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(54) Title: RAPAMYCIN DERIVATIVES

(57) Abstract

Rapamycin derivatives; pharmaceutical compositions comprising such rapamycin derivatives and pharmaceutically acceptable carriers or diluents; and methods of using such derivatives to inhibit pathogenic fungi growth, inhibit immunosuppression or treat carcinogenic tumors are disclosed.

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TITLE

RAPAMYCIN DERIVATIVES

BACKGROUND OF THE INVENTION

This invention relates to rapamycin derivatives, pharmaceutical compositions comprising such derivatives, and methods of treatment of pathogenic fungi, methods of inducing immunosuppression and methods of treating carcinogenic tumors utilizing such rapamycin derivatives.

Rapamycin is a naturally occurring macrocyclic triene antibiotic which can be produced by culturing an organism in an aqueous nutrient medium. Its structure may be illustrated as follows:

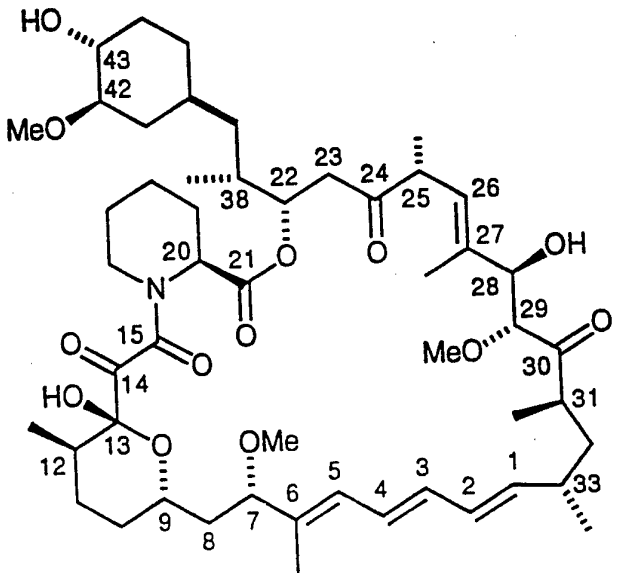
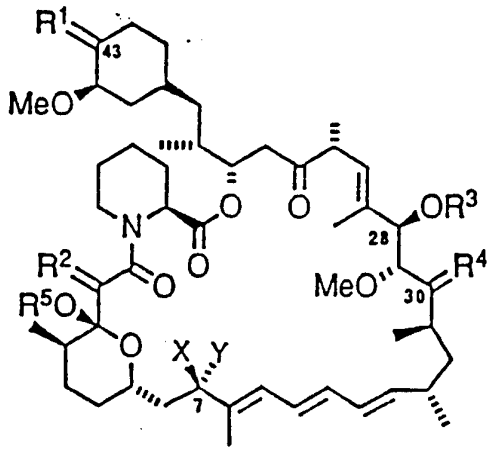


Figure I

At least one rapamycin-producing strain of *Streptomyces hygroscopius* was deposited with the Northern Utilization and Research Division, Agricultural Research Service, U.S. Department of Agriculture, Peoria, Illinois, U.S.A. under accession number NRRL 5491. Rapamycin, and methods for its preparation by culturing NRRL 5491 are disclosed by U.S. Patent 3,929,992, issued December 30, 1975, the entire disclosure of which is hereby incorporated by reference.

SUMMARY OF THE INVENTION

This invention relates to novel rapamycin derivatives of the formula:



Formula II

wherein:

X is selected from the group consisting of H, $-OR^{10}$, $-S(O)_nR^{10}$, $-NR^{10}R^{11}$, alkyl and aryl;

Y is selected from the group consisting of H, $-OR^{10}$, $-S(O)_nR^{10}$, $-NR^{10}R^{11}$, alkyl and aryl;

or X and Y taken together are =O;

n is selected from the group consisting of 0, 1 and 2;

R^1 is selected from the group consisting of =0, ($-OR^6$,H) and (H,H);

R^2 is selected from the group consisting of =0, (H,H), and (H,OH);

R^3 and R^6 are independently selected from the group consisting of -H, C_1-C_4 alkyl, $-C(=0)R^7$, $-C(=0)OR^7$, $-C(=0)NHR^7$, and $-C(=S)OR^7$;

R^4 is selected from the group consisting of =O and (H,OR^6);

or R^3 and R^4 can be taken together to form a bridge of the formula A-$C(R^8)(R^9)$-O-B, where A is a bond to the oxygen bonded to the carbon at the 28-position and B is a bond to the carbon at the 30-position;

R^5 is selected from the group consisting of -H and C_1-C_4 alkyl;

R^7 is selected from the group consisting of C_1-C_{10} alkyl, C_3-C_6 cycloalkyl, aryl groups, and heterocyclic groups;

R^8 and R^9 are independently selected from the group consisting of H, C_1 to C_6 alkyl, or R^8 and R^9 taken together are =O;

R^{10} and R^{11} are independently selected from the group consisting of H, alkyl and aryl;

provided that,

(a) at least one of X and Y is H; and

(b) when Y is $-R^{10}$, then R^{10} is other than CH_3;

and all pharmaceutically acceptable salts, hydrates or solvates thereof.

This invention also relates to a pharmaceutical composition comprising an effective amount of one or more compounds of Formula II and a pharmaceutically acceptable carrier or diluent.

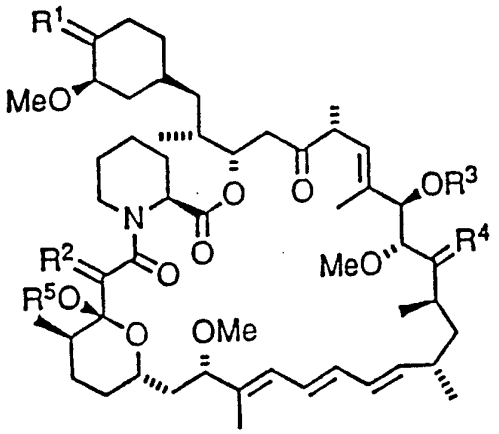
This invention also relates to a method of inhibiting the growth of pathogenic fungi in a human or other animal in need thereof which comprises administering an effective, non-toxic amount of one or

more compounds of Formula II to such human or other animal.

This invention also relates to a method of inducing immunosuppression in a human or other animal in need thereof which comprises administering an effective, non-toxic amount of one or more compounds of Formula II to such human or other animal.

In addition, this invention relates to a method of treating carcinogenic tumors in a human or other animal comprising administering an effective, non-toxic amount of one or more compounds of Formula II to such human or other animal.

Still further, this invention relates to a method of preparing compounds of Formula II comprising contacting a compound of the formula:



with an acid selected from the group consisting of protic acids and Lewis acids, and with an appropriate nucleophile.

DETAILED DESCRIPTION OF THE INVENTION

When any substituent or variable (e.g., aryl, alkoxyl, R^1, R^2, R^3, R^5, R^6, etc.) occurs more than one time in the formula of any of the compounds of Formula II, such variable or substituent definition on each

occurrence is independent of its definition at every other occurrence, unless otherwise indicated. Combinations of substituents and/or variables in a constituent of the compounds of the invention are permissible only if such combinations result in a stable compound.

The parenthetical nomenclature used in the definition of substituents such as R^1 (e.g., (H, OR^6) is intended to reflect the substituents on both valences of the relevant atom. The invention is not limited to particular isomers and the order of moieties in the parentheses does not suggest a particular configuration.

As used herein, except where otherwise noted, the term "alkyl" is intended to include both branched- and straight-chain, cyclic, acyclic and polycyclic, saturated and unsaturated aliphatic hydrocarbon groups. Preferably, alkyl groups have from one to six carbon atoms, unless otherwise noted. Such alkyl group may be optionally substituted by one or more members independently selected from the group consisting of aryl, hydroxyl, protected hydroxyl, C_1-C_6 alkoxyl, acyloxy, amino, N-acylamino, ketone, halogen, cyano, and carboxyl. The term "alkyl" also includes the above-mentioned groups in which a heteroatom selected from oxygen, nitrogen and sulfur is substituted for one or more carbon atoms in the alkyl moiety.

As used herein, the term "aryl" is intended to include cyclic, heterocyclic, polycyclic and heteropolycyclic unsaturated C_4 to C_{14} moieties, especially phenyl or naphthyl. Such aryl moieties may be optionally substituted by one to five members independently selected from the group consisting of C_1-C_6 alkyl, aryl, hydroxyl, protected hydroxyl, C_1-C_6 alkoxyl, acyloxy, amino, N-acylamino, -S(O)n alkyl, nitro, cyano, carboxyl and halogen.

As used herein, the term "alkoxyl" represents an alkyl group as herein defined of the indicated number of

carbon atoms attached through an oxygen bridge.

As used herein, the term "acyloxy" is intended to represent the groups -OC(O)-(alkyl) and OC(O)-(aryl).

As used herein, the term "amino" is intended to represent the groups $-NH_2$, $-N(alkyl)_2$, -NH(alkyl), $-N(aryl)_2$, and -NH(aryl).

As used herein, the term "N-acylamino" is intended to represent the groups -NHC(O)-(alkyl) and -NHC(O)-(aryl).

As used herein, the term "ketone" is intended to mean the moiety -C(O)-.

As used herein, the term "halogen is intended to include fluoro, chloro, bromo and iodo.

As used herein, the term "cycloalkyl" is intended to include saturated and unsaturated hydrocarbon aliphatic ring groups having the specified number of carbon atoms. Such cycloalkyl may be optionally substituted by one or more members independently selected from the group consisting of aryl, hydroxyl, C_1-C_6 alkoxyl, acyloxy, amino, N-acylamino, ketone, and halogen.

As used herein, the term "heterocycle" is intended to include a stable 5- to 7-membered mono- or bicyclic or stable 7- to 10-membered bicyclic heterocyclic ring which is either saturated or unsaturated, and which consists of carbon atoms and from one to three heteroatoms independently selected from the group consisting of N, O and S, and wherein the nitrogen and sulfur heteroatoms may optionally be oxidized, and the nitrogen heteroatom may optionally be quarternized, and including any bicyclic group in which any of the above-defined heterocyclic rings is fused to a benzene ring. The heterocyclic ring may be attached at any heteroatom or carbon atom which results in the creation of a stable structure. Examples of such heterocyclic elements include but are not limited to piperidyl, piperidinyl, piperazinyl, pyrimidinyl, pyridazinyl,

oxazolyl, furyl, and thienyl. The heterocycle may be optionally substituted in a manner such that carbon atoms attached to a heteroatom are not directly substituted by a heteroatom, by from one to four members independently selected from the group consisting of C_1-C_6 alkyl, aryl, hydroxyl, C_1-C_6 alkoxyl, acyloxy, amino, N-acylamino, nitro and halogen.

Naturally occurring rapamycin has a structure in which there is a methoxy group in the S-configuration (---OMe) at the C-7 atom of the molecule. Using the novel synthetic methods disclosed herein, derivatives of rapamycin have been prepared, including compounds derivatized in both the R- (X is other than H) and S- (Y is other than H) configurations at the C-7 atom as well as the compound epirapamycin, in which there is a methoxy group in the R-configuration (OMe) at the C-7 atom.

