Title: METHODS AND COMPOSITIONS FOR INHIBITING IMPDH ISOFORM 1

Abstract: Described herein are methods of inhibiting IMPDH type 1, and treating or preventing a disease or disorder (or symptoms thereof) associated with IMPDH type 1, wherein an IMPDH type 1 inhibitor compound is administered to a subject.
METHODS AND COMPOSITIONS FOR INHIBITING IMPDH ISOFORM 1

Government Support
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Cross-Reference to Related Application
The present application claims the benefit of U.S. provisional application number 60/784,332 filed March 20, 2006, which is incorporated herein by reference in its entirety.

Background of the Invention
Angiogenesis may be defined as the development of a blood supply to a given area of tissue. The development of a blood supply may be part of normal embryonic development, represent the revascularization of a wound bed, or involve the stimulation of vessel growth by inflammatory or malignant cells. Sometimes angiogenesis is defined as the proliferation of new capillaries from pre-existing blood vessels. New growth of soft tissue requires new vascularization, and the concept of angiogenesis is a key component of tissue growth and in particular, a key point of intervention in pathological tissue growth.

Angiogenesis is a fundamental process necessary for embryonic development, subsequent growth, and tissue repair. Angiogenesis is a prerequisite for the development and differentiation of the vascular tree, as well as for a wide variety of fundamental physiological processes including embryogenesis, somatic growth, tissue and organ repair and regeneration, cyclical growth of the corpus luteum and endometrium, and development and differentiation of the nervous system. In the female reproductive system, angiogenesis occurs in the follicle during its development, in the corpus luteum following ovulation and in the placenta to establish
and maintain pregnancy. Angiogenesis additionally occurs as part of the body's repair processes, e.g., in the healing of wounds and fractures.

Both controlled and uncontrolled angiogenesis are thought to proceed in a similar manner. Endothelial cells and pericytes, surrounded by a basement membrane, form capillary blood vessels. Angiogenesis begins with the erosion of the basement membrane by enzymes released by endothelial cells and leukocytes. The endothelial cells, which line the lumen of blood vessels, then protrude through the basement membrane. Angiogenic stimulants induce the endothelial cells to migrate through the eroded basement membrane. The migrating cells form a "sprout" off the parent blood vessel, where the endothelial cells undergo mitosis and proliferate. The endothelial sprouts merge with each other to form capillary loops, creating new blood vessels. Creation of the new microvascular system can initiate or exacerbate disease conditions.

Medical science has recognized that angiogenesis is an important factor in the initiation and/or proliferation of a large number of diverse disease conditions. Under normal physiological conditions, humans and other animals only undergo angiogenesis in very specific, restricted situations. For example, angiogenesis is normally observed in wound healing, fetal and embryonic development, and in the formation of the corpus luteum, endometrium and placenta. The process of angiogenesis has been found to be altered in a number of disease states, and in many instances, the pathological damage associated with the disease is related to uncontrolled angiogenesis. Since it was first put forward over thirty years ago, the hypothesis that angiogenesis is required for tumor growth and metastasis has gained extensive experimental support (Folkman, J. (1971) *N. Engl. J. Med.* 285, 1182-1186, Hanahan, D. & Folkman, J. (1996) *Cell* 86, 353-364). For example, angiogenesis is a factor in tumor growth, since a tumor must continuously stimulate growth of new capillary blood vessels in order to grow. Angiogenesis is an essential part of the growth of human solid cancer, and abnormal angiogenesis is associated with other diseases such as rheumatoid arthritis, psoriasis, and diabetic retinopathy (Folkman, J. and Klagsbrun, M., *Science* 235:442-447,(1987)). In addition to tumor growth and metastasis, angiogenesis has also been implicated in rheumatoid arthritis, diabetic retinopathy and macular degeneration, suggesting that inhibition of angiogenesis may

One way to accelerate the drug discovery process for angiogenesis related disorders/diseases involves finding new uses for existing drugs. Because the toxicity, pharmacokinetics, and clinical properties of existing drugs are well established, compounds that show activity can be rapidly and inexpensively evaluated as new treatments and moved into the clinic if appropriate. Furthermore, the extensive structure/activity data accumulated during the development of each drug can greatly facilitate mechanistic studies for target identification or validation.

Clearly, the development and progress of many disease conditions can be controlled by controlling the process of angiogenesis. However, many materials which appear promising in vitro have proven to be relatively ineffective when applied in vivo. Furthermore, various of such materials have been found to be unstable, toxic, or otherwise difficult to employ. Consequently, there is a need for methods and materials capable of controlling and inhibiting angiogenesis in a reliable manner. It is therefore an object of the invention to provide compounds and pharmaceutical compositions which exhibit activity as inhibitors of angiogenesis.

**Summary of the Invention**

In certain aspects, the invention provides a method of inhibiting IMPDH type 1 in a subject, the method comprising the step of administering to the subject identified as being in need of an IMPDH type 1 inhibitor, an effective amount of an IMPDH type 1 inhibitor compound selected from: an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the subject does not experience immunosuppressive side effects associated with IMPDH type II.

In other aspects, the invention provides a method of inhibiting angiogenesis in a subject, the method comprising the step of administering to the subject identified as being in need of inhibiting angiogenesis, an effective amount of an anti-angiogenic IMPDH type 1 inhibitor compound selected from: an IMPDH type 1 inhibitor, a nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the subject does not experience immunosuppressive side effects associated with IMPDH type II.
In another aspect, the invention provides for the use of an IMPDH type 1 inhibitor compound in the manufacture of a medicament for inhibiting or reducing IMPDH type 1 in a patient, the IMPDH type 1 inhibitor compound being an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In certain aspects, the invention provides a sustained release device for implantation in a patient and sustained release of an IMPDH type 1 inhibitor compound for at least a period of 30 days, wherein the IMPDH type 1 inhibitor compound is an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In another aspect, the invention provides a sustained release drug device adapted for implantation in or adjacent to the eye of a patient, the drug delivery device comprising: (i) an IMPDH type 1 inhibitor drug core comprising an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; (ii) an impermeable coating disposed about the core that is substantially impermeable to the passage of the IMPDH type 1 inhibitor compound, having one or more openings therein which permit diffusion of the IMPDH type 1 inhibitor compound, and which is substantially insoluble and inert in body fluids and compatible with body tissues; and, optionally, (iii) one or more permeable polymer members or coatings disposed in the flow path of the IMPDH type 1 inhibitor compound through said openings in said impermeable coating, said permeable polymer being permeable to the passage of the IMPDH type 1 inhibitor compound, and which is substantially insoluble and inert in body fluids and compatible with body tissues; wherein the impermeable coating and permeable polymer members or coatings are disposed about the drug core so as to produce, when implanted, a substantially constant rate of release of the IMPDH type 1 inhibitor compound from the device; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In another aspect, the invention provides a sustained release formulation for depot injection in a patient and sustained release of an IMPDH type 1 inhibitor
compound for at least a period of 30 days, wherein the formulation includes: an
viscous gel formulation comprising a bioerodible, biocompatible, polymer; and
an anti-angiogenic IMPDH type 1 inhibitor agent dissolved or dispersed therein,
which anti-angiogenic agent is selected from: an IMPDH type 1 inhibitor, an
nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine;
danazol; and asparaginase; wherein the patient does not experience
immunosuppressive side effects associated with IMPDH type II.

In another aspect, the invention provides a kit comprising an effective amount
of an IMPDH type 1 inhibitor compound in unit dosage form, together with
instructions for administering the IMPDH type 1 inhibitor compound to a subject
identified as being in need of treatment with an IMPDH type 1 inhibitor.

**Brief Description of the Drawings**

FIG. 1 shows the inhibition of HUVEC by Mycophenolic acid and its reversal
by guanine.

FIG. 2 shows the selective knockdown of IMPDH-1 and -2 by shRNA in
HUVEC.

FIG. 3 illustrates the inhibition of angiogenesis in vivo.

FIG 4 illustrates the inhibition of tumor-associated angiogenesis.

FIG 5 shows the X-ray structure of human IMPDH-2 in complex with
substrate and cofactor analogs. (A) overall architecture of the tetramer of IMPDH.
Color code: red, SAD, green, 6-CI-IMP. (B) The adenine-binding pocket of IMPDH-
2 with the three residues that differ between IMPDH2 and IMPDH-1 indicated.

**Detailed Description of the Invention**

1. **Definitions**

Before a further description of the present invention, and in order that the
invention may be more readily understood, certain terms are first defined and
collected here for convenience.

As used herein, the term "alkyl" refers to a straight-chained or branched
hydrocarbon group containing 1 to 12 carbon atoms. The term "lower alkyl" refers to
a C1-C6 alkyl chain. Examples of alkyl groups include methyl, ethyl, n-propyl,
isopropyl, tert-butyl, and n-pentyl. Alkyl groups may be optionally substituted with one or more substituents.

The term "alkenyl" refers to an unsaturated hydrocarbon chain that may be a straight chain or branched chain, containing 2 to 12 carbon atoms and at least one carbon-carbon double bond. Alkenyl groups may be optionally substituted with one or more substituents.

The term "alkynyl" refers to an unsaturated hydrocarbon chain that may be a straight chain or branched chain, containing the 2 to 12 carbon atoms and at least one carbon-carbon triple bond. Alkynyl groups may be optionally substituted with one or more substituents.

The term "alkoxy" refers to an -O-alkyl radical.

As used herein, the term "halogen" or "halo" means -F, -Cl, -Br or -I.

As used herein, the term "haloalkyl" means an alkyl group in which one or more (including all) of the hydrogen radicals are replaced by a halo group, wherein each halo group is independently selected from -F, -Cl, -Br, and -I. The term "halomethyl" means a methyl in which one to three hydrogen radical(s) have been replaced by a halo group. Representative haloalkyl groups include trifluoromethyl, bromomethyl, 1,2-dichloroethyl, 4-iodobuty1, 2-fluoropentyl, and the like.

The term "cycloalkyl" refers to a hydrocarbon 3-8 membered monocyclic or 7-14 membered bicyclic ring system having at least one saturated ring. Cycloalkyl groups may be optionally substituted with one or more substituents. In one embodiment, 0, 1, 2, 3, or 4 atoms of each ring of a cycloalkyl group may be substituted by a substituent. Representative examples of cycloalkyl group include cyclopropyl, cyclopentyl, cyclohexyl, cyclobutyl, cycloheptyl, cyclooctyl, cyclononyl, and cyclodecyl.

The term "cycyl" refers to a hydrocarbon 3-8 membered monocyclic or 7-14 membered bicyclic ring system having at least one non-aromatic ring, wherein the non-aromatic ring has some degree of unsaturation. Cycyl groups may be optionally substituted with one or more substituents. In one embodiment, 0, 1, 2, 3, or 4 atoms of each ring of a cycyl group may be substituted by a substituent. Examples of cycyl groups include cyclohexenyl, bicyclo[2.2.1]hept-2-enyl, dihydronaphthalenyl, benzocyclopentyl, cyclopentenyl, cyclopentadienyl, cyclohexenyl, cyclohexadienyl, cycloheptenyl, cycloheptadienyl, cycloheptatrienyl, cyclooctenyl,
cyclooctadienyl, cyclooctatrienyl, cyclooctatetraenyl, cyclononenyl, cyclononadienyl, cyclodecenyl, cyclodecadienyl and the like.

The term “aryl” refers to a hydrocarbon monocyclic, bicyclic or tricyclic aromatic ring system. Aryl groups may be optionally substituted with one or more substituents. In one embodiment, 0, 1, 2, 3, 4, 5 or 6 atoms of each ring of an aryl group may be substituted by a substituent. Examples of aryl groups include phenyl, naphthyl, anthracenyl, fluorenyl, indenyl, azuleny1, and the like.

As used herein, the term “aralkyl” means an aryl group that is attached to another group by a (C1-C6)alkylene group. Aralkyl groups may be optionally substituted, either on the aryl portion of the aralkyl group or on the alkylene portion of the aralkyl group, with one or more substituents. Representative aralkyl groups include benzyl, 2-phenyl-ethyl, naphth-3-yl-methyl and the like.

As used herein, the term “alkylene” refers to an alkyl group that has two points of attachment. The term “(C1-C6)alkylene” refers to an alkylene group that has from one to six carbon atoms. Non-limiting examples of alkylene groups include methylene (-CH2-), ethylene (-CH2CH2-), n-propylene (-CH2CH2CH2-), isopropylene (-CH2CH(CH3)-), and the like.

The term “arylalkoxy” refers to an alkoxy substituted with aryl.

The term “heteroaryl” refers to an aromatic 5-8 membered monocyclic, 8-12 membered bicyclic, or 11-14 membered tricyclic ring system having 1-4 ring heteroatoms if monocyclic, 1-6 heteroatoms if bicyclic, or 1-9 heteroatoms if tricyclic, said heteroatoms selected from O, N, or S, and the remainder ring atoms being carbon (with appropriate hydrogen atoms unless otherwise indicated). Heteroaryl groups may be optionally substituted with one or more substituents. In one embodiment, 0, 1, 2, 3, or 4 atoms of each ring of a heteroaryl group may be substituted by a substituent. Examples of heteroaryl groups include pyridyl, 1-oxo-pyridyl, furanyl, benzo[1,3]dioxolyl, benzo[1,4]dioxinyl, thiienyl, pyrrolyl, oxazolyl, oxadiazolyl, imidazolyl thiazolyl, isoxazolyl, quinolinyl, pyrazolyl, isoazolyl, pyridazinyl, pyrimidinyl, pyrazinyl, triazinyl, triazolyl, thiadiazolyl, isquinolinyl, indazolyl, benzoazolyl, benzofuryl, indolizynyl, imidazopyridyl, tetrazolyl, benzimidazolyl, benzothiazolyl, benzothiadiazolyl, benzoxadiazolyl, indolyl, tetrahydroindolyl, azaindolyl, imidazopyridyl, quinazolinyl, purinyl, pyrrolo[2,3]pyrimidiny1, pyrazolo[3,4]pyrimidiny1, and benzo(b)thienyl, 3H-thiazolo[2,3-c][1,2,4]thiadiazolyl,
imidazo[1,2-d]-1,2,4-thiadiazolyl, imidazo[2,1-b]-1,3,4-thiadiazolyl, 1H,2H-furo[3,4-d]-1,2,3-thiadiazolyl, 1H-pyrazolo[5,1-c]-1,2,4-triazolyl, pyrrolo[3,4-d]-1,2,3-triazolyl, cyclopentatriazolyl, 3H-pyrrolo[3,4-c]isoxazolyl, 1H,3H-pyrrolo[1,2-c]oxazolyl, pyrrolo[2,1-b]oxazolyl, and the like.

As used herein, the term "heteroaralkyl" or "heteroaryllalkyl" means a heteroaryl group that is attached to another group by a (C₁-C₆)alkylene. Heteroaralkyl groups may be optionally substituted, either on the heteroaryl portion of the heteroaralkyl group or on the alkylene portion of the heteroaralkyl group, with one or more substituents. Representative heteroaralkyl groupss include 2-(pyridin-4-yl)-propyl, 2-((thien-3-yl)-ethyl, imidazol-4-yl-methyl and the like.

The term "heterocycloalkyl" refers to a nonaromatic 3-8 membered monocyclic, 7-12 membered bicyclic, or 10-14 membered tricyclic ring system comprising 1-3 heteroatoms if monocyclic, 1-6 heteroatoms if bicyclic, or 1-9 heteroatoms if tricyclic, said heteroatoms selected from O, N, S, B, P or Si, wherein the nonaromatic ring system is completely saturated. Heterocycloalkyl groups may be optionally substituted with one or more substituents. In one embodiment, 0, 1, 2, 3, or 4 atoms of each ring of a heterocycloalkyl group may be substituted by a substituent. Representative heterocycloalkyl groups include piperidinyl, piperazinyl, 2-oxopiperazinyl, 2-oxopiperidinyl, 2-oxopyrrolidinyl, 4-piperidonyl, tetrahydropyranyl, tetrahydrothiopyranyl, tetrahydrothiopyranyl sulfone, morpholinyl, thiomorpholinyl, thiomorpholinyl sulfoxide, thiomorpholinyl sulfone, 1,3-dioxolane, tetrahydrofuranyl, tetrahydrothieryl, thiene.

