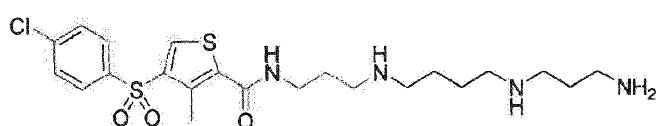
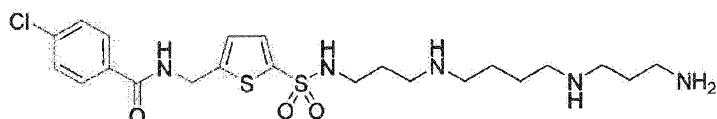
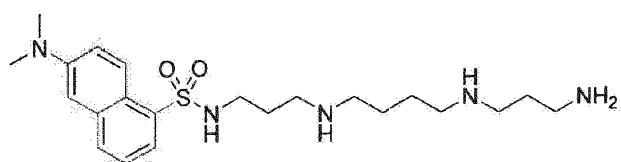
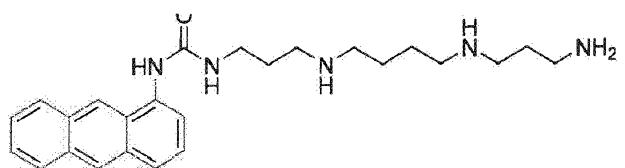
$R_{10}$  and  $R_{11}$  are, independently, H,  $(CH_2)_nNHR_{12}$  or  $(CH_2)_kNH(CH_2)_lNHR_{12}$ ,

where  $n=k=1$  to 10, and  $R_{12}$  is H or  $C(N=H)NH_2$ .

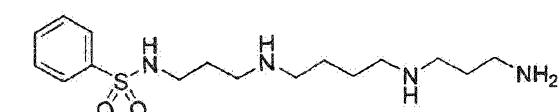
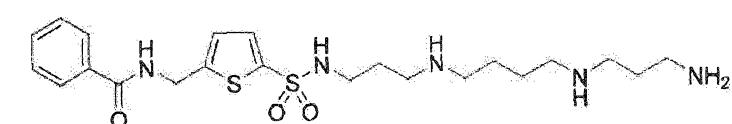
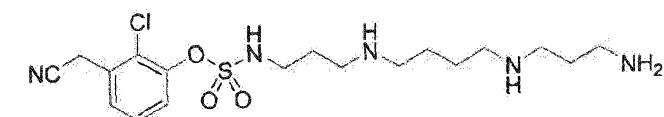
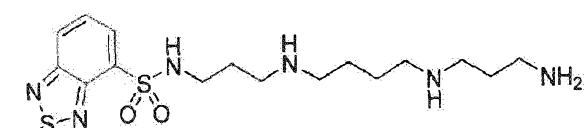
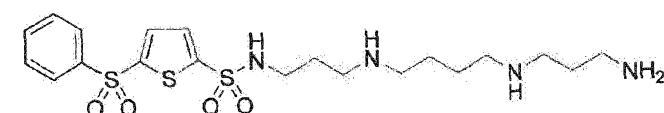
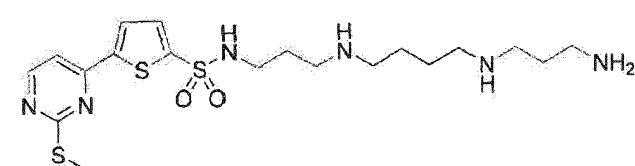
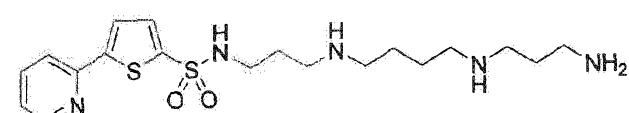
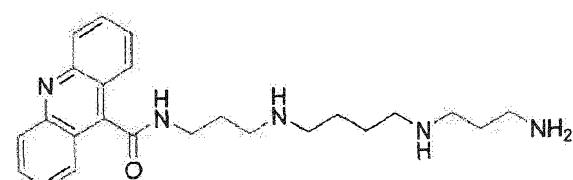
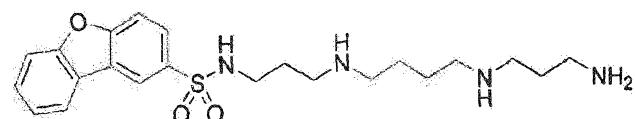
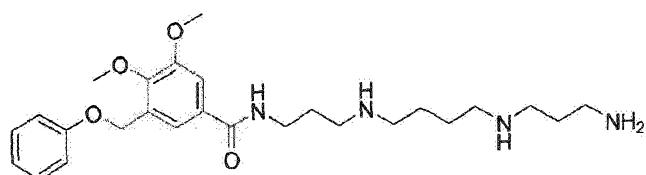
**[0171]** In the above compositions,  $R_2$  is preferably selected from the group consisting of N1-acetylspermamine, N1-acetylspermidine, N8-acetylspermidine, N'-guanidinospermine, cadaverine, aminopropylcadaverine, homo spermidine, caldine (horspermidine), 7-hydroxyspermidine, thermine (norspermine), thermospermine, canavalmine, aminopropylhomospermidine, N,N'-bis(3 -aminopropyl)cadaverine, aminopentylnorspermidine, N4-aminopropylnorspermidine, N4-aminopropylspermidine, caldopentamine, homocaldopentamine, N4-bis(aminopropyl)norspermidine, thermopentamine, N4-bis(aminopropyl)spermidine, caldohexamine, homothermohexamine, homocaldohexamine, N-(3-aminopropyl)-1,3-propanediamine, N,N'-bis(3-aminopropyl)ethylendiamine, N,N'-bis(3 -aminopropyl)-1,4-piperazine, N,N'-bis(3-aminopropyl)-1,3-piperazine, N,N'-bis(3-aminopropyl)-1,3-propanediamine, N,N'-bis(2-aminoethyl)-1,3-propanediamine, tris(3 -aminopropyl)amine, and tris(aminoethyl)amine.

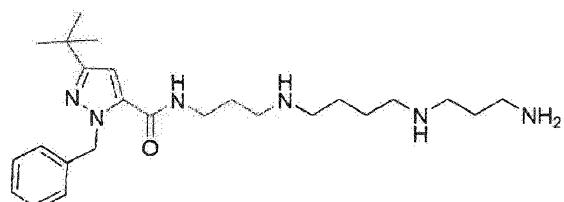
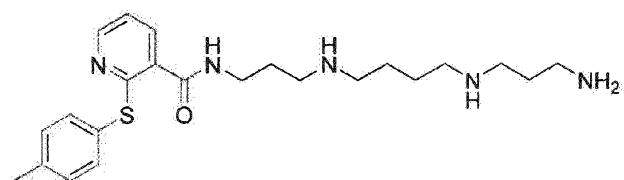
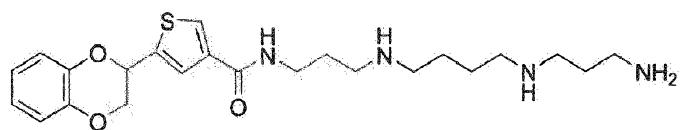
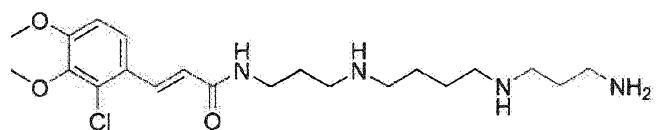
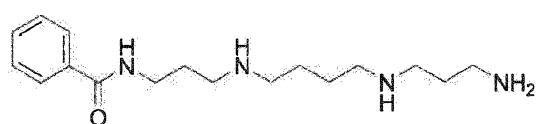
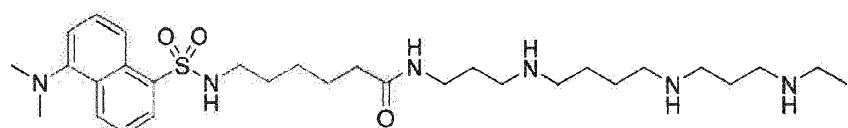
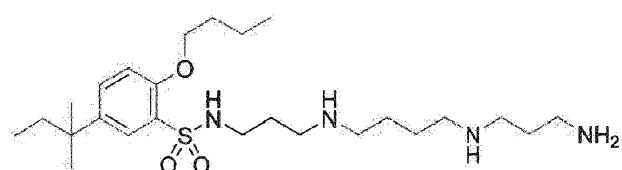
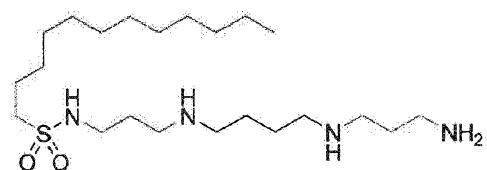
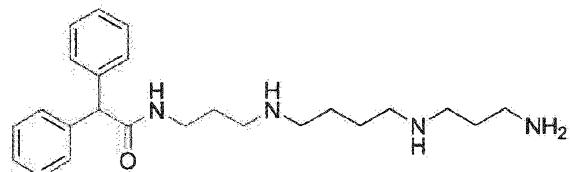
**[0172]** Compounds include, but are not limited to:

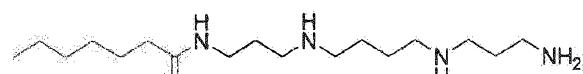
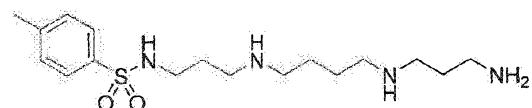
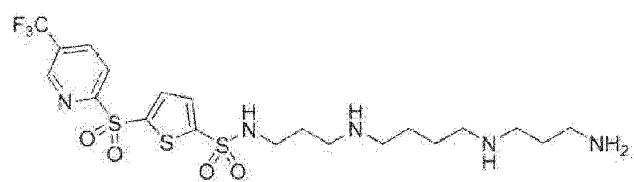
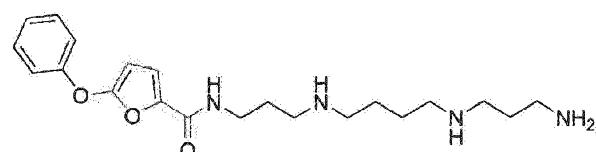
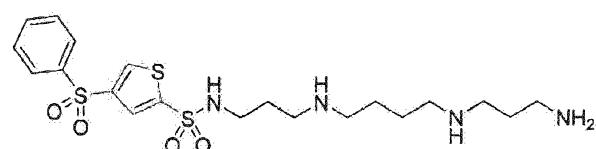
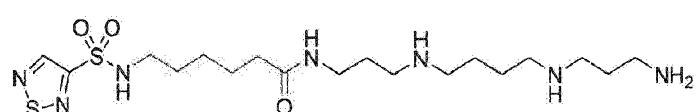
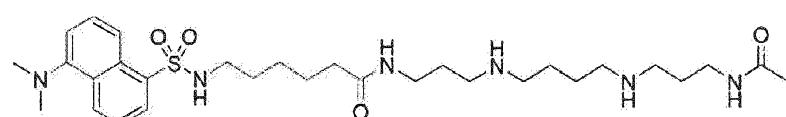
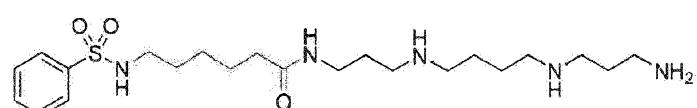
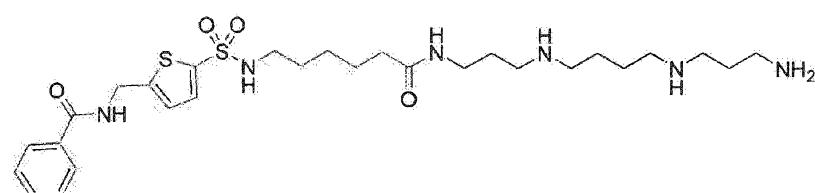
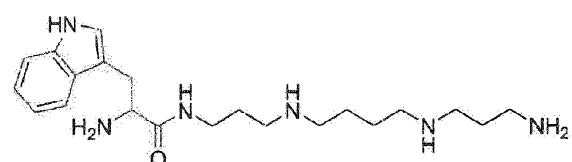
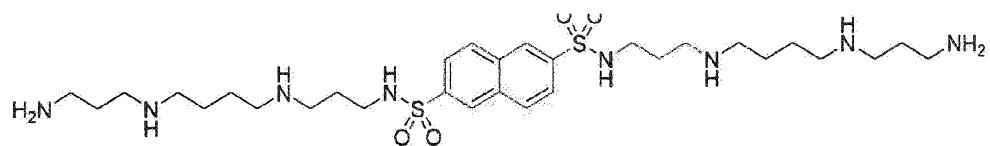


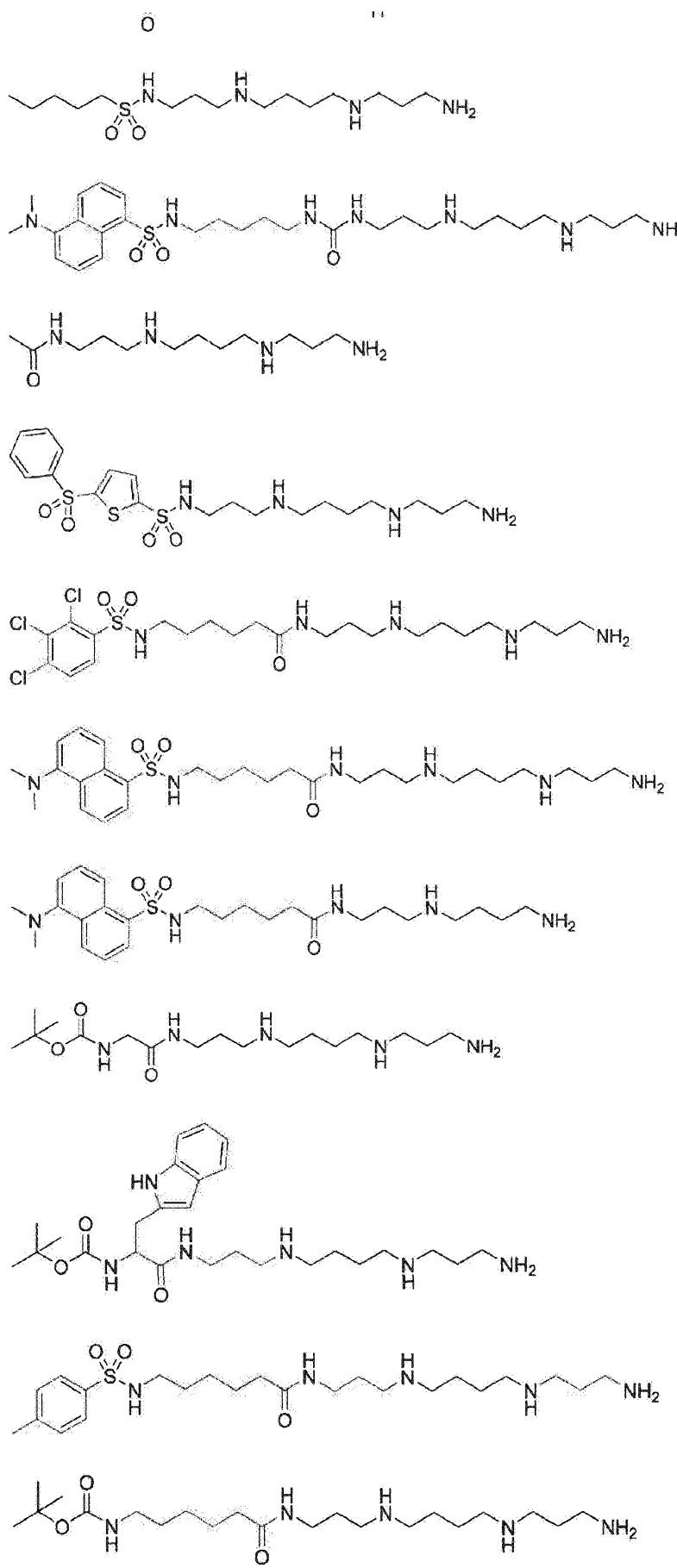


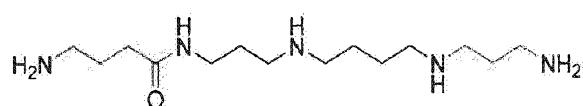
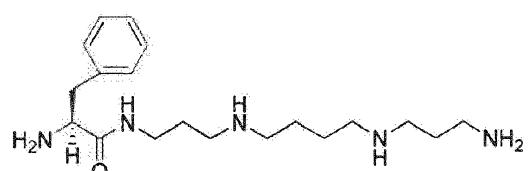
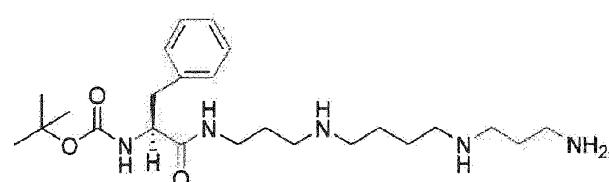
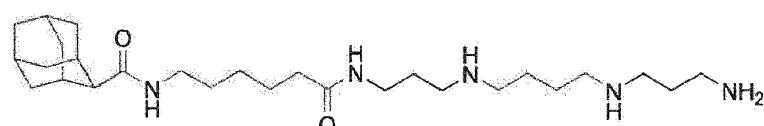
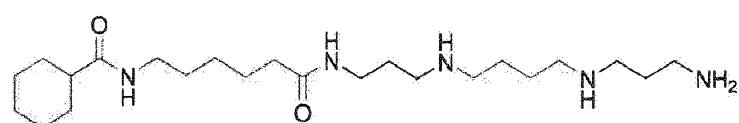
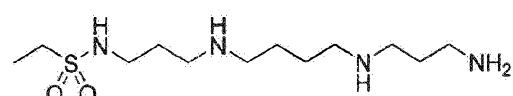
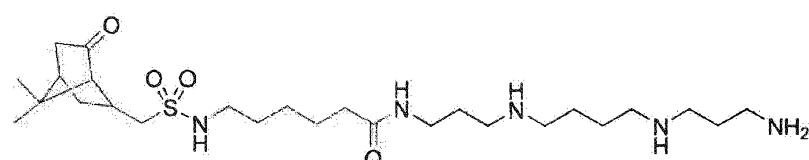
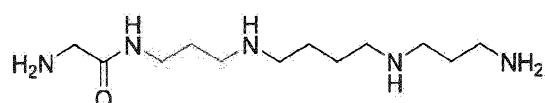
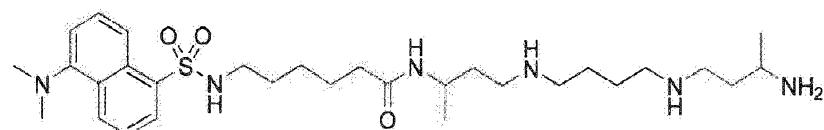
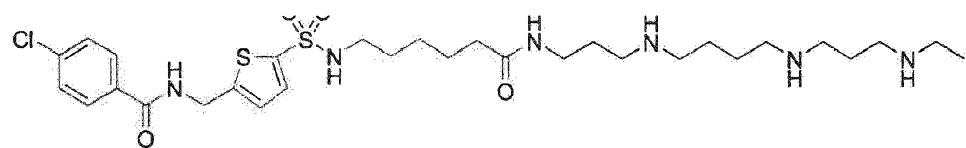
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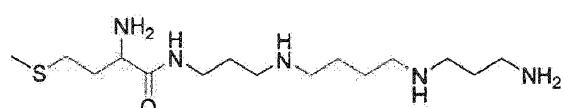
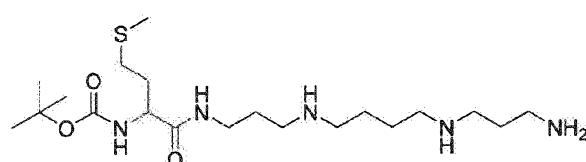
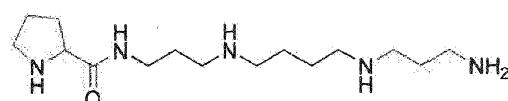
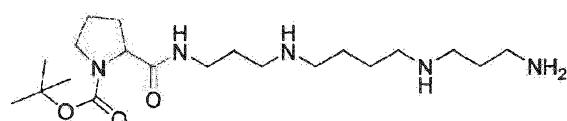
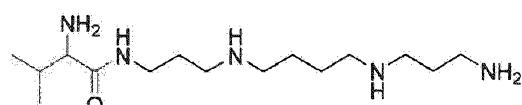
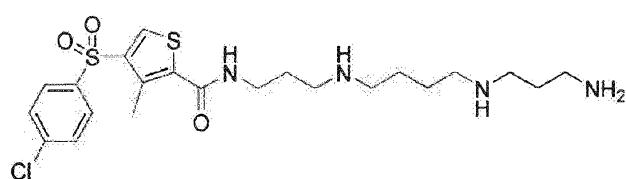
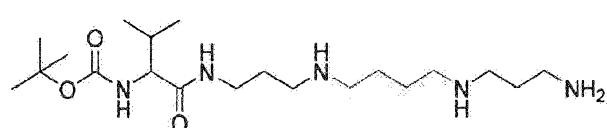
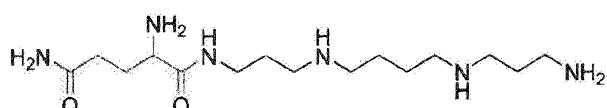
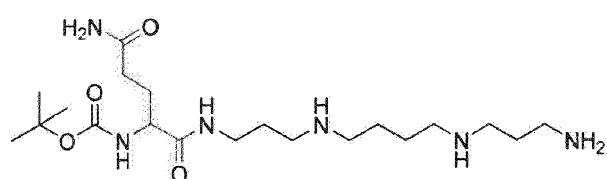
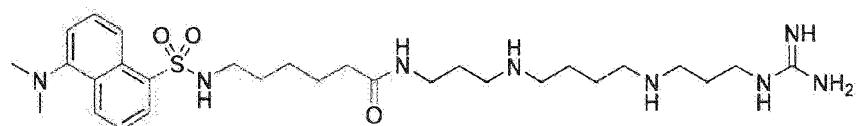