Preferred compounds of the invention include the following compounds wherein, together or independently:

(1) R^1 is (H,OH)

(2) R^2 is =O

(3) R^3 is H

(4) R^4 is =O

(5) R^5 is H

(6) X and Y are each H

(7) One of X and Y is selected from the group consisting of OR^{10} and SR^{10} or where X and Y are taken together as =O

(8) One of X and Y is selected from the group consisting of $-OR^{10}$ and $-SR^{10}$ where R^{10} is selected from the group consisting of optionally substituted C_1-C_3 alkyl, optionally substituted benzyl, and optionally substituted phenyl

(9) One of X and Y is an alkyl group or when one of R^{10} and R^{11} is an alkyl group, said alkyl is selected from the group consisting of branched, straight-chain, cyclic, polycyclic, saturated and unsaturated alkyl

groups which may be substituted with 0, 1 or more substitutents selected from the group consisting of aryl, keto, hydroxyl, alkoxyl, acyloxy, amino, N-acylamino, halogen, cyano and carboxyl substituents, and in which at least one carbon atom may be replaced with a heteroatom selected from the group consisting of O, S and N

(10) One of X and Y is an aryl group or when one of R^{10} or R^{11} is an aryl group, said aryl group is selected from the group consisting of cyclic, heterocyclic, polycyclic and heteropolycyclic C_2 to C_{14} aryl groups, which may be substituted by one to five members selected from the group consisting of alkyl, hydroxyl, alkyoxyl, acyloxy, amino, N-acylamino, halogen, cyano, carboxyl and nitro groups.

Specifically preferred compounds of this invention include those where:

(1) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OCH_3$.

(2) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OC_2H_5$.

(3) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is $-OC_2H_5$.

(4) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OCH_2CH_2OH$.

(5) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is $-OCH_2CH_2OH$.

(6) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is 3,4-dimethoxybenzyloxy.

(7) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is 3,4-dimethoxybenzyloxy.

(8) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is $-SCH_3$.

(9) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is $-SCH_3$.

(10) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is -SPh where Ph is phenyl.

(11) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is -SPh where Ph is phenyl.

(12) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is 2-furanyl.

(13) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is 2-furanyl.

(14) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is O-acetyl.

(15) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, and each of X and Y is H.

(16) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, and X and Y are taken together as =O.

(17) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is allyl.

(18) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is OH.

(19) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is 2,4-dimethoxyphenyl.

(20) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is 2,4-dimethoxyphenyl.

(21) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is -OH.

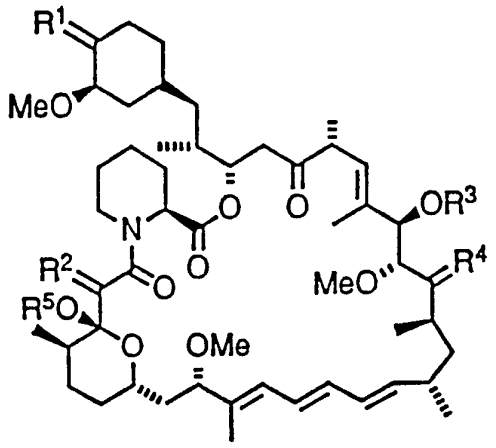
The compounds of this invention can exist in free form or, where appropriate, in salt form. Pharmaceutically acceptable salts and their preparation are well-known to those of skill in the art. The pharmaceutically acceptable salts of the compounds of this invention include the conventional non-toxic salts or the quaternary ammonium salts of such compounds which are formed, for example, from inorganic or organic acids of bases.

The compounds of the invention may form hydrates or solvates. It is known to those of skill in the art that charged compounds form hydrated species when lyophilized with water, or form solvated species when concentrated in a solution with an appropriate organic

solvent.

The compounds of this invention may be prepared by the methods outlined below or conventional variations thereof. The reagents utilized are either described in the literature or are commercially available.

Compounds of Formula II (where X and Y are other than =O) may be prepared from rapamycin or other compounds of the formula:



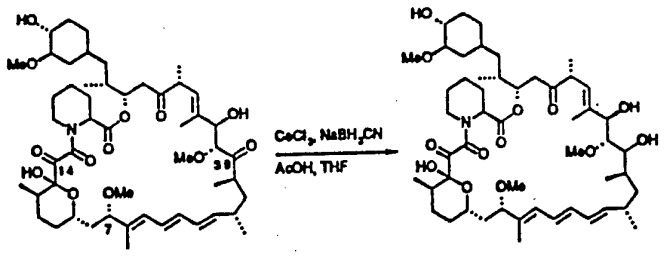
III

where R^1 - R^{11} and n are as defined above, by contacting said compound with an acid selected from the group consisting of protic acids and Lewis acids, and the desired nucleophilic reagent, such as an alcohol, thiol, electron-rich aromatic, allylsilane, etc. Preferred acids are trifluoroacetic acid and titanium tetrachloride, although these are by no means limiting of the acids which may be utilized. The reaction is preferably carried out in an inert atmosphere (e.g., under argon) at a temperature in the range of about -78° to 0° C, preferably about -40° C. Suitable solvents include nonprotic, nonpolar solvents, such as but not limited to dichloromethane.

Compounds of the invention where X and Y are taken together as =O may be prepared from rapamycin or other compounds of formula III by contacting said compounds with a quinone oxidant such as DDQ (2,3-dichloro-5,6- dicyano-1,4-benzoquinone).

Rapamycin derivatives reduced at the C-30 position may be prepared by treatment of rapamycin with a mixture of cerium trichloride and sodium cyanoborohydride. Suitable solvents for this reaction include a mixture of acetic acid and tetrahydrofuran. This reaction is illustrated in Scheme A with rapamycin shown as the starting material; however other rapamycin derivatives may be reduced at the C-30 position using this method.

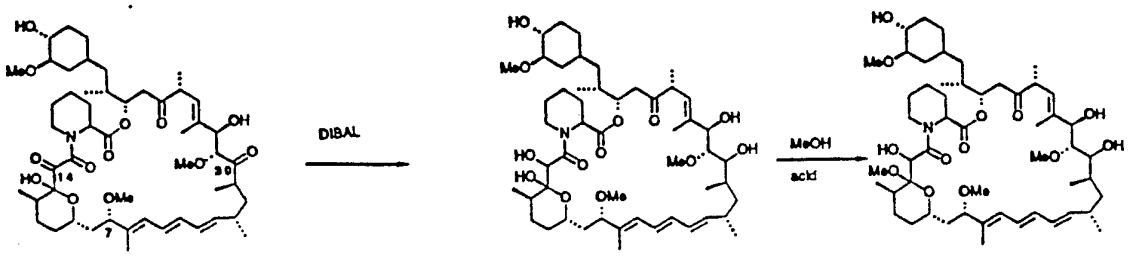
Scheme A



RAPAMYCIN

Compounds of the invention which are reduced at both the C-14 and C-30 positions (i.e., R^2 and R^4 = (H,OH) may be prepared by the action of diisobutylaluminum hydride on rapamycin or a derivative thereof. By appropriate control of the same reduction method (e.g., low temperature, limiting hydride and reaction times), compounds reduced only at the C-14 position can be prepared. These reactions are illustrated, with rapamycin shown as the starting material, in Scheme B.

Scheme B

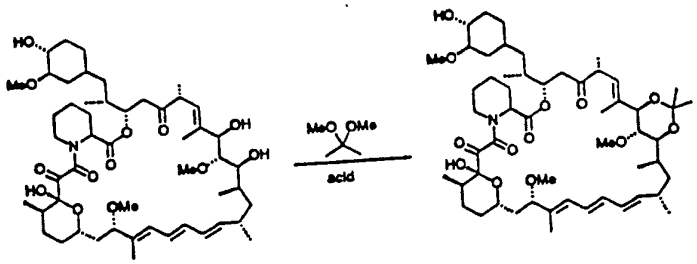


RAPAMYCIN

Also illustrated in Scheme B is the preparation of a C-13 O-methylated derivative, prepared by treatment with acidic methanol.

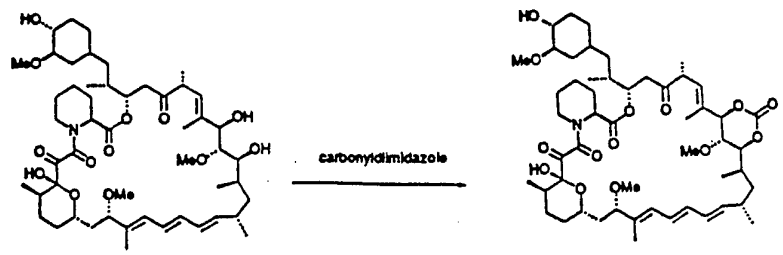
Compounds of the invention having a bridge between the C-28 and C-30 positions may be prepared by methods analogous to that shown in Scheme C. The C-30 reduced derivative is contacted with a dialkoxypropane such as dimethoxypropane to yield the desired compound where R^8 and R^9 are each alkyl.

Scheme C



Another type of compound having a bridge between the C-28 and C-30 positions may be prepared by methods analogous to that shown in Scheme D. The C-30 reduced derivative is contacted with carbonyldiimidazole to yield the desired compound where R^8 and R^9 taken together are =O.

Scheme D



When the derivatives of this invention are compounds derivatized at more than one atom of the rapamycin molecule (e.g., at C-7, C-13, C-14, C-28, C-30 or C-43), the synthetic order of the derivation can vary. The preferred order of reactions will be dependent upon the nature of the modifications at each position and their compatibility with subsequent reaction conditions, and would be obvious to chemists skilled in the art.

The Examples provided below in this specification provide a variety of synthetic methods for preparing compounds of this invention.

This invention also relates to a pharmaceutical compositions comprising a pharmaceutically acceptable carrier or diluent and an effective amount of one or more compounds of Formula II.

A compound of Formula II is administered in conventional dosage form prepared by combining a therapeutically effective amount of the compound ("active ingredient") with standard pharmaceutical carrier or diluents according to conventional procedures. These procedures may involve mixing, granulating and compressing or dissolving the ingredients as appropriate to the desired preparation.

The pharmaceutical carrier employed may be, for example, either a solid or liquid. Exemplary of solid carrier are lactose, terra alba, sucrose, talc, gelatin, agar, pectin, acacia, magnesium stearate, stearic acid and the like. Exemplary of liquid carriers are syrup, peanut oil, olive oil, water and the like. Similarly, the carrier or diluent may include time delay material well known to the art, such as glyceryl monostearate or glyceryl distearate along or with a wax, ethylcellulose, hydroxypropylmethylcellulose, methylmethacrylate and the like.

A wide variety of pharmaceutical forms can be employed. Thus, if a solid carrier is used, the

preparation can be tableted, placed in a hard gelatin capsule in powder or pellet form or in the form of a troche or lozenge. The amount of solid carrier will vary widely but preferably will be from about 25 mg to about 1 g. If a liquid carrier is used, the preparation will be in the form of a syrup, emulsion, soft gelatin capsule, sterile injectable solution or suspension in an ampule or vial or nonaqueous liquid suspension.

To obtain a stable water soluble does form, a pharmaceutically acceptable salt of a compounds of the invention is dissolved in an aqueous solution of an organic or inorganic acid, such as a 0.3M solution of succinic acid, or, preferably, citric acid. If a soluble salt form is not available, the compounds of the invention is dissolved in a suitable cosolvent or combinations thereof. Examples of such suitable cosolvents include, but are not limited to, alcohol, propylene glycol, polyethylene glycol 300, polysorbate 80, glycerin and the like in concentrations ranging from 0-60% of the total volume.

Tests indicate that the compounds of this invention are useful in prophylactically or therapeutically inhibiting the growth of pathogenic fungi in a human or other animal in need thereof. The invention, therefore, includes methods of inhibiting the growth of pathogenic fungi in a human or other animal in need thereof which comprises administering to such human or animal an effective, non-toxic amount of a compound of Formula II.