The term "heterocycl" refers to a nonaromatic 5-8 membered monocyclic, 7-12 membered bicyclic, or 10-14 membered tricyclic ring system comprising 1-3 heteroatoms if monocyclic, 1-6 heteroatoms if bicyclic, or 1-9 heteroatoms if tricyclic, said heteroatoms selected from O, N, S, B, P or Si, wherein the nonaromatic ring system has some degree of unsaturation. Heterocyclyl groups may be optionally substituted with one or more substituents. In one embodiment, 0, 1, 2, 3, or 4 atoms of each ring of a heterocyclyl group may be substituted by a substituent. Examples of these groups include thiirenyl, thiadiazirinyl, dioxaoyl, 1,3-oxathioly, 1,3-dioxoly, 1,3-dithioly, oxathiazinyl, dioxazinyl, dithiazinyl, oxadiazinyl, thiadiazinyl, oxazinyl, thiazinyl, 1,4-oxathiin, 1,4-dioxin, 1,4-dithiin, 1H-pyranyl, oxathiepinyl, 5H-1,4-
dioxpiny1, 5H-1,4-dithiepinyl, 6H-isoazolo[2,3-d]1,2,4-oxadiazolyl, 7aH-oxazolo[3,2-d]1,2,4-oxadiazolyl, and the like.

The term "alkylamino" refers to an amino substituent which is further substituted with one or two alkyl groups. The term "aminoalkyl" refers to an alkyl substituent which is further substituted with one or more amino groups.

The term "hydroxyalkyl" or "hydroxylalkyl" refers to an alkyl substituent which is further substituted with one or more hydroxyl groups.

The term "sulfonylalkyl" refers to an alkyl substituent which is further substituted with one or more sulfonyl groups. The term "sulfonylaryl" refers to an aryl substituent which is further substituted with one or more sulfonyl groups.

The term "alkylcarbonyl" refers to an -C(O)-alkyl. The term "alkylcarbonylalkyl" refers to an alkyl substituent which is further substituted with -C(O)-alkyl. The alkyl or aryl portion of alkylamino, aminoalkyl, hydroxyalkyl, sulfonylaryl, sulfonylalkyl, alkylcarbonyl, and alkylcarbonylalkyl may be optionally substituted with one or more substituents.

As used herein the term "substituent" or "substituted" means that a hydrogen radical on a compound or group (such as, for example, alkyl, alkenyl, alkynyl, alkylene, aryl, aralkyl, heteroaryl, heteroaralkyl, cycloalkyl, cyclyl, heterocycloalkyl, or heterocyclyl group) is replaced with any desired group that do not substantially adversely affect the stability of the compound. In one embodiment, desired substituents are those which do not adversely affect the activity of a compound. The term "substituted" refers to one or more substituents (which may be the same or different), each replacing a hydrogen atom. Examples of substituents include, but are not limited to, halogen (F, Cl, Br, or I), hydroxyl, amino, alkylamino, arylamino, dialkylamino, diarylamino, cyano, nitro, mercapto, oxo (i.e., carbonyl), thio, imino, formyl, carbamido, carbamyl, carboxyl, thioureido, thiocyanato, sulfoamido, sulfonylalkyl, sulfonylaryl, alkyl, alkenyl, alkoxy, mercaptoalkoxy, aryl, heteroaryl, cyclyl, heterocyclyl, wherein alkyl, alkenyl, alkoxy, aryl, heteroaryl, cyclyl, and heterocyclyl are optionally substituted with alkyl, aryl, heteroaryl, halogen, hydroxyl, amino, mercapto, cyano, nitro, oxo (=O), thioxo (=S), or imino (=NR).

In other embodiments, substituents on any group (such as, for example, alkyl, alkenyl, alkynyl, aryl, aralkyl, heteroaryl, heteroaralkyl, cycloalkyl, cyclyl, heterocycloalkyl, and heterocyclyl) can be at any atom of that group, wherein any
group that can be substituted (such as, for example, alkyl, alkenyl, alkynyl, aryl, aralkyl, heteroaryl, heteroaralkyl, cycloalkyl, cycyl, heterocycloalkyl, and heterocycl) can be optionally substituted with one or more substituents (which may be the same or different), each replacing a hydrogen atom. Examples of suitable substituents include, but not limited to alkyl, alkenyl, alkynyl, cycyl, cycloalkyl, heterocycl, heterocycloalkyl, aralkyl, heteroaralkyl, aryl, heteroaryl, halogen, haloalkyl, cyano, nitro, alkoxy, aryloxy, hydroxyl, hydroxylalkyl, oxo (i.e., carbonyl), carboxyl, formyl, alkylcarbonyl, alkyloxycarbonylalkyl, alkoxycarbonyl; alkylcarbonyloxy, aryloxycarbonyl, heteroaryloxy, heteroaryloxycarbonyl, thio, mercapto, mercaptoalkyl, arylsulfonyl, amino, aminoalkyl, dialkylamino, alkylcarbonylamino, alkylaminocarbonyl, or alkoxycarbonylamino; alkylamino, arylamino, diarylamino, alkylcarbonyl, or arylamino-substituted aryl; arylalkylamino, aralkylaminocarbonyl, amido, alkylaminosulfonfyl, aryalkylaminosulfonfyl, dialkylaminosulfonfyl, alkylsulfonfylamino, arylsulfonfylamino, imino, carbamido, carbamyl, thioreido, thiocyanato, sulfoamido, sulfonfylalkyl, sulfonfylaryl, or mercaptoalkoxy.

As used herein, the term “lower” refers to a group having up to six atoms. For example, a “lower alkyl” refers to an alkyl radical having from 1 to 6 carbon atoms, and a “lower alkenyl” or “lower alkynyl” refers to an alkenyl or alkynyl radical having from 2 to 6 carbon atoms, respectively.

The term “pharmaceutically acceptable salt” also refers to a salt prepared from a compound of any one of the formulae disclosed herein having a basic functional group, such as an amino functional group, and a pharmaceutically acceptable inorganic or organic acid. Suitable acids include hydrogen sulfate, citric acid, acetic acid, oxalic acid, hydrochloric acid (HCl), hydrogen bromide (HBr), hydrogen iodide (HI), nitric acid, hydrogen bisulfide, phosphoric acid, lactic acid, salicylic acid, tartaric acid, bitartratic acid, ascorbic acid, succinic acid, maleic acid, benzoic acid, glutamic acid, fumaric acid, gluconic acid, glucaric acid, formic acid, benzoic acid, glutamic acid, methanesulfonic acid, ethanesulfonic acid, benzenesulfonic acid, and p-toluenesulfonic acid.

As used herein and unless otherwise indicated, the term “prodrug” means a derivative of a compound that can hydrolyze, oxidize, or otherwise react under biological conditions (in vitro or in vivo) to provide a compound of this invention.
Prodrugs may only become active upon such reaction under biological conditions, or they may have activity in their unreacted forms. Examples of prodrugs contemplated in this invention include, but are not limited to, analogs or derivatives of compounds of any one of the formulae disclosed herein that comprise biohydrolyzable moieties such as biohydrolyzable amides, biohydrolyzable esters, biohydrolyzable carbamates, biohydrolyzable carbonates, biohydrolyzable ureides, and biohydrolyzable phosphate analogues. Other examples of prodrugs include derivatives of compounds of any one of the formulae disclosed herein that comprise -NO, -NO₂, -ONO, or -ONO₂ moieties. Prodrugs can typically be prepared using well-known methods, such as those described by 1 BURGER’S MEDICINAL CHEMISTRY AND DRUG DISCOVERY (1995) 172-178, 949-982 (Manfred E. Wolff ed., 5th ed).

As used herein, the term “effective amount” refers to an amount of a compound of this invention which is sufficient to reduce or ameliorate the severity, duration, progression, or onset of an inflammatory disorder, immune diseases, or bone loss disease, prevent the advancement of an inflammatory disorder, immune diseases, or bone loss disease, cause the regression of an inflammatory disorder, immune diseases, or bone loss disease, prevent the recurrence, development, onset or progression of a symptom associated with an inflammatory disorder, immune diseases, or bone loss disease, or enhance or improve the prophylactic or therapeutic effect(s) of another therapy.

As used herein, a “nucleoside analog” refers to a glycoside formed by partial hydrolysis of a nucleic acid.

As used herein, a “nicotinamide adenine dinucleotide (NAD) analog” refers to a synthetic or natural compound which resembles nicotinamide adenine dinucleotide in structure and/or function.

II. Methods of Treatment

The present invention is based on the discovery that various classes of compounds that have already been demonstrated as tolerable in human patients as part of other therapies, also have potent anti-angiogenic activities. In general, the compounds of the present invention inhibit endothelial cell proliferation. In certain preferred embodiments, the anti-angiogenic activity derives at least in part from the
ability of the compound to inhibit progression through the G1/S point of the cell cycle.

In one aspect, the invention provides a method of inhibiting or otherwise reducing angiogenesis in a subject using a treatment protocol that includes administering a compound that inhibits inosine monophosphate dehydrogenase type 1 (IMPDH type 1). As described in further detail below, it has been discovered that inhibition of IMPDH type 1 in endothelial cells can prevent their proliferation, and makes IMPDH type 1 inhibitors useful as anti-angiogenic agents.

IMPDH is an enzyme in the de novo synthesis pathway of guanosine nucleotides. IMPDH catalyzes the NAD-dependent oxidation of inosine-5'-monophosphate (IMP) to xanthosine-5'-monophosphate (XMP). Two isoforms of human IMPDH have been identified. Type I is constitutive; Type II is inducible and is important in B- and T-lymphocytes.

As used herein, the terms “type” and “isoform,” when referring to IMPDH, are interchangeable, and refer to either type I and type II or isoform I and isoform II.

In certain aspects, the invention provides a method of inhibiting IMPDH type 1 in a subject, the method comprising the step of administering to the subject identified as being in need of an IMPDH type 1 inhibitor, an effective amount of an IMPDH type 1 inhibitor compound selected from: an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the subject does not experience immunosuppressive side effects associated with IMPDH type II.

In other aspects, the invention provides a method of inhibiting angiogenesis in a subject, the method comprising the step of administering to the subject identified as being in need of inhibiting angiogenesis, an effective amount of an anti-angiogenic IMPDH type 1 inhibitor compound selected from: an IMPDH type 1 inhibitor, a nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the subject does not experience immunosuppressive side effects associated with IMPDH type II.

In another aspect, the invention provides for the use of an IMPDH type 1 inhibitor compound in the manufacture of a medicament for inhibiting or reducing IMPDH type 1 in a patient, the IMPDH type 1 inhibitor compound being an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of
Formula II; trifluridine; danazol; and asparaginase; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In certain aspects, the invention provides a sustained release device for implantation in a patient and sustained release of an IMPDH type 1 inhibitor compound for at least a period of 30 days, wherein the IMPDH type 1 inhibitor compound is an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In another aspect, the invention provides a sustained release drug device adapted for implantation in or adjacent to the eye of a patient, the drug delivery device comprising: (i) an IMPDH type 1 inhibitor drug core comprising an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; (ii) an impermeable coating disposed about the core that is substantially impermeable to the passage of the IMPDH type 1 inhibitor compound, having one or more openings therein which permit diffusion of the IMPDH type 1 inhibitor compound, and which is substantially insoluble and inert in body fluids and compatible with body tissues; and, optionally, (iii) one or more permeable polymer members or coatings disposed in the flow path of the IMPDH type 1 inhibitor compound through said openings in said impermeable coating, said permeable polymer being permeable to the passage of the IMPDH type 1 inhibitor compound, and which is substantially insoluble and inert in body fluids and compatible with body tissues; wherein the impermeable coating and permeable polymer members or coatings are disposed about the drug core so as to produce, when implanted, a substantially constant rate of release of the IMPDH type 1 inhibitor compound from the device; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In another aspect, the invention provides a sustained release formulation for depot injection in a patient and sustained release of an IMPDH type 1 inhibitor compound for at least a period of 30 days, wherein the formulation includes: an viscous gel formulation comprising a bioerodible, biocompatible, polymer; and an anti-angiogenic IMPDH type 1 inhibitor agent dissolved or dispersed therein, which anti-angiogenic agent is selected from: an IMPDH type 1 inhibitor, an
nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

In other aspects, the immunosuppressive side effects in a subject or patient who has been administered an IMPDH type 1 inhibitor of the invention, are modulated relative to the subject or patient who has been administered an IMPDH type II inhibitor or a non-selective IMPDH inhibitor. In certain embodiments, the immunosuppressive side effects are decreased. In other aspects, the subject or patient who has been administered an IMPDH type 1 inhibitor of the invention does not experience immunosuppressive side effects.

In other aspects, gastrointestinal side effects in a subject or patient who has been administered an IMPDH type 1 inhibitor of the invention, are modulated relative to the subject or patient who has been administered an IMPDH type II inhibitor or a non-selective IMPDH inhibitor. In certain embodiments, the gastrointestinal side effects are decreased. In certain aspects, the subject or patient who has been administered an IMPDH type 1 inhibitor of the invention does not experience gastrointestinal side effects.

In one embodiment, the anti-angiogenic IMPDH type 1 inhibitor compound is provided in an amount effective for treatment of retinoblastoma, cystoid macular edema (CME), exudative age-related macular degeneration (AMD), diabetic retinopathy, diabetic macular edema, or ocular inflammatory disorders.

In other embodiments, the invention provides for the treatment of tumor or cancer growth (neoplasia), skin disorders, neovascularization, and inflammatory and arthritic diseases.

In other embodiments, the invention provides for the treatment of dermis, epidermis, endometrium, retina, surgical wound, gastrointestinal tract, umbilical cord, liver, kidney, reproductive system, lymphoid system, central nervous system, breast tissue, urinary tract, circulatory system, bone, muscle, or respiratory tract.

In still other embodiments, the invention provides for eliminating or reducing normal but undesired tissue in a patient. In a further embodiment, the invention provides for the reduction of fat.

In certain embodiments, the IMPDH type 1 inhibitor has a Ki for inhibiting IMPDH type 1 less than about 1 micromolar in endothelial cells.
In certain other embodiments, the IMPDH type 1 inhibitor has a Ki for inhibiting the type 1 isoform of IMPDH (IMPDH Type 1) more than 2 fold less than the Ki of the inhibitor for inhibiting the type II isoform (IMPDH type II) in endothelial cells.

In still other embodiments, the IMPDH type 1 inhibitor has a EC50 for inhibiting the type 1 isoform of IMPDH (IMPDH Type 1) more than 2 fold less than the EC50 of the inhibitor for inhibiting the type II isoform (IMPDH type II) in endothelial cells. In another embodiment, the IMPDH type 1 inhibitor has a IC50 for inhibiting the type 1 isoform of IMPDH (IMPDH Type 1) more than 2 fold less than the IC50 of the inhibitor for inhibiting the type II isoform (IMPDH type II) in endothelial cells.

In certain embodiments, the IMPDH type I is found in endothelial cells.

In other embodiments, the IMPDH type 1 inhibitor is provided in a dose that produces a serum concentration at least 50 percent less than the inhibitor’s EC50 for inhibiting IMPDH type 1 in lymphocytes. In another embodiment, the IMPDH type 1 inhibitor is provided in a dose that produces a serum concentration at least 50 percent less than the inhibitor’s IC50 for inhibiting IMPDH type 1 in lymphocytes.

In some embodiments, the IMPDH type 1 inhibitor is a mycophenolate.

In other embodiments, the IMPDH type 1 inhibitor is a nucleoside analog. In a further embodiment, the IMPDH type 1 inhibitor is a purine derivative. In another further embodiment, the purine derivative is a compound of formula III:

$$
\text{III;}
$$

wherein,

- each X is independently O, S, S(O), S(O)2, N(Rk), C(O), C(S), C(NR), C(NR)NRk, C(O)NRk, C(O)NRkNRk, C(O)O, OC(O), OC(O)O, (C(Rk)(Rk)m, (C(Rk)(Rk)mNRk, (C(Rk)(Rk)m,O, (C(Rk)(Rk)m,S(O)p, NRkC(O), NRkC(O), OC(O)NRk, or absent;

- each Rk is independently H, an optionally substituted alkyl, an optionally substituted cycloalkyl, an optionally substituted cycyl, an optionally substituted heterocycloalkyl, an optionally substituted heterocycyl, an optionally substituted aryl, an optionally substituted heteroaryl, an optionally substituted aralkyl, an optionally substituted...
substituted heteroaralkyl, halo, haloalkyl, aminoalkyl, cyano, nitro, or an optionally substituted alkylcarbonylalkyl;

\( R^8 \), for each occurrence, is independently, \( H \), an optionally substituted alkyl, an optionally substituted alkenyl, an optionally substituted alkylnyl, an optionally substituted cycyl, an optionally substituted cycloalkyl, an optionally substituted heterocyclyl, an optionally substituted heterocycloalkyl, an optionally substituted aralkyl, an optionally substituted heteroaralkyl, an optionally substituted aryl, an optionally substituted heteroaryl, haloalkyl, hydroxylalkyl, alkylcarbonylalkyl, mercaptoalkyl, aminoalkyl, sulfonylalkyl, sulfonylaryl, thioalkoxy, halo, cyano, nitro, nitroso, or azide; and

\( R^k \), for each occurrence, is independently \( H \), an optionally substituted alkyl, an optionally substituted alkenyl, an optionally substituted alkynyl, an optionally substituted cycyl, an optionally substituted cycloalkyl, an optionally substituted heterocyclyl, an optionally substituted heterocycloalkyl, an optionally substituted aralkyl, an optionally substituted heteroaralkyl, an optionally substituted aryl, or an optionally substituted heteroaryl.