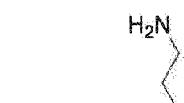
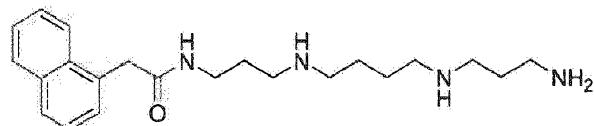
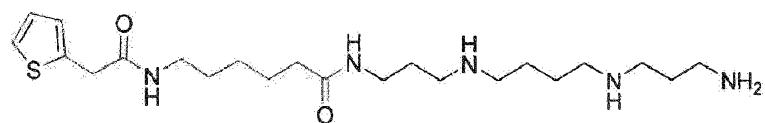
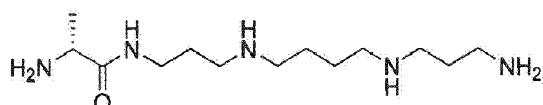
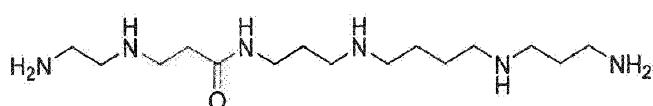
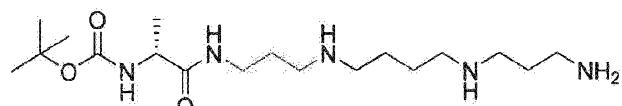
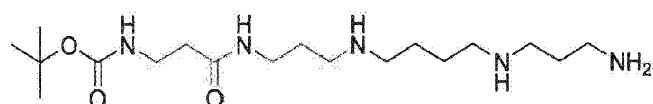
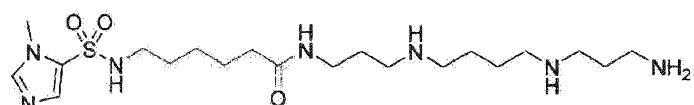
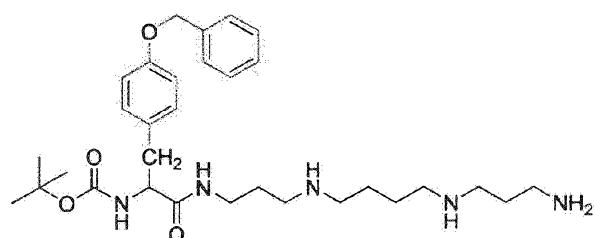
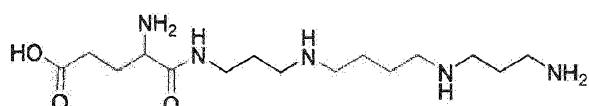
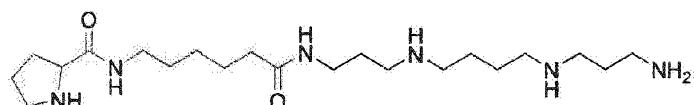
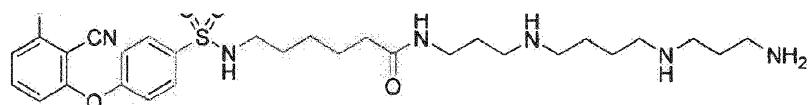


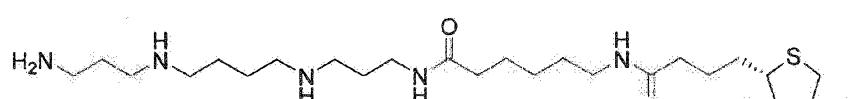
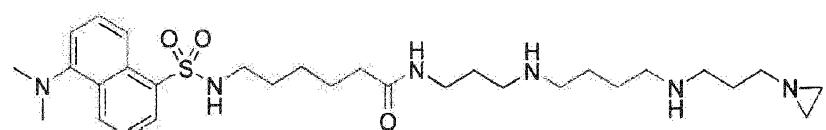
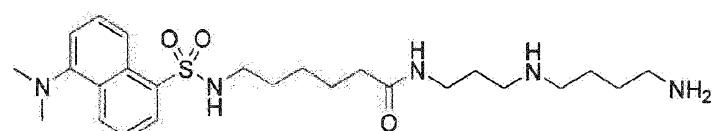
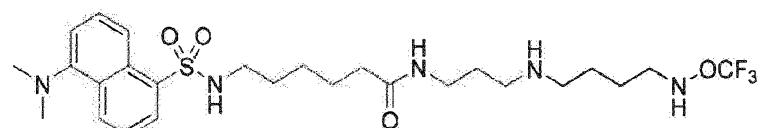
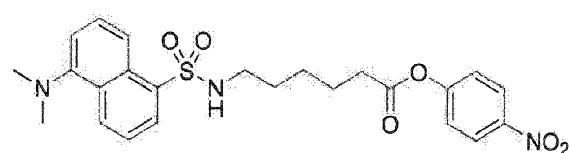
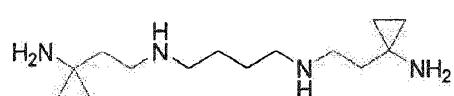
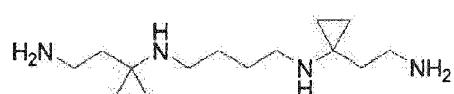
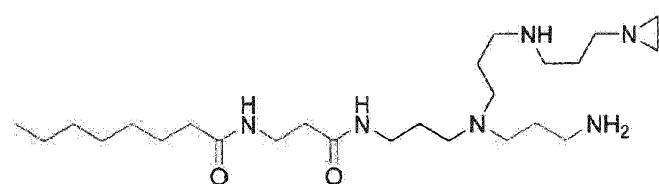
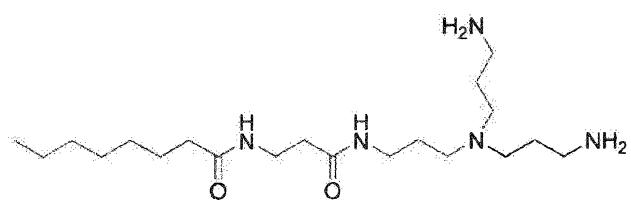
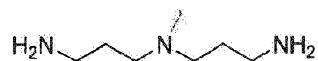


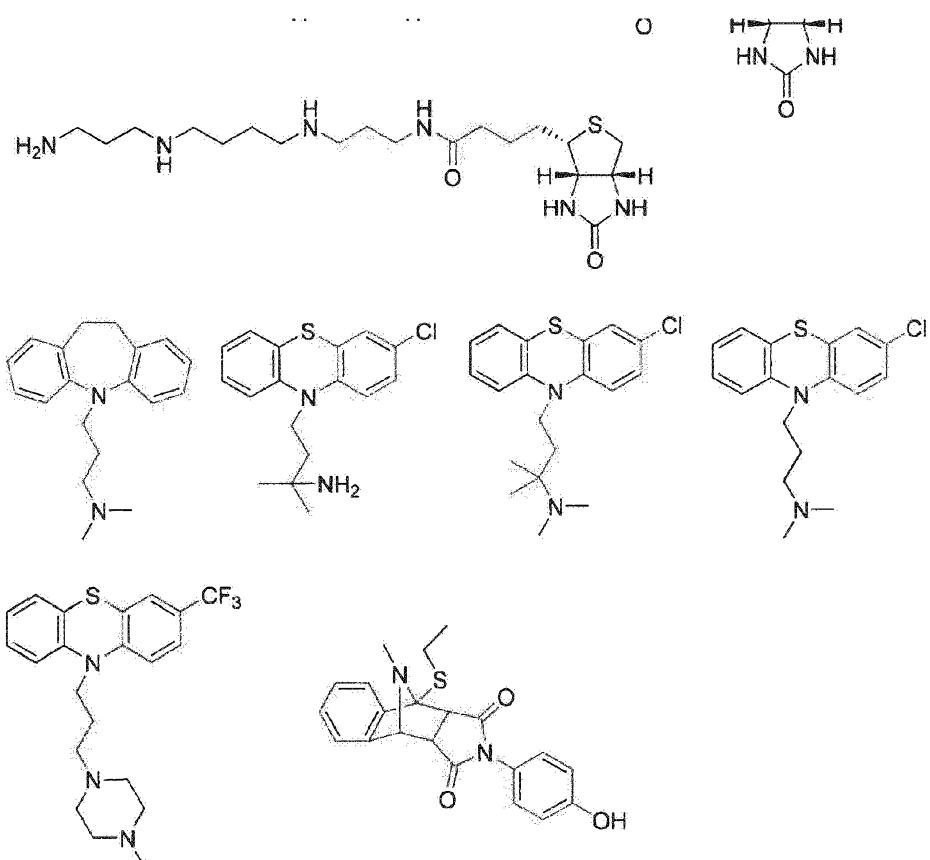
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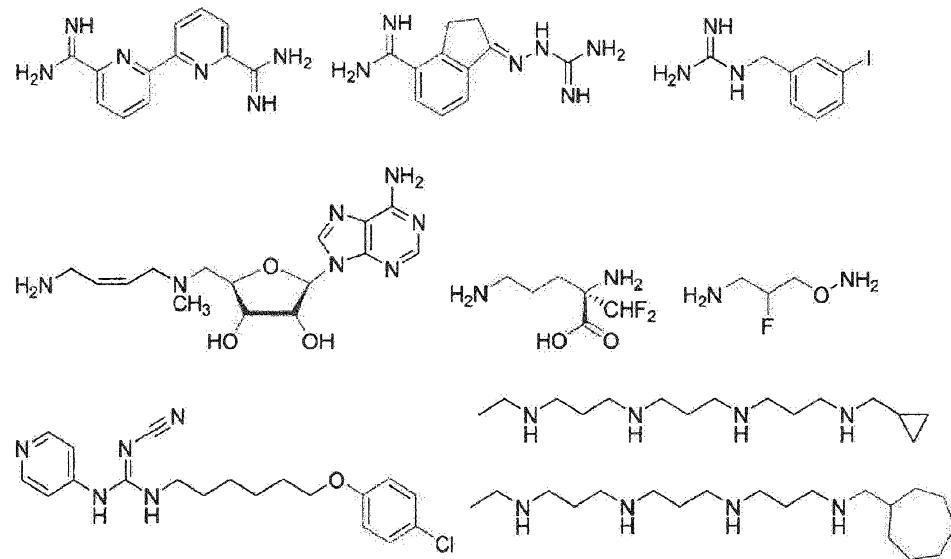






[0173] Additional disclosure may be found in WO99/03823.

[0174] Yet further compounds include, but are not limited to:



[0175] Additional disclosure may be found in: Ackermann, JM; Pegg, AE; McCloskey, DE; Progress in Cell Cycle Research, 2003, Vol. 5, 461-468; Ekelund, S; Nygren, P; Larsson, R; Biochemical Pharmacology, 2001, 61, 1183-1193; Huang, Y; Pledgie, A; Casero Jr, RA;

Davidson, NE; Anti-Cancer Drugs, 2005, 16, 229-241; and Marton, JL; Annu. Rev. Pharmacol Toxicol. 1995, 35, 55-91.

**[0176]** Polyamine analogs depicted above may be prepared both as salts and as free bases. In certain embodiments, the salt is the hydrochloride salt. In certain embodiments, the number of coordinated ion pairs (for example  $\text{H}^+\text{Cl}^-$ ) will be proportional to the number of amino groups in the polyamine. Such coordination typically occurs at said amino groups, forming, for example,  $\text{NH}_3^+\text{Cl}^-$  groups. However, not every amino group may be coordinated. For example, if the amino group is adjacent to an electron-withdrawing group such as carbonyl or sulfonyl, it may not retain sufficient electron density to coordinate an ion. In further embodiments, the number of coordinated ions will be proportional to the number of primary and/or secondary amino groups in the polyamine.

**[0177]** Additional compounds which may be used in the methods and compositions described herein include: naturally occurring polyamines found in prokaryotes and eukaryotic cells, polyamine analogs, polyamine biosynthesis inhibitors, and polyamine transport inhibitors.

**[0178]** Naturally occurring polyamines found in prokaryotes and eukaryotic cells include, but are not limited to: putrescine, spermidine, spermine, diaminopropane, cadaverine, norspermidine, aminopropylcadaverine, homospermine, norspermine, thermospermine, aminopentylnorspermidine, bis(aminopropyl)cadaverine, aminopropylhomospermine, 30 canavalmine, homospermine, caldopentamine, aminopropylcanavaline, bis(aminopropyl)homospermidine, bis(aminobutyl)norspermidine, aminobutylcanavaline, aminopropylhomospermine, homopentamine, N5-aminobutylhomospermine, caldohexamine, thermohexamine, homothermohexamine, agmatine and N6-methylagmatine. See, e.g., Morgan D. M. L., 1999, Molecular Biotechnology 11: 229.

**[0179]** Polyamine analogs include, but are not limited to, BE-4444 [1,19-bis (ethylamino)-5,10,15-triazanonadecane]; BE-3-3-3 [N1,N11-diethylnorspermine; DENSPM; 1,11-bis (ethylamino)-4,8-diazaundecane; thermine; Warner-Parke-Davis]; BE-3-3[N1,N7-bis(ethyl) norspermidine]; BE-3-4[N1,N8-bis(ethyl) spermidine]; BE44 [N1,N9-bis(ethyl) homospermidine]; BE-343 [N1,N12-bis(ethyl) spermine; diethylspermine-N1-N12; DESPM]; BE-373 [N,N'-bis (3-ethylamino) propyl]-1,7-heptane diamine, Merrell-Dow]; BE-4-4-4 [N1,N14-bis(ethyl) homospermine; diethylhomospermine-N1-N1-1]; BE-3-4-4-3 [1,17-bis(ethylamino)-4,9,14triazahedecane]; BE-4-3-3-4[1,17-bis(ethylamino)-5,9,13-triazahedecane]; and 1,12-Mez-SPM[1,12-dimethylspennine]. (WO2007/040535).

**[0180]** Polyamine synthesis inhibitors include but are not limited to: inhibitors of ornithine decarboxylase such as DFMO, aceylenic putrescine, 1-aminoxy-3-aminopropane, antizyme, 2-butylputrescine, cadaverine, L-canaline, 5'-deoxy-5'-[N-methyl-N-[3(aminoxy)ethyl]amino]adenosine, 5'-deoxy-5'-[N-methyl-N-[3-(hydrazinopropyl)amino]adenosine, diaminopropane, 1,3-diamino-2-propanol, 2-difluoromethyl putrescine, difluorophenylethyl(4-aminopropylamidinohydrazone), 2,3-dimethylputrescine, N-

dimethylputrescine, 2-ethylputrescine, (+ or -)-alphafluoromethylornithine, 2-fluoro methylputrescine, 2-hexylputrescine, 2-hydrazinoornithine, ibuprofen, D-methyl acetylenic putrescine, methylglyoxal bis(3aminopropylamininohydrazone), 2-methylornithine, 2-methylputrescine, 2-monofluoromethyl-trans-dehydorornithine, 2-monofluoromethyl dehydroputrescine, monofluoromethylomithine, 2-monofluoromethyl putrescine, neomycin, D-omithine, 2-pentylputrescine, p-phenylenediamine, phosphopeptide MG 25000, phosphothreonine, phosphotyrosine, 2-propylputrescine, putrescine, allo-S-adenosyl-L-methionine, S-ethylthioadenosine, methylthioadenosine, and 5' -methylthioadenosine as discussed in Zollner H. (1993) Handbook of Enzyme Inhibitors, 2nd Ed. Weinheim:Basel(Switzerland); inhibitors of S-adenosylmethionine decarboxylase, such as SAM486A (4-aminoindanon-1(2'amidino)hydrazone dihydrochloride monohydrate), S-adenosyl-1,8-diamino-3thiooctane, S-(5'-adenosyl)methylthio-2-aminoxyethan, S-adenosyl-3-methylthio-i-propylamine, 5'-{[(Z)-4-amino-2-but enyl]methylamino}-5'-deoxyadenosine, 5'-amino-5'deoxyadenosine, 5'-[(aminoiminomethyl)amino]-5']deoxyadenosine dihydrogensulphate, 1-aminoxy-3-aminopropane, [2-(aminoxy)ethyl](5'-deoxyadenosine-5'yl)(methyl)sulphonium, 5'-[(3-aminopropyl]-amino)-5'-deoxyadenosine, 5'-[(3aminopropyl]-methylamino)-5'-deoxyadenosine, 9-[6(RS)-amino-5,6,7-trideoxy-beta-D-ribo-octofuranosyl]-9H-purin-6-amine, borohydride, n-butylglyoxal bis(guanylhydrazone), 9-[6(RS)-c-carboxamido-5,6,7-trideoxy-beta-D-ribo-octofuranosyl]-9H-purin-6-amine, cyanide, cyanoborohydride, S-(5' deoxy-5'adenosyl)methionylethylhydroxylamine, S-(5' deoxy-5'adenosyl)methionylthiohydroxylamine, 5' -deoxy-5'-[N-methyl-N-[2(aminoxy)ethyl]amino]adenosine, 9-[6(S)-diamino-5,6,7,8,9-pentadeoxy-beta-D-ribo-nanofuranosyl] -9H-purin-6-amine, diethylglyoxal bis(guanylhydrazone), difluorophynylethyl (4-aminopropylamidinohydrazone), dimethyl(5' - adenosyl)sulfonium, dimethylglyoxal bis(guanylhydrazone), ethylglyoxal bis(guanylhydrazone), hydroxylamine, 4-hydroxypenal, MDL 73811, 5'[[3-methylamino)propyl] amino]-5'-deoxyadenosine(1,1'-(methylethanediylidine)dinitro)bis(3-aminoguanidne), methylglyoxal bis(3-aminopropylamidinohydrazone), methylglyoxal bis(cyclohexylamidinohydrazone), pentanedraldehyde bis(guanylhydrazone), phenylhydrazine, propanedraldehyde bis(guanylhydrazone), semicarbazide, sodium borohydride, sodium cyanoborohydride, and spermine as discussed in Zollner H. (1993) Handbook of Enzyme Inhibitors, 2nd Ed. Additional disclosure may be found in WO2002/053519.

**[0181]** Additional spermine analogs include N-(2-mercptoethyl)spermine-5-carboxamide (MESC), the disulfide from thereof, namely 2,2 1-dithiobis(N-ethyl-spermine-5-carboxamide) (DESC), and N-[2,2,1-dithio(ethyl 1,1-aminoethyl)]spermine-5-carboxamide (DEASC). (WO98/17623)

**[0182]** Polyamine effectors that are small molecule inhibitors or modulators of key enzymes in the polyamine biosynthetic pathway include, but are not limited to: ODC inhibitors such as difluoromethylomithine (DFMO), alpha-monofluoromethylomithine (MFMO), and methyl acetylenicputrescine (MAP); AdometDC inhibitors such as S-(5-deoxy-5adenoxyl)methylthioethylhydroxylamine (AMA), 5-deoxy-5-[(2aminoxyethyl)methylamino]adenosine (MAOEA), and methylglyoxal bis(guanylhydrazone)

(MGBG); spermidine synthase inhibitors such as S-adenosyll,8-diamino-3-thiooctane (AdoDATO), cyclohexylamine, and butylamine; spermine synthase inhibitors such as S-adenosyl-1,12-diamino-3-thio-9-azadodecane (AdoDATAD) and N-(n-butyl)-1,3-diaminopropane (BDAP).

**[0183]** In certain embodiments, the polyamine effector is a polyamine or arginine analog that carries a functional group that confers a cellular or DNA protective effect to the molecule, or that modulates the polyamine biosynthetic or catabolic pathway. Compounds of this nature include, but are not limited to amifostine, NG-hydroxy-arginine (NORA), N1, N11-bis(ethyl) norspermine (BE-3-3-3), N12-bis(ethyl)spermine (BE-3-4-3), N,N-bis[3-(ethylamino)-propyl]-1,7heptanediamine (BE-3-7-3), BE-3-3-3, BE-3-4-3, BE-3-7-3,N1-ethyl-N11-propargyl 4,8-diazaundecane, and the analogs SL-11141 and SL-II050 (structures set forth in one or more of U.S. Patent 5,889,061, Valasinas et al., 2001, *supra*, WO 00/66587 and WO 02/38105). Additional disclosure may be found in WO03/013245.

**[0184]** As used herein, the terms below have the meanings indicated.

**[0185]** The term "cytokine," as used herein, alone or in combination, means signaling molecules secreted by cells of the immune system which have a local immunoregulatory effect. Cytokines may include, without limitation, IL-1, IL1-Ra, IL-2, IL-6, IL8, IPNy, IP-10, IL-17, MCP-1, MMP-9, MIP-1 $\beta$ , TNF- $\alpha$ , TGF $\beta$ , CRP, OPN, and RANTES.

**[0186]** The term "osteopontin" is used interchangeably with "OPN," "SPP1," "Eta-1," sialoprotein I or 44K BPP (bone phosphoprotein). In general, osteopontin refers to any full length or partial fragment of a full-length osteopontin. Osteopontin can also refer to any modified, e.g., glycosylated, osteopontin.

**[0187]** The term "activity" as used herein in connection with osteopontin refers to both, the biological activity of the polypeptide and to the quantity or level of osteopontin present in the cell. In one embodiment, the term activity refers to the quantity of osteopontin, e.g., present, expressed or produced in the cell. In another embodiment, it refers to the level of osteopontin secreted by the cell, for example, by a mononuclear cell.

**[0188]** When ranges of values are disclosed, and the notation "from n<sub>1</sub> ... to n<sub>2</sub>" or "between n<sub>1</sub> ... and n<sub>2</sub>" is used, where n<sub>1</sub> and n<sub>2</sub> are the numbers, then unless otherwise specified, this notation is intended to include the numbers themselves and the range between them. This range may be integral or continuous between and including the end values. By way of example, the range "from 2 to 6 carbons" is intended to include two, three, four, five, and six carbons, since carbons come in integer units. Compare, by way of example, the range "from 1 to 3 ( $\mu$ M (micromolar)," which is intended to include 1  $\mu$ M, 3  $\mu$ M, and everything in between to any number of significant figures (e.g., 1.255  $\mu$ M, 2.1  $\mu$ M, 2.9999  $\mu$ M, etc.).

**[0189]** The term "about," as used herein, is intended to qualify the numerical values which it modifies, denoting such a value as variable within a margin of error. When no particular margin

of error, such as a standard deviation to a mean value given in a chart or table of data, is recited, the term "about" should be understood to mean that range which would encompass the recited value and the range which would be included by rounding up or down to that figure as well, taking into account significant figures.

**[0190]** The term "substantially" as used herein is intended to mean predominantly or having the overriding characteristic of, such that any opposing or detracting characteristics reach a level of insignificance. By way of example, a composition "substantially" free of water might not be absolutely free of all traces of water, but would be sufficiently anhydrous that any remaining water would not influence the composition in any significant way. By way of further example, "substantially dose-limiting side effects" might be side effects which limited a dose to a level which was below that required for therapeutic efficacy.

**[0191]** The term "disease" as used herein is intended to be generally synonymous, and is used interchangeably with, the terms "disorder," "syndrome," and "condition" (as in medical condition), in that all reflect an abnormal condition of the human or animal body or of one of its parts that impairs normal functioning, is typically manifested by distinguishing signs and symptoms, and causes the human or animal to have a reduced duration or quality of life.

**[0192]** A "proliferative disorder" may be any disorder characterized by dysregulated cellular proliferation. Examples include cancers, psoriasis, and atopic dermatitis.

**[0193]** As used herein, "hyperalgesia" means a heightened sensitivity to pain, and can be considered a type of pain or a measure of pain-related behavior.