By the term "pathogenic fungi" is meant fungi capable of producing disease in a human or other animal. Examples of pathogenic fungi include, but are not limited to *Candida albicans* and other candida species, *Microsporum gypseum*, *Trichophyton mentagrophytes*, *Asperqillus sp.* and *Sporotrichum sp.* The ability of the compounds of this invention to inhibit the growth of pathogenic fungi may be demonstrated or predicted by

standard tests known and used for this purpose, for example, the yeast assay described below.

One skilled in the art would be able, by routine experimentation, to determine what an effective, non-toxic amount of compound would be for the purpose of inhibiting pathogenic fungi growth. Generally, however, an effective dosage will be in the range of about 0.05 to 100 milligrams per kilogram body weight per day.

Tests indicate that the compounds of this invention are also useful for inducing immunosuppression, i.e., inducing a suppression of a human's or animals immune system. This invention therefore relates to a method of prophylactically or therapeutically inducing immunosuppression in a human or other animal in need thereof which comprises administering an effective, non-toxic amount of such a compound of this invention to such human or other animal.

The ability of the compounds of this invention to induce immunosuppression may be demonstrated in standard tests used for this purpose, for example, a mixed lymphocyte reaction test or a test measuring inhibition of T-cell proliferation measured by thimidine uptake.

The fact that the compounds of this invention have utility in inducing immunosuppression means that they are useful in the treatment or prevention of resistance to or rejection of transplanted organs or tissues (e.g., kidney, heart, lung, bone marrow, skin, cornea, etc.); the treatment or prevention of autoimmune, inflammatory, proliferative and hyperproliferative diseases, and of cutaneous manifestations of immunologically mediated diseases (e.g., rheumatoid arthritis, lupus erythematosus, systemic lupus erythematosus, Hashimotos thyroiditis, multiple sclerosis, myasthenia gravis, type 1 diabetes, uveitis, nephrotic syndrome, psoriasis, atopical

dermatitis, contact dermatitis and further eczematous dermatitides, seborrheic dermatitis, Lichen planus, Pemplugus, bullous Pemphigold, Epidermolysis bullosa, uritcaris, angiodemas, vasculitides, erythemas, cutaneous eosinophilias, Alopecia areata, etc.); the treatment of reversible obstructive airways disease, intestinal inflammations and allergies (e.g., Coeliac disease, proctitis, eosinophilia gastroenteritis, mastocytosis, Chrohn's disease and ulcerative colitis) and food related allergies (e.g., migrane, rhinitis, and eczema).

One skilled in the art would be able, by routine experimentation, to determine what an effective, non-toxic amount of compound would be for the purpose of inducing immunosuppression. Generally, however, an effective dosage will be in the range of about 0.05 to 100 milligrams per kilogram body weight per day.

The compounds of this invention should also be useful for treating carcinogenic tumors in a mammal. More specifically, the compounds should be useful for reducing tumor size, inhibiting tumor growth and/or prolonging the survival time of tumor-bearing animals. Accordingly, this invention also relates to a method of treating carcinogenic tumors in a human or other animal comprising administering to such human or animal an effective, non-toxic amount of a compound of Formula II. One skilled in the art would be able, by routine experimentation, to determine what an effective, non-toxic amount of compound would be for the purpose of treating carcinogenic tumors. Generally, however, an effective dosage is expected to be in the range of about 0.05 to 100 milligrams per kilogram body weight per day.

The compounds of the invention may be administered to a human or other animal in accordance with the aforementioned methods of treatment in an amount sufficient to produce such effect to a therapeutic or prophylactic degree. Such compound of the

invention can be administered to such human or other animal in a conventional dosage form prepared by combining the compound of the invention with a conventional pharmaceutically acceptable carrier or diluent according to known techniques. It will be recognized by one of skill in the art that the form and character of the pharmaceutically acceptable carrier or diluent is dictated by the amount of active ingredient with which it is to be combined, the route of administration and other well-known variables.

The route of administration of the compound of the invention may be oral, parenteral, by inhalation or topical. The term parenteral as used herein includes intravenous, intramuscular, subcutaneous, rectal, vaginal or intraperitoneal administration. The subcutaneous and intramuscular forms of parenteral administration are generally preferred.

The daily parenteral and oral dosage regimens for employing compounds of the invention to prophylactically or therapeutically inhibit the growth of pathogenic fungi, to prophylatically or therapeutically induce immunosuppression, or to therapeutically treat carcinogenic tumors will generally be in the range of about 0.05 to 100, but preferably about 0.5 to 10, milligrams per kilogram body weight per day.

The compounds of the invention may also be administered by inhalation. By "inhalation" is meant intranasal and oral inhalation administration. Appropriate dosage forms for such administration, such as an aerosol formulation or a metered dose inhaler, may be prepared by conventional techniques. The preferred dosage amount of a compound of the invention to be employed is generally within the range of about 10 to 100 milligrams.

The compounds of the invention may also be administered topically. By topical administration is

meant non-systemic administration and includes the application of a compounds of the invention externally to the epidermis, to the buccal cavity and instillation of such a compound into the ear, eye and nose, and where the compound does not significantly enter the blood stream. By systemic administration is meant oral, intravenous, intraperitoneal and intramuscular administration. The amount of a compound of the invention (hereinafter referred to as the active ingredient) required for therapeutic or prophylactic effect on pathogenic fungi growth inhibition or immunosuppression induction upon topical administration will, of course, vary with the compound chosen, the nature and severity of the condition being treated and the animal undergoing treatment, and is ultimately at the discretion of the physician. A suitable topical dose of a compound of the invention will generally be within the range of about 1 to 100 milligrams per kilogram body weight daily.

While it is possible for an active ingredient to be administered alone as the raw chemical, it is preferable to present it as a pharmaceutical formulation. The active ingredient may comprise, for topical administration, from 0.001% to 10% w/w, e.g., from 1% to 2% by weight of the formulation although it may comprise as much as 10% w/w but preferably not in excess of 5% w/w and more preferably from 0.1% to 1% w/w of the formulation.

The topical formulations of the present invention, both for veterinary and for human medical use, comprise an active ingredient together with one or more acceptable carrier(s) therefore and optionally any other therapeutic ingredient(s). The carrier(s) must be "acceptable" in the sense of being compatible with the other ingredients of the formulation and no deleterious to the recipient thereof.

Formulations suitable for topical administration

include liquid or semi-liquid preparations suitable for penetration through the skin to the site of where treatment is required such as: liniments, lotions, creams, ointments or pastes, and drops suitable for administration to the eye, ear or nose.

Drops according to the present invention may comprise sterile aqueous or oily solutions or suspensions and may be prepared by dissolving the active ingredient in a suitable aqueous solution of a bactericidal and/or fungicidal agent and/or any other suitable preservative, and preferably including a surface active agent. The resulting solution may then be clarified by filtration, transferred to a suitable container which is then sealed and sterilized by autoclaving or maintaining at 90-100 C for half an hour. Alternatively, the solution may be sterilized by filtration and transferred to the container by an aseptic technique. Examples of bactericidal and fungicidal agents suitable for inclusion in the drops are phenylmercuric nitrate or acetate (0.002%), benzalkonium chloride (0.01%) and chlorhexidine acetate (0.01%). Suitable solvents for the preparation of an oily solution include glycerol, diluted alcohol and propylene glycol.

Lotions according to the present invention include those suitable for application to the skin or eye. An eye lotion may comprise a sterile aqueous solution optionally containing a bactericide and may be prepared by methods similar to those for the preparation of drops. Lotions or liniments for application to the skin may also include an agent to hasten drying and to cool the skin, such as an alcohol or acetone, and/or a moisturizer such as glycerol or an oil such as castor oil or arachis oil.

Creams, ointments or pastes according to the present invention are semi-solid formulations of the active ingredient for external application. They may be

made by mixing the active ingredient in finely-divided or powdered form, alone or in solution or suspension in an aqueous or non-aqueous fluid, with the aid of suitable machinery, with a greasy or non-greasy basis. The basis may comprise hydrocarbons such as hard, soft or liquid paraffin, glycerol, beeswax, a metallic soap; a mucilage; an oil of natural origin such as almond, corn, arachis, castor or olive oil; wool fat or its derivatives, or a fatty acid such as stearic or oleic acid together with an alcohol such as propylene glycol or macrogols. The formulation may incorporate any suitable surface active agent such as an anionic, cationic or non-ionic surface active such as sorbitan esters or polyoxyethylene derivatives thereof. Suspending agents such as natural gums, cellulose derivatives or in organic materials such as silicaceous silicas, and other ingredients such as lanolin, may also be included.

It will be recognized by one of skill in the art that the optimal quantity and spacing of individual dosages of the compound of the invention will be determined by the nature and extent of the condition being treated, the form, route and site of administration, and the particular animal being treated, and that such optimums can be determined by conventional techniques. It will also be appreciated by one of skill in the art that the optimal course of treatment, i.e., the number of doses of the compound of the invention given per day for a defined number of days, can be ascertained by those skilled in the art using conventional course of treatment determination tests.

Without further elaboration, it is believed that one skilled in the art can, using the preceding description, utilize the present invention to its fullest extent. The following Examples are, therefore, to be construed as merely illustrative and not a limitation of the scope of the present invention in any

way.

EXAMPLES

I. COMPOSITION EXAMPLES

EXAMPLE A - CAPSULE COMPOSITION

A pharmaceutical composition of this invention in the form of a capsule is prepared by filling a standard two-piece hard gelatin capsule with 50 mg of a compound of the invention, in powdered form, 100 mg of lactose, 32 mg of talc and 8 mg of magnesium stearate.

EXAMPLE B - INJECTABLE PARENTERAL COMPOSITION

A pharmaceutical composition of this invention in a form suitable for administration by injection is prepared by stirring 1.5% by weight of a compound of the invention in 10% by volume propylene glycol and water. The solution is sterilized by filtration.

EXAMPLE C - OINTMENT COMPOSITION

Compound of the invention 1.0 g
White soft paraffin to 100.0 g

The compound of the invention is dispersed in a small volume of the vehicle and gradually incorporated into the bulk of the vehicle to produce a smooth, homogeneous product. Collapsible metal tubes are then filled with the dispersion.

EXAMPLE D - TOPICAL CREAM COMPOSITION

Compound of the invention 1.0 g
Polawax GP 200 20.0 g
Lanolin Anhydrous 2.0 g
White Beeswax 2.5 g
Methyl hydroxybenzoate 0.1 g
Distilled Water to 100.0 g

The polawax, beeswax and lanolin are heated

together at 60 C. A solution of methyl hydroxybenzoate is added and homogenization is achieved using high speed stirring. The temperature is then allowed to fall to 50 C. The compound of the invention is then added and dispersed throughout, and the composition is allowed to cool with slow speed stirring.

EXAMPLE E - TOPICAL LOTION COMPOSITION

Compound of the invention 1.0 g
Sorbitan Monolaurate 0.6 g
Polysorbate 20 0.6 g
Cetostearyl Alcohol 1.2 g
Glycerin 6.0 g
Methyl Hydroxybenzoate 0.2 g
Purified Water B.P. to 100.00 ml

The methyl hydroxybenzoate and glycerin are dissolved in 70 ml of the water at 75 . The sorbitan monolaurate, polysorbate 20 and cetostearyl alcohol are melted together at 75 C and added to the aqueous solution. The resulting emulsion is homogenized, allowed to cool with continuous stirring and the compound of the invention is added as a suspension in the remaining water. The whole suspension is stirred until homogenized.