In other embodiments, the IMPDH type 1 inhibitor is selected from: Ribavarin; BMS-337197; VX-497 (merimepodib); VX-148; VX-944; Viramidine; Levovirin; Mizoridine; Mizoridine glycone; benzamide riboside; selenazofurin; and Tiazofurin, or an analog thereof that is anabolized to become an NAD analog that inhibits IMPDH type 1.

In some embodiments, the IMPDH type 1 inhibitor is a nicotinamide adenine dinucleotide (NAD) analog that inhibits IMPDH type 1.

In certain embodiments, the nicotinamide adenine dinucleotide (NAD) analog is 5-ethynyl-1-β-D-ribofuranosylimidazole-4-carboxamide (EICAR) or the methylenebis(phosphonate) analogs C2-MAD and C4-MAD.

In other embodiments, the IMPDH type 1 inhibitor is an imidazole compound of formula I:

\[
\begin{array}{c}
\text{N} \\
\text{N} \\
\text{R} \\
\end{array}
\]

wherein \( R \) is selected from:
In a further embodiment, the imidazole compound is Clotrimazole, Ketoconazole, Bifonazole, Miconazole, Sulconazole, or Econazole.

In another embodiment, the IMPDH type 1 inhibitor compound is a triazole compound of formula II:

wherein, \( R_1 \) is

In a further embodiment, the triazole compound is Fluconazol, Voriconazole, Itraconazole, or Terconazole.

In certain embodiments, the IMPDH type 1 inhibitor is capable of interacting with amino acid residues Arg 253, Ile 45, and Tyr 282 of the IMPDH type 1 active site in vitro.

In other embodiments, the IMPDH type 1 compound inhibits endothelial cell proliferation.
In some embodiments, the IMPDH type 1 compound inhibits G1/S cell cycle progression of endothelial cells.

In certain embodiments, the IMPDH type 1 compound decreases new blood vessel formation.

In another embodiment, the invention provides an IMPDH type 1 compound and further comprises an additional therapeutic agent. In a further embodiment, the additional therapeutic agent is an angiogenesis-inhibiting compound. In a further embodiment, the additional therapeutic agent is an anticancer compound.

In other embodiments, the step of administering the IMPDH type 1 inhibitor compound comprises administering the compound orally, topically, parenterally, intravenously or intramuscularly. In a further embodiment, the administration is carried out in a controlled and sustained release.

In other embodiments, the step of administering the IMPDH type 1 inhibitor compound comprises administering the compound in a dosage of between about 0.1 and 100 mg/kg/day.

In some embodiments, the step of administering the IMPDH type 1 inhibitor compound comprises administering the compound in a dosage of less than about 500 mg/day.

In another aspect, the invention provides a kit comprising an effective amount of an IMPDH type 1 inhibitor compound in unit dosage form, together with instructions for administering the IMPDH type 1 inhibitor compound to a subject identified as being in need of treatment with an IMPDH type 1 inhibitor.

In one embodiment, the invention provides the step of administering an effective amount of a composition comprising an IMPDH type 1 inhibitor compound and a pharmaceutically suitable excipient to treat a disease or disorder. In a further embodiment, the disease or disorder is tumor or cancer growth (neoplasia).

In another further embodiment, the disease or disorder is: eye or ocular cancer, rectal cancer, colon cancer, cervical cancer, prostate cancer, breast cancer and bladder cancer, oral cancer, benign and malignant tumors, stomach cancer, liver cancer, pancreatic cancer, lung cancer, corpus uteri, ovary cancer, prostate cancer, testicular cancer, renal cancer, brain/cns cancer (e.g., gliomas), throat cancer, skin melanoma, acute lymphocytic leukemia, acute myelogenous leukemia, Ewing's Sarcoma, Kaposi's Sarcoma, basal cell carcinoma and squamous cell carcinoma, small cell lung
cancer, choriocarcinoma, rhabdomyosarcoma, angiosarcoma, hemangioendothelioma, Wilms Tumor, neuroblastoma, mouth/pharynx cancer, esophageal cancer, larynx cancer, lymphoma, neurofibromatosis, tuberous sclerosis, hemangiomas, and lymphangiogenesis. In a further embodiment, the disease or disorder is a skin disorder. In another further embodiment, the disease or disorder is: psoriasis, acne, rosacea, warts, eczema, hemangiomas, lymphangiogenesis, Sturge-Weber syndrome, venous ulcers, fungal ulcers, herpes simplex infections, herpes zoster infections, protozoan infections, Kaposi's sarcoma, Mooren's ulcer, Terrien's marginal degeneration, marginal keratolysis, trauma, rheumatoid arthritis, systemic lupus, polyarteritis, Wegener's sarcoidosis, scleritis, Stevens-Johnson disease, pemphigoid, radial keratotomy, corneal graft rejection, macular edema, macular degeneration, sickle cell anemia, sarcode, syphilis, pseudoxanthoma elasticum, Paget's disease, vein occlusion, artery occlusion, carotid obstructive disease, chronic uveitis/vitritis, mycobacterial infections, Lyme disease, systemic lupus erythematosus, retinopathy of prematurity, Eales' disease, Behcet's disease, infections causing a retinitis or choroiditis, presumed ocular histoplasmosis, Best's disease, myopia, optic pits, Stargardt's disease, pars planitis, chronic retinal detachment, hyperviscosity syndromes, toxoplasmosis, trauma and post-laser complications, or a disease associated with rubeosis (neovascularization of the ankle).

In a further embodiment, the disease or disorder is rheumatoid arthritis, diabetic retinopathy, macular edema, or macular degeneration.

In another embodiment, the disease or disorder is inflammatory and arthritic disease. In a further embodiment, the disease or disorder is: rheumatoid arthritis, osteoarthritis, lupus, scleroderma, Crohn's disease, ulcerative colitis, psoriasis, sarcoidosis, Sarcoidosis, skin lesions, hemangiomas, Osler-Weber-Rendu disease, hereditary hemorrhagic telangiectasia, and osteoarthritis.
In other embodiments, the subject is a human.

The methods herein include administering to the subject (including a subject identified as in need of such treatment) an effective amount of a compound described herein, or a composition described herein to produce such effect. Identifying a subject in need of such treatment can be in the judgment of a subject or a health care professional and can be subjective (e.g. opinion) or objective (e.g. measurable by a test or diagnostic method).

The preferred therapeutic methods of the invention (which include prophylactic treatment) in general comprise administration of a therapeutically effective amount of the compounds herein, such as a compound of the formulae herein to a subject (e.g., animal, human) in need thereof, including a mammal, particularly a human. Such treatment will be suitably administered to subjects, particularly humans, suffering from, having, susceptible to, or at risk for a cancer or proliferative disease, disorder, or symptom thereof. Determination of those subjects "at risk" can be made by any objective or subjective determination by a diagnostic test or opinion of a subject or health care provider (e.g., genetic test, enzyme or protein marker, Marker (as defined herein), family history, and the like).

In certain embodiments, the method further includes the step of determining a level of a marker (Marker) in the subject. In certain embodiments, the step of determining of the level of Marker is performed prior to administration of the compound of the formulae herein to the subject. In certain embodiments, the determining of the level of Marker is performed subsequent to administration of the compound of the formulae herein to the subject. In certain embodiments, the determining of the level of Marker is performed prior to and subsequent to administration of the compound of the formulae herein to the subject. In certain embodiments, the levels of Marker performed prior to and subsequent to administration of the compound of the formulae herein to the subject are compared. In certain embodiments, the comparison of Marker levels is reported by a clinic, laboratory, or hospital agent to a health care professional. In certain embodiments, when the level of Marker performed prior to administration of the compound of the formulae herein to the subject is lower or higher (depending on the Marker) than the level of Marker performed subsequent to administration of the compound of the formulae herein to the subject, then the amount of compound administered to the
subject is an effective amount. The Marker can be any characteristic or identifier, including for example, a chemical, a fluid, a protein, gene, promoter, enzyme, protein, labeled molecule, tagged molecule, antibody, and the like (e.g., Marker; IMPDH-1, IMPDH-2).

Another aspect of the invention is a compound herein (e.g., IMPDH-1 inhibitor, compound of any of the formulae herein) for use in the treatment or prevention in a subject of a disease, disorder or symptom thereof delineated herein. Another aspect of the invention is the use of an IMPDH-1 inhibitor in the manufacture of a medicament for a disease or disease symptom (e.g., cancer, tumor, any disease or disorder delineated herein) in a subject. Preferably, the medicament is used for treatment or prevention in a subject of a disease, disorder or symptom set forth above.

The IMPDH type 1 inhibitors useful in the practice of the invention preferably have high potency for inhibition of IMPDH type 1 enzyme activity. In preferred embodiments, the IMPDH type 1 inhibitors have a Ki less than about 1 micromolar, and more preferably have Ki less than about 100 nM. In the most preferred embodiments, the IMPDH type 1 inhibitors have Ki less than about 50 nM. While not wishing to be bound by any particular theory, in embodiments wherein the subject IMPDH type 1 inhibitors are used systemically, or otherwise where they are being delivered to tissue in which both isoforms are expressed, the use of inhibitors that are selective for the type I isoform may be preferred. In those embodiments, exemplary IMPDH type 1 inhibitors that can be used have a Ki that is at least 2 fold less for the type I isoform relative to the type II isoform, and more preferably at least 5 fold, 10 fold, 50 fold or even 100 fold less. Where bioavailability of the inhibitor may influence the relative potency, exemplary IMPDH type 1 inhibitors that can be used have an EC50 that is at least 2 fold less for the type I isoform relative to the type II isoform, and more preferably at least 5 fold, 10 fold, 50 fold or even 100 fold less.

In certain embodiments, the use of an IMPDH type 1 inhibitor in treatment involves dosing the patient to produce a serum or local concentration, as appropriate, that is at least 50 percent less than the EC50 for inhibiting IMPDH type 1 in lymphocytes, e.g., such that the dose is sub-immunosuppressive, and even more preferably at least 1 or even 2 orders of magnitude less than the EC50 for inhibiting IMPDH type 1 in lymphocytes.
IMPDH type 1 inhibitors that may be useful in the present invention include Ribavirin; BMS-337197; VX-497 (merimepodib); VX-148; VX-944; Viramidine; Levovirin; Mizoribine; Mizoribine aglycone; benzamide riboside, and selenazofurin. Other IMPDH type 1 inhibitors include Tiazofurin and related compounds. Tiazofurin is anabolized to become an NAD analog that inhibits IMPDH type 1. Tiazofurin may be prepared as described in U.S. Pat. No. 4,680,285 or U.S. Pat. No. 4,451,648.

Still other IMPDH type 1 inhibitors useful according to the present invention include the nicotinamide adenine dinucleotide (NAD) analogs, such as disclosed in Pankiewicz et al. (1997) Pharmacol. Ther. 76(1-3):89-100 and Pankiewicz et al. (2002) J. Med. Chem. 45(3):703-12. These analogues contain 5-beta-D-ribofuranosylnicotinamide (C-NAD), 6-beta-D-ribofuranosylpicolinamide (C-PAD), 3-beta-D-ribofuranosylbenzamide (BAD), and 2-beta-D-ribofuranosylthiazole4-carboxamide (TAD) in place of the nicotinamide riboside moiety, and are reported to have potent inhibitory activity against the enzyme in the form of pyrophosphates, as well as metabolically stable methylene- and difluoromethylene linkages of phosphonate(s). Fluorination at the C2' (ribo and arabino configuration) and C3' (ribo) of the adenosine moiety of TAD yields analogues highly potent against IMPDH type 1. Further, NAD analogues containing difluoromethylene linkage are highly effective in inhibition of K562 cell growth, as well as potent inducers of K562 cell differentiation.

In certain embodiments, the IMPDH type 1 inhibitor is 5-ethynyl-1-β-D-ribofuranosylimidazole-4-carboxamide (EICAR) or the methylenebis(phosphonate) analogs C2-MAD and C4-MAD, which can be obtained by coupling 2',3'-O-isopropylidenedenosine 5'-methylenebis(phosphonate) with mycophenolic alcohols. Imidazole and triazole IMPDH type 1 inhibitor compounds include the following:

Clotrimazole:

Ketoconazole:
Bifonazole:

Miconazole:

Sulconazole:

Econazole:

Fluconazole:

Voriconazole:

Itraconazole:
and Terconazole:

In certain embodiments, the subject IMPDH type 1 inhibitor compounds are used as part of a treatment or prevention for an optic neuropathy. The compounds can be administered, for example, by for intraocular injection or implantation. The IMPDH type 1 inhibitor compound can be administered alone, or in combination with other agents, including anti-inflammatory compounds, neuroprotective agents, agents that reduce intracocular pressure (IOP), and/or immunomodulatory compounds. For instance, the IMPDH type 1 inhibitor compound can be administered as part of therapy that includes treatment with a cholinergic agonists, cholinesterase inhibitors, carbonic anhydrase inhibitors, adrenergic agonists (such as alpha2-selective adrenergic agonists), beta-blockers, prostaglandin analogues, osmotic diuretics, p38 kinase antagonists, Cox-2 inhibitors, corticosteroid (such as triamcinolone, dexamethasone, fluocinolone, cortisone, prednisolone, flumetholone, or derivatives thereof such as triamcinolone acetonide or fluocinolone acetonide), salts thereof, isomers thereof, prodrugs thereof, and mixtures of any of these.

As used herein, the terms "optic neuropathy", or "optic neuropahties" are intended to include diseases, disorders, or damage to the nerves or other structures of the eye. By way of example, such optic neuropathies include uveitis, such as anterior uveitis, intermediate uveitis, posterior uveitis, and diffuse uveitis; uveitic syndromes, such as ankylosing spondylitis, juvenile rheumatoid arthritis, pars planitis, toxoplasmosis, cytomegalovirus, inflammation caused by herpes zoster, inflammation caused by herpes simplex, toxocariasis, birdshot chorioretinopathy, presumed ocular histoplasmosis syndrome, syphilis, tuberculosis, Vogt-Koyanagi-Harada syndrome, sympathetic ophthalmia, ocular sarcoidosis and endophthalmitis; masquerade
syndromes, such as intraocular malignancy, retinitis pigmentosa, and reactions to drugs; vascular retinopathies, such as hypertensive retinopathy, diabetic retinopathy, central retinal artery occlusion, and central retinal vein occlusion; age-related macular degeneration; retinitis pigmentosa; glaucoma; ocular hypertension; optic nerve and pathway disorders, such as papilledema, papillitis, retrobulbar neuritis, toxic amblyopia, optic atrophy, bitemporal hemianopia, and homonymous hemianopia. In certain preferred embodiments, the subject anti-angiogenic compounds are used as part of a treatment for uveitis, Diabetic Macular Edema (DME), Wet ARMD, and CMV retinitis.

There are various sustained release drug delivery devices for implantation in the eye and treating various eye diseases that can be readily adapted for delivery of the subject IMPDH type 1 inhibitor compounds. Examples are found in the following patents, the disclosures of which are incorporated herein by reference: U.S. 2005/0137583 (Renner); U.S. 2004/0219181 (Viscasillas); U.S. 2004/0265356 (Mosack); U.S. 2005/0031669 (Shafiee); U.S. 2005/0137538 (Kunzler); U.S. 2002/0086051A1 (Viscasillas); U.S. 2002/0106395A1 (Brubaker); U.S. 2002/0110591A1 (Brubaker et al.); U.S. 2002/0110592A1 (Brubaker et al.); U.S. 2002/0110635A1 (Brubaker et al.); U.S. Pat. No. 5,378,475 (Smith et al.); U.S. Pat. No. 5,773,019 (Ashton et al.); U.S. Pat. No. 5,902,598 (Chen et al.); U.S. Pat. No. 6,001,386 (Ashton et al.); U.S. Pat. No. 6,726,918 (Wong); U.S. Pat. No. 6,331,313 (Wong); U.S. Pat. No. 5,824,072 (Wong); U.S. Pat. No. 5,632,984 (Wong); U.S. Pat. No. 6,217,895 (Guo et al.); U.S. Pat. No. 6,375,972 (Guo et al.). In certain embodiments, the device include an inner drug core including the anti-angiogenic compound, and some type of holder for the drug core made of an impermeable material such as silicone or other hydrophobic materials. The holder includes one or more openings for passage of the pharmaceutically agent through the impermeable material to eye tissue. Many of these devices include at least one layer of material permeable to the active agent, such as polyvinyl alcohol.