**[0194]** As used herein, reference to "treatment" of a patient is intended to include prophylaxis. Treatment may also be preemptive in nature, i.e., it may include prevention of disease. Prevention of a disease may involve complete protection from disease, for example as in the case of prevention of infection with a pathogen, or may involve prevention of disease progression. For example, prevention of a disease may not mean complete foreclosure of any effect related to the disease at any level, but instead may mean prevention of the symptoms of a disease to a clinically significant or detectable level. Prevention of diseases may also mean prevention of progression of a disease to a later stage of the disease.

**[0195]** The term "combination therapy" means the administration of two or more therapeutic agents to treat a therapeutic condition or disorder described in the present disclosure. Such administration encompasses co-administration of these therapeutic agents in a substantially simultaneous manner, such as in a single capsule having a fixed ratio of active ingredients or in multiple, separate capsules for each active ingredient. In addition, such administration also encompasses use of each type of therapeutic agent in a sequential manner. In either case, the treatment regimen will provide beneficial effects of the drug combination in treating the conditions or disorders described herein.

**[0196]** The term "patient" is generally synonymous with the term "subject" and means an

animal differing from a disease, disorder, or condition treatable in accordance with the methods disclosed herein, including all mammals and humans. Examples of patients include humans, livestock such as cows, goats, sheep, pigs, and rabbits, and companion animals such as dogs, cats, rabbits, and horses. Preferably, the patient is a human.

**[0197]** An "effective amount" or a "therapeutically effective amount" is a quantity of a compound (e.g., MGBG, a polyamine analog, a polyamine biosynthesis inhibitor or any agent) that is sufficient to achieve a desired effect in a subject being treated. For instance, this can be the amount necessary to treat a disease, disorder, condition, or adverse state (such as pain or inflammation) or to otherwise measurably alter or alleviate the symptoms, markers, or mechanisms of the disease, disorder, condition, or adverse state. As just one example, an effective amount for the treatment of pain is an amount sufficient to prevent, delay the onset of, or reduce pain or one or more pain-related symptoms in a subject, as measured by methods known in the art. Similar methods of assessing response to treatment of a number of diseases are well-known in the art. The effective amount of a compound described herein may vary depending upon the route of administration and dosage form. In addition, specific dosages may be adjusted depending on conditions of disease, the age, body weight, general health conditions, sex, and diet of the subject, dose intervals, administration routes, excretion rate, and combinations of agents.

**[0198]** The term "low dose," in reference to a low dose formulation of a drug or a method of treatment specifically employing a "low dose" of a drug, means a dose which for at least one indication is subtherapeutic, or is a fraction of the dose typically given for at least one indication. Take for example the case of a drug for the treatment of proliferative disorders - a low dose formulation for the treatment of, say, chronic psoriasis, might be a fraction of the dose for the treatment of an aggressive cancer. In this way, the dose for one disease might be an amount which would be subtherapeutic for another disease. Alternatively, for a drug which is therapeutic in different individuals or populations at different doses, and is available in a range of doses, a low dose may be simply a dose toward the low end of recognized therapeutic efficacy. Chronic diseases represent an embodiment treatable by low dose formulations and methods. Additionally, a subtherapeutic amount of a drug might be used in combination with one or more other drugs (themselves in either therapeutic or subtherapeutic amounts) to yield a combination formulation or treatment which is potentiated, that is, more efficacious than the expected effects of the sum of the drugs given alone. A low dose for the treatment of one indication may be two-fold, three-fold, four-fold, five-fold, six-fold, seven-fold, eight-fold, nine-fold, ten-fold, fifteen-fold, twenty-fold, thirty-fold, forty-fold, fifty-fold, may be one hundred-fold less than the therapeutic dose for a different indication.

**[0199]** The phrase "therapeutically effective" is intended to qualify the amount of active ingredients used in the treatment of a disease or disorder or on the effecting of a clinical endpoint.

**[0200]** The term "therapeutically acceptable" refers to those compounds (or salts, prodrugs, tautomers, zwitterionic forms, etc.) which are suitable for use in contact with the tissues of

subjects without undue toxicity, irritation, and allergic response, are commensurate with a reasonable benefit/risk ratio, and are effective for their intended use.

**[0201]** The term "drug" is used herein interchangeably with "compound" and "agent."

**[0202]** As used herein, a "polyamine" is any of a group of aliphatic, straight-chain amines derived biosynthetically from amino acids; polyamines are reviewed in Marton et al. (1995) *Ann. Rev. Pharm. Toxicol.* 35:55-91. By "polyamine" is generally meant a naturally-occurring polyamine or a polyamine which is naturally produced in eukaryotic cells. Examples of polyamines include putrescine, spermidine, spermine and cadaverine.

**[0203]** As used herein, a "polyamine analog" is an organic cation structurally similar but non-identical to naturally-occurring polyamines such as spermine and/or spermidine and their precursor, diamine putrescine. Polyamine analogs can be branched or un-branched, or incorporate cyclic moieties. Polyamines may comprise primary, secondary, tertiary, or quaternary amino groups. In one embodiment, all the nitrogen atoms of the polyamine analogs are independently secondary, tertiary, or quaternary amino groups, but are not so limited. Polyamine analogs may include imine, amidine and guanidine groups in place of amine groups. The term "polyamine analog" includes stereoisomers, salts and protected derivatives of polyamine analogs.

**[0204]** A "stereoisomer" is any optical isomer of a compound, including enantiomers and diastereomers. Unless otherwise indicated, structural formulae of compounds are intended to embrace all possible stereoisomers.

**[0205]** A "salt" or "pharmaceutically acceptable salt" is a compound formed by the replacement of one or more hydrogen atoms with elements or groups, which is composed of anions and cations, which usually ionizes in water; a salt is formed, for instance, by neutralization of an acid by a base. Examples of salts include, but are not limited to, halide, for example, chloride, bromide, or iodide, nitrate, sulfate, bisulfate, phosphate, acid phosphate, isonicotinate, acetate, lactate, salicylate, citrate, tartrate, pantothenate, bitartrate, ascorbate, succinate, maleate, gentisinate, fumarate, gluconate, glucuronate, saccharate, formate, benzoate, glutamate, methanesulfonate, ethanesulfonate, benzensulfonate, p-toluenesulfonate and pamoate (i.e., 1,1'-methylene-bis-(2-hydroxy-3-naphthoate)) salts.

**[0206]** "Protected derivative" is used to refer to a compound protected with a protecting group. "Protecting group" refers to a chemical group that exhibits the following characteristics: 1) reacts selectively with the desired functionality in good yield (preferably at least 80%, more preferably at least 90%, more preferably at least 95%, still more preferably at least 99%) to give a protected substrate that is stable to the projected reactions for which protection is desired; 2) is selectively removable from the protected substrate to yield the desired functionality; and 3) is removable in good yield (preferably at least 80%, more preferably at least 90%, more preferably at least 95%, still more preferably at least 99%) by reagents compatible with the other functional group(s) present or generated in such projected reactions.

Examples of suitable protecting groups can be found in Greene et al. (1991) Protective Groups in Organic Synthesis, 2nd Ed. (John Wiley & Sons, Inc., New York). Exemplary protecting groups for the amino functionality include, but are not limited to, mesitylenesulfonyl (MesS02), benzyloxycarbonyl (CBz), t-butyloxycarbonyl (Boc), t-butyldimethylsilyl (TBDIMS), 9-fluorenylmethyloxycarbonyl (Fmoc), or suitable photolabile protecting groups such as 6-nitroveratryloxy carbonyl (Nvoc).

**[0207]** The term "acyl," as used herein, alone or in combination, refers to a carbonyl attached to an alkenyl, alkyl, aryl, cycloalkyl, heteroaryl, heterocycle, or any other moiety where the atom attached to the carbonyl is carbon. An "acetyl" group refers to a -C(O)CH<sub>3</sub> group. An "alkylcarbonyl" or "alkanoyl" group refers to an alkyl group attached to the parent molecular moiety through a carbonyl group. Examples of such groups include methylcarbonyl and ethylcarbonyl. Examples of acyl groups include formyl, alkanoyl and aroyl.

**[0208]** The term "alkenyl," as used herein, alone or in combination, refers to a straight-chain or branched-chain hydrocarbon radical having one or more double bonds and containing from 2 to 20 carbon atoms. In certain embodiments, said alkenyl will comprise from 2 to 6 carbon atoms. The term "alkenylene" refers to a carbon-carbon double bond system attached at two or more positions such as ethenylene [(-CH=CH-),(-C::C-)]. Examples of suitable alkenyl radicals include ethenyl, propenyl, 2-methylpropenyl, 1,4-butadienyl and the like. Unless otherwise specified, the term "alkenyl" may include "alkenylene" groups.

**[0209]** The term "alkoxy," as used herein, alone or in combination, refers to an alkyl ether radical, wherein the term alkyl is as defined below. Examples of suitable alkyl ether radicals include methoxy, ethoxy, n-propoxy, isopropoxy, n-butoxy, iso-butoxy, sec-butoxy, tert-butoxy, and the like.

**[0210]** The term "alkyl," as used herein, alone or in combination, refers to a straight-chain or branched-chain alkyl radical containing from 1 to 20 carbon atoms. In certain embodiments, said alkyl will comprise from 1 to 10 carbon atoms. In further embodiments, said alkyl will comprise from 1 to 6 carbon atoms. Alkyl groups may be optionally substituted as defined herein. Examples of alkyl radicals include methyl, ethyl, n-propyl, isopropyl, n-butyl, isobutyl, sec-butyl, tert-butyl, pentyl, iso-amyl, hexyl, octyl, nonyl and the like. The term "alkylene," as used herein, alone or in combination, refers to a saturated aliphatic group derived from a straight or branched chain saturated hydrocarbon attached at two or more positions, such as methylene (-CH<sub>2</sub>-). Unless otherwise specified, the term "alkyl" may include "alkylene" groups.

**[0211]** The term "alkylamino," as used herein, alone or in combination, refers to an alkyl group attached to the parent molecular moiety through an amino group. Suitable alkylamino groups may be mono- or dialkylated, forming groups such as, for example, N-methylamino, N-ethylamino, N,N-dimethylamino, N,N-ethylmethylamino and the like.

**[0212]** The term "alkynyl," as used herein, alone or in combination, refers to a straight-chain or branched chain hydrocarbon radical having one or more triple bonds and containing from 2 to

20 carbon atoms. In certain embodiments, said alkynyl comprises from 2 to 6 carbon atoms. In further embodiments, said alkynyl comprises from 2 to 4 carbon atoms. The term "alkynylene" refers to a carbon-carbon triple bond attached at two positions such as ethynylene (-C<sub>2</sub>=C-), (-C≡C-). Examples of alkynyl radicals include ethynyl, propynyl, hydroxypropynyl, butyn-1-yl, butyn-2-yl, pentyn-1-yl, 3-methylbutyn-1-yl, hexyn-2-yl, and the like. Unless otherwise specified, the term "alkynyl" may include "alkynylene" groups.

**[0213]** The terms "amido" and "carbamoyl," as used herein, alone or in combination, refer to an amino group as described below attached to the parent molecular moiety through a carbonyl group, or vice versa. The term "C-amido" as used herein, alone or in combination, refers to a -C(O)N(RR') group with R and R' as defined herein or as defined by the specifically enumerated "R" groups designated. The term "N-amido" as used herein, alone or in combination, refers to a RC(O)N(R')- group, with R and R' as defined herein or as defined by the specifically enumerated "R" groups designated. The term "acylamino" as used herein, alone or in combination, embraces an acyl group attached to the parent moiety through an amino group. An example of an "acylamino" group is acetylamino (CH<sub>3</sub>C(O)NH-).

**[0214]** The term "amino," as used herein, alone or in combination, refers to -NRR', wherein R and R' are independently selected from the group consisting of hydrogen, alkyl, acyl, heteroalkyl, aryl, cycloalkyl, heteroaryl, and heterocycloalkyl, any of which may themselves be optionally substituted. Additionally, R and R' may combine to form heterocycloalkyl, either of which may be optionally substituted.

**[0215]** The term "aryl," as used herein, alone or in combination, means a carbocyclic aromatic system containing one, two or three rings wherein such polycyclic ring systems are fused together. The term "aryl" embraces aromatic groups such as phenyl, naphthyl, anthracenyl, and phenanthryl.

**[0216]** The term "arylalkyl" or "aralkyl," as used herein, alone or in combination, refers to an aryl group attached to the parent molecular moiety through an alkyl group. The term "carboxyl" or "carboxy," as used herein, refers to -C(O)OH or the corresponding "carboxylate" anion, such as is in a carboxylic acid salt. An "O-carboxy" group refers to a RC(O)O- group, where R is as defined herein. A "C-carboxy" group refers to a -C(O)OR groups where R is as defined herein.

**[0217]** The term "cyano," as used herein, alone or in combination, refers to -CN.

**[0218]** The term "cycloalkyl," or, alternatively, "carbocycle" or "alicyclic," as used herein, alone or in combination, refers to a saturated or partially saturated monocyclic, bicyclic or tricyclic alkyl group wherein each cyclic moiety contains from 3 to 12 carbon atom ring members and which may optionally be a benzo fused ring system which is optionally substituted as defined herein. In certain embodiments, said cycloalkyl will comprise from 5 to 7 carbon atoms. Examples of such cycloalkyl groups include cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, tetrahydronaphthyl, indanyl, octahydronaphthyl, 2,3-dihydro-1H-indenyl, adamantyl and the like. "Bicyclic" and "tricyclic" as used herein are intended to include both fused ring

systems, such as decahydronaphthalene, octahydronaphthalene as well as the multicyclic (multicentered) saturated or partially unsaturated type. The latter type of isomer is exemplified in general by, bicyclo[1,1,1]pentane, camphor, adamantane, and bicyclo[3,2,1]octane.

**[0219]** The term "halo," or "halogen," as used herein, alone or in combination, refers to fluorine, chlorine, bromine, or iodine.

**[0220]** The term "heteroalkyl," as used herein, alone or in combination, refers to a stable straight or branched chain, or cyclic hydrocarbon radical, or combinations thereof, fully saturated or containing from 1 to 3 degrees of unsaturation, consisting of the stated number of carbon atoms and from one to three heteroatoms selected from the group consisting of O, N, and S, and wherein the nitrogen and sulfur atoms may optionally be oxidized and the nitrogen heteroatom may optionally be quaternized. The heteroatom(s) O, N and S may be placed at any interior position of the heteroalkyl group. Up to two heteroatoms may be consecutive, such as, for example, -CH<sub>2</sub>-NH-OCH<sub>3</sub>.

**[0221]** The term "heteroaryl," as used herein, alone or in combination, refers to a 3- to 15-membered unsaturated heteromonocyclic ring, or a fused monocyclic, bicyclic, or tricyclic ring system in which at least one of the fused rings is aromatic, which contains at least one atom selected from the group consisting of O, S, and N. In certain embodiments, said heteroaryl will comprise from 5 to 7 carbon atoms. The term also embraces fused polycyclic groups wherein heterocyclic rings are fused with aryl rings, wherein heteroaryl rings are fused with other heteroaryl rings, wherein heteroaryl rings are fused with heterocycloalkyl rings, or wherein heteroaryl rings are fused with cycloalkyl rings. Examples of heteroaryl groups include pyrrolyl, pyrrolinyl, imidazolyl, pyrazolyl, pyridyl, pyrimidinyl, pyrazinyl, pyridazinyl, triazolyl, pyranyl, furyl, thienyl, oxazolyl, isoxazolyl, oxadiazolyl, thiazolyl, thiadiazolyl, isothiazolyl, indolyl, isoindolyl, indolizinyl, benzimidazolyl, quinolyl, isoquinolyl, quinoxalinyl, quinazolinyl, indazolyl, benzotriazolyl, benzodioxolyl, benzopyranyl, benzoxazolyl, benzoxadiazolyl, benzothiazolyl, benzothiadiazolyl, benzofuryl, benzothienyl, chromonyl, coumarinyl, benzopyranyl, tetrahydroquinolinyl, tetrazolopyridazinyl, tetrahydroisoquinolinyl, thienopyridinyl, furopyridinyl, pyrrolopyridinyl and the like. Exemplary tricyclic heterocyclic groups include carbazolyl, benzidolyl, phenanthrolinyl, dibenzofuranyl, acridinyl, phenanthridinyl, xanthenyl and the like.

**[0222]** The terms "heterocycloalkyl" and, interchangeably, "heterocycle," as used herein, alone or in combination, each refer to a saturated, partially unsaturated, or fully unsaturated monocyclic, bicyclic, or tricyclic heterocyclic group containing at least one heteroatom as a ring member, wherein each said heteroatom may be independently selected from the group consisting of nitrogen, oxygen, and sulfur. In certain embodiments, said heterocycloalkyl will comprise from 1 to 4 heteroatoms as ring members. In further embodiments, said heterocycloalkyl will comprise from 1 to 2 heteroatoms as ring members. In certain embodiments, said heterocycloalkyl will comprise from 3 to 8 ring members in each ring. In further embodiments, said heterocycloalkyl will comprise from 3 to 7 ring members in each ring. In yet further embodiments, said heterocycloalkyl will comprise from 5 to 6 ring members in each ring. "Heterocycloalkyl" and "heterocycle" are intended to include sulfones, sulfoxides, N-

oxides of tertiary nitrogen ring members, and carbocyclic fused and benzo fused ring systems; additionally, both terms also include systems where a heterocycle ring is fused to an aryl group, as defined herein, or an additional heterocycle group. Examples of heterocycle groups include aziridinyl, azetidinyl, 1,3-benzodioxolyl, dihydroisoindolyl, dihydroisoquinolinyl, dihydrocinnolinyl, dihydrobenzodioxinyl, dihydro[1,3]oxazolo[4,5-b]pyridinyl, benzothiazolyl, dihydroindolyl, dihydropyridinyl, 1,3-dioxanyl, 1,4-dioxanyl, 1,3-dioxolanyl, isoindolinyl, morpholinyl, piperazinyl, pyrrolidinyl, tetrahydropyridinyl, piperidinyl, thiomorpholinyl, and the like. The heterocycle groups may be optionally substituted unless specifically prohibited.

**[0223]** The term "lower," as used herein, alone or in a combination, where not otherwise specifically defined, means containing from 1 to and including 6 carbon atoms.

**[0224]** The term "sulfonyl," as used herein, alone or in combination, refers to  $-S(O)_2^-$ .

**[0225]** Any definition herein may be used in combination with any other definition to describe a composite structural group. By convention, the trailing element of any such definition is that which attaches to the parent moiety. For example, the composite group alkylamido would represent an alkyl group attached to the parent molecule through an amido group, and the term alkoxyalkyl would represent an alkoxy group attached to the parent molecule through an alkyl group.

**[0226]** When a group is defined to be "null," what is meant is that said group is absent.

**[0227]** The term "optionally substituted" means the anteceding group may be substituted or unsubstituted. When substituted, the substituents of an "optionally substituted" group may include, without limitation, one or more substituents independently selected from the following groups or a particular designated set of groups, alone or in combination: lower alkyl, lower alkenyl, lower alkynyl, lower alkanoyl, lower heteroalkyl, lower heterocycloalkyl, lower haloalkyl, lower haloalkenyl, lower haloalkynyl, lower perhaloalkyl, lower perhaloalkoxy, lower cycloalkyl, phenyl, aryl, aryloxy, lower alkoxy, lower haloalkoxy, oxo, lower acyloxy, carbonyl, carboxyl, lower alkylcarbonyl, lower carboxyester, lower carboxamido, cyano, hydrogen, halogen, hydroxy, amino, lower alkylamino, arylamino, amido, nitro, thiol, lower alkylthio, lower haloalkylthio, lower perhaloalkylthio, arylthio, sulfonate, sulfonic acid, trisubstituted silyl,  $N_3$ ,  $SH$ ,  $SCH_3$ ,  $C(O)CH_3$ ,  $CO_2CH_3$ ,  $CO_2H$ , pyridinyl, thiophene, furanyl, lower carbamate, and lower urea. Two substituents may be joined together to form a fused five-, six-, or seven-membered carbocyclic or heterocyclic ring consisting of zero to three heteroatoms, for example forming methylenedioxy or ethylenedioxy. An optionally substituted group may be unsubstituted (e.g.,  $-CH_2CH_3$ ), fully substituted (e.g.,  $-CF_2CF_3$ ), monosubstituted (e.g.,  $-CH_2CH_2F$ ) or substituted at a level anywhere in-between fully substituted and monosubstituted (e.g.,  $-CH_2CF_3$ ). Where substituents are recited without qualification as to substitution, both substituted and unsubstituted forms are encompassed. Where a substituent is qualified as "substituted," the substituted form is specifically intended. Additionally, different sets of optional substituents to a particular moiety may be defined as needed; in these cases, the optional substitution will be as

defined, often immediately following the phrase, "optionally substituted with."

**[0228]** The term R or the term R', appearing by itself and without a number designation, unless otherwise defined, refers to a moiety selected from the group consisting of hydrogen, alkyl, cycloalkyl, heteroalkyl, aryl, heteroaryl and heterocycloalkyl, any of which may be optionally substituted. Such R and R' groups should be understood to be optionally substituted as defined herein. Whether an R group has a number designation or not, every R group, including R, R' and R<sup>n</sup> where n=(1, 2, 3, ...n), every substituent, and every term should be understood to be independent of every other in terms of selection from a group. Should any variable, substituent, or term (e.g. aryl, heterocycle, R, etc.) occur more than one time in a formula or generic structure, its definition at each occurrence is independent of the definition at every other occurrence. Those of skill in the art will further recognize that certain groups may be attached to a parent molecule or may occupy a position in a chain of elements from either end as written. Thus, by way of example only, an unsymmetrical group such as -C(O)N(R)- may be attached to the parent moiety at either the carbon or the nitrogen.

**[0229]** Asymmetric centers exist in the compounds disclosed herein. These centers are designated by the symbols "R" or "S," depending on the configuration of substituents around the chiral carbon atom. It should be understood that the present disclosure encompasses all stereochemical isomeric forms, including diastereomeric, enantiomeric, and epimeric forms, as well as d-isomers and 1-isomers, and mixtures thereof. Individual stereoisomers of compounds can be prepared synthetically from commercially available starting materials which contain chiral centers or by preparation of mixtures of enantiomeric products followed by separation such as conversion to a mixture of diastereomers followed by separation or recrystallization, chromatographic techniques, direct separation of enantiomers on chiral chromatographic columns, or any other appropriate method known in the art. Starting compounds of particular stereochemistry are either commercially available or can be made and resolved by techniques known in the art. Additionally, the compounds disclosed herein may exist as geometric isomers. The present disclosure includes all cis, trans, syn, anti, entgegen (E), and zusammen (Z) isomers as well as the appropriate mixtures thereof. Additionally, compounds may exist as tautomers; all tautomeric isomers are provided by this disclosure. Additionally, the compounds disclosed herein can exist in unsolvated as well as solvated forms with pharmaceutically acceptable solvents such as water, ethanol, and the like. In general, the solvated forms are considered equivalent to the unsolvated forms.