EXAMPLE F - EYE DROP COMPOSITION

Compound of the invention 0.5 g
Methyl Hydroxybenzoate 0.01 g
Propyl Hydroxybenzoate 0.04 g
Purified water B.P. to 100.00 ml (B.P.=British Pharmacopia)

The methyl and propyl hydroxybenzoates are dissolved in 70 ml purified water at 75 C and the resulting solution is allowed to cool. The compound of the invention is then added, and the solution is sterilized by filtration through a membrane filter (0.22 mu m pore size) and packed aseptically into suitable

sterile containers.

EXAMPLE G - COMPOSITION FOR ADMINISTRATION BY INHALATION

For an aerosol container with a capacity of 15-20 ml: Mix 10 mg of a compound of the invention with 0.2-0.2% of a lubricating agent, such as polysorbate 85 or oleic acid, and disperse such mixture in a propellant, such as freon, preferably in a combination of (1,2 dichlorotetrafluoroethane) and difluorochloromethane and put into an appropriate aerosol container adapted for either intranasal or oral inhalation administration

EXAMPLE H - COMPOSITION FOR ADMINISTRATION BY INHALATION

For an aerosol container with a capacity of 15-20 ml: Dissolve 10 mg of a compound of the invention in ethanol (6- 8 ml), add 0.1-0.2% of a lubricating agent, such as polysorbate 85 or oleic acid; and disperse such in a propellant, such as freon, preferably a combination of (1.2 dichlorotetrafluoroethane) and difluorochloromethane, and put into an appropriate aerosol container adapted for either intranasal or oral inhalation administration.

II. SYNTHETIC EXAMPLES

In the following Examples, all starting materials and chemical reagents were obtained from commercial suppliers unless otherwise indicated. Rapamycin was obtained from fermentation as described above.

General Procedure for C-7 Solvolysis

The following Methods A and B were used to prepare compounds of Formula II.

Method A: Rapamycin (91.4mg, 0.10 mmol) in dry dichloromethane (5mL) was treated with trifluoroacetic

acid (0.5mL) at -40° C under argon. A bright yellow color developed immediately and the resulting solution was stirred at -40° C for 30 to 60 min. After that time, an excess of the appropriate nucleophile was added dropwise and stirring was continued for 30 min at -40° C. The mixture was then partitioned between ethyl acetate and saturated aqueous sodium bicarbonate. Layers were separated and the organic layer successively washed with saturated sodium bicarbonate, brine and dried over anhydrous sodium sulfate. The resulting crude material was subjected to preparative HPLC purification to afford the desired 7-demethoxy-7-substituted rapamycins.

Method B: Rapamycin (100 mg, 0.109 mmol) in dry dichloromethane (2.5mL) was treated with trifluoroacetic acid (100μ L) at -40° C under argon. A bright yellow color developed immediately and the resulting solution was stirred at -40° C for 10 min. After that time, ten to fifteen equivalents of the appropriate nucleophile was added dropwise and stirring was continued for 15 min at -40° C. The mixture was then partitioned between dichloromethane and 5% aqueous sodium bicarbonate. Layers were separated and the organic layer successively washed with saturated sodium bicarbonate, brine and dried over anhydrous sodium sulfate. The resulting crude material was subjected to preparative HPLC purification to afford the desired 7-demethoxy-7-substituted rapamycins.

Example 1. 7-Demethoxy-7(S)-ethoxyrapamycin

Method A was used to prepare the title compound: ^{1}H NMR($CDCl_3$, 400 MHz, 4:1 mixture of trans:cis amide rotamers; data for the trans-rotamer); _ 6.375 (dd, J=14.8, 10.5Hz, 1H), 6.300 (dd, J=14.8, 9.8Hz, 1H), 6.134 (dd, J=14.9, 9.5Hz, 1H), 5.940 (d, J=10.0 Hz, 1H), 5.533 (dd, J=14.9, 9.2Hz, 1H), 5.408 (d, J=9.7 Hz, 1H),

5.29 (m, 1H), 5.17 (m, 1H), 4.799 (s, 1H), 4.173 (d, J=5.9 Hz, 1H), 3.87 (m, 1H), 3.775 (dd, J=7.8, 7.0Hz, 1H) 3.714 (d, J=5.9 Hz, 1H), 3.576 (br d, J=14.0 Hz, 1H), 3.407 (s, 3H), 3.338 (s, 3H), 3.161 (dq, J=9.8, 6.9Hz, 1H), 2.587 (dd, J=17.0, 6.3Hz, 1H), 1.751 (s, 3H), 1.662 (s, 3H), 1.152 (t, J=6.9 Hz, 3H), 1.098 (d, J=6.7 Hz, 3H), 1.047 (d, J=6.5 Hz, 3H), 0.991 (d, J=6.6 Hz, 3H), 0.951 (d, J=6.6 Hz, 3H), 0.917 (d, J=6.7 Hz, 3H), 0.668 (q, J=12.0 Hz, 1H); MS (ESI^+/NH_4OAc) m/z 950 ($M+Na^+$), 945 ($M+NH_4^+$); MS (ESI^-/NH_4COOH) m/z 972 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm.

Example 2. 7-Demethoxy-7(R)-ethoxyrapamycin

Method A was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 2.5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.365 (dd, J=14.2, 11.0Hz, 1H), 6.185 (dd, J=14.2, 10.4Hz, 1H), 6.12-6.05 (m, 2H), 5.471 (dd, J=14.4, 9.1Hz, 1H), 5.421 (d, J=10.3 Hz, 1H), 5.25-5.20 (m, 2H), 4.260 (br s, 1H), 4.10-4.04 (m, 1H), 4.050 (s, 1H), 4.012 (d, J=3.6 Hz, 1H), 3.698 (dd, J=8.1, 1.5Hz, 1H), 3.394 (s, 3H), 3.332 (s, 3H), 3.232 (dt, J=13.4, 7.0Hz, 1H), 2.718 (dd, J=17.4, 2.7Hz, 1H), 2.376 (dd, J=17.4, 8.5Hz, 1H), 1.750 (s, 3H), 1.654 (s, 3H), 1.180 (t, J=7.0 Hz, 3H), 1.059 (d, J=6.7 Hz, 3H), 1.005 (d, J=6.6 Hz, 3H), 0.941 (d, J=6.5 Hz, 3H), 0.925 (d, J=6.5 Hz, 3H), 0.863 (d, J=6.6 Hz, 3H), 0.646 (q, J=12.0 Hz, 1H); MS (ESI^+/NH_4OAc) m/z 950 ($M+Na^+$), 945 ($M+NH_4^+$); MS (ESI^-/NH_4COOH) m/z 972 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm.

Example 3. 7-Demethoxy-7(S)-(3,4-dimethoxybenzyl-oxy)rapamycin

Method A was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer); _ 6.87-6.79 (m, 3H), 6.412 (dd, J=14.7, 10.9Hz, 1H), 6.291 (dd, J=14.7, 10.4Hz, 1H), 6.148 (dd, J=14.9, 10.4Hz, 1H), 5.926 (d,

J=10.9 Hz, 1H), 5.503 (dd, J=14.9, 9.1Hz, 1H), 5.394 (d, J=9.9 Hz, 1H), 5.211 (d, J=4.8 Hz, 1H), 5.142 (td, J=6.0, 4.0Hz, 1H), 4.744 (s, 1H), 4.418 (d, J=12.0 Hz, 1H), 4.157 (d, J=6.0, Hz, 1H), 4.110 (d, J=12.0 Hz, 1H), 3.884 (s, 3H), 3.867 (s, 3H), 3.824 (t, J=7.6 Hz, 1H), 3.502 (br d J=13.4 Hz, 1H), 3.404 (s, 3H), 3.325 (s, 3H), 2.732 (dd, J=17.0, 5.8Hz, 1H), 2.539 (dd, J=17.0, 6.3Hz, 1H), 1.735 (s, 6H), 1.081 (d, J=6.7 Hz, 3H), 1.054 (d, J=6.6 Hz, 3H), 0.988 (d, J=6.6 Hz, 3H), 0.931 (d, J=6.6 Hz, 3H), 0.893 (d, J=6.7 Hz, 3H), 0.661 (q, J=11.9 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100.6 MHz, 5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 215.7, 208.2, 192.7, 169.3, 166.7, 149.1, 148.5, 140,5, 136.0, 135.9, 133.3, 131.3, 130.4, 129.7, 126.8, 126.5, 119.8, 111.0, 110.7, 98.6, 84.9, 84.4, 81.3, 77.2, 75.5, 74.0, 69.3, 67.3, 59.5, 56.6, 55.9, 55.8, 51.4, 46.5, 44.3, 41.4; MS (ESI^+/NH_4OAc) m/z 1072 ($M+Na^+$), 1049 ($M+NH_4^+$); UV (MeOH) max 267, 278, 289 nm.

Example 4. 7-Demethoxy-7(S)-(2-hydroxyethoxy)rapamycin

Method B was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 4:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.371 (dd, J=14.6, 10.0Hz, 1H), 6.303 (dd, J=14.6, 9.6Hz, 1H), 6.138 (dd, J=15.0, 9.6Hz, 1H), 5.962 (d, J=9.7 Hz, 1H), 5.538 (dd, J=15.0, 8.9Hz, 1H), 5.408 (d, J=9.9 Hz, 1H), 5.282 (d, J=4.7 Hz, 1H), 5.164 (td, J=6.2, 4.0Hz, 1H), 4.849 (s, 1H), 4.170 (d, J=6.0 Hz, 1H), 3.822 (dd, J=8.0, 7.2Hz, 1H), 3.711 (d, J=6.0 Hz, 1H), 3.683 (t, J=4.0 Hz, 1H), 3.582 (br d, J=14.0 Hz, 1H), 3.408 (s, 3H), 3.336 (s, 3H), 3.244 (ddd, J=10.5, 5.5, 4.0Hz, 1H), 2.739 (dd, J=16.8, 6.0Hz, 1H), 2.595 (dd, J=16.8, 6.3Hz, 1H), 1.748 (s, 3H), 1.668 (s, 3H), 1.103 (d, J=6.8 Hz, 3H), 1.050 (d, J=6.7 Hz, 3H), 0.994 (d, J=6.5 Hz, 3H), 0.950 (d, J=6.6 Hz, 3H), 0.919 (d, J=6.7 Hz, 3H), 0.668 (q, J=11.9 Hz, 1H); MS (ESI^+/NH_4OAc) m/z 961 ($M+NH_4^+$), 882, 864; MS (ESI^-/NH_4COOH) m/z 988 ($M+HCOO^-$); UV (MeOH)

max 267, 277, 289 nm.