A screening of a clinical drug library in an endothelial cell proliferation assay was carried out, and several hits were identified with IC_{50} values below 1 μM. It is demonstrated here that MPA inhibits endothelial cells by the same mechanism as it does T and B lymphocytes. Like T and B cells, treatment of endothelial cells with MPA leads to cell cycle arrest in the G1 phase, which can be reversed by addition of
guanine or guanosine, suggesting that inosine monophosphate dehydrogenase is the target. Of the two isoforms of IMPDH expressed in humans, siRNA knockdown of IMPDH-1 is sufficient to cause G1 cell cycle arrest while knockdown of IMPDH-2 led to a delay in the progression within the S phase. MPA effectively blocks new blood vessel formation in response to bFGF and VEGF in a matrigel plug assay and inhibits angiogenesis in a mouse renal cell carcinoma model in vivo.

The selectivity of MPA for T and B lymphocytes was thought to be due to dependence on the de novo nucleotide synthesis pathway for proliferation. Endothelial cells, along with smooth muscle cells, fibroblasts, and epithelial cells, were thought to be less sensitive to blockade of de novo nucleotide synthesis as inhibition of proliferation is seen at relatively high concentrations of MPA compared to T and B cells (Mohacsi, P. J., Tuller, D., Hulliger, B. & Wijngaard, P. L. (1997) J. Heart. Lung. Transplant. 16, 484-492, Eugui, E. M., Almquist, S. J., Muller, C. D. & Allison, A. C. (1991) Scand. J. Immunol. 33, 161-173). The reduced sensitivity of these cell types to MPA was attributed to use of the nucleotide salvage pathway. It is demonstrated here the unequivocal role of de novo nucleotide synthesis in regulating endothelial cell proliferation. MPA is as potent at inhibiting the proliferation of HUVEC as Jurkat T-cells (Fig. 1B vs. 1C) as well as primary T and B cells as reported previously (Eugui, E. M., Almquist, S. J., Muller, C. D. & Allison, A. C. (1991) Scand. J. Immunol. 33, 161-173). Unlike T cells, however, it was found that much higher concentrations of guanosine are required to completely reverse inhibition in HUVEC (Fig. 1B), indicating de novo nucleotide synthesis plays a more essential role in endothelial cell proliferation than in T and B cells. This difference in dependence on de novo nucleotide synthesis between endothelial and T cells raised the possibility that angiogenesis may be more susceptible to inhibition by MPA in vivo.

MPA and its prodrug form, mycophenolate mofetil, have been in clinical use for a number of years as an immunosuppressant. Its peak plasma level in human renal transplant patients undergoing chronic oral treatment is 75 μM with a half life of 18 hours, which is nearly 750-fold higher than the IC50 values for inhibition in endothelial cells (Roche Laboratories, I. (2003), 1-34). MPA has been shown to inhibit the growth of tumor cells in vitro and in mouse xenographs (Carter, S. B., Franklin, T. J., Jones, D. F., Leonard, B. J., Mills, S. D., Turner, R. W. & Turner, W.
B. (1969) *Nature* 223, 848-850). This observation led to testing of MPA in small
cohorts of patients (<35) with a variety of cancers in the 1970s (Knudtzon, S. &

As the diethanolamine salt of MPA was used rather than the currently prescribed
mofetil prodrug, considerable dose-limiting gastrointestinal toxicity occurred
the mouse Matrigel angiogenesis assay and the RENCA tumor model, it was
demonstrated that MPA blocks VEGF and bFGF stimulated angiogenesis and tumor
associated angiogenesis. This suggests that in addition to a direct anti-proliferative
effect on cancer cells, MPA can block tumor growth by inhibiting new blood vessel
formation. Given the current understanding of angiogenesis, MPA would be expected
to slow the rate of tumor progression, which was not the endpoint used in previous
(1972) *Cancer Chemother. Rep.* 56, 83-87). Furthermore, our demonstration of the *in
vivo* anti-angiogenic effect of MPA suggests this drug could potentially be used to
treat other diseases characterized by inappropriate new blood vessel formation.

Although the inhibitory effects of MPA on endothelial cells have been
observed previously, it was not clear whether the same molecular mechanism
underlies the anti-angiogenic and the immunosuppressive effects. It is established for
the first time that the anti-angiogenic activity of MPA in endothelial cells share the
same molecular basis as its immunosuppressive effects in T and B cells by multiple
criteria including G1/S cell cycle arrest and reversion of inhibition by guanine or
guanosine. More importantly, IMPDH is validated as the target for MPA in
endothelial cells using RNA interference delivered through the non-invasive lentivirus
transduction of primary endothelial cells. With isoform-specific siRNA constructs, it
was found that knockdown of IMPDH-1 is sufficient to cause the same cell cycle
effect in endothelial cells as MPA treatment. The physiological functions of both
isoforms of IMPDH have been investigated using a gene knockout approach in mice.

Of the two isoforms, IMPDH-2 appears to be more essential, as the homozygous
knockout of murine IMPDH-2 led to early embryonic lethality (Gu, J. J., Stegmann,
Cl.
Invest. 106, 599-606). In contrast, the IMPDH-1 null animal developed normally and exhibited no obvious defects (Gu, J. J., Tolin, A. K., Jain, J., Huang, H., Santiago, L. & Mitchell, B. S. (2003) Mol. Cell. Biol. 23, 6702-6712). Importantly, the function of T cells from proliferation to cytokine production was largely intact in IMPDH-1 knockout mice, suggesting that this isoform is dispensable for T cell development and function. Together, these findings raise the exciting possibility that isoform-specific inhibitors of IMPDH-1 may be selective for endothelial cells while minimally affecting the immune system and reduced or eliminated of the side effects of MPA and other existing non-selective IMPDH inhibitors.

Immunosuppressive side effects include disorders associated with metabolic disturbances such as hypertension, hyperlipidemia, loss of bone density, nephrotoxicity, diabetes, changes in appearance, gingival hyperplasia, hirsutism, alopecia, weight gain, anemia, gastrointestinal disorders and hematologic disorders. These include leukopenia, diarrhea, and vomiting. There also is an increased incidence of some infections, especially sepsis associated with cytomegalovirus. Other immunosuppressive side effects are disclosed in Fulton, B., and Markham, A. Mycophenolate mofetil. A review of its pharmacodynamic and pharmacokinetic properties and clinical efficacy in renal transplantation. Drugs, 1996, 51:278-298; and Bardsley-Elliot, A., Noble, S., and Foster, R.H. Mycophenolate mofetil: a review of its use in the management of solid organ transplantation. BioDrugs, 1999, 12:363-410; both of which are incorporated herein by reference.


This instant invention is based on the premise that there exist unappreciated physiological activities among known clinical drugs. This premise was proven by the identification of multiple known drugs with unexpected inhibitory effects on endothelial cell proliferation in vitro and angiogenesis in vivo. In addition to the endothelial cell proliferation assay, a library in a number of other cellular assays was screened. It was found that the hit rates with this drug library are significantly higher than commercially available small molecule libraries on more than half a dozen
cellular screens. The molecular basis of these high hit rates may lie in the shared genome and largely overlapping proteome of all human cell types and tissues. Significant redundancy exists in the usage of individual genes in different physiological as well as pathological processes. Thus, there is great potential in screening existing drugs for novel biological and therapeutic activities.

III. Treatment of Diseases

The term "effective amount" is used throughout the specification to describe concentrations or amounts of compounds according to the present invention which may be used to produce a favorable change in the disease or condition treated, whether that change is a remission, a decrease in growth or size of cancer, tumor or other growth, a favorable physiological result including the clearing up of skin or tissue, or the like, depending upon the disease or condition treated.

As used herein, the terms "prevent," "preventing," "prevention," and the like refer to reducing the probability of developing a disorder or condition in a subject, who does not have, but is at risk of or susceptible to developing a disorder or condition.

The term "subject" is used throughout the specification to describe an animal, preferably a human, to whom treatment, including prophylactic treatment, with the compounds according to the present invention is provided. For treatment of those infections, conditions or disease states which are specific for a specific animal such as a human patient, the term patient refers to that specific animal. In most instances, the term patient refers to a human patient.

As used herein, the terms "treat," "treating," "treatment," and the like refer to reducing or ameliorating a disorder and/or symptoms associated therewith. It will be appreciated that, although not precluded, treating a disorder or condition does not require that the disorder, condition or symptoms associated therewith be completely eliminated.

As used herein, the terms "anti-angiogenic compound" and "angiogenesis inhibiting compound" may be used interchangeably.

The term "tumor" is used to describe an abnormal growth in tissue which occurs when cellular proliferation is more rapid than normal tissue and continues to grow after the stimuli that initiated the new growth cease. Tumors generally exhibit
partial or complete lack of structural organization and functional coordination with
the normal tissue, and usually form a distinct mass of tissue which may be benign
(benign tumor) or malignant (carcinoma). Tumors tend to be highly vascularized.

The term "cancer" is used as a general term herein to describe malignant
tumors or carcinoma. These malignant tumors may invade surrounding tissues, may
metastasize to several sites and are likely to recur after attempted removal and to
cause death of the patient unless adequately treated. As used herein, the terms
carcinoma and cancer are subsumed under the term tumor. Methods of treating
tumors and/or cancer according to the present invention comprise administering to a
patient in need thereof an effective amount of one or compounds according to the
present invention.

Angiogenesis is used throughout the specification to describe the biological
processes which result in the development of blood vessels or increase in the
vascularity of tissue in an organism. Persistent, unregulated angiogenesis occurs in a
multiplicity of disease states, tumor metastasis and abnormal growth by endothelial
cells and supports the pathological damage seen in these conditions. The diverse
pathological states created due to unregulated angiogenesis have been grouped
together as angiogenic dependent or angiogenic associated diseases. Therapies
directed at control of the angiogenic processes could lead to the abrogation or
mitigation of these diseases.

Diseases or disorders treated, ameliorated or prevented by the instant
invention include the following: neoplasia, internal malignancies such as eye or ocular
cancer, rectal cancer, colon cancer, cervical cancer, prostate cancer, breast cancer and
bladder cancer, benign and malignant tumors, including various cancers such as, anal
and oral cancers, stomach, rectal, liver, pancreatic, lung, cervix uteri, corpus uteri,
ovary, prostate, testis, renal, mouth/pharynx, esophageal, larynx, kidney, brain/cns
dependent or angiogenic associated diseases. Therapies
directed at control of the angiogenic processes could lead to the abrogation or
mitigation of these diseases.

Diseases or disorders treated, ameliorated or prevented by the instant
invention include the following: neoplasia, internal malignancies such as eye or ocular
cancer, rectal cancer, colon cancer, cervical cancer, prostate cancer, breast cancer and
bladder cancer, benign and malignant tumors, including various cancers such as, anal
and oral cancers, stomach, rectal, liver, pancreatic, lung, cervix uteri, corpus uteri,
ovary, prostate, testis, renal, mouth/pharynx, esophageal, larynx, kidney, brain/cns
e.g., gliomas), head and neck, throat, skin melanoma, acute lymphocytic leukemia,
acute myelogenous leukemia, Ewing's Sarcoma, Kaposi's Sarcoma, basal cell
carinoma and squamous cell carcinoma, small cell lung cancer, choriocarcinoma,
rhabdomyosarcoma, angiosarcoma, hemangioendothelioma, Wilms Tumor,
neuroblastoma, lymphoma, neurofibromatosis, tuberous sclerosis (each of which
conditions produces benign tumors of the skin), hemangiomas, lymphangiogenesis,
rhabdomyosarcomas, retinoblastoma, osteosarcoma, acoustic neuroma, neurofibroma,
trachoma, pyogenic granulomas, blood-born tumors such as leukemias, any of various acute or chronic neoplastic diseases of the bone marrow in which unrestrained proliferation of white blood cells occurs, usually accompanied by anemia, impaired blood clotting, and enlargement of the lymph nodes, liver, and spleen, psoriasis, acne, rosacea, warts, eczema, neurofibromatosis, Sturge-Weber syndrome, venous ulcers of the skin, tuberous sclerosis, chronic inflammatory disease, arthritis, lupus, scleroderma, diabetic retinopathy, retinopathy of prematurity, corneal graft rejection, neovascular glaucoma and retrolental fibroplasias, epidemic keratoconjunctivitis; vitamin A deficiency, contact lens overwear, atopic keratitis, superior limbic keratitis, pterygium, keratitis sicca, Sjogren's, phlyctenulosis, syphilis, Mycobacteria infections, lipid degeneration, chemical burns, bacterial ulcers, fungal ulcers, herpes simplex infections, herpes zoster infections, protozoan infections, Mooren's ulcer, Terrien's marginal degeneration, marginal keratolysis, trauma, rheumatoid arthritis, systemic lupus, polyarteritis, Wegener's sarcoidosis, scleritis, Stevens-Johnson disease, pemphigoid, radial keratotomy, corneal graft rejection, diabetic retinopathy, macular edema, macular degeneration, sickle cell anemia, sarcoid, pseudoxanthoma elasticum, Paget's disease, vein occlusion, artery occlusion, carotid obstructive disease, chronic uveitis/vitritis, Lyme disease, systemic lupus erythematosus, Eales' disease, Behcet's disease, infections causing a retinitis or choroiditis, presumed ocular histoplasmosis, Best's disease, myopia, optic pits, Stargardt's disease, pars planitis, chronic retinal detachment, hyperviscosity syndromes, toxoplasmosis, trauma, post-laser complications, rubeosis (neovascularization of the ankle), diseases caused by the abnormal proliferation of fibrovascular or fibrous tissue including all forms of proliferative vitreoretinopathy, whether or not associated with diabetes, neovascular disease, pannus, diabetic macular edema, vascular retinopathy, retinal degeneration, inflammatory diseases of the retina, proliferative vitreoretinopathy, diseases associated with rubeosis (neovascularization of the ankle), diseases caused by the abnormal proliferation of fibrovascular or fibrous tissue including all forms of proliferative vitreoretinopathy, Crohn's disease and ulcerative colitis, sarcoidosis, osteoarthritis, inflammatory bowel diseases, skin lesions, Osler-Weber-Rendu disease, or hereditary hemorrhagic telangiectasia, osteoarthritis, Sarcoidosis, skin lesions, acquired immune deficiency syndrome, and small bowel obstruction.
The inhibition of angiogenesis in treating or reversing the disease state or condition is an important aspect of the present invention. More particularly, the present invention relates to methods for inhibiting the growth of neoplasia, including a malignant tumor or cancer comprising exposing the neoplasia to an inhibitory or therapeutically effective amount or concentration of at least one of the disclosed compounds. This method may be used therapeutically, in the treatment of neoplasia, including cancer or in comparison tests such as assays for determining the activities of related analogs as well as for determining the susceptibility of a patient's cancer to one or more of the compounds according to the present invention. Treatment of internal malignancies such as eye or ocular cancer, rectal cancer, colon cancer, cervical cancer, prostate cancer, breast cancer and bladder cancer, among numerous others, and oral malignancies are also contemplated by the present invention.

Angiogenesis inhibiting compounds of the present invention are used to treat, ameliorate or prevent benign and malignant tumors, including various cancers such as, cervical, anal and oral cancers, stomach, colon, bladder, rectal, liver, pancreatic, lung, breast, cervix uteri, corpus uteri, ovary, prostate, testis, renal, brain/cns (e.g., gliomas), head and neck, eye or ocular, throat, skin melanoma, acute lymphocytic leukemia, acute myelogenous leukemia, Ewing's Sarcoma, Kaposi's Sarcoma, basal cell carcinoma and squamous cell carcinoma, small cell lung cancer, choriocarcinoma, rhabdomyosarcoma, angiosarcoma, hemangioendothelioma, Wilms Tumor, neuroblastoma, mouth/pharynx, esophageal, larynx, kidney and lymphoma, among others. In addition, conditions such as neurofibromatosis, tuberous sclerosis (each of which conditions produces benign tumors of the skin), hemangiomas and lymphangiogenesis, among others, may be treated effectively with compounds according to the present invention.