**[0230]** The term "bond" refers to a covalent linkage between two atoms, or two moieties when the atoms joined by the bond are considered to be part of larger substructure. A bond may be single, double, or triple unless otherwise specified. A dashed line between two atoms in a drawing of a molecule indicates that an additional bond may be present or absent at that position.

**[0231]** A "macrophage" is a phagocytic cell, some are fixed and others circulate in the blood stream. Macrophages are regulatory and effector cell of the immune response. These cells are susceptible to infection by viruses. As used herein, the terms "macrophage" and "monocyte"

are used interchangeably, as it is understood that in the art the term "monocyte" is often used to describe a circulating mononuclear cell that expresses the CD14 cell surface marker, and when in a tissue this cell is also classified as a macrophage.

**[0232]** A "macrophage-associated condition" is a condition, disorder, or indication that is associated with an elevated or abnormal level of macrophage proliferation or activation as compared to control sample(s). Such disorders include, but are not limited to, AIDS-associated dementia, Alzheimer's disease (AD), Amyotrophic Lateral Sclerosis (ALS) AIDS lymphoma, follicular lymphoma, mycoses fungoides, T cell and B cell lymphomas with significant macrophage compartments, age related macular degeneration (ARMD), wet and dry forms, atherosclerosis, kidney disease such as focal segmental glomerulosclerosis, and membrane proliferative glomerulonephropathy, lupus, psoriaform dermatitis, AIDS-associated diarrhea, prelymphomatous autoimmune disease such as AILD (angioimmunoblastic lymphadenopathy with dysproteinemia), chronic hepatitis viral diseases (HBV and HCV), peripheral sensory neuropathy associated with HIV infection or diabetes mellitus and herpes virus associated diseases such as Castleman's disease and Kaposi's sarcoma. In one embodiment, they include invasive breast cancer and pancreatic cancer. The terms "condition," "disorder," and "disease" are used interchangeably herein. "Macrophage-associated dementia" is a dementia that is associated with an elevated, or abnormal, level of macrophage proliferation or activation as compared to control sample(s). Such dementias include, but are not limited to, AD. A macrophage-associated disorder, disease or dementia can be HIV-mediated or non-HIV-mediated, or HIV-associated or non-HIV associated. A "non-HIV-mediated" disease or dementia is a disease or dementia which is not caused by HIV, either directly or indirectly. A "non-HIV-associated" disease or dementia is not normally associated with or secondary to HIV infection. An "HIV-mediated" disease, dementia, or indication is directly or indirectly caused by (and/or linked to) HIV infection. An "HIV-associated" disease, dementia or indication is defined more broadly as generally associated with or secondary to an HIV infection; "HIV-mediated" diseases, for example, are included in those considered to be "HIV-associated." The terms "HIV neuropathy" and "HIV-associated neurodegeneration" ("HAND") may be used herein interchangeably. HIV-associated dementia may also be used interchangeably with HAD, HIV dementia, AIDS dementia, AIDS dementia complex, ADC, and neuroAIDS.

**[0233]** A "virus" is a microscopic infectious organism that reproduces inside living cells. A virus consists essentially of a core of a single nucleic acid surrounded by a protein coat, and has the ability to replicate inside a living cell. The term virus includes retroviruses, which are RNA viruses wherein the viral genome is RNA, and lentiviruses, a classification which describes a genus of viruses containing reverse transcriptase.

**[0234]** HIV is a retrovirus that causes immunosuppression in humans (HIV disease), and leads to a disease complex known as the acquired immunodeficiency syndrome (AIDS). "HIV disease" refers to a well-recognized group of signs and symptoms in persons infected by an HIV virus.

**[0235]** "Viral load" is a measure of the severity of a viral infection, and can be estimated by

calculating the amount of virus in a body fluid or in infected cells. Viral load may be employed as a surrogate marker for disease progression. Viral load is typically measured by PCR and bDNA tests and is generally expressed in number of virus copies or equivalents per milliliter. For example, "HIV viral load" may be measured by determining the level of HIV-RNA (measured in copies per ml) detectable by polymerase chain reaction (PCR) in the plasma of an HIV-infected subject.

**[0236]** A "clinically significant" reduction in HIV viral load includes a reduction greater than or equal to about 80% (a half log) relative to a baseline value. Similarly, a "clinically significant" reduction in the number of HIV-infected CD14/CD16<sup>+</sup> blood macrophages includes a reduction of at least about 80% relative to a baseline value.

**[0237]** The term "prodrug" refers to a compound that is made more active *in vivo*. Certain compounds disclosed herein may also exist as prodrugs, as described in *Hydrolysis in Drug and Prodrug Metabolism : Chemistry, Biochemistry, and Enzymology* (Testa, Bernard and Mayer, Joachim M. Wiley-VHCA, Zurich, Switzerland 2003). Prodrugs of the compounds described herein are structurally modified forms of the compound that readily undergo chemical changes under physiological conditions to provide the compound. Additionally, prodrugs can be converted to the compound by chemical or biochemical methods in an *ex vivo* environment. For example, prodrugs can be slowly converted to a compound when placed in a transdermal patch reservoir with a suitable enzyme or chemical reagent. Prodrugs are often useful because, in some situations, they may be easier to administer than the compound, or parent drug. They may, for instance, be bioavailable by oral administration whereas the parent drug is not. The prodrug may also have improved solubility in pharmaceutical compositions over the parent drug. A wide variety of prodrug derivatives are known in the art, such as those that rely on hydrolytic cleavage or oxidative activation of the prodrug. An example, without limitation, of a prodrug would be a compound which is administered as an ester (the "prodrug"), but then is metabolically hydrolyzed to the carboxylic acid, the active entity. Additional examples include peptidyl derivatives of a compound.

**[0238]** The compounds disclosed herein can exist as therapeutically acceptable salts. Described herein are compounds listed above in the form of salts, including acid addition salts. Suitable salts include those formed with both organic and inorganic acids. Such acid addition salts will normally be pharmaceutically acceptable. However, salts of non-pharmaceutically acceptable salts may be of utility in the preparation and purification of the compound in question. Basic addition salts may also be formed and be pharmaceutically acceptable. For a more complete discussion of the preparation and selection of salts, refer to *Pharmaceutical Salts: Properties, Selection, and Use* (Stahl, P. Heinrich. Wiley-VCHA, Zurich, Switzerland, 2002).

**[0239]** The term "therapeutically acceptable salt," as used herein, represents salts or zwitterionic forms of the compounds disclosed herein which are water or oil-soluble or dispersible and therapeutically acceptable as defined herein. The salts can be prepared during the final isolation and purification of the compounds or separately by reacting the appropriate

compound in the form of the free base with a suitable acid. Representative acid addition salts include acetate, adipate, alginate, L-ascorbate, aspartate, benzoate, benzenesulfonate (besylate), bisulfate, butyrate, camphorate, camphorsulfonate, citrate, digluconate, formate, fumarate, gentisate, glutarate, glycerophosphate, glycolate, hemisulfate, heptanoate, hexanoate, hippurate, hydrochloride, hydrobromide, hydroiodide, 2-hydroxyethansulfonate (isethionate), lactate, maleate, malonate, DL-mandelate, mesitylenesulfonate, methanesulfonate, naphthylenesulfonate, nicotinate, 2-naphthalenesulfonate, oxalate, pamoate, pectinate, persulfate, 3-phenylpropionate, phosphonate, picrate, pivalate, propionate, pyroglutamate, succinate, sulfonate, tartrate, L-tartrate, trichloroacetate, trifluoroacetate, phosphate, glutamate, bicarbonate, para-toluenesulfonate (p-tosylate), and undecanoate. Also, basic groups in the compounds disclosed herein can be quaternized with methyl, ethyl, propyl, and butyl chlorides, bromides, and iodides; dimethyl, diethyl, dibutyl, and diamyl sulfates; decyl, lauryl, myristyl, and steryl chlorides, bromides, and iodides; and benzyl and phenethyl bromides. Examples of acids which can be employed to form therapeutically acceptable addition salts include inorganic acids such as hydrochloric, hydrobromic, sulfuric, and phosphoric, and organic acids such as oxalic, maleic, succinic, and citric. Salts can also be formed by coordination of the compounds with an alkali metal or alkaline earth ion. Hence, the present disclosure contemplates sodium, potassium, magnesium, and calcium salts of the compounds disclosed herein, and the like.

**[0240]** Basic addition salts can be prepared during the final isolation and purification of the compounds by reacting a carboxy group with a suitable base such as the hydroxide, carbonate, or bicarbonate of a metal cation or with ammonia or an organic primary, secondary, or tertiary amine. The cations of therapeutically acceptable salts include lithium, sodium, potassium, calcium, magnesium, and aluminum, as well as nontoxic quaternary amine cations such as ammonium, tetramethylammonium, tetraethylammonium, methylamine, dimethylamine, trimethylamine, triethylamine, diethylamine, ethylamine, tributylamine, pyridine, *N,N*-dimethylaniline, *N*-methylpiperidine, *N*-methylmorpholine, dicyclohexylamine, procaine, dibenzylamine, *N,N*-dibenzylphenethylamine, 1-ephenamine, and *N,N*'-dibenzylethylenediamine. Other representative organic amines useful for the formation of base addition salts include ethylenediamine, ethanolamine, diethanolamine, piperidine, and piperazine.

**[0241]** While it may be possible for the compounds disclosed herein to be administered as the raw chemical, it is also possible to present them as a pharmaceutical formulation. Accordingly, provided herein are pharmaceutical formulations which comprise one or more of certain compounds disclosed herein, or one or more pharmaceutically acceptable salts, esters, prodrugs, amides, or solvates thereof, together with one or more pharmaceutically acceptable carriers thereof and optionally one or more other therapeutic ingredients. The carrier(s) must be "acceptable" in the sense of being compatible with the other ingredients of the formulation and not deleterious to the recipient thereof. Proper formulation is dependent upon the route of administration chosen. Any of the well-known techniques, carriers, and excipients may be used as suitable and as understood in the art; e.g., in Remington's Pharmaceutical Sciences. The pharmaceutical compositions disclosed herein may be manufactured in any manner known in

the art, e.g., by means of conventional mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping or compression processes.

**[0242]** The agent - a polyamine analog, polyamine biosynthesis inhibitor, polyamine transport inhibitor, or agent that inhibits S-adenosyl methionine decarboxylase - may also be administered in combination with one or more entities. In one embodiment, the entity is a therapeutic entity, including, but not limited to, an anti-viral or anti-retroviral agent, a steroid or other anti-inflammatory agent. In another embodiment, the entity is a pharmaceutically acceptable carrier.

**[0243]** The effective amount of an agent that inhibits S-adenosyl methionine decarboxylase, e.g., in a cell or a subject, can be any amount that is sufficient to decrease the level or activity of osteopontin, e.g., in the cell or the subject, typically by about 25%, 30%, 40%, 50%, 60%, 70%, 75%, 80%, 85%, 90%, 95% or more. In one embodiment, the effective amount of an agent is an amount that is sufficient to decrease the activity of osteopontin by 70% or more. In another embodiment, the effective amount of an agent is an amount that is sufficient to decrease the activity of osteopontin by 80% or more. In yet another embodiment, the agent inhibits S-adenosyl methionine decarboxylase and the effective amount is an amount sufficient to activate adenosine deaminase ("ADA").

**[0244]** The optimal dose, frequency of administration, and duration of treatment with the agent in a subject may vary from subject to subject, depending on the disease to be treated or clinical endpoint to be reached (for example, decrease in the level or activity of osteopontin, inhibition of infiltration of macrophages to a tissue, or mitigation of pain) the subject's condition, the subject's age, weight, response to the treatment, and the nature of the therapeutic entity. Determination of the optimal dose and duration of treatment is within the scope of one of skill in the art. The optimal dose and duration of treatment may be best determined by monitoring the subject's response during the course of the treatment. In some instances, the administration of higher doses may permit less frequent administration, and lower doses may require more frequent administration in order to achieve a clinically significant improvement in the subject's condition. The agent(s) described herein may be administered as a single dose or in multiple doses.

**[0245]** Generally, a therapeutically effective dose of the agent in accordance with the present methods will be one or more doses of from about 10 to about 1100 mg/m<sup>2</sup>. Lower dose regimens include doses of 10-200, 10-100, 10-50 and 20-200 mg/m<sup>2</sup>. Higher dose regimens include 200-400, 250-500, 400-600, 500-800 600-1000 and 800-1100 mg/m<sup>2</sup>. In one embodiment, the dose regimens range from 200-400 mg/m<sup>2</sup>. In another embodiment, the dose regimens range from 250-500 mg/m<sup>2</sup>. In yet another embodiment, the dose regimens range from 600-1 000 mg/m<sup>2</sup>. In some embodiments the agent is administered daily, once per week, once every other week, or once per month. In one embodiment, a dose regimen ranging from 200-400 mg/m<sup>2</sup> is administered once a week. In another embodiment, a dose regimen ranging

from 250-500 mg/m<sup>2</sup> is administered once every other week.

**[0246]** The doses may be constant over the entire treatment period, or they may increase or decrease during the course of the treatment. In one embodiment, the agent is administered once a week and starts with the administration of 200 mg/m<sup>2</sup>, and increases to 300 mg/m<sup>2</sup> and 400 mg/m<sup>2</sup> in the second and third weeks, respectively. In another embodiment, the agent is administered once every other week and is kept constant for the entire duration of treatment with the administration of 250 mg/m<sup>2</sup>. The doses of the agent may be administered for at least one week, at least two weeks, at least three weeks, at least four weeks, at least 6 weeks, or even at least 8 weeks. Adjusting the dose of the agent within these ranges for a particular subject is well within the skill of the ordinary clinician.

**[0247]** The agent may be administered via any conventional route normally used to administer a medicament including, but not limited to, oral, parenteral (including subcutaneous, intradermal, intramuscular, intravenous, intraarticular, and intramedullary), intraperitoneal, transmucosal (including nasal), transdermal, rectal and topical (including dermal, buccal, sublingual and intraocular) routes. Intravenous delivery may take place via a bolus injection or via infusion; infusion may be done over a period ranging from less than a minute to several hours to continuously. In certain embodiments, a course of treatment will involve administration by a combination of routes.

**[0248]** For example, the agent may be administered via a combination of intravenous and oral routes for the treatment of pain or another disorder. In one embodiment, a "loading" dose may be administered IV in order to bring the concentration of drug to the desired therapeutic level, followed by one or more maintenance doses via the oral route to keep it there. In a further embodiment, a combination of oral and IV delivery may be used to mitigate pain in a surgery patient. The agent may be delivered pre-, peri-, and post- surgically by a combination of IV and oral routes. In one embodiment, the patient may be administered or may self-administer the drug orally prior to surgery, be administered the drug via IV infusion during surgery and just after, and may thereafter be administered or may self-administer the drug orally after surgery. In another embodiment, the patient may be administered the drug IV prior to surgery, be administered the drug via IV infusion during surgery and just after, and may thereafter be administered or may self-administer the drug orally after surgery.

**[0249]** The agent may be administered as a pharmaceutical composition in a variety of forms including, but not limited to, liquid, powder, suspensions, tablets, pills, capsules, sprays and aerosols. The pharmaceutical compositions may include various pharmaceutically acceptable additives including, but not limited to, carriers, excipients, binders, stabilizers, antimicrobial agents, antioxidants, diluents and/or supports. Examples of suitable excipients and carriers are described, for example, in "Remington's Pharmaceutical Sciences," Mack Pub. Co., New Jersey (1991). In some embodiments, the agent may be administered via an IV infusion in an aqueous sugar solution. The agent may also be associated with another substance that facilitates agent delivery. For example, the agent may be associated into liposomes. The

liposomes, in turn, may be conjugated with targeting substance(s), such as IgGFc receptors.

**[0250]** Formulations of the compounds disclosed herein suitable for oral administration may be presented as discrete units such as capsules, cachets or tablets each containing a predetermined amount of the active ingredient; as a powder or granules; as a solution or a suspension in an aqueous liquid or a non-aqueous liquid; or as an oil-in-water liquid emulsion or a water-in-oil liquid emulsion. The active ingredient may also be presented as a bolus, electuary or paste.

**[0251]** Pharmaceutical preparations which can be used orally include tablets, push-fit capsules made of gelatin, as well as soft, sealed capsules made of gelatin and a plasticizer, such as glycerol or sorbitol. Tablets may be made by compression or molding, optionally with one or more accessory ingredients. Compressed tablets may be prepared by compressing in a suitable machine the active ingredient in a free-flowing form such as a powder or granules, optionally mixed with binders, inert diluents, or lubricating, surface active or dispersing agents. Molded tablets may be made by molding in a suitable machine a mixture of the powdered compound moistened with an inert liquid diluent. The tablets may optionally be coated or scored and may be formulated so as to provide slow or controlled release of the active ingredient therein. All formulations for oral administration should be in dosages suitable for such administration. The push-fit capsules can contain the active ingredients in admixture with filler such as lactose, binders such as starches, and/or lubricants such as talc or magnesium stearate and, optionally, stabilizers. In soft capsules, the active compounds may be dissolved or suspended in suitable liquids, such as fatty oils, liquid paraffin, or liquid polyethylene glycols. In addition, stabilizers may be added. Dragee cores are provided with suitable coatings. For this purpose, concentrated sugar solutions may be used, which may optionally contain gum arabic, talc, polyvinyl pyrrolidone, carbopol gel, polyethylene glycol, and/or titanium dioxide, lacquer solutions, and suitable organic solvents or solvent mixtures. Dyestuffs or pigments may be added to the tablets or dragee coatings for identification or to characterize different combinations of active compound doses.

**[0252]** The compounds may be formulated for parenteral administration by injection, e.g., by bolus injection or continuous infusion. Formulations for injection may be presented in unit dosage form, e.g., in ampoules or in multi-dose containers, with an added preservative. The compositions may take such forms as suspensions, solutions or emulsions in oily or aqueous vehicles, and may contain formulatory agents such as suspending, stabilizing and/or dispersing agents. The formulations may be presented in unit-dose or multi-dose containers, for example sealed ampoules and vials, and may be stored in powder form or in a freeze-dried (lyophilized) condition requiring only the addition of the sterile liquid carrier, for example, saline or sterile pyrogen-free water, immediately prior to use. Extemporaneous injection solutions and suspensions may be prepared from sterile powders, granules and tablets of the kind previously described.

**[0253]** Formulations for parenteral administration include aqueous and non-aqueous (oily) sterile injection solutions of the active compounds which may contain antioxidants, buffers,

bacteriostats and solutes which render the formulation isotonic with the blood of the intended recipient; and aqueous and non-aqueous sterile suspensions which may include suspending agents and thickening agents. Suitable lipophilic solvents or vehicles include fatty oils such as sesame oil, or synthetic fatty acid esters, such as ethyl oleate or triglycerides, or liposomes. Aqueous injection suspensions may contain substances which increase the viscosity of the suspension, such as sodium carboxymethyl cellulose, sorbitol, or dextran. Optionally, the suspension may also contain suitable stabilizers or agents which increase the solubility of the compounds to allow for the preparation of highly concentrated solutions.

**[0254]** In addition to the formulations described previously, the compounds may also be formulated as a depot preparation. Such long acting formulations may be administered by implantation (for example subcutaneously or intramuscularly) or by intramuscular injection. Thus, for example, the compounds may be formulated with suitable polymeric or hydrophobic materials (for example as an emulsion in an acceptable oil) or ion exchange resins, or as sparingly soluble derivatives, for example, as a sparingly soluble salt.

**[0255]** For buccal or sublingual administration, the compositions may take the form of tablets, lozenges, pastilles, or gels formulated in conventional manner. Such compositions may comprise the active ingredient in a flavored basis such as sucrose and acacia or tragacanth.

**[0256]** The compounds may also be formulated in rectal compositions such as suppositories or retention enemas, e.g., containing conventional suppository bases such as cocoa butter, polyethylene glycol, or other glycerides.

**[0257]** Certain compounds disclosed herein may be administered topically, that is by non-systemic administration. This includes the application of a compound disclosed herein externally to the epidermis or the buccal cavity and the instillation of such a compound into the ear, eye and nose, such that the compound does not significantly enter the blood stream. In contrast, systemic administration refers to oral, intravenous, intraperitoneal and intramuscular administration.

**[0258]** Formulations suitable for topical administration include liquid or semiliquid preparations suitable for penetration through the skin to the site of inflammation such as gels, liniments, lotions, creams, ointments or pastes, and drops suitable for administration to the eye, ear or nose. The active ingredient for topical administration may comprise, for example, from 0.001 % to 10% w/w (by weight) of the formulation. In certain embodiments, the active ingredient may comprise as much as 10% w/w. In other embodiments, it may comprise less than 5% w/w. In certain embodiments, the active ingredient may comprise from 2% w/w to 5% w/w. In other embodiments, it may comprise from 0.1% to 1% w/w of the formulation.

**[0259]** For administration by inhalation, compounds may be conveniently delivered from an insufflator, nebulizer pressurized packs or other convenient means of delivering an aerosol spray. Pressurized packs may comprise a suitable propellant such as dichlorodifluoromethane, trichlorofluoromethane, dichlorotetrafluoroethane, carbon dioxide or other suitable gas. In the

case of a pressurized aerosol, the dosage unit may be determined by providing a valve to deliver a metered amount. Alternatively, for administration by inhalation or insufflation, the compounds described herein may take the form of a dry powder composition, for example a powder mix of the compound and a suitable powder base such as lactose or starch. The powder composition may be presented in unit dosage form, in for example, capsules, cartridges, gelatin or blister packs from which the powder may be administered with the aid of an inhalator or insufflator.

**[0260]** Exemplary unit dosage formulations are those containing an effective dose, as herein below recited, or an appropriate fraction thereof, of the active ingredient.

**[0261]** Fillers to be used in the compositions herein include all those now known and in use, as well as those developed in the future. Examples of fillers, or diluents, include, without limitation, lactose, mannitol, xylitol, dextrose, sucrose, sorbitol, compressible sugar, microcrystalline cellulose (MCC), powdered cellulose, cornstarch, pregelatinized starch, dextrates, dextran, dextrin, dextrose, maltodextrin, calcium carbonate, dibasic calcium phosphate, tribasic calcium phosphate, calcium sulfate, magnesium carbonate, magnesium oxide, poloxamers such as polyethylene oxide, and hydroxypropyl methyl cellulose. Fillers may have complexed solvent molecules, such as in the case where the lactose used is lactose monohydrate. Fillers may also be proprietary, such in the case of the filler PROSOLV® (available from JRS Pharma). PROSOLV is a proprietary, optionally high-density, silicified microcrystalline cellulose composed of 98% microcrystalline cellulose and 2% colloidal silicon dioxide. Silicification of the microcrystalline cellulose is achieved by a patented process, resulting in an intimate association between the colloidal silicon dioxide and microcrystalline cellulose. ProSolv comes in different grades based on particle size, and is a white or almost white, fine or granular powder, practically insoluble in water, acetone, ethanol, toluene and dilute acids and in a 50g/l solution of sodium hydroxide.