Example 5. 7-Demethoxy-7(R)-(2-hydroxyethoxy)rapamycin

Method A was used to prepare the title compound: ^{1}H NMR($CDCl_3$, 400 MHz 6:1 mixture of trans:cis amide rotamers; data for the trans-rotamer):_ 6.359 (dd, J=14.2, 11.1Hz, 1H), 6.174 (dd, J=14.2, 10.7Hz, 1H), 6.099 (dd, J=14.5, 10.7Hz, 1H), 6.055 (d, J=12.0 Hz, 1H), 5.448 (d, J=10.0 Hz, 1H), 5.377 (dd, J=14.5, 9.6Hz, 1H), 5.34 (m, 1H), 5.084 (dd, J=6.1, 2.7Hz, 1H), 4.295 (t, J=10.0 Hz, 1H), 4.327 (d, J=4.0 Hz, 1H), 4.065 (d, J=4.0 Hz, 1H), 4.026 (dd, J=10.8 2.3Hz, 1H), 3.781 (br d, J=12.8 Hz, 1H), 3.70-3.64 (m, 1H), 3.541 (br d, J=14.0 Hz, 1H), 3.465 (br d, J=11.5 Hz, 1H), 3.394 (s, 3H), 3.342 (s, 3H), 3.234 (dd, J=9.9, 6.7Hz, 1H), 3.092 (br t, J=9.2 Hz, 1H), 2.715 (dd, J=17.8, 5.7Hz, 1H), 2.560 (dd, J=17.8, 6.8Hz, 1H), 1.833 (s, 3H), 1.677(s, 3H), 1.049 (d, J=6.6 Hz, 3H), 0.979 (d, J=6.5 Hz, 3H), 0.963 (d, J=6.5 Hz, 3H), 0.932 (d, J=6.5 Hz, 3H), 0.883 (d, J=6.8 Hz 3H), 0.591 (q, J=11.9 Hz, 1H); MS (ESI^+/NH_4OAc) m/z 966 ($M+Na^+$), 961 ($M+NH_4^+$), 882; MS (ESI^-/NH_4COOH) m/z 988 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm.

Example 6. 7-Demethoxy-7(R)-acetoxyrapamycin

Method A was used to prepare the title compound: ^{1}H NMR($CDCl_3$, 400 MHz, 4:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.337 (dd, J=14.6, 10.7Hz, 1H), 6.233 (dd, J=14.6, 10.5Hz, 1H), 6.097 (d, J=10.7 Hz, 1H), 6.086(dd, J=14.9, 10.5Hz, 1H), 5.459 (dd, J=14.9, 9.0Hz, 1H), 5.423(d, J=9.7 Hz, 1H), 5.331 (d, J=10.3 Hz, 1H), 5.23-5.20 (m, 2H), 4.270 (d, J=3.7 Hz, 1H), 4.014 (d, J=3.7 Hz, 1H), 3.897 (t, J=10.0 Hz, 1H), 3.562 (br d, J=14.5 Hz, 1H), 3.392 (s, 3H), 3.333 (2, 3H), 2.708 (dd, J=17.5, 3.8Hz, 1H), 2.405 (dd, J=17.5, 8.0Hz, 1H), 2.067 (s, 3H), 1.751 (s, 3H), 1.727 (s, 3H), 1.062 (d, J=6.5 Hz, 3H), 1.018 (d, J=6.6

Hz, 3H), 0.952 (d, J=6.4 Hz, 3H), 0.906 (d, J=6.5 Hz, 3H), 0.873 (d, J=6.7 Hz, 3H), 0.642 (q, J=11.8 Hz, 1H); ^{13}C NMR ($CDCl_3$, 3:1 mixture of trans:cis amide rotamers; data for trans-rotamer) _ 212.4, 207.5, 195.5, 169.7, 166.2, 139.1, 137.7, 131.1, 130.4, 128.2, 125.9, 124.4, 98.7, 84.4, 84.0, 74.6, 73.9, 69.6; MS (FAB/NaCL) m/z 964 ($M+Na^+$); UV (MeOH) max 267, 277, 289 nm.

Example 7. 7-Demethoxy-7(S)-(2'-furanyl)rapamycin

Method A was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 3:1 mixture of trans:cis amide rotamers; data for the trans-rotamer); _ 7.290 (d, J=1.6 Hz, 1H), 6.289 (dd, J=3.2, 1.6Hz, 1H), 6.006 (d, J=3.2 Hz, 1H), 5.615 (dd, J=15.1, 8.2Hz, 1H), 5.424 (d, J=9.8 Hz, 1H), 5.336 (d, J=4.9 Hz, 1H), 5.191 (q, J=5.5 Hz, 1H), 5.096 (1H, s), 4.200 (d, J=6.0 Hz, 1H), 3.734 (d, J=6.0 Hz, 1H), 3.697 (dd, J=8.5, 8.1Hz, 1H), 3.408 (s, 3H), 3.357 (s, 3H), 2.689 (d, J=6.2 Hz, 2H), 1.775 (s, 3H), 1.559 (s, 3H), 1.136 (d, J=6.8 Hz, 3H), 1.051 (d, J=6.6 Hz, 3H), 1.006(d, J=6.5 Hz, 3H), 0.990 (d, J=6.5 Hz, 3H), 0.929 (d, J=6.8 Hz, 3H), 0.669 (q, J=12.0 Hz, 1H); MS (ESI^+/NH_4OAc) m/z 967 ($M+NH_4^+$), 932; MS (ESI^-/NH_4COOH) m/z 994 ($M+HCOO^-$); UV (MeOH) max 268, 278, 290 nm.

Example 8. 7-Demethoxyrapamycin

Method B (trimethylsilane as the nucleophile) was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 3:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.353 (dd, J=14.0, 11.1Hz, 1H), 6.151 (dd, J=14.1, 10.5Hz, 1H), 6.075 (dd, J=14.1, 10.4Hz, 1H), 5.874 (d, J=11.0 Hz, 1H), 5.387 (d, J=10.5 Hz, 1H), 5.343 (dd, J=14.4, 10.5Hz, 1H), 5.24-5.18 (m, 2H), 4.248 (br t, 1H), 4.197 (s, 1H), 4.005 (d, J=4.0 Hz, 1H), 3.91-3.87 (m, 1H), 3.457 (br d, J=11.5 Hz, 1H), 3.397 (s, 3H), 3.336 (s, 3H), 2.719 (dd, J=17.6, 4.6 Hz, 1H), 2.445(dd, J=17.6, 7.4 Hz, 1H), 1.793 (s, 3H), 1.722

(s, 3H), 1.0535 (d, J=6.6 Hz, 3H), 0.997 (d, J=6.7 Hz, 3H), 0.940(d, J=6.5 Hz, 3H), 0.913(d, J=6.7 Hz, 3H), 0.891 (d, J=6.9 Hz, 3H), 0.626 (q, J=11.9 Hz, 1H); ^{13}C NMR ($CDCl_3$, 3:1 mixture of trans:cis amide rotamers; data for trans-rotamer) _ 212.4, 207.5, 195.5, 169.7, 166.2, 139.1, 137.7, 131.1, 130.4, 128.2, 125.9, 124.4, 98.7, 84.4, 84.0, 74.6, 73.9, 69.6; MS(ESI^+/NH_4OAc) m/z 906 ($M+Na^+$), 901($M+NH_4^+$); MS (ESI^-/NH_4COOH) m/z 928 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm.

Example 9. 7-Demethoxy-7(S)-methylthiorapamycin

Method B was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 3:1 mixture of trans:cis amide rotamer; data for the trans-rotamer): _ 6.363 (dd, J=15.4, 10.6Hz, 1H), 6.32-6.09 (m, 2H), 5.816 (d, J=10.5 Hz, 1H), 5.537 (dd, J=15.0, 9.4Hz, 1H), 5.403 (d, J=9.8 Hz, 1H), 5.289 (d, J=5.7 Hz, 1H), 5.20-5.14 (m, 2H), 5.020 (br s, 1H), 4.158 (d, J=7.0, 1H), 3.88-3.76 (m, 1H), 3.617 (d, J=7.0 Hz, 1H), 3.59-3.45 (m, 2H), 3.407 (s, 3H), 3.341 (s, 3H), 2.99-2.90 (m, 1H), 2.780 (dd, J=16.9, 6.5Hz, 1H), 2.641 (dd, J=16.9, 6.5Hz, 1H), 1.883 (s, 3H), 1.739 (s, 3H), 1.720 (s, 3H), 1.128 (d, J=6.8 Hz, 3H), 1.156 (d, J=6.5 Hz, 3H), 1.015 (d, J=6.5 Hz, 3H), 0.966 (d, J=6.5 Hz, 3H), 0.932 (d, J=6.7 Hz, 3H), 0.667 (q, J=11.4 Hz, 1H); ^{13}C NMR ($CDCl_3$, 3:1 mixture of trans:cis amide rotamers; data for trans-rotamer) _ 215.7, 208.4, 190.7, 169.2, 166.9, 140.3, 136.3, 134.3, 133.3, 130.1, 129.4, 127.1, 126.4, 98.3, 85.0, 84.4, 75.9, 73.9, 67.4; MS (FAB/NaCl) m/z 952 ($M+Na^+$); MS (ESI^+/NH_4OAc) m/z 952 ($M+Na^+$), 947, ($M+NH_4^+$), 882, 864; MS (ESI^-/NH_4COOH) m/z 974 ($M+HCOO^-$); UV (MeOH) max 266, 277, 288 nm.

Example 10. 7-Demethoxy-7(R)-methylthiorapamycin

Method B was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 2.5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.388 (dd,

J=14.5, 11.0Hz, 1H), 6.22-6.06 (m, 2H), 5.884 (d, J=10.9 Hz, 1H), 5.45-5.38 (m, 2H), 5.22-5.14 (m, 2H), 4.278(br t, 1H), 4.065 (d, J=3.4 Hz, 1H), 4.030 (s, 1H), 3.48-3.41 (m, 1H) 3.395 (s, 3H), 3.334 (s, 3H), 3.18-3.09 (br t, J=9.8, 1H), 2.710 (dd, J=17.5, 3.2Hz, 1H), 2.384 (dd, J=17.6, 7.8Hz, 1H), 1.814 (s, 6H), 1.754 (s, 3H), 1.715 (s, 3H), 1.059 (d, J=6.5 Hz, 3H), 0.989 (d, J=6.5 Hz, 3H), 0.939 (d, J=6.5 Hz, 3H), 0.915 (d, J=6.8 Hz, 3H), 0.877 (d, J=6.9 Hz, 3H), 0.636 (q, J=11.7 Hz, 1H); MS (FAB/NaCl) m/z 952 ($M+Na^+$); MS (ESI^+/NH_4OAc) m/z 952 ($M+Na^+$), 947 ($M+NH_4^+$), 882, 864; MS (ESI^-/NH_4COOH) m/z 974 ($M+HCOO^-$); UV (MeOH) max 266, 277, 288 nm.

Example 11. 7-Demethoxy-7(S)-phenylthiorapamycin

Method B was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 4:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 7.35-7.15 (m, 5H), 6.300 (dd, J=14.4, 11.2Hz, 1H), 6.17-6.06 (m, 2H), 5.552 (d, J=11.1 Hz, 1H), 5.510 (dd, J=14.0, 8.1Hz, 1H), 5.378(d, J=9.8 Hz, 1H), 5.19-5.09 (m, 2H), 4.111 (d, J=7.2 Hz, 1H), 3.824 (br t, 1H), 3.711 (dd, J=12.0, 4.9Hz, 1H), 3.549 (d, J=7.2 Hz, 1H), 3.537 (br d, 1H), 3.409 (s, 3H), 3.307 (s, 3H), 2.99- 2.90 (m, 1H), 2.785 (dd, J=17.0, 7.2Hz, 1H), 2.611 (dd, J=16.9, 5.5Hz, 1H), 1.814 (s, 3H), 1.707 (s, 3H), 1.131 (d, J=6.7 Hz, 3H), 1.033 (d, J=6.5 Hz, 3H), 0.994 (d, J=6.5 Hz, 3H), 0.950 (d, J=6.5 Hz, 3H), 0.921 (d, J=6.8, 3H), 0.669 (q, J=15.8 Hz, 1H); ^{13}C NMR ($CDCl_3$, 4:1 mixture of trans:cis amide rotamers, data for trans-rotamer) _ 214.8, 208.7, 193.1, 169.0, 166.4, 140.5, 136.5, 133.4, 132.3, 130.4, 128.5, 126.4, 125.4, 98.4, 85.2, 84.4, 75.9, 73.9, 67.5; MS (ESI^+/NH_4OAc) m/z 1009 ($M+NH_4^+$); MS (ESI^-/NH_4COOH) m/z 1036 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm.