Angiogenesis is prominent in solid tumor formation and metastasis. Angiogenic factors have been found associated with several solid tumors such as rhabdomyosarcomas, retinoblastoma, Ewing sarcoma, neuroblastoma, and osteosarcoma. A tumor cannot expand without a blood supply to provide nutrients and remove cellular wastes. Tumors in which angiogenesis is important include solid tumors, and benign tumors such as acoustic neuroma, neurofibroma, trachoma and pyogenic granulomas. Prevention of angiogenesis could halt the growth of these tumors and the resultant damage to the animal due to the presence of the tumor.
It should be noted that angiogenesis has been associated with blood-born tumors such as leukemias, any of various acute or chronic neoplastic diseases of the bone marrow in which unrestrained proliferation of white blood cells occurs, usually accompanied by anemia, impaired blood clotting, and enlargement of the lymph nodes, liver, and spleen. It is believed that angiogenesis plays a role in the abnormalities in the bone marrow that give rise to leukemia-like tumors.

Angiogenesis is important in two stages of tumor metastasis. The first stage where angiogenesis stimulation is important is in the vascularization of the tumor, which allows tumor cells to enter the blood stream and to circulate throughout the body. After the tumor cells have left the primary site, and have settled into the secondary, metastasis site, angiogenesis must occur before the new tumor can grow and expand. Therefore, prevention or control of angiogenesis could lead to the prevention of metastasis of tumors and possibly contain the neoplastic growth at the primary site.

Knowledge of the role of angiogenesis in the maintenance and metastasis of tumors has led to a prognostic indicator for breast cancer. The amount of neovascularization found in the primary tumor was determined by counting the microvessel density in the area of the most intense neovascularization in invasive breast carcinoma. A high level of microvessel density was found to correlate with tumor recurrence. Control of angiogenesis by therapeutic means can lead to cessation of the recurrence of the tumors.

One of the most frequent angiogenic diseases of childhood is the hemangioma. In most cases, the tumors are benign and regress without intervention. In more severe cases, the tumors progress to large cavernous and infiltrative forms and create clinical complications. Systemic forms of hemangiomas, the hemangiomatose, have a high mortality rate. Therapy-resistant hemangiomas exist that cannot be treated with therapeutics currently in use.

Angiogenic disease, angiogenic disorder and angiogenic skin disorder are used throughout the specification to describe a disorder, generally a skin disorder or related disorder which occurs as a consequence of or which results in increased vascularization in tissue. Any skin disorder which has as a primary or secondary characterization, increased vascularization, is considered an angiogenic skin disorder.
for purposes of the present invention and is amenable to treatment with compounds according to the present invention.

Methods for treating, ameliorating, or preventing angiogenic skin disorders such as psoriasis, acne, rosacea, warts, eczema, hemangiomas, lymphangiogenesis, neurofibromatosis, Sturge-Weber syndrome, venous ulcers of the skin, tuberous sclerosis, chronic inflammatory disease and arthritis, as well as inflammation such as chronic inflammatory disease, including arthritis, lupus and scleroderma are also contemplated by the present invention, such methods comprising administering a therapeutically effective amount of one or more of the disclosed compounds to a patient in need of such treatment.

Diseases associated with neovascularization include optic disc neovascularization, iris neovascularization, retinal neovascularization, choroidal neovascularization, corneal neovascularization, and intravitreal neovascularization.

Diseases associated with corneal neovascularization and retinal/choroidal neovascularization that can be treated, ameliorated, or prevented, according to the present invention include but are not limited to, diabetic retinopathy, retinopathy of prematurity, corneal graft rejection, neovascular glaucoma and retrolental fibroplasias, epidemic keratoconjunctivitis, vitamin A deficiency, contact lens overwear, atopic keratitis, superior limbic keratitis, pterygium keratitis sicca, Sjogren's, acne rosacea, phlyctenulosis, syphilis, Mycobacteria infections, lipid degeneration, chemical burns, bacterial ulcers, fungal ulcers, herpes simplex infections, herpes zoster infections, protozoan infections, Kaposi's sarcoma, Moor's ulcer, Terrien's marginal degeneration, marginal keratolysis, trauma, rheumatoid arthritis, systemic lupus, polyarteritis, Wegener's sarcoidosis, scleritis, Stevens-Johnson disease, pemphigoid, radial keratotomy, macular edema, macular degeneration, sickle cell anemia, sarcoid, syphilis, pseudoxanthoma elasticum, Paget's disease, vein occlusion, artery occlusion, carotid obstructive disease, chronic uveitis/vitritis, mycobacterial infections, Lyme disease, systemic lupus erythematosus, retinopathy of prematurity, Eales' disease, Behcet's disease, infections causing a retinitis or choroiditis, presumed ocular histoplasmosis, Best's disease, myopia, optic pits, Stargardt's disease, pars planitis, chronic retinal detachment, hyperviscosity syndromes, toxoplasmosis, trauma and post-laser complications. Other diseases include, but are not limited to, diseases associated with rubeosis (neovascularization...
of the ankle), diseases caused by the abnormal proliferation of fibrovascular or fibrous tissue including all forms of proliferative vitreoretinopathy, whether or not associated with diabetes, and corneal graft rejection.

In some embodiments, the corneal neovascularization to be treated or inhibited is caused by trauma, chemical burns or corneal transplantation. In other particular embodiments, the iris neovascularization to be treated or inhibited is caused by diabetic retinopathy, vein occlusion, ocular tumor or retinal detachment. In still other particular embodiments, the retinal or intravitreal neovascularization to be treated or inhibited is caused by diabetic retinopathy, vein occlusion, sickle cell retinopathy, retinopathy of prematurity, retinal detachment, ocular ischemia or trauma. Additional diseases associated with choroidal neovascularization to be treated or inhibited are caused by retinal or subretinal disorders of age-related macular degeneration, diabetic macular edema, presumed ocular histoplasmosis syndrome, myopic degeneration, angioid streaks or ocular trauma.

One example of a disease mediated by angiogenesis is ocular neovascular disease. This disease is characterized by invasion of new blood vessels into the structures of the eye such as the retina or cornea. It is the most common cause of blindness and is involved in approximately twenty eye diseases. In age-related macular degeneration, the associated visual problems are caused by an ingrowth of chorioidal capillaries through defects in Bruch's membrane with proliferation of fibrovascular tissue beneath the retinal pigment epithelium.

Diseases associated with chronic inflammation and arthritis can be treated, ameliorated or prevented by the compositions and methods of the present invention. Diseases with symptoms of chronic inflammation include inflammatory bowel diseases such as Crohn's disease and ulcerative colitis, psoriasis, sarcoidosis, rheumatoid arthritis, osteoarthritis, lupus and scleroderma. Angiogenesis is a key element that these chronic inflammatory diseases have in common. The chronic inflammation depends on continuous formation of capillary sprouts to maintain an influx of inflammatory cells. The influx and presence of the inflammatory cells produce granulomas and thus, maintains the chronic inflammatory state.

The compositions and methods of the present invention can be used to treat, ameliorate or prevent disease in patients with inflammatory bowel diseases such as Crohn's disease and ulcerative colitis. Both Crohn's disease and ulcerative colitis are
characterized by chronic inflammation and angiogenesis at various sites in the gastrointestinal tract. Crohn's disease is characterized by chronic granulomatous inflammation throughout the gastrointestinal tract consisting of new capillary sprouts surrounded by a cylinder of inflammatory cells. Prevention of angiogenesis by the compositions and methods of the present invention inhibits the formation of the sprouts and prevents the formation of granulomas.

Chronic inflammation may also involve pathological angiogenesis. Such disease states as ulcerative colitis and Crohn's disease show histological changes with the ingrowth of new blood vessels into the inflamed tissues. Bartonellosis, a bacterial infection found in South America, can result in a chronic stage that is characterized by proliferation of vascular endothelial cells. Another pathological role associated with angiogenesis is found in atherosclerosis. The plaques formed within the lumen of blood vessels have been shown to have angiogenic stimulatory activity.

Crohn's disease occurs as a chronic transmural inflammatory disease that most commonly affects the distal ileum and colon but may also occur in any part of the gastrointestinal tract from the mouth to the anus and perianal area. Patients with Crohn's disease generally have chronic diarrhea associated with abdominal pain, fever, anorexia, weight loss and abdominal swelling. Ulcerative colitis is also a chronic, nonspecific, inflammatory and ulcerative disease arising in the colonic mucosa and is characterized by the presence of bloody diarrhea.

The inflammatory bowel diseases also show extraintestinal manifestations such as skin lesions. Such lesions are characterized by inflammation and angiogenesis and can occur at many sites other than the gastrointestinal tract. The compositions and methods of the present invention are also capable of treating these lesions by preventing the angiogenesis, thus reducing the influx of inflammatory cells and the lesion formation.

Sarcoidosis is another chronic inflammatory disease that is characterized as a multisystem granulomatous disorder. The granulomas of this disease may form anywhere in the body and thus the symptoms depend on the site of the granulomas and whether the disease is active. The granulomas are created by the angiogenic capillary sprouts providing a constant supply of inflammatory cells.

The compositions and methods of the present invention can also treat the chronic inflammatory conditions associated with psoriasis. Psoriasis, a skin disease, is
another chronic and recurrent disease that is characterized by papules and plaques of various sizes. Prevention of the formation of the new blood vessels necessary to maintain the characteristic lesions leads to relief from the symptoms.

Another disease (or symptoms thereof) which can be treated, ameliorated or prevented according to the present invention is rheumatoid arthritis. Rheumatoid arthritis is a chronic inflammatory disease characterized by nonspecific inflammation of the peripheral joints. It is believed that the blood vessels in the synovial lining of the joints undergo angiogenesis. In addition to forming new vascular networks, the endothelial cells release factors and reactive oxygen species that lead to pannus growth and cartilage destruction. The factors involved in angiogenesis may actively contribute to, and help maintain, the chronically inflamed state of rheumatoid arthritis. Another disease that can be treated according to the present invention are Osler-Weber-Rendu disease, or hereditary hemorrhagic telangiectasia, and acquired immune deficiency syndrome.

Factors associated with angiogenesis may also have a role in osteoarthritis. The activation of the chondrocytes by angiogenic-related factors contributes to the destruction of the joint. At a later stage, the angiogenic factors would promote new bone formation. Therapeutic intervention that prevents the bone destruction could halt the progress of the disease and provide relief for persons suffering with arthritis.

Angiogenesis is also responsible for damage found in hereditary diseases such as Osler-Weber-Rendu disease, or hereditary hemorrhagic telangiectasia. This is an inherited disease characterized by multiple small angiomas, tumors of blood or lymph vessels. The angiomas are found in the skin and mucous membranes, often accompanied by epistaxis (nosebleeds) or gastrointestinal bleeding and sometimes with pulmonary or hepatic arteriovenous fistula.

Angiogenesis is also involved in normal physiological processes such as reproduction and wound healing. Angiogenesis is an important step in ovulation and also in implantation of the blastula after fertilization. Prevention of angiogenesis could be used to induce amenorrhea, to block ovulation or to prevent implantation by the blastula, thereby preventing conception.

In wound healing, excessive repair or fibroplasia can be a detrimental side effect of surgical procedures and may be caused or exacerbated by angiogenesis.
Adhesions are a frequent complication of surgery and lead to problems such as small bowel obstruction.

The present compounds may be used to treat subjects including animals, and in particular, mammals, including humans, as patients. Thus, humans and other animals, and in particular, mammals, suffering from diseases or disorders related to angiogenesis, can be treated, ameliorated or prevented by administering to the patient an effective amount of one or more of the compounds according to the present invention or its derivative or a pharmaceutically acceptable salt thereof optionally in a pharmaceutically acceptable carrier or diluent, either alone, or in combination with other known pharmaceutical agents (depending upon the disease to be treated). Treatment according to the present invention can also be administered in conjunction with other conventional therapies, e.g., cancer therapy, such as radiation treatment or surgery.

IV. Pharmaceutical Compositions/Methods of Administration

The present invention is also directed to pharmaceutical compositions comprising an effective amount of one or more compounds according to the present invention (including a pharmaceutically acceptable salt, thereof), optionally in combination with a pharmaceutically acceptable carrier, excipient or additive.

A “pharmaceutically acceptable derivative or prodrug” means any pharmaceutically acceptable salt, ester, salt of an ester, or other derivative of a compound of this invention which, upon administration to a recipient, is capable of providing (directly or indirectly) a compound of this invention. Particularly favored derivatives and prodrugs are those that increase the bioavailability of the compounds of this invention when such compounds are administered to a mammal (e.g., by allowing an orally administered compound to be more readily absorbed into the blood) or which enhance delivery of the parent compound to a biological compartment (e.g., the brain or lymphatic system) relative to the parent species.

While the angiogenesis inhibiting compounds of the invention can be administered as the sole active pharmaceutical agent, they can also be used in combination with one or more compounds of the invention or other agents. When administered as a combination, the therapeutic agents can be formulated as separate
compositions that are given at the same time or different times, or the therapeutic agents can be given as a single composition.

The angiogenesis inhibiting compounds of the present invention may be administered orally, parenterally, by inhalation spray, rectally, vaginally, or topically in dosage unit formulations containing conventional pharmaceutically acceptable carriers, adjuvants, and vehicles. The term parenteral as used herein includes, subcutaneous, intravenous, intramuscular, intrasternal, infusion techniques, intraperitoneally, eye or ocular, intrabuccal, transdermal, intranasal, into the brain, including intracranial and intradural, into the joints, including ankles, knees, hips, shoulders, elbows, wrists, directly into tumors, and the like, and in suppository form.

The pharmaceutically active compounds of this invention can be processed in accordance with conventional methods of pharmacy to produce medicinal agents for administration to patients, including humans and other mammals.

Modifications of the active compound can affect the solubility, bioavailability and rate of metabolism of the active species, thus providing control over the delivery of the active species. Further, the modifications can affect the anti-angiogenesis activity of the compound, in some cases increasing the activity over the parent compound. This can easily be assessed by preparing the derivative and testing its activity according to known methods well within the routineer's skill in the art.

Pharmaceutical compositions based upon these chemical compounds comprise the above-described compounds in a therapeutically effective amount for treating diseases and conditions which have been described herein, optionally in combination with a pharmaceutically acceptable additive, carrier and/or excipient. One of ordinary skill in the art will recognize that a therapeutically effective amount of one of more compounds according to the present invention will vary with the infection or condition to be treated, its severity, the treatment regimen to be employed, the pharmacokinetics of the agent used, as well as the patient (animal or human) treated.

To prepare the pharmaceutical compositions according to the present invention, a therapeutically effective amount of one or more of the compounds according to the present invention is preferably intimately admixed with a pharmaceutically acceptable carrier according to conventional pharmaceutical compounding techniques to produce a dose. A carrier may take a wide variety of forms depending on the form of preparation desired for administration, e.g., oral,
topical or parenteral, including gels, creams, ointments, lotions and time released implantable preparations, among numerous others. In preparing pharmaceutical compositions in oral dosage form, any of the usual pharmaceutical media may be used. Thus, for liquid oral preparations such as suspensions, elixirs and solutions, suitable carriers and additives including water, glycols, oils, alcohols, flavouring agents, preservatives, colouring agents and the like may be used. For solid oral preparations such as powders, tablets, capsules, and for solid preparations such as suppositories, suitable carriers and additives including starches, sugar carriers, such as dextrose, mannitol, lactose and related carriers, diluents, granulating agents, lubricants, binders, disintegrating agents and the like may be used. If desired, the tablets or capsules may be enteric-coated or sustained release by standard techniques.

The active compound is included in the pharmaceutically acceptable carrier or diluent in an amount sufficient to deliver to a patient a therapeutically effective amount for the desired indication, without causing serious toxic effects in the patient treated.

Oral compositions will generally include an inert diluent or an edible carrier. They may be enclosed in gelatin capsules or compressed into tablets. For the purpose of oral therapeutic administration, the active compound or its prodrug derivative can be incorporated with excipients and used in the form of tablets, troches, or capsules. Pharmaceutically compatible binding agents, and/or adjuvant materials can be included as part of the composition.

The tablets, pills, capsules, troches and the like can contain any of the following ingredients, or compounds of a similar nature: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a dispersing agent such as alginic acid or corn starch; a lubricant such as magnesium stearate; a glidant such as colloidal silicon dioxide; a sweetening agent such as sucrose or saccharin; or a flavoring agent such as peppermint, methyl salicylate, or orange flavoring. When the dosage unit form is a capsule, it can contain, in addition to material of the above type, a liquid carrier such as a fatty oil. In addition, dosage unit forms can contain various other materials which modify the physical form of the dosage unit; for example, coatings of sugar, shellac, or enteric agents.
Formulations of the present invention 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 emulsion and as a bolus, etc.