**[0262]** Disintegrants to be used in the compositions herein include all those now known and in use, as well as those developed in the future. Examples of disintegrants include, without limitation, sodium starch glycolate, sodium carboxymethyl cellulose, calcium carboxymethyl cellulose, croscarmellose sodium, povidone, crospovidone (polyvinylpolypyrrolidone), methyl cellulose, microcrystalline cellulose, powdered cellulose, low-substituted hydroxy propyl cellulose, starch, pregelatinized starch, and sodium alginate.

**[0263]** Lubricants to be used in the compositions herein include all those now known and in use, as well as those developed in the future. Examples of lubricants include, without limitation, calcium stearate, glyceryl monostearate, glyceryl palmitostearate, hydrogenated vegetable oil, light mineral oil, magnesium stearate, mineral oil, polyethylene glycol, sodium benzoate, sodium lauryl sulfate, sodium stearyl fumarate, stearic acid, talc, and zinc stearate.

**[0264]** Glidants to be used in the compositions herein include all those now known and in use, as well as those developed in the future. Examples of glidants include, without limitation, silicon dioxide (SiO<sub>2</sub>), talc cornstarch, and poloxamers. Poloxamers (or LUTROL®, available from the

BASF Corporation) are A-B-A block copolymers in which the A segment is a hydrophilic polyethylene glycol homopolymer and the B segment is hydrophobic polypropylene glycol homopolymer.

**[0265]** Tablet binders to be used in the compositions herein include all those now known and in use, as well as those developed in the future. Examples of tablet binders include, without limitation, acacia, alginic acid, carbomer, carboxymethyl cellulose sodium, dextrin, ethylcellulose, gelatin, guar gum, hydrogenated vegetable oil, hydroxyethylcellulose, hydroxypropyl cellulose, hydroxypropylmethyl cellulose, copolyvidone, methyl cellulose, liquid glucose, maltodextrin, polymethacrylates, povidone, pregelatinized starch, sodium alginate, starch, sucrose, tragacanth, and zein.

**[0266]** Examples of surfactants include, without limitation, fatty acid and alkyl sulfonates; commercial surfactants such as benzethonium chloride (HYAMINE® 1622, available from Lonza, Inc., Fairlawn, N.J.); DOCUSATE SODIUM® (available from Mallinckrodt Spec. Chem., St. Louis, MO); polyoxyethylene sorbitan fatty acid esters (TWEEN®, available from ICI Americas Inc., Wilmington, DE; LIPOSORB® P-20, available from Lipochem Inc., Patterson NJ; CAPMUL® POE-0, available from Abitec Corp., Janesville, WI), polyoxyethylene (20) sorbitan monooleate (TWEEN 80®, available from ICI Americas Inc., Wilmington, DE); and natural surfactants such as sodium taurocholic acid, 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine, lecithin, and other phospholipids and mono- and diglycerides. Such materials can advantageously be employed to increase the rate of dissolution by facilitating wetting, thereby increasing the maximum dissolved concentration, and also to inhibit crystallization or precipitation of drug by interacting with the dissolved drug by mechanisms such as complexation, formation of inclusion complexes, formation of micelles or adsorbing to the surface of solid drug

**[0267]** Drug complexing agents and solubilizers to be used in the compositions herein include all those now known and in use, as well as those developed in the future. Examples of drug complexing agents or solubilizers include, without limitation, the polyethylene glycols, caffeine, xanthene, gentisic acid and cyclodextrins.

**[0268]** The addition of pH modifiers such as acids, bases, or buffers may also be beneficial, retarding or enhancing the rate of dissolution of the composition, or, alternatively, helping to improve the chemical stability of the composition. Suitable pH modifiers to be used in the compositions herein include all those now known and in use, as well as those developed in the future.

**[0269]** It should be understood that in addition to the ingredients particularly mentioned above, the formulations provided herein may include other agents conventional in the art having regard to the type of formulation in question. Proper formulation is dependent upon the route of administration chosen. Any of the well-known techniques, carriers, and excipients may be used as suitable and as understood in the art; e.g., Remington, *supra*. The pharmaceutical compositions may be manufactured in a manner that is itself known, e.g., by means of

conventional mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping or compression processes.

**[0270]** Compounds may be administered orally or via injection at a dose of from 0.1 to 500 mg/kg per day. The dose range for adult humans is generally from 5 mg to 2 g/day. Tablets or other forms of presentation provided in discrete units may conveniently contain an amount of one or more compounds which is effective at such dosage or as a multiple of the same, for instance, units containing 5 mg to 500 mg, usually around 10 mg to 200 mg.

**[0271]** The precise amount of compound administered to a subject will be the responsibility of the attendant physician. The specific dose level for any particular subject will depend upon a variety of factors including the activity of the specific compound employed, the age, body weight, general health, sex, diets, time of administration, route of administration, rate of excretion, drug combination, the precise disorder being treated, and the severity of the indication or condition being treated. Also, the route of administration may vary depending on the condition and its severity. Dosing frequency may also be selected or adjusted based on factors including those above as well as the formulation of the compound delivered. Dosing may occur, for example: once daily, twice daily, three or four times daily, every other day, weekly, bi-weekly, or monthly; or in cycles comprising a sustained dosing period followed by a non-dosing period; or on an as-needed basis.

**[0272]** In certain instances, it may be appropriate to administer at least one of the compounds described herein (or a pharmaceutically acceptable salt, ester, or prodrug thereof) in combination with another therapeutic agent. By way of example only, if one of the side effects experienced by a subject upon receiving one of the compounds herein is hypertension, then it may be appropriate to administer an antihypertensive agent in combination with the initial therapeutic agent. Or, by way of example only, the therapeutic effectiveness of one of the compounds described herein may be enhanced by administration of an adjuvant (i.e., by itself the adjuvant may only have minimal therapeutic benefit, but in combination with another therapeutic agent, the overall therapeutic benefit to the subject is enhanced). Or, by way of example only, the benefit experienced by a subject may be increased by administering one of the compounds described herein with another therapeutic agent (which also includes a therapeutic regimen) that also has therapeutic benefit. By way of example only, in a treatment for neuropathy involving administration of one of the compounds described herein, increased therapeutic benefit may result by also providing the subject with another therapeutic agent for neuropathy. In any case, regardless of the disease, disorder or condition being treated, the overall benefit experienced by the subject may simply be additive of the two therapeutic agents or the subject may experience a synergistic benefit.

**[0273]** In certain embodiments, the other therapeutic agent is an antiviral agent. In one embodiment, the antiviral agent is an antiretroviral agent, e.g. nucleoside reverse transcriptase inhibitors, nucleotide reverse transcriptase inhibitors, nonnucleoside reverse transcriptase inhibitors, protease inhibitors, entry inhibitors, integrase inhibitors or gp41, CXCR4, or gp120 inhibitors. Examples of nucleoside reverse transcriptase inhibitors for the treatment of HIV

infections include amdoxovir, elvucitabine, alovudine, racivir ( $\pm$ -FTC), phosphazide, fozivudine tidoxil, apricitabine (AVX754), amdoxovir, zidovudine (AZT), didanosine (ddI), lamivudine (3TC), stavudine (d4T), zalcitabine (ddC), emtricitabine (FTC), and abacavir (ABC). Examples of nucleotide reverse transcriptase inhibitors include tenofovir (TDF) and adefovir. Examples of non-nucleoside reverse transcriptase inhibitors include capravirine, emivirine, calanolide A, etravirine, efavirenz (EFV), nevirapine (NVP) and delavirdine (DLV). Examples of protease inhibitors include amprenavir (APV), tipranavir (TPV), lopinavir (LPV), fosamprenavir (FPV), atazanavir (ATV), darunavir, brecanavir, mozenavir, indinavir (IDV), nelfinavir (NFV), ritonavir (RTV), and saquinavir (SQV). Examples of entry inhibitors include SPOIA. Examples of a HIV integrase inhibitor include curcumin, derivatives of curcumin, chicoric acid, derivatives of chicoric acid, 3,5-dicaffeoylquinic acid, derivatives of 3,5dicaffeoylquinic acid, aurintricarboxylic acid, derivatives of aurintricarboxylic acid, caffeic acid phenethyl ester, derivatives of caffeic acid phenethyl ester, tyrphostin, derivatives of tyrphostin, quercetin, derivatives of quercetin, S-1360, zintevir (AR-177), L-870812, and L-25 870810, MK-0518, BMS-538158, GSK364735C, Examples of a gp41 inhibitor include enfuvirtide (ENF). Examples of a CXCR4 inhibitor include AMD-070, Examples of a gp120 inhibitor include BMS-488043.

**[0274]** In another embodiment, the polyamine analog is administered concurrently with a highly active antiretroviral therapy (HAART), *i.e.*, a combination of a protease inhibitor, a non-nucleoside reverse transcriptase inhibitor and a nucleoside reverse transcriptase inhibitor, or a combination of two non-nucleoside reverse transcriptase inhibitors and a nucleoside reverse transcriptase inhibitor. In general, the polyamine analog may be administered simultaneously or sequentially (*i.e.*, before or after) with the administration of antiviral or antiretroviral agents. Administration of the antiviral and antiretroviral agents to subjects in need thereof can be made in accordance with regimens and dosages well known in the art.

**[0275]** In yet other embodiments, the antiviral agent is an agent that is capable of reducing the immunodeficiency viral load in T-cells. T-cells, particularly CD4+ T-cells, also serve as a viral reservoir for immunodeficiency viruses such as HIV. Thus, combination treatments of polyamine analogs with agents that reduce the immunodeficiency viral load in T-cells are particularly desirable for flushing or destroying viral reservoirs of immunodeficiency virus. Suitable agents that reduce the immunodeficiency viral load in T-cells are reviewed in Pierson et al. (Annu. Rev. Immunol. (2000), 18:665-708) and include, without limitation, T-cell activating cytokines, anti-CD3 antibodies, and anti-CD45RO-toxin conjugates. For example, T-cell activating cytokine such as IL-2, IL-6, TNF- $\alpha$ , and any two or more combinations thereof may be used in the present methods.

**[0276]** In other embodiments, the other therapeutic agent is a TNF inhibitor. The TNF inhibitor may be: a monoclonal antibody such as, for example, infliximab (Remicade), adalimumab (Humira), certolizumab pegol (Cimzia), or golimumab (Simponi); a circulating receptor fusion protein such as etanercept (Enbrel); or a small molecule, such as pentoxifylline or bupropion (Zyban, Wellbutrin).

**[0277]** In other embodiments, the other therapeutic agent is a disease-modifying anti-

rheumatic drug (DMARD). Examples of DMARDs include azathioprine, cyclosporin (cyclosporine A), D-penicillamine, gold salts, hydroxychloroquine, leflunomide, methotrexate (MTX), minocycline, sulfasalazine (SSZ), and cyclophosphamide.

**[0278]** In further embodiments, the other therapeutic agent is methotrexate.

**[0279]** Other agents for used in combination include interleukin 1 (IL-1) blockers such as anakinra (Kineret), T-cell costimulation blockers such as abatacept (Orencia), interleukin 6 (IL-6) blockers such as tocilizumab (an anti-IL-6 receptor antibody; RoActemra, Actemra), monoclonal antibodies against B cells such as rituximab (Rituxan), and other biologics (eg. Ocrelizumab, Ofatumumab, Golimumab, and Certolizumab pegol).

**[0280]** In other embodiments, the other therapeutic agent is a glucocorticoid or a non-steroidal anti-inflammatory drug (NSAID). NSAIDS include propionic acid derivatives such as ibuprofen, naproxen, fenoprofen, ketoprofen, flurbiprofen, and oxaprozin; acetic acid derivatives such as indomethacin, sulindac, etodolac, and diclofenac; enolic acid (oxicam) derivatives such as piroxicam and meloxicam; fenamic acid derivatives such as mefenamic acid and meclofenamic acid; selective COX-2 inhibitors (Coxibs) such as celecoxib (Celebrex), rofecoxib, valdecoxib, parecoxib, lumiracoxib, and etoricoxib.

**[0281]** In any case, the multiple therapeutic agents (at least one of which is a compound disclosed herein) may be administered in any order or even simultaneously. If simultaneously, the multiple therapeutic agents may be provided in a single, unified form, or in multiple forms (by way of example only, either as a single pill or as two separate pills). One of the therapeutic agents may be given in multiple doses, or both may be given as multiple doses. If not simultaneous, the timing between the doses of the multiple therapeutic agents may be any duration of time ranging from a few minutes to four weeks.

**[0282]** Thus, in another aspect, certain embodiments provide methods for treating disorders in a human or animal subject in need of such treatment comprising administering to said subject an amount of a compound disclosed herein effective to reduce or prevent said disorder in the subject, optionally in combination with at least one additional agent for the treatment of said disorder that is known in the art. Specific diseases to be treated by the compounds, compositions, and methods disclosed herein, singly or in combination, include, without limitation: pain; neuropathy; inflammation and related disorders; arthritis; metabolic inflammatory disorders; respiratory disorders; autoimmune disorders; neurological disorders; and proliferative disorders, including cancer and non-cancerous diseases.

**[0283]** The compounds disclosed herein are useful to treat patients with pain, including neuropathy and/or neuropathic pain, and inflammatory pain. Pain indications include, but are not limited to, treatment or prophylaxis of surgical or post-surgical pain for various surgical procedures including amputation, post-cardiac surgery, dental pain/dental extraction, pain resulting from cancer, muscular pain, mastalgia, pain resulting from dermal injuries, lower back pain, headaches of various etiologies, including migraine, menstrual cramps, and the like. The

compounds are also useful for the treatment of pain-related disorders such as tactile allodynia and hyperalgesia. The pain may be somatogenic (either nociceptive or neuropathic), acute and/or chronic.

Peripheral neuropathies which can be treated with the compounds disclosed herein include mono-neuropathies, mono-multiplex neuropathies, and poly-neuropathies, including axonal and demyelinating neuropathies. Both sensory and motor neuropathies are encompassed. The neuropathy or neuropathic pain may be associated with a number of peripheral neuropathies of varying etiologies, including but not limited to:

- trauma-induced neuropathies, including those caused by physical injury (such as blunt trauma, abrasion, or burns) or disease state, physical damage to the brain, physical damage to the spinal cord, or stroke associated with brain damage; neurological disorders related to neurodegeneration; and post-surgical neuropathies and neuropathic pain (such as from tumor resection, mastectomy, and the like)
- infectious and viral neuropathies, including those caused by leprosy, Lyme disease, a herpes virus (and more particularly by a herpes zoster virus, which may lead to post-herpetic neuralgia), human immunodeficiency virus (HIV, which may lead to HIV neuropathy), or a papilloma virus, or any other pathogen-induced nerve damage;
- toxin-induced neuropathies (including but not limited to neuropathies induced by alcoholism, vitamin B6 intoxication, hexacarbon intoxication, amiodarone, chloramphenicol, disulfiram, isoniazide, gold, lithium, metronidazole, misonidazole, nitrofurantoin);
- drug-induced neuropathies, including therapeutic-drug-induced neuropathy, particularly a) chemotherapy-induced neuropathies caused by anti-cancer agents such as taxol, taxotere, cisplatin, nocodazole, vincristine, vindesine and vinblastine, and b) anti-viral neuropathies caused by anti-viral agents such as ddl, DDC, d4T, foscarnet, dapsone, metronidazole, and isoniazid);
- vitamin-deficiency-induced neuropathies including those resulting from vitamin B12 deficiency, vitamin B6 deficiency, and vitamin E deficiency);
- hereditary neuropathy (including but not limited to Friedreich ataxia, familial amyloid polyneuropathy, Tangier disease, Fabry disease);
- diabetic neuropathy and neuropathy caused by metabolic disorders such as renal insufficiency and hypothyroidism;
- neuropathy secondary to tumor infiltration,
- auto-immune neuropathies, including those resulting from Guillain-Barre syndrome, chronic inflammatory de-myelinating polyneuropathy, monoclonal gammopathy of undetermined significance and polyneuropathy, and multiple sclerosis;
- other neuropathies and neuropathic pain syndromes including inflammation-induced nerve damage, neurodegeneration, post-traumatic neuralgia, central neuropathic pain syndromes such as phantom limb pain, pain, complex regional pain syndromes (including but not limited to reflex sympathetic dystrophy, causalgia), neoplasia-associated pain, vasculitic/angiopathic neuropathy, and sciatica; and
- idiopathic neuropathies,

**[0284]** In certain embodiments, neuropathic pain may alternatively be manifested as allodynia, hyperalgesic pain, thermal hyperalgesia, or phantom pain. In another embodiment, neuropathy may instead lead to loss of pain sensitivity. Additional sub-categories of neuropathic pain are discussed in Dworkin, Clin J Pain (2002) vol. 18(6) pp. 343-9.

**[0285]** Furthermore, the compounds disclosed herein can be used in the treatment or prevention of opiate tolerance in patients needing protracted opiate analgesics, and benzodiazepine tolerance in patients taking benzodiazepines, and other addictive behavior, for example, nicotine addiction, alcoholism, and eating disorders. Moreover, the compounds disclosed herein are useful in the treatment or prevention of drug withdrawal symptoms, for example treatment or prevention of symptoms of withdrawal from opiate, alcohol, or tobacco addiction.

**[0286]** The compounds disclosed herein are useful in therapeutic methods to treat or prevent respiratory disease or conditions, including therapeutic methods of use in medicine for preventing and treating a respiratory disease or condition including: asthmatic conditions including allergen-induced asthma, exercise-induced asthma, pollution-induced asthma, cold-induced asthma, and viral-induced-asthma; chronic obstructive pulmonary diseases including chronic bronchitis with normal airflow, chronic bronchitis with airway obstruction (chronic obstructive bronchitis), emphysema, asthmatic bronchitis, and bullous disease; and other pulmonary diseases involving inflammation including bronchioectasis cystic fibrosis, hypersensitivity pneumonitis, farmer's lung, acute respiratory distress syndrome, pneumonia, aspiration or inhalation injury, fat embolism in the lung, acidosis inflammation of the lung, acute pulmonary edema, acute mountain sickness, acute pulmonary hypertension, persistent pulmonary hypertension of the newborn, perinatal aspiration syndrome, hyaline membrane disease, acute pulmonary thromboembolism, heparin-protamine reactions, sepsis, status asthmaticus, hypoxia, hyperoxic lung injuries, and injury induced by inhalation of certain injurious agents including cigarette smoking, leading up to complications thereof such as lung carcinoma.

**[0287]** Other disorders or conditions which can be advantageously treated by the compounds disclosed herein include inflammation and inflammatory conditions. Inflammatory conditions include, without limitation: arthritis, including sub-types and related conditions such as rheumatoid arthritis, spondyloarthropathies, gouty arthritis, osteoarthritis, systemic lupus erythematosus, juvenile arthritis, acute rheumatic arthritis, enteropathic arthritis, neuropathic arthritis, psoriatic arthritis, and pyogenic arthritis; osteoporosis, tendonitis, bursitis, and other related bone and joint disorders; gastrointestinal conditions such as reflux esophagitis, diarrhea, inflammatory bowel disease, Crohn's disease, gastritis, irritable bowel syndrome, ulcerative colitis, acute and chronic inflammation of the pancreas; pulmonary inflammation, such as that associated with viral infections and cystic fibrosis; skin-related conditions such as psoriasis, eczema, burns, sunburn, dermatitis (such as contact dermatitis, atopic dermatitis, and allergic dermatitis), and hives; pancreatitis, hepatitis, pruritis and vitiligo. In addition, compounds described herein are also useful in organ transplant patients either alone or in

combination with conventional immunomodulators.

**[0288]** Autoimmune disorders may be ameliorated by the treatment with compounds disclosed herein. Autoimmune disorders include Crohns disease, ulcerative colitis, dermatitis, dermatomyositis, diabetes mellitus type 1, Goodpasture's syndrome, Graves' disease, Guillain-Barre syndrome (GBS), autoimmune encephalomyelitis, Hashimoto's disease, idiopathic thrombocytopenic purpura, lupus erythematosus, mixed connective tissue disease, multiple sclerosis (MS), myasthenia gravis, narcolepsy, pemphigus vulgaris, pernicious anemia, psoriasis, psoriatic arthritis, polymyositis, primary biliary cirrhosis, rheumatoid arthritis, Sjögren's syndrome, scleroderma, temporal arteritis (also known as "giant cell arteritis"), vasculitis, and Wegener's granulomatosis. The compounds disclosed herein may regulate TH-17 (T-helper cells producing interleukin 17) cells or IL-17 levels.

**[0289]** In addition, the compounds disclosed herein can be used to treat metabolic disorders that are typically associated with an exaggerated inflammatory signaling, such as insulin resistance, diabetes (type I or type II), metabolic syndrome, nonalcoholic steatohepatitis, atherosclerosis, cardiovascular disease, congestive heart failure, myocarditis, atherosclerosis, and aortic aneurysm..

**[0290]** The compounds disclosed herein are also useful in treating organ and tissue injury injury associated with severe burns, sepsis, trauma, wounds, and hemorrhage- or resuscitation-induced hypotension, and also in such diseases as vascular diseases, migraine headaches, periarteritis nodosa, thyroiditis, aplastic anemia, Hodgkin's disease, sclerodoma, rheumatic fever, type I diabetes, neuromuscular junction disease including myasthenia gravis, white matter disease including multiple sclerosis, sarcoidosis, nephritis, nephrotic syndrome, Behcet's syndrome, polymyositis, gingivitis, periodontitis, swelling occurring after injury, ischemias including myocardial ischemia, cardiovascular ischemia, and ischemia secondary to cardiac arrest, and the like.

**[0291]** The compounds described herein are also useful for the treatment of certain diseases and disorders of the nervous system. Central nervous system disorders in which nitric oxide inhibition is useful include cortical dementias including Alzheimer's disease, central nervous system damage resulting from stroke, ischemias including cerebral ischemia (both focal ischemia, thrombotic stroke and global ischemia (for example, secondary to cardiac arrest), and trauma. Neurodegenerative disorders in which nitric oxide inhibition is useful include nerve degeneration or nerve necrosis in disorders such as hypoxia, hypoglycemia, epilepsy, and in cases of central nervous system (CNS) trauma (such as spinal cord and head injury), hyperbaric oxygen-induced convulsions and toxicity, dementia e.g. pre-senile dementia, and AIDS-related dementia, cachexia, Sydenham's chorea, Huntington's disease, Parkinson's Disease, amyotrophic lateral sclerosis (ALS), Korsakoff's disease, cognitive disorders relating to a cerebral vessel disorder, hypersensitivity, sleeping disorders, schizophrenia, depression, depression or other symptoms associated with Premenstrual Syndrome (PMS), and anxiety.