Example 12. 7-Demethoxy-7(R)-phenylthiorapamycin

Method B was used to prepare the title compound: ^{1}NMR($CDCl_3$, 400 MHz, 4:1 mixture of trans:cis amide

rotamers; data for the trans-rotamer): _ 7.38-7.18 (m, 5H), 6.37-6.28 (m, 14.2, 1H), 6.13-5.89 (m, 2H), 5.644 (d, J=10.8 Hz, 1H), 5.421 (d, J=10.3 Hz, 1H), 5.25-5.20 (m, 2H), 4.260 (br s, 1H), 5.40-5.28 (m, 2H), 4.385 (br t, J=11.0, 1H), 4.226 (br t, 1H), 4.078 (d, J=3.3, 1H), 3.563 (dd, J=10.7, 2.3Hz, 1H), 3.386 (s, 3H), 3.323 (s, 3H), 3.200 (dd, J=10.5, 6.6Hz, 1H), 3.16-3.06 (m, 1H), 2.98-2.87 (m, 2H), 2.698 (dd, J=17.6, 3.0Hz, 1H), 2.263 (dd, J=17.6, 8.4Hz, 1H), 1.898 (s, 3H), 1.770 (s, 3H), 1.062 (d, J=6.6 Hz, 3H), 0.971 (d, J=6.6 Hz, 3H), 0.926 (d, J=6.4 Hz, 3H), 0.883 (d, J=6.5 Hz, 3H), 0.845 (d, J=6.8 Hz, 3H), 0.623 (q, J=12.0 Hz, 1H); MS (ESI^+/NH_4OAc) m/z 1009 ($M+NH_4^+$); MS (ESI^-/NH_4COOH) m/z 1036 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm

Example 13. 7-Demethoxy-7(S)-allylrapamycin

Method B; ^1H NMR($CDCl_3$, 400 MHz, 3:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.368 (dd, J=15.6, 11.4 Hz, 1H), 6.257 (dd, J=15.4, 11.2 Hz, 1H), 6.133 (dd, J=15.7, 11.1 Hz, 1H), 5.785 (d, J=11.4 Hz, 1H), 5.69-5.60 (m, 1H), 5.564 (dd, J=15.9, 9.4 Hz, 1H), 5.413 (d, J=10.7 Hz, 1H), 5.294 (d, J=6.2 Hz, 1H), 5.21- 5.16 (m, 1H), 5.02-4.87 (m, 2H), 4.168 (d, J=7.2 Hz, 1H), 3.774 (br. t, J=11.4, Hz, 1H), 3.642 (d, J=7.2 Hz, 1H), 3.562 (s, 1H), 3.502 (br d J=13.4 Hz, 1H), 3.410 (s, 3H), 3.350 (s, 3H), 2.99-2.86 (m, 2H), 2.755 (dd, J = 17.4, 7.0, 1H), 2.663 (dd, J = 17.4, 6.2, 1H), 1.761 (s, 3H), 1.607 (s, 3H), 1.135 (d, J=6.7 Hz, 3H), 1.044 (d, J=6.6 Hz, 3H), 1.013 (d, J=6.6 Hz, 3H), 0.972 (d, J=6.6 Hz, 3H), 0.924 (d, J=6.8 Hz, 3H), 0.671 (q, J=12.0 Hz, 1H); ^{13}C NMR ($CDCl_3$, 3:1 mixture of trans:cis amide rotamers; data for the trans- rotamer) _ 215.6, 208.7, 192.0, 169.4, 167.1, 139.1, 137.6, 137.2, 136.4, 132.1, 130.1, 128.4, 127.2, 126.9, 115.3, 98.2, 84.8, 84.4, 77.7, 76.0, 73.9, 67.2;, MS (ESI^+/NH_4OAc) m/z 946 ($M+Na^+$), 941 ($M+NH_4^+$); UV (MeOH) max 267, 278, 289 nm.

Example 14. 7-Demethoxy-7(R)-(2'-furanyl)rapamycin

Method A was used to prepare the title compound: ^1H NMR ($CDCl_3$, 400MHz, 3.5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 7.302 (d, J=1.8 Hz, 1H), 6.356 (dd, J=14.6, 10.9Hz, 1H), 6.292 (dd, J=3.0, 1.8Hz, 1H), 6.190 (dd, J=14.6, 10.4Hz, 1H), 6.044 (br d, J=10.9 Hz, 1H), 6.019 (d, J=3.0 Hz, 1H), 5.390 (d, J=10.3 Hz, 1H), 5.327 (dd, J=14.7, 9.4Hz, 1H), 5.24-5.09 (m, 2H), 4.294 (br s), 4.116 (d, J=3.1 Hz, 1H), 3.805 (s, 1H), 3.524 (br d, J=15.0 Hz, 1H), 3.469 (dd, J=11.7, 3.6Hz, 1H), 3.391 (s, 3H), 3.335 (s, 3H), 3.245 (dq, J=10.3, 6.6 Hz,1H), 2.673 (dd, J=17.7, 2.8 Hz, 1H), 2.360 (dd, J=17.7, 8.4Hz, 1H), 1.825 (s, 3H), 1.662 (s, 3H), 1.058 (d, J=6.4 Hz, 3H), 0.973 (d, J=6.5 Hz, 3H), 0.924 (d, J=6.4 Hz, 3H), 0.888 (d, J=6.4 Hz, 3H), 0.883 (d, J=6.5 Hz, 3H), 0.633 (q, J=11.6 Hz, 1H); MS (ESI+/NH_4OAc) m/z 972 ($M+Na^+$), 967 ($M+NH_4^+$); MS (ESI$^-$ /NH_4COOH) m/z 994 ($M+HCOO^-$); UV (MeOH) $_{max}$ 268, 278, 290 nm.

Example 15. 7-Demethoxy-7(S)-(2'4'-dimethoxyphenyl)-rapamycin

Method A was used to prepare the title compound: ^1H NMR ($CDCl_3$, 400MHz, 2.5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 7.087 (d, J=8.4 Hz, 1H), 6.450 (dd, J=8.4, 2.2Hz, 1H), 6.422 (d, J=2.2 Hz, 1H), 6.326 (dd, J=14.6, 10.5Hz, 1H), 6.232 (dd, J=14.6, 10.4Hz, 1H), 6.14-6.07 (m, 2H), 5.465 (dd, J=14.9, 9.8Hz, 1H), 5.421 (d, J=10.5 Hz, 1H), 5.312 (d, J=5.3 Hz, 1H), 5.177 (q, J=5.4 Hz, 1H), 4.202 (br d, J=5.0 Hz, 1H), 3.912 (t, J=8.4 Hz, 1H), 3.792 (s, 3H), 3.738 (s, 3H), 3.565 (br d, J=13.4 Hz, 1H), 3.410 (s, 3H), 3.353 (s, 3H), 2.772 (dd, J=17.1, 5.5Hz, 1H), 2.596 (dd, J=17.1, 6.4 Hz, 1H), 1.817 (s, 3H), 1.500 (s, 3H), 1.096 (d, J=6.7 Hz, 3H), 1.038 (d, J=6.7 Hz, 3H), 0.980 (d, J=6.5 Hz, 3H), 0.935 (d, J=6.5 Hz, 3H), 0.924 (d,

J=6.7 Hz, 3H), 0.674 (q, J=11.8 Hz, 1H); MS (ESI+/NH_4OAc) m/z 1037 ($M+NH_4^+$); MS (ESI$^-$/NH_4COOH) m/z 1064 ($M+HCOO^-$).

Example 16. 7-Demethoxy-7(R)-(2',4'-dimethoxyphenyl)-rapamycin

Method A was used to prepare the title compound: ^1H NMR ($CDCl_3$, 400MHz, 10:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.921 (d, J=9.1 Hz, 1H), 6.47-6.42 (m, 2H), 6.371 (dd, J=14.7, 11.0Hz, 1H), 6.225 (dd, J=14.7, 10.5Hz, 1H), 6.090 (br d, J=11.0 Hz, 1H), 6.059 (dd, J=14.9, 10.5Hz, 1H), 5.369 (d, J=10.4 Hz, 1H), 5.29-5.23 (m, 2H), 5.191 (br d, J=2.8 Hz, 1H), 4.323 (br s), 4.205 (d, J=2.7 Hz, 1H), 3.906 (br d, J=11.4 Hz, 1H), 3.790 (s, 3H), 3.743 (s, 3H), 3.579 (d, J=1.0 Hz, 1H), 3.408 (br t, J=10.0 Hz, 1H), 3.396 (s, 3H), 3.341 (s, 3H), 3.246 (dq, J=10.3, 6.4Hz, 1H), 2.775 (d, J=18.0 Hz, 1H), 2.274 (dd, J=18.0, 9.1Hz, 1H), 1.884 (s, 3H), 1.511 (s, 3H), 1.069 (d, J=6.5 Hz, 3H), 0.926 (d, J=6.6 Hz, 3H), 0.906 (d, J=6.5 Hz, 3H), 0.890 (d, J=6.5 Hz, 3H), 0.857 (d, J=6.6 Hz, 3H), 0.651 (q, J=11.8 Hz, 1H); MS (ESI+/NH_4OAc) m/z 1037 ($M+NH_4^+$); 1019 ($M+Na^+$); MS (ESI$^-$/NH_4COOH) m/z 1064 ($M+HCOO^-$).

Example 17. 7-Demethoxy-7-oxorapamycin

To a solution of rapamycin (5 mg, 0.055 mmol) in dichloromethane (0.1 mL) was added water (0.02 mL) followed by DDQ (2.4 mg, 0.011 mmol), and the resulting dark brown suspension was stirred at room temperature for 45 min. The mixture was filtered through celite and then partitioned between dichloromethane and brine; the organic extracts were dried over anhydrous magnesium sulfate and the resulting crude material was purified by prep TLC. The desired product was isolated as a colorless oil (1.2 mg). ^1H NMR ($CDCl_3$, 400MHz, 3:1

mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 7.145 (d, J=10.4 Hz, 1H), 6.635 (dd, J=14.9, 10.6Hz, 1H), 6.57-6.48 (m, 2H), 6.224 (dd, J=15.1, 10.6Hz, 1H), 5.662 (dd, J=15.1, 8.6Hz, 1H), 5.447 (d, J=11.2 Hz, 1H), 4.305 (br d, J=5.4 Hz, 1H), 3.983 (d, J=5.4 Hz, 1H), 3.385 (s, 3H), 3.346 (s, 3H),; MS (ESI+/NH_4OAc) $_{m/z}$ 920 ($M+Na^+$), 915 ($M+NH_4^+$), 898, 880; UV (MeOH) $_{max}$ 318nm.