A tablet 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 a binder, lubricant, inert diluent, preservative, surface-active or dispersing agent. 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 optionally may be coated or scored and may be formulated so as to provide slow or controlled release of the active ingredient therein.

Methods of formulating such slow or controlled release compositions of pharmaceutically active ingredients, are known in the art and described in several issued US Patents, some of which include, but are not limited to, US Patent Nos. 3,870,790; 4,226,859; 4,369,172; 4,842,866 and 5,705,190, the disclosures of which are incorporated herein by reference in their entireties. Coatings can be used for delivery of compounds to the intestine (see, e.g., U.S. Patent Nos. 6,638,534, 5,541,171, 5,217,720, and 6,569,457, and references cited therein).

The active compound or pharmaceutically acceptable salt thereof may also be administered as a component of an elixir, suspension, syrup, wafer, chewing gum or the like. A syrup may contain, in addition to the active compounds, sucrose or fructose as a sweetening agent and certain preservatives, dyes and colorings and flavors.

Solutions or suspensions used for parenteral, intradermal, subcutaneous, or topical application can include the following components: a sterile diluent such as water for injection, saline solution, fixed oils, polyethylene glycols, glycerine, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl parabens; antioxidants such as ascorbic acid or sodium bisulfite; chelating agents such as ethylenediaminetetraacetic acid; buffers such as acetates, citrates or phosphates and agents for the adjustment of tonicity such as sodium chloride or dextrose.
In one embodiment, the active compounds are prepared with carriers that will protect the compound against rapid elimination from the body, such as a controlled release formulation, including implants and microencapsulated delivery systems. Biodegradable, biocompatible polymers can be used, such as ethylene vinyl acetate, polyanhydrides, polyglycolic acid, collagen, polyorthoesters, and polylactic acid. Methods for preparation of such formulations will be apparent to those skilled in the art.

A skilled artisan will recognize that in addition to tablets, other dosage forms can be formulated to provide slow or controlled release of the active ingredient. Such dosage forms include, but are not limited to, capsules, granulations and gel-caps.

Liposomal suspensions may also be pharmaceutically acceptable carriers. These may be prepared according to methods known to those skilled in the art. For example, liposomal formulations may be prepared by dissolving appropriate lipid(s) in an inorganic solvent that is then evaporated, leaving behind a thin film of dried lipid on the surface of the container. An aqueous solution of the active compound are then introduced into the container. The container is then swirled by hand to free lipid material from the sides of the container and to disperse lipid aggregates, thereby forming the liposomal suspension. Other methods of preparation well known by those of ordinary skill may also be used in this aspect of the present invention.

The formulations may conveniently be presented in unit dosage form and may be prepared by conventional pharmaceutical techniques. Such techniques include the step of bringing into association the active ingredient and the pharmaceutical carrier(s) or excipient(s). In general, the formulations are prepared by uniformly and intimately bringing into association the active ingredient with liquid carriers or finely divided solid carriers or both, and then, if necessary, shaping the product.

Formulations and compositions suitable for topical administration in the mouth include lozenges comprising the ingredients in a flavored basis, usually sucrose and acacia or tragacanth; pastilles comprising the active ingredient in an inert basis such as gelatin and glycerin, or sucrose and acacia; and mouthwashes comprising the ingredient to be administered in a suitable liquid carrier.

Formulations suitable for topical administration to the skin may be presented as ointments, creams, gels and pastes comprising the ingredient to be administered in
a pharmaceutical acceptable carrier. A preferred topical delivery system is a transdermal patch containing the ingredient to be administered.

Formulations for rectal administration may be presented as a suppository with a suitable base comprising, for example, cocoa butter or a salicylate.

Formulations suitable for nasal administration, wherein the carrier is a solid, include a coarse powder having a particle size, for example, in the range of 20 to 500 microns which is administered in the manner in which snuff is administered, i.e., by rapid inhalation through the nasal passage from a container of the powder held close up to the nose. Suitable formulations, wherein the carrier is a liquid, for administration, as for example, a nasal spray or as nasal drops, include aqueous or oily solutions of the active ingredient.

Formulations suitable for vaginal administration may be presented as pessaries, tampons, creams, gels, pastes, foams or spray formulations containing in addition to the active ingredient such carriers as are known in the art to be appropriate.

The parenteral preparation can be enclosed in ampoules, disposable syringes or multiple dose vials made of glass or plastic. If administered intravenously, preferred carriers include, for example, physiological saline or phosphate buffered saline (PBS).

For parenteral formulations, the carrier will usually comprise sterile water or aqueous sodium chloride solution, though other ingredients including those which aid dispersion may be included. Of course, where sterile water is to be used and maintained as sterile, the compositions and carriers must also be sterilized. Injectable suspensions may also be prepared, in which case appropriate liquid carriers, suspending agents and the like may be employed.

Formulations suitable for parenteral administration include aqueous and non-aqueous sterile injection solutions 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. The formulations may be presented in unit-dose or multi-dose containers, for example, sealed ampules and vials, and may be stored in a freeze-dried (lyophilized) condition requiring only the addition of the sterile liquid carrier, for example, water for injections, immediately prior to use.
Extemporaneous injection solutions and suspensions may be prepared from sterile powders, granules and tablets of the kind previously described.

Administration of the active compound may range from continuous (intravenous drip) to several oral administrations per day (for example, Q.I.D.) and may include oral, topical, eye or ocular, parenteral, intramuscular, intravenous, subcutaneous, transdermal (which may include a penetration enhancement agent), buccal and suppository administration, among other routes of administration, including through an eye or ocular route.

Application of the subject therapeutics may be local, so as to be administered at the site of interest. Various techniques can be used for providing the subject compositions at the site of interest, such as injection, use of catheters, trocars, projectiles, pluronic gel, stents, sustained drug release polymers or other device which provides for internal access. Where an organ or tissue is accessible because of removal from the patient, such organ or tissue may be bathed in a medium containing the subject compositions, the subject compositions may be painted onto the organ, or may be applied in any convenient way.

The angiogenesis-inhibiting compound may be administered through a device suitable for the controlled and sustained release of a composition effective in obtaining a desired local or systemic physiological or pharmacological effect. The method includes positioning the sustained released drug delivery system at an area wherein release of the agent is desired and allowing the agent to pass through the device to the desired area of treatment.

More specifically, the angiogenesis-inhibiting compound is administered through an ocular device suitable for direct implantation into the vitreous of the eye. Such devices of the present invention are surprisingly found to provide sustained controlled release of various compositions to treat the eye without risk of detrimental local and systemic side effects. An object of the present ocular method of delivery is to maximize the amount of drug contained in an intraocular device while minimizing its size in order to prolong the duration of the implant. See, e.g., U.S. Patents 5,378,475; 5,773,019; 6,001,386; 6,217,895, 6,375,972, and 6,756,058 and U.S. Publications 20050096290 and 200501269448.

Other methods of delivery include: an ocular delivery system that could be applied to an intra-ocular lens to prevent inflammation or posterior capsular
opacification, an ocular delivery system that could be inserted directly into the vitreous, under the retina, or onto the sclera, and wherein inserting can be achieved by injecting the system or surgically implanting the system, a sustained release drug delivery system, and a method for providing controlled and sustained administration of an agent effective in obtaining a desired local or systemic physiological or pharmacological effect comprising surgically implanting a sustained release drug delivery system at a desired location.

Examples include, but are not limited to the following: a sustained release drug delivery system comprising an inner reservoir comprising an effective amount of an agent effective in obtaining a desired local or systemic physiological or pharmacological effect, an inner tube impermeable to the passage of said agent, said inner tube having first and second ends and covering at least a portion of said inner reservoir, said inner tube sized and formed of a material so that said inner tube is capable of supporting its own weight, an impermeable member positioned at said inner tube first end, said impermeable member preventing passage of said agent out of said reservoir through said inner tube first end, and a permeable member positioned at said inner tube second end, said permeable member allowing diffusion of said agent out of said reservoir through said inner tube second end; a method for administering a compound of the invention to a segment of an eye, the method comprising the step of implanting a sustained release device to deliver the compound of the invention to the vitreous of the eye or an implantable, sustained release device for administering a compound of the invention to a segment of an eye; a sustained release drug delivery device comprising: a) a drug core comprising a therapeutically effective amount of at least one first agent effective in obtaining a diagnostic effect or effective in obtaining a desired local or systemic physiological or pharmacological effect; b) at least one unitary cup essentially impermeable to the passage of said agent that surrounds and defines an internal compartment to accept said drug core, said unitary cup comprising an open top end with at least one recessed groove around at least some portion of said open top end of said unitary cup; c) a permeable plug which is permeable to the passage of said agent, said permeable plug is positioned at said open top end of said unitary cup wherein said groove interacts with said permeable plug holding it in position and closing said open top end, said permeable plug allowing passage of said agent out of said drug core, through said permeable plug, and out said open top end of
said unitary cup; and d) at least one second agent effective in obtaining a diagnostic effect or effective in obtaining a desired local or systemic physiological or pharmacological effect; or a sustained release drug delivery device comprising: an inner core comprising an effective amount of an agent having a desired solubility and a polymer coating layer, the polymer layer being permeable to the agent, wherein the polymer coating layer completely covers the inner core.

The methods are particularly suitable for treating ocular conditions such as glaucoma, proliferative vitreoretinopathy, macular edema, including diabetic macular edema, age-related macular degeneration, diabetic retinopathy, uveitis, ocular neovascularization, and ocular infection. The devices are also particularly suitable for use as an ocular device in treating subjects suffering from ocular histoplasmosis, wherein the device is surgically implanted within the vitreous of the eye.

The angiogenesis-inhibiting compound may be utilized in combination with at least one known other therapeutic agent, or a pharmaceutically acceptable salt of said agent. Examples of known therapeutic agents which can be used for combination therapy include, but are not limited to, corticosteroids (e.g., cortisone, prednisone, dexamethasone), non-steroidal anti-inflammatory drugs (NSAIDS) (e.g., ibuprofen, celecoxib, aspirin, indomethicin, naproxen), alkylating agents such as busulfan, cisplatin, mitomycin C, and carboplatin; antimitotic agents such as colchicine, vinblastine, paclitaxel, and docetaxel; topo I inhibitors such as camptothecin and topotecan; topo II inhibitors such as doxorubicin and etoposide; RNA/DNA antimetabolites such as 5-azacytidine, 5-fluorouracil and methotrexate; DNA antimetabolites such as 5-fluoro-2'-deoxy-uridine, ara-C, hydroxyurea and thioguanine; antibodies such as Herceptin® and Rituxan®. Other known anti-cancer agents which can be used for combination therapy include melphalan, chlorambucil, cyclophosphamide, ifosfamide, vincristine, mitoguazone, epirubicin, aclacinubicin, bleomycin, mitoxantrone, elliptinium, fludarabine, octreotide, retinoic acid, tamoxifen and alanosome.

The active compound or pharmaceutically acceptable salts thereof can also be mixed with other active materials that do not impair the desired action, or with materials that supplement the desired action, such as other anticancer agents, and in certain instances depending upon the desired therapy or target, antibiotics,
antifungals, antinflammatories, antiviral compounds or other agents having a distinct pharmacological effect.

Alternatively, the compound of the invention may be administered apart from the at least one known cancer chemotherapeutic agent. In one embodiment, the compound of the invention and the at least one known cancer chemotherapeutic agent are administered substantially simultaneously, i.e., the compounds are administered at the same time or one after the other, so long as the compounds reach therapeutic levels in the blood at the same time. On another embodiment, the compound of the invention and the at least one known cancer chemotherapeutic agent are administered according to their individual dose schedule, so long as the compounds reach therapeutic levels in the blood.

It should be understood that in addition to the ingredients particularly mentioned above, the formulations of the present invention may include other agents conventional in the art having regard to the type of formulation in question, for example, those suitable for oral administration may include flavoring agents.

In certain pharmaceutical dosage forms, the pro-drug form of the compounds may be preferred. One of ordinary skill in the art will recognize how to readily modify the present compounds to pro-drug forms to facilitate delivery of active compounds to a targeted site within the host organism or patient. The routinee also will take advantage of favorable pharmacokinetic parameters of the pro-drug forms, where applicable, in delivering the present compounds to a targeted site within the host organism or patient to maximize the intended effect of the compound.

189-214. The prodrug forms may be active themselves, or may be those such that when metabolized after administration provide the active therapeutic agent in vivo.

Pharmacologically acceptable salt forms may be the preferred chemical form of compounds according to the present invention for inclusion in pharmaceutical compositions according to the present invention.

Certain of the compounds, in pharmaceutical dosage form, may be used as agents for preventing a disease or condition from manifesting itself. In certain pharmaceutical dosage forms, the pro-drug form of the compounds according to the present invention may be preferred. In particular, prodrug forms which rely on C₁ to C₂₀ ester groups or amide groups (preferably a hydroxyl, free amine or substituted nitrogen group) which may be transformed into, for example, an amide or other group may be particularly useful in this context.

The present compounds or their derivatives, including prodrug forms of these agents, can be provided in the form of pharmaceutically acceptable salts. As used herein, the term pharmaceutically acceptable salts or complexes refers to appropriate salts or complexes of the active compounds according to the present invention which retain the desired biological activity of the parent compound and exhibit limited toxicological effects to normal cells. Nonlimiting examples of such salts are (a) acid addition salts formed with inorganic acids (for example, hydrochloric acid, hydrobromic acid, sulfuric acid, phosphoric acid, nitric acid, and the like), and salts formed with organic acids such as acetic acid, oxalic acid, tartaric acid, succinic acid, malic acid, ascorbic acid, benzoic acid, tannic acid, pamoic acid, alginic acid, and polyglutamic acid, among others; (b) base addition salts formed with metal cations such as zinc, calcium, sodium, potassium, and the like, among numerous others.

The compounds herein are commercially available or can be synthesized. As can be appreciated by the skilled artisan, further methods of synthesizing the compounds of the formulae herein will be evident to those of ordinary skill in the art. Additionally, the various synthetic steps may be performed in an alternate sequence or order to give the desired compounds. Synthetic chemistry transformations and protecting group methodologies (protection and deprotection) useful in synthesizing the compounds described herein are known in the art and include, for example, those such as described in R. Larock, Comprehensive Organic Transformations, 2nd. Ed., Wiley-VCH Publishers (1999); T.W. Greene and P.G.M. Wuts, Protective Groups in

The compounds of this invention may contain one or more asymmetric centers and thus occur as racemates and racemic mixtures, single enantiomers, individual diastereomers and diastereomeric mixtures. All such isomeric forms of these compounds are expressly included in the present invention. The compounds of this invention may also be represented in multiple tautomeric forms, in such instances, the invention expressly includes all tautomeric forms of the compounds described herein (e.g., alkylation of a ring system may result in alkylation at multiple sites, the invention expressly includes all such reaction products). All such isomeric forms of such compounds are expressly included in the present invention. All crystal forms of the compounds described herein are expressly included in the present invention.

Preferred unit dosage formulations are those containing a daily dose or unit, daily sub-dose, as hereinabove recited, or an appropriate fraction thereof, of the administered ingredient.

The dosage regimen for treating a disorder or a disease with the angiogenesis inhibiting compounds of this invention and/or compositions of this invention is based on a variety of factors, including the type of disease, the age, weight, sex, medical condition of the patient, the severity of the condition, the route of administration, and the particular compound employed. Thus, the dosage regimen may vary widely, but can be determined routinely using standard methods.

The amounts and dosage regimens administered to a subject will depend on a number of factors, such as the mode of administration, the nature of the condition being treated, the body weight of the subject being treated and the judgment of the prescribing physician.

The amount of compound included within therapeutically active formulations according to the present invention is an effective amount for treating the infection or condition. In general, a therapeutically effective amount of the present preferred compound in dosage form usually ranges from slightly less than about 0.025 mg/kg/day to about 2.5 g/kg/day, preferably about 0.1 mg/kg/day to about 100 mg/kg/day of the patient or considerably more, depending upon the compound used,
the condition or infection treated and the route of administration, although exceptions
to this dosage range may be contemplated by the present invention. In its most
preferred form, compounds according to the present invention are administered in
amounts ranging from about 1 mg/kg/day to about 100 mg/kg/day. The dosage of the
compound will depend on the condition being treated, the particular compound, and
other clinical factors such as weight and condition of the patient and the route of
administration of the compound. It is to be understood that the present invention has
application for both human and veterinary use.