**[0292]** Still other disorders or conditions advantageously treated by the compounds described

herein include the prevention or treatment of (hyper)proliferative diseases, especially cancers, either alone or in combination of standards of care especially those agents that target tumor growth by re-instating the aberrant apoptotic machinery in the malignant cells. Hematological and non-hematological malignancies which may be treated or prevented include but are not limited to multiple myeloma, acute and chronic leukemias including Acute Lymphocytic Leukemia (ALL), Chronic Lymphocytic Leukemia (CLL), and Chronic Myelogenous Leukemia(CLL), lymphomas, including Hodgkin's lymphoma and non-Hodgkin's lymphoma (low, intermediate, and high grade), as well as solid tumors and malignancies of the brain, head and neck, breast, lung, reproductive tract, upper digestive tract, pancreas, liver, renal, bladder, prostate and colorectal. The present compounds and methods can also be used to treat the fibrosis, such as that which occurs with radiation therapy. The present compounds and methods can be used to treat subjects having adenomatous polyps, including those with familial adenomatous polyposis (FAP). Additionally, the present compounds and methods can be used to prevent polyps from forming in patients at risk of FAP. Non-cancerous proliferative disorders additionally include psoriasis, eczema, and dermatitis.

**[0293]** Compounds disclosed herein may also be used in the treatment of polycystic kidney disease, as well as other diseases of renal dysfunction.

**[0294]** The compounds described herein can be used in the treatment of ophthalmic diseases, such as glaucoma, retinal ganglion degeneration, ocular ischemia, corneal neovascularization, optic neuritis, retinitis, retinopathies such as glaucomatous retinopathy and/or diabetic retinopathy, uveitis, ocular photophobia, dry eye, Sjogren's syndrome, seasonal and chronic allergic conjunctivitis, and of inflammation and pain associated with chronic ocular disorders and acute injury to the eye tissue. The compounds can also be used to treat post-operative inflammation or pain as from ophthalmic surgery such as cataract surgery and refractive surgery.

**[0295]** The present compounds may also be used in co-therapies, partially or completely, in place of other conventional anti-inflammatory therapies, such as together with steroids, NSAIDs, COX-2 selective inhibitors, 5-lipoxygenase inhibitors, LTB<sub>4</sub> antagonists and LTA<sub>4</sub> hydrolase inhibitors. The compounds described herein may also be used to prevent tissue damage when therapeutically combined with antibacterial or antiviral agents.

**[0296]** It has been shown that osteopontin ("OPN") can be regulated by regulating AMD-I, polyamine biosynthesis, adenosine or a pathway containing either AMD-I or adenosine. Osteopontin, also known as secreted phosphoprotein 1 ("SPP1 "), early T-lymphocyte activation marker ("Eta-I"), sialoprotein I or 44K BPP (bone phosphoprotein), is a glycosylated phosphoprotein found in plasma, other bodily fluids, and extracellular matrices. The protein is composed of approximately 300 amino acids residues and has about 30 carbohydrate residues, including 10 sialic acid residues, attached to it. OPN is an acidic protein which exhibits a high amino acid homology between species (e.g., mouse, rat, human and pig) with several conserved elements including a stretch of 7 to 9 Asp or Glu residues.

**[0297]** Osteopontin is biosynthesized by a variety of tissue types including preosteoblasts, osteoblasts, osteocytes, extraosseous cells in the inner ear, brain, kidney, deciduum, placenta, odontoblasts, some bone marrow cells, hypertrophic chondrocytes, macrophages, smooth muscle, and endothelial cells. In the bone, the protein is primarily made by cells of the osteoblastic lineage and deposited on mineralized matrix. It is abundant in bone mineral matrix and accelerates bone regeneration and remodeling. Osteopontin is a multifunctional protein with an ability to bind several proteins, including integrin proteins and variants of the protein CD44.

**[0298]** Osteopontin is associated with, and plays a role in, the regulation and progression of many diseases. OPN is known to be increased in a number of autoimmune disorders and is overexpressed in a variety of cancers. Plasma levels of OPN are also elevated in individuals with coronary artery disease and elevated levels of OPN are found in the synovial fluid of individuals with rheumatoid arthritis. Modulation of OPN may, therefore, confer significant therapeutic benefits to subjects having these conditions. Accordingly, disclosed herein is a method for modulating OPN levels in a subject, comprising administering to the subject an amount of a compound described herein, e.g., MGBG, sufficient to modulate OPN. Such modulation may be of total body or plasma OPN, or it may be of OPN in a target organ or tissue. In certain embodiments, said modulation is downwards, i.e., leading to reduced levels of OPN or reduced activity of OPN.

**[0299]** Additionally, MGBG has been examined in HIV and related chronic and progressive conditions such as HIV neuropathy and HIV dementia. In the case of HIV and related disorders and complications, polyamine analogs or polyamine biosynthesis inhibitors such as MGBG may work via an additional mechanism. HIV is an RNA retrovirus, that upon successful infection of a host cell, reverse transcribes its genomic RNA into DNA, which then, in a double stranded form, integrates into susceptible host cells. The major targets for infection *in vivo* are the CD4 expressing T cells and macrophages. Whereas T cells, upon activation of the HIV DNA into an infectious RNA form, generally get killed, the virus-expressing macrophages persist after infection and likely serve as the long term HIV DNA reservoir *in vivo*.

**[0300]** Several studies have confirmed the long lived nature of HIV infected macrophages *in vivo*, and the consequences for subjects with AIDS-related neurological conditions. At least one study on the HIV reservoir has provided half life estimates of 4 years for infected blood macrophages and less than 2 years for infected T cells. Both values help explain the reason for the failure of highly active antiretroviral therapy (HAART) to clear the virus *in vivo*. Additionally, studies on the HIV DNA sequence *in vivo* showed that in HIV plasma viral load negative subjects on HAART HIV replication continued to occur *in vivo* within macrophages but not T cells. Therefore, the longest lived reservoir of HIV *in vivo* is the macrophage. Furthermore, one study showed that the ancestral form of HIV *in vivo* in a subject who died of AIDS related dementia resided within macrophages in the outer membrane covering of the brain (meningeal layer). Viral sequences present in this long lived reservoir gave rise to all of the sequences residing in other portions of the brain as well as the peripherally located seminal vesicles and lymph nodes. Yet another study has suggested a mechanism for the long lived

nature of HIV infected macrophages. This study mapped HIV DNA insertion sites within macrophages in tissues from subjects with late stage AIDS. All of the insertion sites were within genes near activation genetic loci that, if activated through an HIV insertional process, would keep the infected macrophages in a persistently activated and essentially immortal state.

**[0301]** Ideally, drugs would target only HIV-infected macrophages and spare normal macrophages. In fact, it has been shown that in AIDS dementia subjects, infected macrophages express CD 14 as well as CD 16 and elevated levels of the activation marker, HLA-DR, as well as the proliferation marker, proliferating cell nuclear antigen (PCNA). Polyamine analogs and polyamine biosynthesis inhibitors, such as MGBG, can be used to decrease the proviral load in a subject by destroying macrophages that act as proviral reservoirs.

**[0302]** Exemplary embodiments of the present methods are provided in the following examples. The following examples are presented to illustrate the present invention and to assist one of ordinary skill in using the same, and are not to be construed as limiting the scope of the invention.

### **MGBG Oral Activity Assays**

**[0303]** The following standard abbreviations are used to represent the associated pharmacokinetic parameters.

AUC

Area under the curve up to the last measurable concentration plus the AUC extrapolated from the last measurable concentration ( $C_{last}$  at  $t_{last}$ ) to infinity:  $AUC_{INFobs} = AUC_{0-t_{last}} + C_{last}/\lambda_z$  (where  $\lambda_z$  is the first order rate constant associated with the terminal (log-linear) portion of the curve)

$AUC_{0-12}$

Area under the curve between the time of dose and the 12 h time point

$AUC_{0-24}$

Area under the curve between the time of dose and the 24 h time point

F

Fraction available (bioavailability):

$$F = [AUC_{oral}] \cdot dose_{iv} / [AUC_{iv}] \cdot dose_{oral}$$

$Cl_{obs}$

Observed clearance

$V_{ss,obs}$

Steady state volume of distribution

$V_d$

Volume of distribution (often used with oral)

$Cl/F_{obs}$

Apparent total body clearance as a function of bioavailability

$t_{1/2}$

Terminal half-life ( $HL_{\lambda Z}$ )

$C_{max}$

The maximum observed concentration

$T_{max}$

The time at which  $C_{max}$  occurred

### **Rhesus Macaque Single-Dose**

**[0304]** Two groups of three male rhesus monkeys were fasted overnight before being administered the test article, MGBG, as either a single bolus intravenous dose of 1 mg/kg (Group 1) or as a single oral gavage dose of 10 mg/kg (Group 2). Dose formulation analysis verified administered dose solutions as within 14% of targeted concentrations of 1 and 10 mg/kg for Groups 1 and 2, respectively.

**[0305]** Blood samples were collected into tubes containing lithium heparin from the femoral vein/artery (approximately 1.0 mL) for plasma MGBG concentration measurement from all intravenously dosed animals prior to dosing and at approximately  $T=0.083$  (5 min), 0.25 (15 min), 0.5 (30 min), 1, 2, 4, 8, and 24 hours after dosing. Blood samples for plasma MGBG concentration measurement were collected from all orally dosed animals prior to dosing at approximately  $T=1$ , 2, 4, 8, 12, 24, and 36 hours after dosing. Food was also withheld through the first four hours of blood sample collection.

**[0306]** The samples were centrifuged under refrigerated conditions following completion of sample collection at each interval. The resulting plasma was separated and stored frozen at approximately -70°C until analysis.

**[0307]** PK analysis was performed on the individual plasma concentration-time profiles for MGBG using the WinNonlin non-compartmental approach (linear trapezoidal rule for AUC calculations). Nominal dose values and sampling times were used for calculations. All MGBG plasma concentration measurements reported as BQL (< 2.51 ng/mL) were set equal to zero for the purpose of analysis. Following IV and PO administration of MGBG, plasma PK disposition parameters were calculated using the WinNonlin default selection criteria for the selection of the Lambda Z.

**[0308]** Evidence of systemic plasma MGBG exposure was observed at all collected plasma time points following IV and PO administration of MGBG. Hemolysis was noted in one animal in Group 1 at a single time point, which may have negatively impacted the MGBG plasma concentration analysis for this animal. Consequently, a model-dependent two-compartmental

analysis was used to calculate bioavailability.

#### Dog Single-Dose

**[0309]** Two groups of three male beagle dogs weighing 9.0-10.7 kg and aged 8-30 months were fasted overnight before being administered the test article, MGBG, as either a single bolus intravenous dose of 1 mg/kg (Group 1) or as a single oral gavage dose of 10 mg/kg (Group 2). Dose formulation analysis verified administered dose solutions as within 17% of targeted concentrations of 1 and 10 mg/kg for Groups 1 and 2, respectively.

**[0310]** Blood samples (approximately 2.0 mL) were collected for plasma MGBG concentration measurement from all intravenously dosed animals prior to dosing and at approximately T=0.083 (5 min), 0.25 (15 min), 0.5 (30 min), 1, 2, 4, 8, and 24 hours after dosing. A similar procedure was used with orally dosed animals was used, except that collection took place at T=1, 2, 4, 8, 12, 24, and 36 hours after dosing. The samples were centrifuged under refrigerated conditions following completion of sample collection at each interval. The resulting plasma was separated and stored frozen at approximately -70°C until analysis.

**[0311]** Analysis was performed by LC/MS/MS, and plasma PK disposition parameters were calculated using the last five plasma concentrations for IV (1-24 h) and PO (4-36 h) administration for the selection of the Lambda Z. Due to inter-animal variability and limited terminal phase data, these results should be interpreted with caution.

**[0312]** No clinically abnormal findings followed IV or oral administration. Systemic exposure was observed at all time points.

#### Rat Single-Dose

**[0313]** Eighteen male Sprague Dawley rats (Charles River) weighing 217-263 g and aged 8-9 weeks were administered the test article, MGBG as either a single bolus intravenous dose of 1 mg/kg (Group 1) or as a single oral gavage dose of 10 mg/kg. A cohort of three animals was sacrificed via CO<sub>2</sub> inhalation anaesthesia after final blood collection at each of T = 2, 4, 12, 24, 36, and 48 hours post-dose. Dose formulation analysis verified administered dose solutions as within 17% of targeted concentration of 10 mg/kg.

**[0314]** Analysis was performed by LC/MS/MS. Pharmacokinetic analyses were performed on the mean MGBG plasma concentration versus time data using the non-compartmental approach (linear trapezoidal rule for AUC calculations). The WinNonlin sparse sampling tool was used for PK calculations. All samples reported as BQL (Below the Limit of Quantitation, in plasma 2.50 ng/mL) were changed to 0.00 ng/mL for the purpose of analysis. Dose formulation analysis revealed that formulations were within 15% of the targeted dose concentration of 10

mg/kg.

**[0315]** Abnormal clinical findings were not noted following dosing. A single PO administration of 10 mg/kg of MGBG resulted in evidence of measurable MGBG levels in plasma through the 12 hour time point; beyond that point, certain samples began measure BLQ.

### **Mouse Single-Dose**

**[0316]** Twenty-four male DBA/1 mice weighing 19.5-24.7 g and aged 7-9 weeks administered the test article, MGBG, as either a single bolus intravenous dose via a lateral tail vein of 1 mg/kg (Group 1, n = 12) or as a single oral gavage dose of 10 mg/kg (Group 2, n = 12). Each dose group consisted of 4 cohorts of 3 animals each. Group 1 was sampled at 5, 15, and 30 minutes after dosing; and 1, 2, 4, 8, and 24 hours after dosing. Group 2 was sampled at 1, 2, 4, 8, 12, 24, and 36 hours after dosing. Starting with the first time point, a new cohort was sampled at each successive time point up to the 1-hour (Group 1) or 12-hour (Group 2) time point. The order of sampling among the cohorts was repeated for the subsequent time points (some cohorts may have been bled only once). The second bleed for each cohort was terminal. Animals were sacrificed via CO<sub>2</sub> inhalation anaesthesia after final blood collection.

**[0317]** The samples were centrifuged under refrigerated conditions following completion of sample collection at each interval. The resulting plasma was separated and stored frozen at approximately -70°C until analysis. Analysis was performed by LC/MS/MS. Pharmacokinetic analyses were performed on the mean MGBG plasma concentration versus time data using the non-compartmental approach (linear trapezoidal rule for AUC calculations). The WinNonlin sparse sampling tool was used for PK calculations. Dose formulation analysis revealed that the IV and PO formulations were within 15% of their targeted concentrations.

**[0318]** Abnormal clinical findings were not noted following dosing. Evidence of systemic plasma MGBG exposure was observed at all collected plasma time points following IV and PO administration of MGBG.

**[0319]** Results of the foregoing assays are shown below in Tables 2 and 3. Values reported are mean across treatment groups without standard deviation.

**Table 2**

IV	t <sub>1/2</sub> (h)	T <sub>max</sub> (h)	C <sub>max</sub> (ng/mL)	Cl <sub>obs</sub> (mL/min/kg)	V <sub>SSobs</sub> (L/kg)	AUC (h*ng/mL)
rhesus	30.8	0.139	757	13.7	13.2	1660
rat	17.4	0.083	684	13.4	15.4	1250
mouse	13	0.083	181	49.6	38.3	336
dog	15.8	0.083	1180	13.7	14.6	1410

**Table 3**

ORAL	t <sub>1/2</sub> (h)	T <sub>max</sub> (h)	C <sub>max</sub> (ng/mL)	AUC (h*ng/mL)	Cl <sub>obs</sub> (mL/min/kg)	V <sub>d</sub> (L/kg)	F %
rhesus	24.2	3.33	192	4240	6.63	13.2	35.0%
rat	28.1	4.67	55.8	1280**	3.18	15.4	11.6%
mouse	11.8	1	106	1420	6.68	38.3	44.3%
dog	15.5	1	616	6290	8.29	14.6	49.0%

**[0320]** In Table 2 above, the double asterisk indicates that the rat AUC reported is the AUC<sub>all</sub>, computed from time zero to the time of the last plasma concentration measurement. Each of these values carries the caveat that terminal measurements are subject to different methods of extrapolation.

#### MULTI-DOSE RAT PHARMACOKINETIC AND TOLERABILITY STUDY

**[0321]** The purpose of this study was to determine the pharmacokinetic (PK) properties and tolerability of MGBG in rats. Additionally, recovery from any toxic effects was assessed after a seven day non-dosing period. Tolerability was demonstrated in test article-treated animals by body weight changes similar to the control group and a lack of adverse clinical observations.

**[0322]** Three per group of male Sprague Dawley (CD® IGS, Charles River) aged 7-9 weeks and weighing 222.7-252.0 g were administered by oral (PO) gavage, twice daily, at 10, 20, or 30 mg/kg/dose (20, 40, or 60 mg/kg/day) for seven consecutive days. A washout period of seven days followed. Collection of approximately 200 µL of whole blood was collected from the tail vein of all animals in Groups 5, 6, and 7 were bled at six (Day 1), seven (Day 7), or one (Days 9 through 15) time point(s), respectively. Whole blood samples were collected in a lithium heparin microtainer and processed to plasma by centrifugation. Plasma was frozen at -70°C. Pharmacokinetic analyses were performed on the individual animal plasma concentration versus time data for MGBG using WinNonlin (linear trapezoidal rule for AUC calculations). Nominal dose values and sampling times were used for calculations. For Study Day 7, the reported values for MGBG concentrations at time zero were used in the calculations of AUC. Study Day 1 disposition parameters were not reported due to insufficient terminal phase data to adequately characterize these parameters. Following PO administration of MGBG on Study Day 7, plasma PK disposition parameters were calculated on plasma concentrations obtained following the second administered dose (T=12-192 h) using the WinNonlin default selection criteria for the selection of the Lambda Z, the elimination rate constant, upon which half-life, AUC<sub>INFobs</sub>, and Cl/F<sub>obs</sub>, were based; inter-animal variability was noted.

**[0323]** Plasma samples collected from test article-treated animals on Day 1 and Day 7 were subjected to bioanalysis and confirmed systemic exposure to the test article at all time points.

Over the dose range evaluated,  $T_{max}$  values were dose-dependent and ranged from 3.33 to 14.0 h, and indicated absorption was slightly delayed on Study Day 7 compared to Study Day 1. Systemic exposure (as assessed by  $C_{max}$  and  $AUC_{all}$ ) increased with increasing dose, and the increase in both parameters was slightly less than dose-proportional at each evaluation interval. Repeat, twice-daily PO dosing of MGBG was associated with 3.77-, 4.03-, and 3.68-fold increases in mean  $AUC_{all}$  values compared to Study Day 1 for the 20, 40, and 60 mg/kg/day dose groups, respectively. On Study Day 7, evidence of dose-dependent dispositions for  $Cl/F_{obs}$  and elimination half-life were observed as mean parameter values for  $Cl/F_{obs}$  and elimination half-life increased and decreased, respectively, with increasing dose levels.

**[0324]** Differences between the control group and the 60 mg/kg/day dose group were noted for a few hematology parameters (lower reticulocyte count and percentage) and some serum chemistry parameters (e.g., osmolality and electrolyte changes consistent with slight dehydration). However, these changes were not thought to be adverse as they did not coincide with other signs of frank toxicity and the serum chemistry changes were demonstrated to be reversible. No gross or microscopic lesions were observed in test article-treated animals at terminal sacrifice, and no gross lesions were observed in test article-treated animals at recovery sacrifice.

**[0325]** Based on the findings of this exploratory study, the no observable adverse effect level (NOAEL) for MGBG administered by PO gavage twice daily for seven consecutive days to male Sprague Dawley rats is 30 mg/kg/dose (60 mg/kg/day).

**Table 4**

PO Dose Day 1 (mg/kg/day)	$C_{max}$ (ng/mL)	$T_{max}$ (h)	$AUC$ (h*ng/mL)	$AUC_{0-12}$ (h*ng/mL)	$AUC_{0-24}$ (h*ng/mL)	$Cl/F_{obs}$
20	124	10	NC	773	1750	NC
40	175	7.33	NC	1380	2980	NC
60	296	3.33	NC	2170	4380	NC

**Table 5**

PO Dose Day 7 (mg/kg/day)	$C_{max}$ (ng/mL)	$T_{max}$ (h)	$AUC$ (h*ng/mL)	$AUC_{0-12}$ (h*ng/mL)	$AUC_{0-24}$ (h*ng/mL)	$Cl/F_{obs}$
20	117	14	7930	1030	2190	24.3
40	194	9.33	13700	1800	3690	28.2
60	337	7.33	17100	3130	6040	35.8

#### ALLOMETRIC SCALING AND PREDICTED HUMAN EFFICACY

**[0326]** Multi-species allometric scaling based on pharmacokinetic parameters disclosed in

Tables 2 and 3 was employed to calculate predicted pharmacokinetic parameters in humans according to methods known in the art. See, e.g., Ings RM, "Interspecies scaling and comparisons in drug development and toxicokinetics," *Xenobiotica*, 1990 Nov;20(11):1201-31 and Khor, SP et al., "Dihydropyrimidine dehydrogenase inactivation and 5-fluorouracil pharmacokinetics: allometric scaling of animal data, pharmacokinetics and toxicodynamics of 5-fluorouracil in humans," *Cancer Chemother Pharmacol* (1997) 39(3): 833-38. Expected values are given below in Tables 6 and 7.

**Table 6**

IV	t <sub>1/2</sub> (h)	CL (mL/min/kg)	V <sub>SS</sub> (L/kg)
Based on Mouse, Rat, Dog, Rhesus	13.4	7.7	9.0
Based on Mouse, Dog, Rhesus	13.3	7.9	9.1

**Table 7**

ORAL	t <sub>1/2</sub> (h)	CL (mL/min/kg)	V <sub>SS</sub> (L/kg)
Based on Mouse, Rat, Dog, Rhesus	23.3	21.0	42.4
Based on Mouse, Dog, Rhesus	23.0	20.9	41.6

**[0327]** In both the murine carrageenan-induced paw edema and hyperalgesia models, the top efficacious dose of MGBG is 30 mg/kg PO BID (totaling 60 mg/kg/day). Based upon this dosing paradigm in mice, at least two methods to estimate the equivalent dosing in humans may be used.

**[0328]** The first method is based upon body surface area (BSA) normalization (described in Reagen-Shaw et al. (2007) *FASEB J.* 22, 659-661), as the authors note that BSA correlates well across species for various biological parameters, including basal metabolic rate, blood volume, caloric expenditure, plasma protein levels, and renal function. Using this method, a 60 mg/kg/day dose in mice would convert to about **4.9 mg/kg/day** in humans.