Example 18. 7-O-Demethylrapamycin

Method A was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 2.5:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.356 (dd, J=14.8, 11.2Hz, 1H), 6.241 (dd, J=14.8, 10.6Hz, 1H), 6.089 (dd, J=14.9, 10.6Hz, 1H), 5.382 (dd, J=14.9, 9.7Hz, 1H), 5.337 (d, J=11.0 Hz, 1H), 5.28-5.21 (m, 2H), 4.580 (s, 1H), 4.239 (br s, 1H), 4.07-4.03 (m, 1H), 4.001 (d, J=4.0 Hz, 1H), 3.539 (br d, J=13.0 Hz, 1H), 3.380 (s, 3H), 3.333 (s, 3H), 3.192 (d, J=2.8 Hz, 1H), 2.563 (dd, J=17.7, 5.0Hz, 1H), 1.819 (s, 3H), 1.684 (s, 3H), 1.054 (d, J=6.6 Hz, 3H), 0.991 (d, J=6.6 Hz, 3H), 0.970 (d, J=6.6 Hz, 3H), 0.934 (d, J=6.4 Hz, 3H), 0.879 (d, J=6.8 Hz, 3H), 0.552 (q, J=11.8 Hz, 1H); MS (FAB/NaCl) m/z 922 ($M+Na^+$), MS (ESI^+/NH_4OAc) m/z 922 ($M+Na^+$) 917 ($M+NH_4^+$), 882, 864; MS(ESI^-/NH_4COOH) m/z 944 ($M+HCOO^-$); UV (MeOH) max 266, 277, 288nm

Example 19. 7-O-Demethyl-7-epirapamycin

Method A was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 4:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 6.397 (dd, J=14.6, 10.9Hz, 1H), 6.270 (dd, J=14.6, 10.7Hz, 1H), 6.160 (d, J=10.9 Hz, 1H), 6.131 (dd, J=15.0, 10.7Hz, 1H), 5.470 (dd, J=15.0, 8.7Hz, 1H), 5.401 (d, J=9.8 Hz, 1H), 5.271 (d, J=4.8 Hz, 1H), 5.193 (td, J=6.4, 3.5Hz, 1H), 4.799 (s, 1H), 4.200 (br d, J=5.0 Hz, 1H), 3.816 (d, J=5.4 Hz, 1H), 3.573 (br d, J=13.0 Hz, 1H), 3.396

(s, 3H), 3.337 (s, 3H), 3.251 (d, J=2.5 Hz, 1H), 2.729 (dd, J=17.4, 6.4Hz, 1H), 2.631 (dd, J=17.4, 6.2Hz, 1H), 1.765 (s, 3H), 1.715 (s, 3H), 1.076 (d, J=6.8 Hz, 3H), 1.055 (d, J=6.7 Hz, 3H), 0.978 (d, J=6.4 Hz, 3H), 0.948 (d, J=6.6 Hz, 3H), 0.914 (d, J=6.8 Hz, 3H), 0.627 (q, J=11.7 Hz, 1H); MS (FAB/NaCl) m/z 922 ($M+Na^+$); MS (ESI^+/NH_4OAc) m/z 922 (M+Na4+), 917 ($M+NH_4^+$), 882, 864; MS (ESI^-/NH_4COOH) m/z 944 ($M+HCOO^-$); UV (MeOH) max 266, 277, 288 nm.

Example 20. 7-Epirapamycin

Method A was used to prepare the title compound: ^1H NMR($CDCl_3$, 400 MHz, 3:1 mixture of trans:cis amide rotamers; data for the trans-rotamer):_ 6.377 (dd, J=14.4, 11.1Hz, 1H), 6.23-6.07 (3H, m), 5.481 (dd, J=14.9, 8.7Hz, 1H), 5.425 (d, J=10.2 Hz, 1H), 5.24-5.20 (m, 2H), 4.258 (br s, 1H), 4.076 (d, J=0.9 Hz, 1H), 4.005 (d, J=4.7 Hz, 1H), 3.618 (dd, J=9.3, 2.7Hz, 1H), 3.450 (br d, J=14.8 Hz, 1H), 3.394 (s, 3H), 3.332 (s, 3H), 3.202 (s, 3H), 2.709 (dd, J=17.4, 3.4Hz, 1H), 2.411(dd, J=17.4, 8.4Hz, 1H), 1.756 (s, 3H), 1.657(s, 3H), 1.061 (d, J=6.6 Hz, 3H), 1.009 (d, J=6.6 Hz, 3H), 0.944 (d, J=6.6 Hz 3H), 0.923 (d, J=6.6 Hz, 3H),0.865 (d, J=6.7 Hz, 3H), 0.648 (q, J=11.9 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100.6 MHz, 3:1 mixture of trans:cis amide rotamers; data for the trans-rotamer): _ 211.9, 207.5, 196.1, 169.6, 166.2, 138.4, 138.1, 134.9, 132.3, 130.6, 127.3, 125.5, 125.2, 98.7, 84.4, 82.8, 76.2, 74.2, 73.9, 68.9; MS (ESI^+/NH_4OAc) m/z 936 ($M+Na^+$), 931 ($M+NH_4^+$); MS (ESI^-/NH_4COOH) m/z 958 ($M+HCOO^-$); UV (MeOH) max 267, 277, 289 nm.

III. BIOLOGICAL EXAMPLES

Compounds of the invention were analyzed for antifungal and immunosuppressive activity using the following assays.

Assay for Antifungal Activity

Yeast organism (*Saccharomyces cerevisiae*) in logarithmic growth were plated on complete agar medium (YPD). Compounds dissolved in an appropriate aqueous or organic solvent were placed in wells punched in the agar. Plates were incubated for 48 hours and zones of inhibition were measured. All of the compounds of the invention which were tested in this assay exhibited antifungal activity.

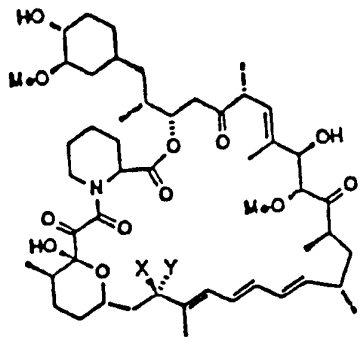
Mitogenesis Assay for Immunosuppressive Activity

Spleen cells from BDF1 female mice were established in RPMI with 10% fetal calf serum at 5 x 10^6/mL. One hundred mL aliquots of this suspension (5 x 10^5 cells) were dispensed into 96-well round-bottomed microtiter plates (Linbro, Flow Laboratories). Concanavalin A (5 μ g/ml)was added as the mitogenic stimulus, and the final volume in the microtiter wells was adjusted to 200 μ L with RPMI. Cell cultures were incubated for 72 hours at 37° C in a 5% CO_2 atmosphere and pulsed with 0.5 μ Ci ^{3}H-thymidine (specific activity 2.00 Ci/mole) for the last 18 hours of the 72 hour culture. The cells were harvested on an automated multiple sample harvester and cell-associated radioactivity counted in a Beckman liquid scintillation counter. The results are expressed as the mean values derived from quadruplicate measurements. Cell viability was determined by trypan blue exclusion after 72 hours of incubation. Compounds to be tested were added to the microtiter plates at the appropriate dilutions prior to the addition of cells. All of the compounds of the invention which were tested in this assay exhibited immunosuppressive activity.

Results of these two assays, i.e., antifungal activity assay and the mitogenesis assay for immunosuppressive activity, for compounds of this invention are provided in Table 1.

TABLE 1

Biological Activity



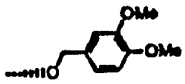
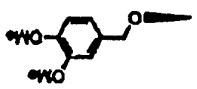
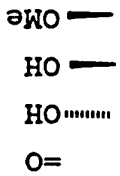
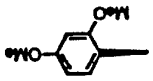
Compound of Example #	X or Y	Antifungal IC_{12} ng/mL	Mitogenesis (IC_{50} nM)
1	····· OEt	<500	<500
2	— OEt	<500	<500
3		<500	<500
4	····· $O(CH_2)_2OH$	<500	<500
5	— $O(CH_2)_2OH$	<500	<500
6	— OAc	<500	<500
7		<500	<500
8	— H_2	<500	<500
9	····· SMe	<500	<500
10	— SMe	<500	<500
11	····· SPh	<500	<500
13	····· allyl	<500	<500

Table I - continued

Compound of Example #	X or Y	Antifungal IC_{12} ng/mL	Mitogenesis (IC_{50} nM)
14		<500	<500
15		<500	<500
16		<500	<1000
17		<500	<500
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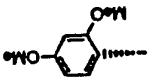
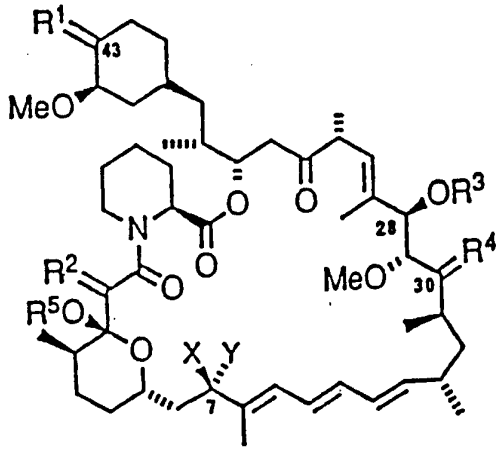
20		<500	<500
		<500	

While the above descriptions and Examples fully describe the invention and the preferred embodiments thereof, it is understood that the invention is not limited to the particular disclosed embodiments coming within the scope of the following claims.

CLAIMS

What is claimed is:

1. A compound of the formula



wherein:

X is selected from the group consisting of H, $-OR^{10}$, $-S(O)_nR^{10}$, $-NR^{10}R^{11}$, alkyl and aryl;

Y is selected from the group consisting of H, $-OR^{10}$, $-S(O)_nR^{10}$, $-NR^{10}R^{11}$, alkyl and aryl;

or X and Y taken together are =O;

n is selected from the group consisting of 0, 1 and 2;

R^1 is selected from the group consisting of =O, $(-OR^6,H)$ and (H,H);

R^2 is selected from the group consisting of =O, (H,H), and (H,OH);

R^3 and R^6 are independently selected from the group consisting of -H, C_1-C_4 alkyl, $-C(=O)R^7$, $-C(=O)OR^7$, $-C(=O)NHR^7$, and $-C(=S)OR^7$;

R^4 is selected from the group consisting of =O and (H,OR^6);

or R^3 and R^4 can be taken together to form a bridge of the formula $A-C(R^8)(R^9)-O-B$, where A is a bond to the oxygen bonded to the carbon at the 28-position and B is a bond to the carbon at the 30-position;

R^5 is selected from the group consisting of -H

and C_1-C_4 alkyl;

R^7 is selected from the group consisting of C_1-C_{10} alkyl, C_3-C_6 cycloalkyl, aryl groups, and heterocyclic groups;

R^8 and R^9 are independently selected from the group consisting of H, C_1 to C_6 alkyl, or R^8 and R^9 taken together are =O;

R^{10} and R^{11} are independently selected from the group consisting of H, alkyl and aryl;

provided that,

(a) at least one of X and Y is H; and

(b) when Y is $-R^{10}$, then R^{10} is other than CH_3; and all pharmaceutically acceptable salts, hydrates or solvates thereof.

2. A compound of Claim 1 where R^1 is (H,OH).

3. A compound of Claim 1 where R^2 is =O.

4. A compound of Claim 1 where R^3 is H.

5. A compound of Claim 1 where R^4 is =O.

6. A compound of Claim 1 where R^5 is H.

7. A compound of Claim 1 where R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H.

8. A compound of Claim 1 where one of X and Y is selected from the group consisting of OR^{10} and SR^{10} or where X and Y are taken together as =O.

9. A compound of Claim 8 where X and Y are taken together as =O.

10. A compound of Claim 8 where one of X and Y is selected from the group consisting of $-OR^{10}$ and $-SR^{10}$

where R^{10} is selected from the group consisting of optionally substituted C_1-C_3 alkyl, optionally substituted benzyl, and optionally substituted phenyl.