For oral administration to humans, a dosage of between approximately 0.1 to
100 mg/kg/day, preferably between approximately 1 and 100 mg/kg/day, is generally
sufficient.

Where drug delivery is systemic rather than topical, this dosage range
generally produces effective blood level concentrations of active compound ranging
from less than about 0.04 to about 400 micrograms/cc or more of blood in the patient.

The compound is conveniently administered in any suitable unit dosage form,
including but not limited to one containing 1 to 3000 mg, preferably 5 to 500 mg of
active ingredient per unit dosage form. An oral dosage of 10-250 mg is usually
convenient.

The concentration of active compound in the drug composition will depend on
absorption, distribution, inactivation, and excretion rates of the drug as well as other
factors known to those of skill in the art. It is to be noted that dosage values will also
vary with the severity of the condition to be alleviated. It is to be further understood
that for any particular subject, specific dosage regimens should be adjusted over time
according to the individual need and the professional judgment of the person
administering or supervising the administration of the compositions, and that the
concentration ranges set forth herein are exemplary only and are not intended to limit
the scope or practice of the claimed composition. The active ingredient may be
administered at once, or may be divided into a number of smaller doses to be
administered at varying intervals of time.

In certain embodiments, the compound is administered once daily; in other
embodiments, the compound is administered twice daily; in yet other embodiments,
the compound is administered once every two days, once every three days, once every
four days, once every five days, once every six days, once every seven days, once
every two weeks, once every three weeks, once every four weeks, once every two months, once every six months, or once per year. The dosing interval can be adjusted according to the needs of individual patients. For longer intervals of administration, extended release or depot formulations can be used.

The compounds of the invention can be used to treat diseases and disease conditions that are acute, and may also be used for treatment of chronic conditions. In certain embodiments, the compounds of the invention are administered for time periods exceeding two weeks, three weeks, one month, two months, three months, four months, five months, six months, one year, two years, three years, four years, or five years, ten years, or fifteen years; or for example, any time period range in days, months or years in which the low end of the range is any time period between 14 days and 15 years and the upper end of the range is between 15 days and 20 years (e.g., 4 weeks and 15 years, 6 months and 20 years). In some cases, it may be advantageous for the compounds of the invention to be administered for the remainder of the patient's life. In preferred embodiments, the patient is monitored to check the progression of the disease or disorder, and the dose is adjusted accordingly. In preferred embodiments, treatment according to the invention is effective for at least two weeks, three weeks, one month, two months, three months, four months, five months, six months, one year, two years, three years, four years, or five years, ten years, fifteen years, twenty years, or for the remainder of the subject's life.

Still other objects, features, and attendant advantages of the present invention will become apparent to those skilled in the art from a reading of the preceding detailed description of embodiments constructed in accordance therewith, taken in conjunction with the accompanying drawings.

The invention also provides a pharmaceutical pack or kit comprising one or more containers filled with one or more of the ingredients of the pharmaceutical compositions of the invention. Associated with such container(s) can be a notice in the form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceuticals or biological products, which notice reflects approval by the agency of manufacture, use or sale for human administration. In addition, the polypeptide of the present invention may be employed on conjunction with other therapeutic compounds.
The invention also provides kits for treatment or prevention of a disease or disorder (or symptoms) thereof associated with angiogenesis. In one embodiment, the kit includes an effective amount of an angiogenesis-inhibiting compound in unit dosage form, together with instructions for administering the angiogenesis-inhibiting compound to a subject suffering from or susceptible to a disease or disorder or symptoms thereof associated with angiogenesis, wherein the effective amount of an angiogenesis-inhibiting compound is less than 500 mg of the compound. In preferred embodiments, the kit comprises a sterile container which contains the angiogenesis-inhibiting compound; such containers can be boxes, ampules, bottles, vials, tubes, bags, pouches, blister-packs, or other suitable container form known in the art. Such containers can be made of plastic, glass, laminated paper, metal foil, or other materials suitable for holding medicaments. The instructions will generally include information about the use of the angiogenesis-inhibiting compound for treatment of a disease or disorder or symptoms thereof associated with angiogenesis; in preferred embodiments, the instructions include at least one of the following: description of the angiogenesis-inhibiting compound; dosage schedule and administration for treatment of a disease or disorder or symptoms thereof associated with angiogenesis; precautions; warnings; indications; counter-indications; overdose information; adverse reactions; animal pharmacology; clinical studies; and/or references. The instructions may be printed directly on the container (when present), or as a label applied to the container, or as a separate sheet, pamphlet, card, or folder supplied in or with the container.

The invention will be further described in the following examples. It should be understood that these examples are for illustrative purposes only and are not to be construed as limiting this invention in any manner.

**EXAMPLES**

**Materials and Methods**

**Library construction.** 32,000 FDA drug approvals obtained by Freedom of Information Act requests were condensed to 3,400 unique drug formulations. Drugs were purchased from Sigma, Spectrum Chemicals, MP Biomedicals, The Johns Hopkins Hospital pharmacy, Biomol, and Tocris. 10 mM stock solutions were made
using DMSO, water, or ethanol as solvents. Drugs were arrayed in 96-well plates and screened at a final concentration of 10 μM. Cells were incubated with drug for 36 h and proliferation was measured by pulsing with 1 μCi [³H]- thymidine for 8 h followed by absorption onto glass filters and scintillation counting.

**Statistical analysis.** IC₅₀ measurements were carried out in triplicate, and analyzed using four parameter logarithmic analysis. Values are presented +/- the standard error of the mean. P-values were calculated using the Student’s T-test. IC₅₀ measurements are carried out using standard assays, and include assays described in Gilbert HJ, Lowe CR, Drabble WT. Biochem J. 1979,183:481-94; and Xiang, B., Taylor, J. C., and Markham, G. D. J. Biol. Chem., 271, 1435 (1996); the contents of which are incorporated herein by reference.

**Semi-quantitative RT-PCR.** RNA from subconfluent proliferating HUVEC (1 x 10⁶) cells was prepared using a commercial kit (Roche) and 1 μg of total RNA was used in the reverse transcription reaction (SuperScript II, Invitrogen). cDNA was amplified by incorporating equal amounts of gene specific primers and Taq polymerase (Promega). 15 μL of this reaction was visualized on a 1% agarose gel containing 1:40,000 ethidium bromide.

**IMPDH-1 and -2 siRNA knockdown.** pLVV-H1 lentivirus vector was a modified version based on our previous pFUP-U6 lentivirus vector (Pan, F., Ye, Z., Cheng, L. & Liu, J. O. (2004) J. Biol. Chem. 279, 14477-14480.). It was named double-copy design of siRNA lentivirus vector. Among the features of the new vector are, (1) the vector contains an internal marker EGFP under the control of CMV promoter; (2) a cassette containing H1 promoter was cloned in the 3' U3 region of pFUP. During reverse transcription, the U3 region of the 5' LTR is synthesized by using its 3' homologue as a template, which results in a duplication of the siRNA cassette in the provirus integrated in the genome of transduced cells. For the RNA interference of IMPDH, oligos were designed and tested for three different regions of each IMPDH isoform. The best one was chosen. The oligonucleotide sequences for shRNA knockdown are as follows. For IMPDH-1, Sense: 5'-
CTATCTTCAGTGCTTACttcaagagaGTAAGACCACTGAAGATAGTTTTTT-3'
(The first 19 nucleotides correspond to 544-562 of human IMPDH-1 cDNA
(NM_00883.2), which is followed by a 9-nucleotide linker (small letters), the
complementary sequence and five T's). Antisense: 5'-
AAAAAAACTATCTTCAGTGCTTACttctttgaaGTAAGACCACTGAAGATAGG
GCC-3'.

For IMPDH-2, Sense: 5'-
GATGATGAGCTTGTGGCCAttcagagaTGGCCACAAGCTCATCCTTTTTT-3'
(The first 19 nucleotides correspond to 683-701 of human IMPDH-2 cDNA
(BC006124.1)). Antisense: 5'-
AAAAAAAGATGATGAGCTTGTGGCCAttcctttgaaTGGCCACAAGCTCATCCTCAG
GCC-3'.

293T cell line was cultured in DMEM medium supplemented with 10% FBS.
Recombinant lentiviruses were produced by transient transfection of 293T cells as
follows. Briefly, subconfluent 293T cells (each well of 6-well plate) were
cotransfected with 2µg of lentivirus vector, 1.5µg of pCMV-ΔR8.91, and 0.5µg of
pMD2G-VSVG by lipofectamine 2000. Supernatant containing the recombinant
lentivirus was collected at 24h and 48h post-transfection.

For transduction, HUVEC were plated on 6-well plates (8 x 10^5 cells/ well)
and incubated at 37 °C for 12 h before medium containing recombinant lentivirus was
added. Following 24 h incubation, the cells were re-transduced one more time. Five
days later the cells were harvested and analyzed by RT-PCR, Western blotting and
cell cycle analysis.
**In vivo angiogenesis.** Female C57BL/6NCr 5-week old, 25-30 g mice were purchased from NCI and treated in accordance with Johns Hopkins ACUC procedures. In all animal experiments the MPA prodrug mycophenolate mofetil i.v. formulation (Roche) was used. Mice were pretreated with drug for three days and then implanted with 0.5 mL of Matrigel (BD Biosciences) containing 100 ng/mL VEGF and 150 ng/mL bFGF. Drug treatment was continued daily for 10 days, mice were sacrificed, and plugs were harvested, fixed in neutral buffered formalin, and processed for histology using MAS-trichrome staining. The entire Matrigel plug was photographed at 100X and erythrocyte-filled blood vessels were counted per field.

**Renal cell tumor model.** Female BALB/C 5-6 week old, 25-30 g mice were purchased from NCI. Logarithmically growing RENCA cells (5 x 10⁵) in 50 μL 50% PBS, 50% Matrigel were injected into the renal subcapsule through an incision in the left flank (Dreves, J., Hofmann, I., Hugenschmidt, H., Wittig, C., Madjar, H., Muller, M., Wood, J., Martiny-Baron, G., Unger, C. & Marne, D. (2000) *Cancer Res.* 60, 4819-4824.). The mice were sacrificed 3.5 weeks after surgery and the primary tumors were dissected and fixed for immunohistochemistry. For CD31 analysis eight fields with the highest level of angiogenesis were selected per tumor for quantification using ImagePro Plus software (Media Cybernetics), and represented as mean percentage area occupied by CD31 positive blood vessels.

**Results**

**FIG. 1** Inhibition of HUVEC by MPA and its reversal by guanine. (A) Structure of mycophenolic acid. (B) Inhibition of HUVEC proliferation by MPA as measured by[^3]H]-thymidine incorporation. MPA inhibition (◻) is partially reversed by addition of exogenous guanine at 10 μM (■) and 20 μM (▲), however some inhibition is present even at the highest concentrations tested, 50 μM (★). (C) Inhibition of Jurkat T-cell proliferation by MPA as measured by[^3]H]-thymidine incorporation. MPA inhibition (◻) is unaffected by addition of 10 μM exogenous guanine (■), but is completely reversed by the addition of 20 μM (▼) and 50 μM (◆) guanine, in contrast to HUVEC. (D) Cell cycle analysis of HUVEC treated with
DMSO vehicle, 1 μM MPA showing G1/S cell cycle arrest (E), or 1 μM MPA + 50 μM guanosine (F). Addition of exogenous guanosine reverses MPA-induced G1/S cell cycle arrest.

FIG. 2  Selective knockdown of IMPDH-1 and -2 by shRNA in HUVEC. (A) RT-PCR of IMPDH-1 and -2 knockdown in HUVEC. The IMPDH-1 and -2 shRNA vectors completely and selectively abolish IMPDH gene expression in HUVEC. (B) Cell cycle analysis of HUVEC transfected with empty vector, sh-IMPDH-1 (C), sh-IMPDH-2 (D), and sh-IMPDH-1 + -2 (E) show a G1/S arrest similar to that observed with MPA treatment. This G1/S arrest is more pronounced with the shIMPDH-1 construct.

FIG. 3  MPA inhibits angiogenesis in vivo. (A) Representative images of Matrigel plugs in mice treated with vehicle (n = 7) or 60 mg/kg/day MPA (n = 7) (B). (C) MAS-trichrome stain of Matrigel plug from a mouse treated with vehicle demonstrating new blood vessel formation (100X magnification, 200X inset) or MPA (60 mg/kg/day s.c.) (D) shows a 69% decrease in new blood vessel formation (p < 0.002) (E).

FIG. 4  MPA inhibits tumor-associated angiogenesis. (A) Representative primary RENCA tumors from mice treated with vehicle (n = 7), left, or 120 mg/kg/day MPA (n = 8), right, showing decrease in tumor size. (B) H&E cross section (100x) of representative RENCA tumor from control mice shows extensive angiogenesis, with blood vessels highlighted by CD31 staining. In contrast, representative tumor sections from mice treated with 120 mg/kg/day MPA stained with H&E and CD31 show decreased angiogenesis compared to control. (C) Quantification of CD31 positive area per 200X microscope field shows a 49% decrease in mice treated with 120 mg/kg/day MPA (p < 0.001).

A library of 1,850 FDA-approved drugs and 600 drugs that entered the clinic via approval in a foreign country or phase II clinical trials was created. To discover new angiogenesis inhibitors among known drugs, the library was screened for inhibition of human umbilical vein endothelial cells (HUVEC) using [3H]-thymidine incorporation. An initial screen of the drugs at 10 μM (final concentration) revealed over 100 drugs that exhibited at least 50% inhibition. After eliminating known cytotoxic drugs such as taxol and colchicine or drugs that are restricted to topical use,
several drugs remained. One of the most potent inhibitors was mycophenolic acid (MPA) (Fig. 1A). The IC\textsubscript{50} for MPA on HUVEC proliferation was 99.2 (± 5.2) nM (Fig. 5B). Although MPA has been previously shown to inhibit endothelial cells (15, 16), the molecular basis of this inhibition remains unclear. Nor has it been shown whether MPA affected angiogenesis \textit{in vivo}. A series of experiments to address these questions was thus initiated.

MPA is an immunosuppressive drug widely used to prevent rejection of transplanted organs (Lipsky, J. J. (1996) \textit{Lancet} 348, 1357-1359.). The mechanism of action in the immune system is well established—MPA inhibits \textit{de novo} biosynthesis of purines in T and B lymphocytes (Allison, A. C. & Eugui, E. M. (2000) \textit{Immunopharmacology} 47, 85-118.). As the alternative nucleotide salvage pathway is absent in T and B cells, MPA was thought to specifically inhibit their proliferation by causing cell cycle arrest in the G1/S transition (Eugui, E. M., Almquist, S. J., Muller, C. D. & Allison, A. C. (1991) \textit{Scand. J. Immunol.} 33, 161-173.). Thus, a comparison of HUVEC to Jurkat T cells for sensitivity to MPA in the presence and absence of guanine was carried out. As shown in Fig. 1C, Jurkat T cells are inhibited by MPA with an IC\textsubscript{50} value of 128 (±6.1) nM, which is comparable to IC\textsubscript{50} values previously reported in human peripheral blood T and B lymphocytes (Eugui, E. M., Almquist, S. J., Muller, C. D. & Allison, A. C. (1991) \textit{Scand. J. Immunol.} 33, 161-173.). Addition of guanine at or above 20 μM rendered Jurkat T cells resistant to MPA. Similar to Jurkat T cells, inhibition of HUVEC proliferation is also reversed by guanine in a dose-dependent manner (Fig. 1B). Unlike Jurkat T cells, however, HUVECs are less sensitive to guanine, the cause of which remains unknown. Similar reversal of MPA inhibition is also seen in HUVEC and Jurkat T-cells with the addition of exogenous guanosine and deoxyguanosine (data not shown).

It has been shown that MPA causes cell cycle arrest in activated T and B lymphocytes at G1 (Laliberte, J., Yee, A., Xiong, Y. & Mitchell, B. S. (1998) \textit{Blood} 91, 2896-2904.). Therefore, the effect of MPA on endothelial cell cycle progression was examined. Treatment of HUVEC with 1 μM of MPA led to, in comparison to the control cells, an increase in the population of cells in G1 phase (81 vs. 71%) and a corresponding decrease in the population of cells in S (6.5 vs. 12%) and G2/M (12.4 vs. 17.8%) phases, indicating that MPA also causes a G1 blockade in HUVEC similar
to T and B cells (Fig. 1D, E). Addition of 50 μM guanosine completely reversed the cell cycle effect of MPA in HUVEC (Fig. 1F). Together, these results strongly suggest that blockade of purine biosynthesis is responsible for the inhibition of HUVEC by MPA, similar to T and B cells.