**[0329]** The second method used to convert the efficacious 60 mg/kg/day dose in mice to an equivalent dose in humans was based more directly on allometric scaling. Data from an MGBG pharmacokinetic study consisting of a 10 mg/kg oral dose in mice was modeled in a simulation to determine the theoretical AUC<sub>INF</sub> value for a dosing regimen of 30 mg/kg PO BID, which was 9050 h\*ng/mL. Next, predicted human clearance values as determined by single- and multi-species allometric scaling were used to estimate doses likely to produce an exposure in humans (AUC<sub>INF</sub>) similar to that of the 60 mg/kg/day in mice. Using single-species allometric scaling and a range of predicted human clearance values, a human equivalent dose would be in the range of **1.73 mg/kg/day to 4.51 mg/kg/day**. Using multi-species allometric scaling, the predicted human equivalent dose is about **4.2 mg/kg/day**.

**[0330]** In the murine carrageenan models, we also observed efficacy of MGBG at lower doses, including 3 mg/kg PO BID and 10 mg/kg PO BID, which would proportionally convert to human doses of **~0.42 mg/kg/day and ~1.2 mg/kg/day**.

**[0331]** The average body weight of a normal male human is often presumed to be 70 kg. Thus, daily doses based on the predictions above could be estimated to range from **about 25mg/day to about 350 mg/day**.

**[0332]** The proper dose depends, of course, on a number of factors. The patient may weigh much more or much less, or be female, elderly, or juvenile, requiring a lower or higher dose. The patient may exhibit a drug metabolic profile which might counsel for a lower or higher dose, such as a low expression level or activity of metabolizing enzymes such as cytochromes P450 (CYPs). This low expression or activity level may be due to a number of factors. Polymorphic expression of one or more CYPs (for example CYP2C19 and CYP2D6, though polymorphisms have been described for nearly all the CYPs) is known to be responsible for some populations to be "deficient" as compared to the population at large, leading to a "poor metabolizer" phenotype, requiring a lower dose. Additionally, exposure to an infectious agent or xenobiotic may cause repression of CYP expression or inhibition of existing CYPs. Alternatively, the patient may be physically weak, injured, or immunocompromised, all of which might counsel a lower dose. The patient may be taking a number of other drugs which compete with metabolic systems (including CYPs as discussed above) for disposal; this well-known polypharmaceutical effect may call for a lower dose. The dose also depends, as discussed above, on the condition and its severity. The efficacious dose for one disease or clinical endpoint will not necessarily be the same as the dose for another, and a severe, chronic, or otherwise serious case may call for a higher dose. However, a chronic case may also call for a lower dose administered over a longer or even indefinite period of time. All of these are discussed by way of example to illustrate the variability of ideal dosing; it is within the capacity of the skilled artisan to select an appropriate dosing range for a disease, population, or individual.

**[0333]** With these factors in mind, it should be clear that it is possible that the daily human dose may be as low as 1 mg/day, and as high as a 1g/day. In certain embodiments, the human dose may range: from 10 mg/day to 500 mg/day, from 20 mg/day to 400 mg/day, or from 25 mg/day to 350 mg/day. In further embodiments, the human dose may range: from 120 mg/day to 350 mg/day, from 150 mg/day to 350 mg/day, from 200 mg/day to 350 mg/day, or from 250 mg/day to 350 mg/day. In certain embodiments, the human dose may be any one of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70 75, 80, 85, 90, 95, 100, 110, 120, 125, 130, 140, 150, 160, 170, 175, 180, 190, 200, 210, 220, 225, 230, 240, 250, 260, 270, 275, 280, 290, 300, 310, 320, 325, 330, 240 or 350 mg/day.

**[0334]** In certain embodiments, the human dose may be any one of 275, 280, 285, 290, 295, 300, 305, 310, 315, 320, 325, 330, 335, 340, 350, 355, 360, 365, 370, or 375 mg/day. In one embodiment, the dose may be 275 mg/day. In another embodiment, the dose may be 300 mg/day. In another embodiment, the dose may be 305 mg/day. In another embodiment, the dose may be 310 mg/day. In another embodiment, the dose may be 315 mg/day. In another embodiment, the dose may be 320 mg/day. In another embodiment, the dose may be 325 mg/day. In another embodiment, the dose may be 330 mg/day. In another embodiment, the

dose may be 335 mg/day. In another embodiment, the dose may be 340 mg/day. In another embodiment, the dose may be 345 mg/day. In another embodiment, the dose may be 350 mg/day.

**[0335]** In certain embodiments, the human dose may be any one of 350, 375, 400, 425, 450, 475, 500, 525, 550 or 600 mg/day. In one embodiment, the dose may be 375 mg/day. In another embodiment, the dose may be 400 mg/day. In another embodiment, the dose may be 450 mg/day. In another embodiment, the dose may be 500 mg/day.

**[0336]** In certain embodiments, the human dose may be any one of 25, 50, 75, 100, or 125 mg/day. In one embodiment, the dose may be 375 mg/day. In another embodiment, the dose may be 25 mg/day. In another embodiment, the dose may be 50 mg/day. In another embodiment, the dose may be 75 mg/day. In another embodiment, the dose may be 100 mg/day. In another embodiment, the dose may be 125 mg/day.

#### **IN VIVO CARRAGEENAN TESTS**

##### **Carrageenan Paw Test for Edema and Hyperalgesia**

**[0337]** Injection of carrageenan subcutaneously into the hind foot (paw) of a rat or mouse induces robust inflammation and pain. The inflammatory response begins 1-2 hrs post-carrageenan injection and persists for at least five hours following inoculation. In addition, the animal's inflamed hind paw is sensitive to noxious (hyperalgesia) or innocuous (allodynia) stimuli, compared to the contralateral hind paw. Compounds can be evaluated in this model for anti-hyperalgesia and anti-inflammatory activity. A general increase in threshold or time to respond following drug administration suggests analgesic efficacy. A general decrease in paw swelling following drug administration suggests anti-inflammatory efficacy. It is possible that some compounds will affect the inflamed paw and not affect the responses of the contralateral paw.

**[0338]** Embodiments of the carrageenan foot edema test are performed with materials, reagents and procedures essentially as described by Winter, et al., (Proc. Soc. Exp. Biol. Med., 111, 544 (1962)). Prophylactic and therapeutic embodiments have been developed, and are known in the art. The animals are evaluated for their responsiveness to noxious (paw pinch, plantar test) or innocuous (cold plate, von Frey filaments) stimuli. In the following protocol, mice were used.

**[0339]** Animals, compounds, and dosing. Healthy young male Swiss Webster mice in which weight variation of the mice will not exceed  $\pm$  20% of the mean were used for the study. Animals were divided into four groups of forty, and each group was dosed by oral gavage with either MGBG (BID, 12 hours apart at 30 mg/kg in 5 mL/kg normal saline), dexamethasone as

positive control (QD, 1 mg/kg in 5mL/kg 0.5% methylcellulose), or saline vehicle (BID, 5 mL/kg). A fourth group served as naïve control (no carrageenan, no treatment). Treatment with MGBG took place on each of three days prior to carrageenan, one hour prior to carrageenan, and 11 hours post-carrageenan. Paw edema is developed by injecting carrageenan (Sigma: λ-carrageenan) subcutaneously in the subplantar region of the right paw of the mouse at a volume of 50  $\mu$ l of 1% carrageenan (w/v) in saline. The contra-lateral paw (left paw) received the same volume (50  $\mu$ l) of saline and serve as control. Mice will be anesthetized using light dose of ketamine before carrageenan injection.

**[0340] Paw Edema.** Immediately before sub-plantar administration of carrageenan and after 2, 3, 5 and 24 hours post carrageenan, mouse paw volume was measured using the plethysmometer (Ugo Basile). The assessment of edema was expressed as the mean increase in paw volume relative to control.

**[0341] Assessment of Paw Withdrawal Latency.** Prior to sub-plantar administration of carrageenan and after 0.5, 2, 3, 5, and 24 hours post- carrageenan, the latency of withdrawal response was determined by placing mice on a hot plate analgesia meter with surface temperature maintained at 51°C. A cut-off period of 30s was maintained to avoid any thermal injury to paw. Immediately after testing, all paws were immersed in ice-cold water before returning to the cage. Paw withdrawal latency is calculated as  $\Delta t$  = right paw withdrawal - left paw withdrawal.

**[0342] Serum, Plasma, and Histological Collection.** Prior to first drug dose on day 0 and at peak disease times (5 and 24 hours post-carrageenan challenge for serum, prior to first drug dose on day 0 and at conclusion of study), serum or plasma was collected from eight mice per group (each) and stored at -70°C until cytokine level determination or MGBG drug level determination. For serum collection, whole blood samples are collected in a serum separator tube, processed by centrifugation and frozen at -70°C. For drug level determination, whole blood samples are collected in a lithium heparin microtainer, processed to plasma by centrifugation and plasma frozen at -70°C. Additionally, paws are collected and preserved in 10% formalin for histology.

**[0343] Alternative Protocol.** In an alternative embodiment of this assay, MGBG was dosed PO, BID at 3, 10, and 30 mg/kg (with dexamethasone as positive control, saline as negative, and a treatment/carrageenan-naïve group, n = 16 each).

**[0344] Results.** Results are given in Figures 1 and 2. Statistically significant reduction in paw volume was observed in the MGBG-treated mice. Statistically significant reduction in paw edema was seen at all doses at several timepoints each. Statistically significant increases in paw withdrawal latency was seen at all doses for at least one timepoint each.

#### **IN VIVO MURINE COLLAGEN-INDUCED ARTHRITIS**

**Collagen-Induced Arthritis Models of Arthritis and Rheumatoid Arthritis**

**[0345]** The collagen-induced arthritis (CIA) model is considered a suitable model for studying potential drugs active in human arthritis because of the many immunological and pathological similarities to human rheumatoid arthritis (RA), the involvement of localized major histocompatibility, complete class-II-restricted T helper lymphocyte activation, and the similarity of histological lesions. See, e.g., Roslonec EF et al., "Collagen-Induced Arthritis," Current Protocols in Immunology, Unit 15.5 (1993). See also the model using a monoclonal antibody to CD18 and VLA-4 integrins described in Issekutz, A.C. et al., Immunology (1996) 88:569. Features of this CIA model that are similar to that found in RA patients include, without limitation: erosion of cartilage and bone at joint margins (as can be seen in radiographs), proliferative synovitis, symmetrical involvement of small and medium-sized peripheral joints in the appendicular, but not the axial, skeleton. The following procedure was followed to assess the efficacy of MGBG in the treatment of arthritic diseases.

**[0346]** Animals and dosing. Inbred male DBA/1 mice (DBA/1OlaHsd, Harlan Laboratories), at least 7 weeks old, may be used in the following collagen-induced arthritis model. Twenty animals per compound or vehicle are assigned to the arthritis and saline groups, 4 to the control group. To induce an arthritic state, mice are anesthetized with isoflurane and given 1501  $\mu$ l of bovine type II collagen in Freund's complete adjuvant injections (day 0 and day 21). Mice are randomized by body weight into treatment groups on study day 7. Treatment consists of 25 mg/kg MGBG, 0.2 mg/kg dexamethasone as positive control, or saline as vehicle control, all given as oral gavage beginning on study day 0 and continuing daily (PO, BID twice daily/12 hours apart). Twenty mice per group may be used, in which serum is collected from 15 animals, and plasma from five. Four additional animals serve as normal (untreated, non-arthritic) control group. The in-life portion of the study may proceed for 35 days.

**[0347]** Compounds. MGBG solution may be made from the hydrated dihydrochloride salt; other salts could be used, and in any case a salt/hydrate correction factor should be implemented. Solid MGBG can be stored at room temperature, but dose formulations should be made fresh for each administration. Dexamethasone is commercially available.

**[0348]** Data. On days 21-35, onset of arthritis typically occurs. During this time clinical scores for paw edema and swelling were given for each of the paws (right front, left front, right rear, left rear). Plasma draws are taken on days 0, 14, and 25 to assess pharmacokinetics, and blood draws taken on days 0 and 28 for disease analysis. Edema is measured on days 18-20, 22-27, and 29-34. Inflammation is assessed by infiltration of inflammatory cells and edema. Post-euthanasia, terminal blood draws are collected, heparinized, and frozen at -70°C until analyzed for cytokines such as osteopontin, TNFalpha, IL-1, CRP, MCP1, MIP-1beta, RANTES, IFNgamma, TGFbeta, IP-10, IL-17, and MMP9. Fore and hind paws and knees are collected, and following 1-2 days in fixative and then 4-5 days in decalcifier, processed, embedded, sectioned and stained with toluidine blue for histological analysis. Bone resorption is quantified

by presence of osteoclasts, defects in or loss of medullary trabecular or cortical bone. Cartilage damage is assessed by examining the severity and spread of chondrocyte loss and collagen disruption. Pannus tissue formation and the severity and spread of other evidence of destruction of joint architecture is followed.

**[0349]** Statistical Analysis. Clinical data for paw scores (means for animal) are analyzed by determining the area under the dosing curve (AUC) for days 1-15. For calculation of AUC, the daily mean scores for each mouse are entered into Microsoft Excel and the area between the treatment days after the onset of disease to the termination day is computed. Means for each group are determined and % inhibition from arthritis controls calculated by comparing values for treated and normal animals. Paw scores and histologic parameters (mean $\pm$ SE) for each group are analyzed for differences using a Student's t-test with significance is set at p 0.05. Percent inhibition of histologic parameters and AUC is calculated as  $[(\text{mean disease control} - \text{mean normal}) - (\text{mean treated} - \text{mean normal})] / [(\text{mean disease control} - \text{mean normal}) + (\text{mean treated} - \text{mean normal})] \cdot 100$ .

**[0350]** Expectations. It is expected that MGBG, as well as other polyamine analogs and polyamine biosynthesis inhibitors and compounds disclosed herein, will be efficacious in this model as shown by prevention of, reduction of, or delay of onset of arthritic symptoms as discussed above, reduced inflammation and related measures as discussed above, reduced measures of pain, and other related sequelae. The protocols above may be varied according to methods known in the art.

#### **ADDITIONAL *IN VIVO* MODELS OF THERAPEUTIC EFFICACY**

**[0351]** The following models, presented by way of example, may be used to evaluate compounds disclosed herein for efficacy in the treatment of a number of diseases and indications. It is within the capacity of one skilled in the art to modify these models to suit the needs of the study. Additionally, those skilled in the art will be familiar with additional models of disease which may be employed. It is expected that MGBG, as well as other polyamine analogs and polyamine biosynthesis inhibitors and compounds disclosed herein, will be efficacious in these models.

##### **Neuropathy and Neuropathic Pain Models**

##### **Bennett Model of Neuropathic Pain:**

**[0352]** A peripheral mononeuropathy is produced in adult rats by placing loosely constrictive ligatures around the common sciatic nerve. The postoperative behavior of these rats indicates that hyperalgesia, allodynia and, possibly, spontaneous pain (or dysesthesia) were produced.

Hyperalgesic responses to noxious radiant heat are typically evident on the second postoperative day and lasted for over 2 months. Hyperalgesic responses to chemogenic pain were also present. The presence of allodynia may be inferred from nocifensive responses evoked by standing on an innocuous, chilled metal floor or by innocuous mechanical stimulation (e.g., with von Frey filaments), and by the rats' persistence in holding the hind paw in a guarded position. The presence of spontaneous pain is suggested by a suppression of appetite and by the frequent occurrence of apparently spontaneous nocifensive responses. The affected hind paw is typically abnormally warm or cool in about one-third of the rats. About one-half of the rats develop grossly overgrown claws on the affected side. In compound efficacy models, test compound is typically delivered prior to stimulation and vehicle serves as control. Experiments with this animal model may advance understanding of the neural mechanisms of neuropathic pain disorders in humans. Bennett GJ, Xie YK, 1988 "A peripheral mononeuropathy in rat that produces disorders of pain sensation like those seen in man," Pain, Apr; 33(1):87-107 (PMID: 2837713).

### **Chung Model of Neuropathic Pain**

**[0353]** Since its introduction in 1992, the spinal nerve ligation (SNL) model of neuropathic pain has been widely used for various investigative works on neuropathic pain mechanisms as well as in screening tests for the development of new analgesic drugs. This model was developed by tightly ligating one (L5) or two (L5 and L6) segmental spinal nerves in the rat. The operation results in long-lasting behavioral signs of mechanical allodynia, heat hyperalgesia, cold allodynia, and ongoing pain. In the process of widespread usage, many different variations of the SNL model have been produced, either intentionally or unintentionally, by different investigators. Although the factors that cause these variations themselves are interesting and important topics to be studied, the pain mechanisms involved in these variations are likely different from the original model. The method for producing the spinal nerve ligation model that will minimally induce potential factors that may contribute to these variations is described in detail in Chung JM, Kim HK, and Chung K, "Segmental spinal nerve ligation model of neuropathic pain," Methods Mol Med.; 2004 99:35-45 (PMID: 15131327).

### **Chung Model in NHP**

**[0354]** In a model of painful neuropathy in the primate (*Macaca fascicularis*), a neuropathic state is induced by tight ligation of the L7 spinal nerve, just distal to the L7 dorsal root ganglion. Sensory testing may be done on the ventral surface of the foot, a region that includes the L7 dermatome. Within 1 week following surgery, primates typically develop a marked sensitivity to mechanical stimulation (e.g., with von Frey hairs), indicating the presence of mechanical allodynia. Increased sensitivity to mechanical stimulation is sometimes also observed on the contralateral side. The threshold for withdrawal to a heat stimulus decreases, indicating the presence of heat hyperalgesia. Presentation of various cooling stimuli, such as acetone and

cold water baths, indicates that cold allodynia also develops. Observed behavioral phenomena are similar to those seen in humans diagnosed with peripheral neuropathic pain. Thus, the model is useful for assessing a number of parameters relevant to human neuropathy and neuropathic pain disorders, and for evaluating the efficacy of drug candidates as treatments for related disorders. See, e.g., Carlton SM et al., "Behavioral manifestations of an experimental model for peripheral neuropathy produced by spinal nerve ligation in the primate," Pain 1994 Feb;56(2):155-66 (PMID: 8008406).

#### **Tactile Allodynia Assessment with Von Frey Filaments**

**[0355]** The following quantitative allodynia assessment technique may be modified to measure tactile allodynia in any of the various animal models of neuropathic pain. The following summary is given by way of example and refers to a rat surgical neuropathy model wherein nocifensive behaviors are evoked by light touch to the paw. Employing von Frey hairs from 0.41 to 15.1 g, the percent response at each stimulus intensity may first be characterized. A smooth log-linear relationship is typically observed. Additionally or alternatively, a paradigm using stimulus oscillation around the response threshold may be employed, which allows more rapid, efficient measurements. Correlation coefficient between the two methods is typically high. In neuropathic rats, good intra- and inter-observer reproducibility is found for the up-down paradigm; some variability may be seen in normal rats, attributable to extensive testing. The fact that thresholds in a sizable group of neuropathic rats show insignificant variability over 20 days, and after 50 days, 61% still met strict neuropathy criteria (using survival analysis), indicates that threshold measurement using the up-down paradigm, in combination with the neuropathic pain model, represents a powerful tool for analyzing the effects of manipulations of the neuropathic pain state. See, e.g., Chaplan SR et al., "Quantitative assessment of tactile allodynia in the rat paw," J Neurosci Methods, 1994 Jul;53(1):55-63 (PMID: 7990513).

#### **Hargreaves Method of Assessing Thermal Nociception**

**[0356]** Alternatively, a method to measure cutaneous hyperalgesia to thermal stimulation in unrestrained animals has been described. The testing paradigm uses an automated detection of the behavioral end-point; repeated testing does not contribute to the development of the observed hyperalgesia. Carrageenan-induced inflammation results in significantly shorter paw withdrawal latencies as compared to saline-treated paws and these latency changes corresponded to a decreased thermal nociceptive threshold. This sensitive thermal method detects dose-related hyperalgesia and its blockade by test compounds and allows for the measurement of other behavioral parameters in addition to the nociceptive threshold. See, e.g., Hargreaves K, et al., "A new and sensitive method for measuring thermal nociception in cutaneous hyperalgesia," Pain, 1988 Jan;32(1):77-88 (PMID: 3340425).

#### **HIV Dementia Models**

### **HIV Dementia Macaque Models**

**[0357]** Human Immunodeficiency virus (HIV), the virus that causes acquired immunodeficiency syndrome (AIDS), also manifests neurological complications. HIV-associated dementia (HAD) is the most severe form of HIV-induced neurocognitive disorders. HIV encephalitis (HIVE), the pathological correlate of HAD, is characterized by the formation of multinucleated giant cells and microglial nodules, astrocytosis, and neuronal damage and loss. Pathological evaluation of HAD disease progression in humans is not possible, with the only data collected being from individuals who have succumbed to the disorder, a snap shot of end-stage disease at best. Therefore, pertinent animal models have been developed to alleviate this gap of knowledge in the field of neurovirology and neuroinflammation. In general, the most widely used animal models are the simian immunodeficiency virus (SIV) and the chimeric simian/human immunodeficiency virus (SHIV) macaque model systems. Although both SIV and SHIV model systems are able to potentiate neuroinvasion and the concomitant neuropathology similar to that seen in the human syndromes, the innate differences between the two in disease pathogenesis and progression make for two separate, yet effective, systems for the study of HIV-associated neuropathology. For a thorough comparison of these two models, see Williams R et al., "Nonhuman primate models of NeuroAIDS," *J Neurovirol.* 2008 Aug;14(4):292-300 (PMID: 18780230). An exemplary SIV model is given below.

### **Simian Immunodeficiency Virus (SIV) Model**

**[0358]** The neuropathogenesis of human immunodeficiency virus (HIV)-associated dementia has remained elusive, despite identification of HIV as the causal agent. Although a number of contributing factors have been identified, the series of events that culminate in motor and cognitive impairments after HIV infection of the central nervous system (CNS) are still not known. Rhesus monkeys infected with simian immunodeficiency virus (SIV) manifest immunosuppression and CNS disease that is pathologically [L. R. Sharer et al. (1991) *J. Med. Primatol.* 20, 211-217] and behaviorally [E. A. Murray et al. (1992) *Science* 255, 1246-1249] similar to humans. The SIV model of HIV-associated dementia (HAD, HIV dementia, AIDS dementia, AIDS dementia complex, ADC, neuroAIDS, HIV-associated neurodegeneration, HAND) is widely recognized as a highly relevant model in which to investigate neuropathogenesis. With better understanding of neuropathogenesis comes the opportunity to interrupt progression and to design better treatments for HAD. This becomes increasingly important as patients live longer yet still harbor HIV-infected cells in the CNS. The use of the SIV model has allowed the identification of neurochemical markers of neuropathogenesis important not only for HAD, but also for other inflammatory neurological diseases.