The molecular target for MPA in T and B cells has been unambiguously established as inosine monophosphate dehydrogenase (IMPDH), which catalyzes the NAD⁺-dependent conversion of inosine 5'-monophosphate to xanthosine 5'-monophosphate (Allison, A. C. & Eugui, E. M. (2000) Immunopharmacology 47, 85-118.). Two isoforms of IMPDH are known in humans, named type 1 and type 2 enzymes. Whereas IMPDH-1 is constitutively expressed, IMPDH-2 is induced in a number of tumor cell types undergoing active proliferation (Collart, F. R., Chubb, C. B., Mirkin, B. L. & Huberman, E. (1992) Cancer Res. 52, 5826-5828.). The expression of the two isoforms in actively proliferating HUVEC was assessed using RT-PCR. It was found that IMPDH-1 is the predominantly expressed isoform in HUVEC, although IMPDH-2 mRNA is also detected (data not shown). This expression pattern is similar to that in peripheral leukocytes but distinct from those in most other tissues in which IMPDH-2 is more abundantly expressed (Senda, M. & Natsumeda, Y. (1994) Life Sci. 54, 1917-1926.). To determine whether inhibition of either isoform of IMPDH accounts for the effect of MPA on HUVEC, each isoform was knocked down by RNA interference. To avoid subjecting the primary HUVEC to the relatively harsh conditions of conventional transfection methods, lentiviruses were used to deliver isoform-specific shRNAs to HUVEC (Pan, F., Ye, Z., Cheng, L. & Liu, J. O. (2004) J. Biol. Chem. 279, 14477-14480, Lois, C., Hong, E. J., Pease, S., Brown, E. J. & Baltimore, D. (2002) Science 295, 868-872). Upon testing three different regions of each IMPDH cDNA, at least one shRNA construct was found that efficiently blocked the expression of either IMPDH-1 or IMPDH-2 with high specificity (Fig. 2A). Transduction of HUVEC with a mixture of lentiviruses carrying shRNAs for both IMPDH-1 and IMPDH-2 led to the knockdown of mRNA (Fig. 2A) and protein (data not shown) for both isoforms. The effects of knockdown of the two isoforms of IMPDH on the cell cycle of HUVEC were then determined. As shown in Fig. 2B-C, knockdown of IMPDH-1 is sufficient to cause a cell cycle arrest in G1. Interestingly, knockdown of IMPDH-2 appeared to cause a significant delay in S phase progression rather than a G1 blockade (Fig 2D). Not surprisingly, knockdown
of both isoforms of IMPDH also led to accumulation of HUVEC in G1 (Fig 2E).
These observations validated IMPDH-1 as the target for MPA in endothelial cells.

To test the *in vivo* efficacy of MPA as an anti-angiogenic inhibitor, the
Matrigel plug angiogenesis assay in mice was performed using doses previously used
to demonstrate immunosuppression in murine transplant models (van Leeuwen, L.,
*Transplantation* 64, 1097-1101, Fahmy, R. G., Dass, C. R., Sun, L. Q., Chesterman,
containing VEGF and bFGF were subcutaneously implanted into mice. The control
group was treated with saline vehicle and the MPA group was treated with 60 or 120
mg/kg/day of drug. After 10 days, plugs from control mice showed extensive
neovascularization that is visible both macroscopically (Fig. 3A) and microscopically
(Fig. 3C). In contrast, MPA treated mice had significantly less new blood vessel
formation (Fig. 3B, D). To quantify these differences we counted erythrocyte-filled
blood vessels per 100X field (Fig. 3E) (Fahmy, R. G., Dass, C. R., Sun, L. Q.,
Chesterman, C. N. & Khachigian, L. M. (2003) *Nat. Med.* 9, 1026-1032), and
observed a 69% decrease in new blood vessel formation in mice treated with MPA in
comparison with vehicle control (p < 0.002). Thus, MPA is capable of inhibiting
angiogenesis at a therapeutically achievable dose *in vivo*.

Angiogenesis has been implicated in tumor growth among a number of other
diseases. Although MPA inhibits fast-growing tumor cell lines in culture and in
J. Cancer* 57, 568-573, Carter, S. B., Franklin, T. J., Jones, D. F., Leonard, B. J.,
whether MPA also affects tumor-associated angiogenesis. We determined the
efficacy of MPA in inhibiting tumor-associated angiogenesis in a murine renal cell
carcinoma (RENCA) model (Salup, R. R. & Wiltrout, R. H. (1986) *Cancer Res.* 46,
3358-3363). MPA inhibited the growth of the primary tumor in a dose-dependent
fashion, causing a 34% and 27% decrease in volume and weight, respectively, at 60
mg/kg/day and a 64% decrease in both volume and weight at 120 mg/kg/day (p <
0.001) (Fig. 4A). The decrease in tumor growth caused by MPA at 120 mg/kg/day
was accompanied by a 48% decrease in the area of CD31 positive staining blood
vessels per 200X field (p < 0.001). As shown in Fig. 4B, C, whereas CD31 positive
blood vessels were abundant in tumors from control animals, MPA treatment at 120 mg/kg/day led to a significant reduction in CD31 positive vessels in the primary tumor. These results demonstrate that MPA is capable of decreasing tumor-induced angiogenesis \textit{in vivo}. 

All references cited herein, whether in print, electronic, computer readable storage media or other form, are expressly incorporated by reference in their entirety, including but not limited to, abstracts, articles, journals, publications, texts, treatises, technical data sheets, internet web sites, databases, patents, patent applications, and patent publications.

Certain compounds of the present invention may exist in particular geometric or stereoisomeric forms. The present invention contemplates all such compounds, including cis- and trans-isomers, R- and S-enantiomers, diastereomers, (D)-isomers, (L)-isomers, the racemic mixtures thereof, and other mixtures thereof, as falling within the scope of the invention. Additional asymmetric carbon atoms may be present in a substituent such as an alkyl group. All such enriched isomers, as well as racemic mixtures thereof, are intended to be included in this invention.

A number of embodiments of the invention have been described. Embodiments herein include those recited alone or in combination with other delineated embodiments herein. Nevertheless, it will be understood that various modifications may be made without departing from the spirit and scope of the invention. Accordingly, other embodiments are within the scope of the following claims.
What is claimed:

1. A method of inhibiting IMPDH type 1 in a subject, the method comprising the step of administering to the subject identified as being in need of an IMPDH type 1 inhibitor, an effective amount of an IMPDH type 1 inhibitor compound selected from: an IMPDH type 1 inhibitor, a nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase;
   wherein the subject does not experience immunosuppressive side effects associated with IMPDH type II.

2. A method of inhibiting angiogenesis in a subject, the method comprising the step of administering to the subject identified as being in need of inhibiting angiogenesis, an effective amount of an anti-angiogenic IMPDH type 1 inhibitor compound selected from: an IMPDH type 1 inhibitor, a nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase;
   wherein the subject does not experience immunosuppressive side effects associated with IMPDH type II.

3. Use of an IMPDH type 1 inhibitor compound in the manufacture of a medicament for inhibiting or reducing IMPDH type 1 in a patient, the IMPDH type 1 inhibitor compound being an IMPDH type 1 inhibitor, a nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase;
   wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

4. A sustained release device for implantation in a patient and sustained release of an IMPDH type 1 inhibitor compound for at least a period of 30 days, wherein the IMPDH type 1 inhibitor compound is an IMPDH type 1 inhibitor, a nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase;
wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

5. A sustained release drug device adapted for implantation in or adjacent to the eye of a patient, the drug delivery device comprising: (i) an IMPDH type 1 inhibitor drug core comprising an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase; (ii) an impermeable coating disposed about the core that is substantially impermeable to the passage of the IMPDH type 1 inhibitor compound, having one or more openings therein which permit diffusion of the IMPDH type 1 inhibitor compound, and which is substantially insoluble and inert in body fluids and compatible with body tissues; and, optionally, (iii) one or more permeable polymer members or coatings disposed in the flow path of the IMPDH type 1 inhibitor compound through said openings in said impermeable coating, said permeable polymer being permeable to the passage of the IMPDH type 1 inhibitor compound, and which is substantially insoluble and inert in body fluids and compatible with body tissues; wherein the impermeable coating and permeable polymer members or coatings are disposed about the drug core so as to produce, when implanted, a substantially constant rate of release of the IMPDH type 1 inhibitor compound from the device;

wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.

6. A sustained release formulation for depot injection in a patient and sustained release of an IMPDH type 1 inhibitor compound for at least a period of 30 days, wherein the formulation includes:

an viscous gel formulation comprising a bioerodible, biocompatible, polymer; and

an anti-angiogenic IMPDH type 1 inhibitor agent dissolved or dispersed therein, which anti-angiogenic agent is selected from: an IMPDH type 1 inhibitor, an nucleoside analog; an imidazole of Formula I; a triazole of Formula II; trifluridine; danazol; and asparaginase;

wherein the patient does not experience immunosuppressive side effects associated with IMPDH type II.
7. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the subject or patient does not experience gastrointestinal side effects.

8. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the anti-angiogenic IMPDH type 1 inhibitor compound is provided in an amount effective for treatment of retinoblastoma, cystoid macular edema (CME), exudative age-related macular degeneration (AMD), diabetic retinopathy, diabetic macular edema, or ocular inflammatory disorders.

9. The method of claim 1 or 2, use of claim 3; device of claim 4 or 5, or formulation of claim 6, for the treatment of tumor or cancer growth (neoplasia), skin disorders, neovascularization, and inflammatory and arthritic diseases.

10. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, for the treatment of dermis, epidermis, endometrium, retina, surgical wound, gastrointestinal tract, umbilical cord, liver, kidney, reproductive system, lymphoid system, central nervous system, breast tissue, urinary tract, circulatory system, bone, muscle, or respiratory tract.

11. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, for eliminating or reducing normal but undesired tissue in a patient.

12. The method, use, device or formulation of 11, for the reduction of fat.

13. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type 1 inhibitor has a Ki for inhibiting IMPDH type 1 less than about 1 micromolar in endothelial cells.

14. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type 1 inhibitor has a Ki for inhibiting
the type I isoform of IMPDH (IMPDH Type I) more than 2 fold less than the Ki of the inhibitor for inhibiting the type II isoform (IMPDH type II) in endothelial cells.

15. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type I inhibitor has a EC50 for inhibiting the type I isoform of IMPDH (IMPDH Type I) more than 2 fold less than the EC50 of the inhibitor for inhibiting the type II isoform (IMPDH type II) in endothelial cells.

16. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type I is found in endothelial cells.

17. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type I inhibitor is provided in a dose that produces a serum concentration at least 50 percent less than the inhibitor’s EC50 for inhibiting IMPDH type I in lymphocytes.

18. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type I inhibitor is a nucleoside analog.

19. The method, use, device, or formulation of claim 18, wherein the IMPDH type 1 inhibitor is a purine derivative.

20. The method, use, device, or formulation of claim 19, wherein the purine derivative is a compound of formula III:

\[
\begin{align*}
\text{III;}
\end{align*}
\]

wherein,

each X is independently O, S, S(O), S(O)\_2, N(R\_k), C(O), C(S), C(NR), C(NR)NR\_k, C(O)NR\_k, C(O)NR\_kNR\_k, C(O)O, OC(O), OC(O)O, (C(R^8)(R^9))_m, (C(R^8)(R^9))_mNR\_k, (C(R^8)(R^9))_mO, (C(R^9))_mS(O)\_p, NR\_kC(O), NR\_kC(O)O, OC(O)NR\_k, or absent;
each Rₙ is independently H, an optionally substituted alkyl, an optionally substituted cycloalkyl, an optionally substituted cycyl, an optionally substituted heterocycloalkyl, an optionally substituted heterocycl, an optionally substituted aryl, an optionally substituted heteroaryl, an optionally substituted aralkyl, an optionally substituted heteroaralkyl, halo, haloalkyl, aminoalkyl, cyano, nitro, or an optionally substituted alkylcarbonylalkyl;

Rᵦ, for each occurrence, is independently, H, an optionally substituted alkyl, an optionally substituted alkenyl, an optionally substituted alkynyl; an optionally substituted cycyl, an optionally substituted cycloalkyl, an optionally substituted heterocycloalkyl, an optionally substituted heterocycl, an optionally substituted aralkyl, an optionally substituted heteroaralkyl, an optionally substituted aryl, an optionally substituted heteroaryl, haloalkyl, hydroxylalkyl, alkylcarbonylalkyl, mercaptoalkyl, aminoalkyl, sulfonylalkyl, sulfonylaryl, thioalkoxy, halo, cyano, nitro, nitroso, or azide; and

Rᵦ, for each occurrence, is independently H, an optionally substituted alkyl, an optionally substituted alkenyl, an optionally substituted alkynyl, an optionally substituted cycyl, an optionally substituted cycloalkyl, an optionally substituted heterocycloalkyl, an optionally substituted heterocycl, an optionally substituted aralkyl, an optionally substituted heteroaralkyl, an optionally substituted aryl, or an optionally substituted heteroaryl.

21. The method of claim 1 or 2, use of claim 3, device of claim 4 or 5, or formulation of claim 6, wherein the IMPDH type 1 inhibitor is capable of interacting with amino acid residues Arg 253, Ile 45, and Tyr 282 of the IMPDH type 1 active site in vitro.

22. The method, use, device or formulation of any of the preceding claims, wherein the IMPDH type 1 compound inhibits endothelial cell proliferation.

23. The method, use, device or formulation of any of the preceding claims, wherein the IMPDH type 1 compound inhibits G1/S cell cycle progression of endothelial cells.
24. The method, use, device or formulation of any of the preceding claims, wherein the IMPDH type 1 compound decreases new blood vessel formation.

25. The method, use, device or formulation of any of the preceding claims, further comprising an additional therapeutic agent.

26. The method, use, device or formulation of claim 25, wherein the additional therapeutic agent is an angiogenesis-inhibiting compound.

27. The method, use, device or formulation of claim 25, wherein the additional therapeutic agent is an anticancer compound.

28. The method, use, device or formulation of any of the preceding claims, wherein the step of administering the IMPDH type 1 inhibitor compound comprises administering the compound orally, topically, parentally, intravenously or intramuscularly.

29. The method, use, device or formulation of claim 28, wherein the administration is carried out in a controlled and sustained release.

30. The method, use, device or formulation of any of the preceding claims, wherein the step of administering the IMPDH type 1 inhibitor compound comprises administering the compound in a dosage of between about 0.1 and 100 mg/kg/day.

31. The method, use, device or formulation of any of the preceding claims, wherein the step of administering the IMPDH type 1 inhibitor compound comprises administering the compound in a dosage of less than about 500 mg/day.

32. A kit comprising an effective amount of an IMPDH type 1 inhibitor compound in unit dosage form, together with instructions for administering the IMPDH type 1 inhibitor compound to a subject identified as being in need of treatment with an IMPDH type 1 inhibitor.
33. The method, use, device or formulation of any of the preceding claims, comprising the step of administering an effective amount of a composition comprising an IMPDH type 1 inhibitor compound and a pharmaceutically suitable excipient to treat a disease or disorder.

34. The method of claim 33, wherein the disease or disorder is tumor or cancer growth (neoplasia).

35. The method of claim 33, wherein the disease or disorder is: eye or ocular cancer, rectal cancer, colon cancer, cervical cancer, prostate cancer, breast cancer and bladder cancer, oral cancer, benign and malignant tumors, stomach cancer, liver cancer, pancreatic cancer, lung cancer, corpus uteri, ovary cancer, prostate cancer, testicular cancer, renal cancer, brain/cns cancer (e.g., gliomas), throat cancer, skin melanoma, acute lymphocytic leukemia, acute myelogenous leukemia, Ewing's Sarcoma, Kaposi's Sarcoma, basal cell carcinoma and squamous cell carcinoma, small cell lung cancer, choriocarcinoma, rhabdomyosarcoma, angiosarcoma, hemangioendothelioma, Wilms Tumor, neuroblastoma, mouth/pharynx cancer, esophageal cancer, larynx cancer, lymphoma, neurofibromatosis, tuberous sclerosis, hemangiomas, and lymphangiogenesis.
Mycophenolic Acid

FIG. 1A

FIG. 1B
FIG. 1C

% Control CPM

Log [MPA] (M)

FIG. 1D

Control

G1: 70.6 %
S: 11.8 %
G2/M: 17.6 %

Counts

DNA Content
FIG. 5B