**[0359]** The SIV model offers an ideal opportunity to investigate the neuropathogenesis of HAD. SIV is genetically, antigenically, and morphologically similar to HIV. In the CNS, SIV

predominantly infects MG/MP, whereas it does not productively infect neurons. Rhesus monkeys (*Macaca mulatta*) inoculated with SIV display neuropathological features similar to those seen with HAD, namely, multinucleated giant cells containing SIV, white matter lesions, and astrocytosis, although there are some pathological differences. The pattern of disease progression in monkeys resembles that in humans, with an initial burst of viral replication, followed by a latency period before development of immunosuppression, opportunistic infections, and death. The time course of disease progression is more rapid than that observed in humans, thus expediting experimental analysis. The premier advantage of the SIV model for the study of HAD is the opportunity to investigate neurochemical and neuropathological changes in relation to the onset of behavioral impairments in a nonhuman primate with a behavioral repertoire close to humans. Such investigations are accomplished by killing animals at various points during disease progression, especially at early stages. An additional advantage of the SIV model is the elimination of the confounding factor of treatment of HIV-infected individuals at end-stage disease. In short, the SIV model lends itself to probing the temporal, and ultimately causal, sequence of CNS changes induced by infection to determine the neuropathogenesis of HAD. The neuropathological correlates of impairments uncovered by this model have utility for HAD and for other neurological diseases. See, e.g., Rausch DM et al., "The SIV-infected rhesus monkey model for HIV-associated dementia and implications for neurological diseases," *J Leukoc Biol.* 1999 Apr;65(4):466-74 (PMID: 10204575).

#### **Measuring HIV Dementia and HIV Encephalopathy in SIV Models**

**[0360]** A battery of tasks may be chosen by the skilled investigator to assess encephalopathy and dementia in the SIV model. The tasks may be selected, for example, for their breadth in capturing neuropsychological impairments, including those described in HIV-infected humans, and for their ability to target neural substrates well characterized in primate lesioning experiments and implicated in HAD.

#### **HIV Dementia**

**[0361]** For example, in Rausch et al., above, the tasks assessed cognitive and motor function, and included: (1) delayed matching-to-sample with new stimuli on every trial to test visual recognition memory; (2) delayed matching-to-sample with two repeatedly used stimuli to test recent memory, (3) a visual discrimination learning and retention task to test stimulus-response association, and (4) a spatial scene learning task, which measured long-term memory for spatial locations. The motor task assessed the ability of each monkey to retrieve food from a rotating table (via the turntable speed at which animals successfully retrieved food in 50% of trials).

**[0362]** In one paradigm, a cohort of animals is trained on the battery of tasks, inoculated with SIV (optionally one or more isolated or molecularly cloned strains selected for properties such

as neurovirulence), and then evaluated on the tasks to detect changes in performance over a defined period. Alternatively, a separate cohort of animals is inoculated first and then trained and evaluated for their ability to learn the tasks. Varying the timing of SIV inoculation relative to neurobehavioral testing distinguishes between acquisition and retention of tasks. Infected animals may be considered impaired, for example, when their scores were more than two standard deviations from the mean score of uninfected control animals, which are sham-inoculated. While animals are alive, their clinical status, blood, and CSF may be assayed periodically to determine disease progression, immune status, and viral load. Neurochemistry and neuropathology may be performed at death. The range, extent, and variability of the cognitive and motor impairments resemble findings with HIV-infected humans.

### **HIV Encephalopathy**

**[0363]** An alternative model of HIV encephalopathy allows a more focused inquiry into the pathology of HIV infection in the brain. Following inoculation with SIV and development of moderate to severe encephalitis, viral load may be measured in cerebrospinal fluid (CSF) examined longitudinally to onset of AIDS and in brain tissue at necropsy to examine the relationship of systemic and central nervous system (CNS) viral replication to the development of encephalitis. Persistent high levels of CSF viral RNA after the acute phase of infection correlate with the development of encephalitis, and the level of both viral RNA and antigen in the brain correlate with the severity of the CNS lesions. In contrast, plasma levels of viral RNA do not correlate to the development or severity of encephalitis. Thus, CSF viral load measurements in the postacute phase of SIV infection serve as a marker for encephalitis and CNS viral replication, an effective way to measure the efficacy of test compounds in preventing or attenuating HIV encephalitis and related disorders. See, e.g., Zink MC et al., "High viral load in the cerebrospinal fluid and brain correlates with severity of simian immunodeficiency virus encephalitis,": J Virol. 1999 Dec;73(12):10480-8 (PMID: 10559366).

**[0364]** Additionally, metabolic markers such as NAA/Cr may also be used to quantitate disease severity in SIV/macaque models of encephalitis. In vivo MR spectroscopy (MRS) studies have shown reductions in NAA/Cr levels in human patients with severe neurocognitive deficits due to AIDS dementia complex (ADC), also known as neuroAIDS. High-field proton MRS was performed on extracted metabolites from frontal cortex tissue samples of 29 rhesus macaques (6 healthy, 23 moribund with AIDS). Neuropathologic determination of encephalitis severity for each animal was completed and was found to correlate with NAA/Cr levels. Decreases in Glu/Cr and GABA/Cr may indicate that both excitatory and inhibitory neurons are affected. Highly significant correlations between NAA/Cr, Glu/Cr, and GABA/Cr were observed. These neuronal metabolites were also decreased in the absence of classical SIV encephalitis (SIVE). These studies indicate that metabolic markers may serve as indicators of disease progression, and the efficacy of treatment at preventing or delaying the development of NeuroAIDS. See Lentz MR et al., "Metabolic markers of neuronal injury correlate with SIV CNS disease severity and inoculum in the macaque model of neuroAIDS," Magn Reson Med. 2008 Mar;59(3):475-84 (PMID: 18306400).

### FIV Model of HIV Dementia

**[0365]** Feline immunodeficiency virus (FIV) is a neurotropic lentivirus that produces a protracted state of immunodeficiency and encephalopathy in the cat. Recent evidence has shown several similarities to the natural progression of human immunodeficiency virus infection (HIV-1) associated degenerative effects on the central and peripheral nervous systems. Similar to HIV-1, FIV-induced encephalopathy neurovirulence is strain dependent, results in progressive immunodeficiency and increasing early peripheral but not brain viral load, preferentially affects the developing nervous system, produces quantifiable behavioral and neurophysiological impairment that is not directly linked to neuronal infectivity, and induces neuronal injury and loss both *in vivo* and *in vitro*. FIV has the added benefit of being non-communicable to human researchers. For these reasons, the FIV model is useful as a platform for assessing the efficacy of compounds in the treatment of HIV dementia and related disorders. See, e.g., Podell M et al., "The feline model of neuroAIDS: understanding the progression towards AIDS dementia," *J Psychopharmacol.* 2000;14(3):205-13 (PMID: 11106298).

### Inflammatory and Autoimmune Models

#### Contact Dermatitis and Related Disorders

**[0366]** Contact hypersensitivity is a simple delayed type hypersensitivity *in vivo* assay of cell-mediated immune function, which can be used to assess potential therapeutic efficacy in a number of disorders having an inflammatory and/or autoimmune component. Such diseases include contact dermatitis, atopic dermatitis, psoriasis, allergic dermatitis, and dermal irritation. Compounds may be topically applied, optionally in a topical formulation, or may be delivered by a non-topical (e.g., oral, IV, etc) route.

#### Murine Model

**[0367]** In one procedure, cutaneous exposure to exogenous haptens gives rise to a delayed type hypersensitivity reaction which is measured and quantitated. Contact sensitivity involves an initial sensitizing phase followed by an elicitation phase. The elicitation phase occurs when the T lymphocytes encounter an antigen to which they have had previous contact. Swelling and inflammation occur, making this an excellent model of human allergic contact dermatitis. Murine models also typically have the additional benefit of being economical to run. A suitable procedure is described in detail in Gaspari AA and Katz SI, "Contact Hypersensitivity," *Current Protocols in Immunology*, Unit 4.2, John Wiley & Sons, Inc. (1994). See also Grabbe S and

Schwarz T. "Immunoregulatory mechanisms involved in elicitation of allergic contact hypersensitivity," *Immun. Today* 19 (1): 37-44 (1998).

### **Porcine Model**

**[0368]** The choice of animal can be important in dermatological studies intended to predict human response. For this reason, pigs and in particular minipigs are favored due to the similarities between human and pig skin (particularly follicular density). See, for example, an exemplary model in Bilski AJ and Thomson DS, "Allergic contact dermatitis in the domestic pig. A new model for evaluating the topical anti-inflammatory activity of drugs and their formulations," *Br J Dermatol*, 1984 Jul;111 Suppl 27:143 (PMID: 6743545).

### **Hairless Guinea Pig Model**

**[0369]** Allergic and irritant contact reactions have also been evaluated in the recently identified hairless guinea pig, Crl:IAF(HA)BR, a mutant from the Hartley strain. The irritant contact dermatitis may be induced by croton oil, 2,4-dinitrochlorobenzene (DNCB), or anthralin. Both hairless and hairy guinea pigs develop similar reactions to these chemicals. Photoallergic contact sensitization may be also induced with tetrachlorosalicylanilide (TCSA), or with cyclophosphamide before sensitization with tribromosalicylanilide (TBS). Cutaneous changes are observed macro- and microscopically according to methods known in the art. Thus, hairless guinea pigs can be used as animal models for assessment of test compounds in the treatment of immunologic and nonimmunologic contact reactions and related disorders. See, e.g., Miyauchi H and Horio T, "A new animal model for contact dermatitis: the hairless guinea pig," *J Dermatol*. 1992 Mar; 19(3):140-5(PMID: 1640019).

**[0370]** Simple dermal irritation may also be studied in hairless guinea-pigs. In an exemplary model, test compounds are delivered in one or more topical formulations for 30 min daily exposure for 4 days. Scoring is performed daily; evaporimetry (total epidermal water loss (TEWL)), hydration and colorimetry are measured at baseline (day 0) in the middle and at the end of treatment. Test compounds are applied twice daily. See, e.g., Andersen F et al., "The hairless guinea-pig as a model for treatment of cumulative irritation in humans," *Skin Res Technol*. 2006 Feb;12(1):60-7 (PMID: 16420540).

### **Psoriasis Murine Chimera Model**

**[0371]** Additionally, the compounds disclosed herein can be tested in animal models for psoriasis-like diseases. Research into the cause and pathophysiological mechanisms underlying expression of psoriatic skin lesions has been hampered by lack of an appropriate animal model for this common and enigmatic cutaneous disease. One suitable model is the

human skin/scid mouse chimera prepared as described by Nickoloff BJ et al., "Severe combined immunodeficiency mouse and human psoriatic skin chimeras. Validation of a new animal model," Am J Pathol., 1995 Mar;146(3): 580-8 (PMID: 7887440) . The methods described therein characterize normal skin, pre-psoriatic skin, and psoriatic plaque skin samples transplanted onto severe combined immunodeficiency mice. Either normal, prepsoriatic, or psoriatic plaque keratome skin samples are transplanted onto severe combined immunodeficiency mice reliably with high rates of graft survival (> 85%) and with reproducible changes consistently observed over prolonged periods of engraftment. After transplantation, by clinical assessment and routine light microscopy, normal skin remains essentially normal whereas pre-psoriatic skin became thicker, and psoriatic plaque skin retains its characteristic plaque-type elevation and scale. By using a panel of antibodies and immunohistochemical analysis, the overall phenotype of human cell types (including immunocytes) that persisted in the transplanted skin was remarkably similar to the immunophenotype of pretransplanted skin samples. Additionally, clearly recognized interface zones between human and murine skin within the epidermal and dermal compartments can be identified by routine microscopy and immunostaining, with focal areas of chimerism. The many similarities between pre- and post-transplanted human samples of normal and psoriatic skin that are grafted onto severe combined immunodeficiency mice make this animal model appropriate for use in evaluating test compounds for efficacy in treating psoriasis and related disorders.

#### **Psoriasis Murine scid/scid Model**

**[0372]** Alternatively, the compounds disclosed herein can be tested in the scid/scid mouse model described by Schön MP et al., "Murine psoriasis-like disorder induced by naive CD4+ T cells," Nat Med., 1997 Feb;3(2):183-8 (PMID: 9018237). In this model, reconstitution of scid/scid mice with minor histocompatibility mismatched naive CD4+ T lymphocytes results in skin alterations that strikingly resemble human psoriasis clinically, histopathologically and in cytokine expression.

#### **Asthma**

**[0373]** Compounds may additionally be evaluated for efficacy in the treatment of asthma and related pulmonary disorders. In one murine model of asthma, wild-type control [C57BL/6J, (+/)] and ICAM-1 (intercellular adhesion molecule-1) knockout [C57BL/6J-ICAM-1, (-/-)] mice are sensitized to ovalbumin (OVA), and challenged with OVA delivered by aerosol (OVA-OVA) to induce a phenotype consistent with an asthmatic response. Bronchial responsiveness to methacholine and counts of cell numbers and measurements of eosinophil content and cytokine levels in bronchoalveolar lavage fluid (BALF) may be measured. Additionally, lymphocyte proliferation in response to antigen, eosinophil migration into the airways, and the development of airway hyperreactivity (AHR) in allergen-sensitized and -challenged mice may

all be measures in vivo or ex vivo according to methods known in the art. See Wolyniec WW et al., "Reduction of antigen-induced airway hyperreactivity and eosinophilia in ICAM-1-deficient mice," Am J Respir Cell Mol Biol., 1998 Jun;18(6):777-85 (PMID: 9618382).

### **Inflammatory Bowel Disease, Crohn's Disease, and Ulcerative Colitis**

**[0374]** The compounds disclosed herein can also be evaluated for activity in animal models of inflammatory bowel disease, Crohn's disease, and ulcerative colitis. The protocol described by Scheiffele F, Fuss IJ, "Induction of TNBS colitis in mice," Curr Protoc Immunol, 2002 Aug; Chapter 15:Unit 15.19 (PMID: 18432874), is one of several that have been used to study the immunopathogenesis of these diseases. The model employs the use of 2,4,6-trinitrobenzenesulfonic acid (TNBS), which induces severe colonic inflammation when administered intrarectally in SJL/J mice. The colitis which results from this procedure presents clinical and histopathological findings that resemble those seen in Crohn's disease. Scheiffle and Fuss discuss the critical parameters needed for successful induction of TNBS-colitis as well methods for monitoring and grading disease levels, and give a support protocol for isolating lamina propria mononuclear cells from mouse colons. See also Morris GP et al.. "Hapten-induced model of chronic inflammation and ulceration in the rat colon," Gastroenterology, 1989 Mar;96(3):795-803 (PMID: 2914642), describing the original rat model of chronic colonic inflammation by the intraluminal instillation of a solution containing a "barrier breaker" (e.g., 0.25 ml of 50% ethanol) and a hapten (e.g., TNBS, 5-30 mg) At a dose of 30 mg, trinitrobenzenesulfonic acid/ethanol-induced ulceration and marked thickening of the bowel wall persisted for at least 8 weeks. Histologically, the inflammatory response included mucosal and submucosal infiltration by polymorphonuclear leukocytes, macrophages, lymphocytes, connective tissue mast cells, and fibroblasts. Granulomas (3 wk after induction of inflammation), Langhan's-type giant cells, segmental ulceration and inflammation. The characteristics and relatively long duration of inflammation and ulceration induced in these models afford an opportunity to study the pathophysiology of colonic inflammatory disease in a specifically controlled fashion, and to evaluate new treatments potentially applicable to inflammatory bowel disease in humans.

### **EXEMPLARY ORAL PHARMACEUTICAL FORMULATIONS**

**[0375]** The following are examples of compositions which may be used to orally deliver compounds disclosed herein as a capsule.

**[0376]** A solid form of a compound of Formula VI may be passed through one or more sieve screens to produce a consistent particle size. Excipients, too, may be passed through a sieve. Appropriate weights of compounds, sufficient to achieve the target dosage per capsule, may be measured and added to a mixing container or apparatus, and the blend is then mixed until uniform. Blend uniformity may be done by, for example, sampling 3 points within the container

(top, middle, and bottom) and testing each sample for potency. A test result of 95-105% of target, with an RSD of 5%, would be considered ideal; optionally, additional blend time may be allowed to achieve a uniform blend. Upon acceptable blend uniformity results, a measured aliquot of this stock formulation may be separated to manufacture the lower strengths. Magnesium stearate may be passed through a sieve, collected, weighed, added to the blender as a lubricant, and mixed until dispersed. The final blend is weighed and reconciled. Capsules may then be opened and blended materials flood fed into the body of the capsules using a spatula. Capsules in trays may be tamped to settle the blend in each capsule to assure uniform target fill weight, then sealed by combining the filled bodies with the caps.

#### COMPOSITION EXAMPLE 1

**[0377] 300 mg Capsule:** Total fill weight of capsule is 500 mg, not including capsule weight. Target compound dosage is 300 mg per capsule, but may be adjusted to account for the weight of counterions and/or solvates if given as a salt or solvated polymorph thereof. In such a case the weight of the other excipients, typically the filler, is reduced.

Ingredient	Quantity per Capsule, mg
MGBG	300.00
Lactose monohydrate	179.00
Silicon dioxide	3.00
Crospovidone	15.00
Magnesium stearate (vegetable grade)	3.00

#### COMPOSITION EXAMPLE 2

**[0378] 150 mg Capsule:** Total fill weight of capsule is 300 mg, not including capsule weight. Target compound dosage is 150 mg per capsule, but may be adjusted to account for the weight of counterions and/or solvates if given as a salt or solvated polymorph thereof. In such a case the weight of the other excipients, typically the filler, is reduced.

Ingredient	Quantity per Capsule, mg
MGBG	150
Microcrystalline cellulose (MCC)	147
Magnesium stearate (vegetable grade)	3

## REFERENCES CITED IN THE DESCRIPTION

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**Patentkrav**

1. Oral farmaceutisk sammensætning, omfattende MGBG sammen med mindst en oral farmaceutisk acceptabel excipiens, til anvendelse til behandling af smerte ved 5 oral indgivelse til et individ.
2. Oral farmaceutisk sammensætning til anvendelse ifølge krav 1, omfattende 25 mg til 350 mg MGBG.
- 10 3. Oral farmaceutisk sammensætning til anvendelse ifølge krav 1 eller krav 2, formuleret som en tablet eller kapsel.
- 15 4. Oral farmaceutisk sammensætning til anvendelse ifølge et hvilket som helst af kravene 1 til 3, hvor smerten er valgt fra gruppen bestående af inflammatorisk smerte, smerte forårsaget af nerveskade, kronisk smerte, intraktabel kræftsmerte, komplekst regionalt smertesyndrom, neuropatisk smerte, smerter under eller efter kirurgisk indgreb, tandsmærter, smerter forårsaget af hudskader, lændesmerter, hovedpine, migræne, taktil allodyni og hyperalgesi.
- 20 5. Oral farmaceutisk sammensætning til anvendelse ifølge et hvilket som helst af kravene 1 til 4, hvor individet er et menneske.
- 25 6. Oral farmaceutisk sammensætning til anvendelse ifølge et hvilket som helst af kravene 1 til 5, hvor smerten er kronisk.
7. Oral farmaceutisk sammensætning til anvendelse ifølge et hvilket som helst af kravene 1 til 5, hvor smerten er akut.
- 30 8. MGBG til anvendelse til behandling af smerte.
9. MGBG til anvendelse ifølge krav 8, hvor MGBG'en indgives oralt.
10. MGBG til anvendelse ifølge krav 8 eller krav 9, hvor smerten er valgt fra gruppen bestående af inflammatorisk smerte, smerte forårsaget af nerveskade, 35 kronisk smerte, intraktabel kræftsmerte, komplekst regionalt smertesyndrom,

neuropatisk smerte, smerter under eller efter kirurgisk indgreb, tandsmarter, smerter forårsaget af hudskader, lændesmerter, hovedpine, migræne, taktil allodyni og hyperalgesi.

5 **11.** MGBG til anvendelse ifølge et hvilket som helst af kravene 8 til 10, hvor smerten er kronisk.

**12.** MGBG til anvendelse ifølge et hvilket som helst af kravene 8 til 10, hvor smerten er akut.

## DRAWINGS

Fig. 1/2

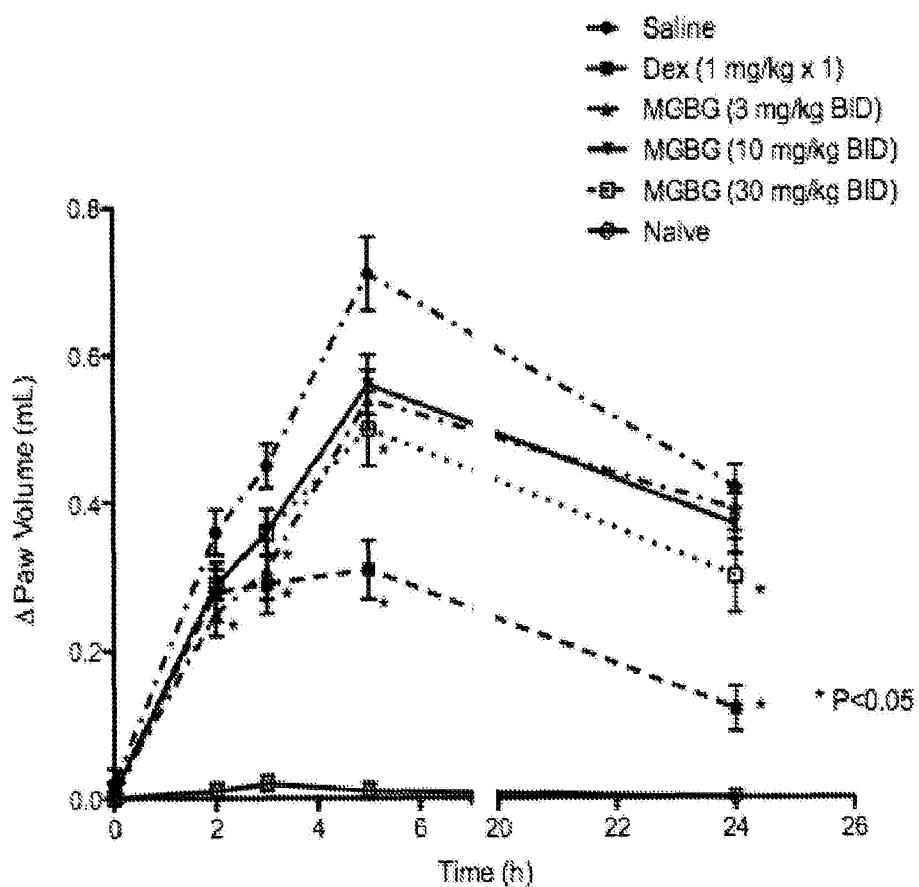


Fig. 2/2