11. A compound of Claim 1 where, when one of X and Y is an alkyl group or when one of R^{10} and R^{11} is an alkyl group, said alkyl is selected from the group consisting of branched, straight-chain, cyclic, polycyclic, saturated and unsaturated alkyl groups which may be substituted with 0, 1 or more substitutents selected from the group consisting of aryl, keto, hydroxyl, alkoxyl, acyloxy, amino, N-acylamino, halogen, cyano and carboxyl substituents, and in which at least one carbon atom may be replaced with a heteroatom selected from the group consisting of O, S and N.

12. A compound of Claim 8 where, when one of X and Y is an aryl group or when one of R^{10} or R^{11} is an aryl group, said aryl group is selected from the group consisting of cyclic, heterocyclic, polycyclic and heteropolycyclic C_2 to C_{14} aryl groups, which may be substituted by one to five members selected from the group consisting of alkyl, hydroxyl, alkyoxyl, acyloxy, amino, N- acylamino, halogen, cyano, carboxyl and nitro groups.

13. A compound of Claim 7 where one of X and Y is selected from the group consisting of OR^{10} and SR^{10} or where X and Y are taken together as =O.

14. A compound of Claim 13 where X and Y are taken together as =O.

15. A compound of Claim 7 where one of X and Y is selected from the group consisting of $-OR^{10}$ and $-SR^{10}$ where R^{10} is selected from the group consisting of optionally substituted C_1-C_3 alkyl, optionally

substituted benzyl, and optionally substituted phenyl.

16. A compound of Claim 7 where, when one of X and Y is an alkyl group or when one of R^{10} and R^{11} is an alkyl group, said alkyl is selected from the group consisting of branched, straight-chain, cyclic, polycyclic, saturated and unsaturated alkyl groups which may be substituted with 0, 1 or more substitutents selected from the group consisting of aryl, keto, hydroxyl, alkoxyl, acyloxy, amino, N-acylamino, halogen, cyano and carboxyl substituents, and in which at least one carbon atom may be replaced with a heteroatom selected from the group consisting of O, S and N.

17. A compound of Claim 7 where, when one of X and Y is an aryl group or when one of R^{10} or R^{11} is an aryl group, said aryl group is selected from the group consisting of cyclic, heterocyclic, polycyclic and heteropolycyclic C_2 to C_{14} aryl groups, which may be substituted by one to five members selected from the group consisting of alkyl, hydroxyl, alkyoxyl, acyloxy, amino, N- acylamino, halogen, cyano, carboxyl and nitro groups.

18. The compound of Claim 1 wherein:

(1) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OCH_3$;

(2) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OC_2H_5$;

(3) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is $-OC_2H_5$;

(4) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OCH_2CH_2OH$;

(5) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is $-OCH_2CH_2OH$;

(6) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is 3,4-dimethoxybenzyloxy;

(7) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is 3,4-dimethoxybenzyloxy;

(8) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is $-SCH_3$;

(9) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is $-SCH_3$;

(10) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is -SPh where Ph is phenyl;

(11) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is -SPh where Ph is phenyl;

(12) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is 2-furanyl;

(13) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is 2-furanyl;

(14) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is O-acetyl;

(15) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, and each of X and Y is H;

(16) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, and X and Y are taken together as =O;

(17) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is allyl;

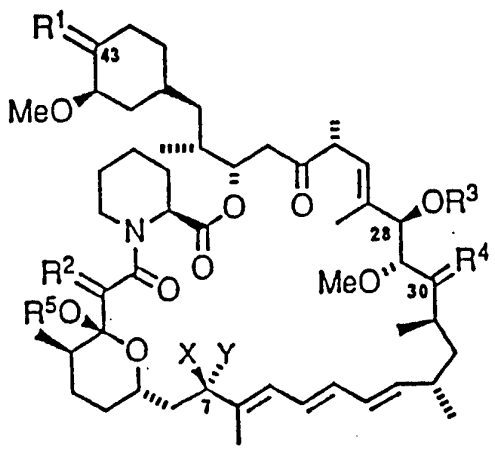
(18) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is OH;

(19) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is 2,4-dimethoxyphenyl;

(20) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is 2,4-dimethoxyphenyl; or

(21) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is -OH.

19. A pharmaceutical composition comprising a pharmaceutically acceptable carrier or diluent, and an effective therapeutic or prophylatic amount of one or more compounds of the formula



wherein:

X is selected from the group consisting of H, $-OR^{10}$, $-S(O)_nR^{10}$, $-NR^{10}R^{11}$, alkyl and aryl;

Y is selected from the group consisting of H, $-OR^{10}$, $-S(O)_nR^{10}$, $-NR^{10}R^{11}$, alkyl and aryl;

or X and Y taken together are =O;

n is selected from the group consisting of 0, 1 and 2;

R^1 is selected from the group consisting of =O, $(-OR^6,H)$ and (H,H);

R^2 is selected from the group consisting of =O, (H,H), and (H,OH);

R^3 and R^6 are independently selected from the group consisting of -H, C_1 - C_4 alkyl, $-C(=O)R^7$, $-C(=O)OR^7$, $-C(=O)NHR^7$, and $-C(=S)OR^7$;

R^4 is selected from the group consisting of =O and (H,OR^6);

or R^3 and R^4 can be taken together to form a bridge of the formula $A-C(R^8)(R^9)-O-B$, where A is a bond to the oxygen bonded to the carbon at the 28-position and B is a bond to the carbon at the 30-position;

R^5 is selected from the group consisting of -H and C_1-C_4 alkyl;

R^7 is selected from the group consisting of C_1-C_{10} alkyl, C_3-C_6 cycloalkyl, aryl groups, and heterocyclic groups;

R^8 and R^9 are independently selected from the group consisting of H, C_1 to C_6 alkyl, or R^8 and R^9 taken together are =O;

R^{10} and R^{11} are independently selected from the group consisting of H, alkyl and aryl;

provided that,

(a) at least one of X and Y is H; and

(b) when Y is $-OR^{10}$, then R^{10} is other than CH_3;

and all pharmaceutically acceptable salts, hydrates or solvates thereof.

20. The pharmaceutical composition of Claim 19 wherein:

(1) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OCH_3$;

(2) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OC_2H_5$;

(3) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is $-OC_2H_5$;

(4) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is $-OCH_2CH_2OH$;

(5) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is $-OCH_2CH_2OH$;

(6) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, Y is H and X is 3,4-dimethoxybenzyloxy;

(7) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is 3,4-dimethoxybenzyloxy;

(8) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is $-SCH_3$;

(9) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is $-SCH_3$;

(10) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is -SPh where Ph is phenyl;

(11) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O,

R^5 is H, X is H and Y is -SPh where Ph is phenyl;

(12) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is 2-furanyl;

(13) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is 2-furanyl;

(14) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is O-acetyl;

(15) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, and each of X and Y is H;

(16) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, and X and Y are taken together as =O;

(17) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is allyl;

(18) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is OH;

(19) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, Y is H and X is 2,4-dimethoxyphenyl;

(20) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is=O, R^5 is H, X is H and Y is 2,4-dimethoxyphenyl; or

(21) R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, R^5 is H, X is H and Y is -OH.

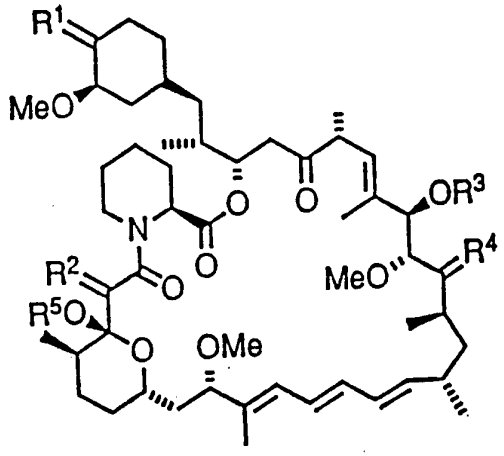
21. A method of inhibiting the growth of pathogenic fungi in a human or other animal in need thereof which comprises administering to such human or other animal an effective, non-toxic amount of a composition of Claim 19.

22. A method of inducing immunosuppression in a human or other animal in need thereof which comprises administering to such human or other animal an effective, non-toxic amount of a composition of Claim 19.

23. A method of treating carcinogenic tumors in a human or other animal comprising administering to such human or animal an effective, non-toxic amount of a

composition of Claim 19.

24. A method of preparing compounds of Claim 1 comprising contacting a compound of the formula:



where

R^1 is selected from the group consisting of =0, ($-OR^6$,H) and (H,H);

R^2 is selected from the group consisting of =0, (H,H), and (H,OH);

R^3 and R^6 are independently selected from the group consisting of -H, C_1 - C_4 alkyl, $-C(=0)R^7$, $-C(=0)OR^7$, $-C(=0)NHR^7$, and $-C(=S)OR^7$;

R^4 is selected from the group consisting of =O and (H,OR^6);

or R^3 and R^4 can be taken together to form a bridge of the formula $A-C(R^8)(R^9)-O-B$, where A is a bond to the oxygen bonded to the carbon at the 28-position and B is a bond to the carbon at the 30-position;

R^5 is selected from the group consisting of -H and C^1-C^4 alkyl;

R^7 is selected from the group consisting of C_1-C_{10} alkyl, C_3-C_6 cycloalkyl, aryl groups, and heterocyclic groups;

R^8 and R^9 are independently selected from the group consisting of H, C_1 to C_6 alkyl, or R^8 and R^9 taken together are =O;

with an acid selected from the group consisting of protic acids and Lewis acids, and with an appropriate

nucleophile.

25. The method of Claim 24 where said acid is selected from the group consisting of trifluoroacetic acid and titanium tetrachloride.

26. The method of Claim 25 where said acid is trifluoroacetic acid.

27. The method of Claim 25 where R^1 is (H,OH), R^2 is =O, R^3 is H, R^4 is =O, and R^5 is H.

28. The method of Claim 27 where said acid is trifluoroacetic acid.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US93/06678

A. CLASSIFICATION OF SUBJECT MATTER

IPC(5) :498/12; A61K 31/395
US CL :540/456 514/291
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. :

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

NONE

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

NONE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US, A, 5,118,677 (Caufield) 02 June 1992. See entire document.	1-8, 10-13, 15-28
X Y	US, A, 5,102,876 (Caufield) 07 April 1992. See entire document.	1-2,7,13, 15-28 1-8, 10-13, 15-28
X Y	US, A, 5,023,263 (Von Burg) 11 June 1991. See entire document.	1, 3-6, 8, 10-12, 19, 21-22, 24 25-28

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be part of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

Date of the actual completion of the international search
02 SEPTEMBER 1993

Date of mailing of the international search report
OCT 14 1993

Name and mailing address of the ISA/US
Commissioner of Patents and Trademarks
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Washington, D.C. 20231
Facsimile No. NOT APPLICABLE

Authorized officer
MARIANNE CINTINS
Telephone No. (703) 308-1235

Form PCT/ISA/210 (second sheet)(July 1992)*

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US93/06678

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

Please See Extra Sheet.

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☒ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

BOX II. OBSERVATIONS WHERE UNITY OF INVENTION WAS LACKING
This ISA found multiple inventions as follows:

Group I, claims 1-20, 21 & 24-28, drawn to Rapamycin derivatives, their method of use in inhibition of the growth of fungi, and a method for their synthesis, respectively, classified in Class 540, subclass 456 and Class 514, subclass 183.

Group II, claim 22, drawn to a method of inducing immunosuppression utilizing a Rapamycin derivative, classified in Class 514, subclass 183.

Group III, claim 23, drawn to a method of treating carcinogenic tumors utilizing a Rapamycin derivative, classified in Class 514, subclass 183.

PCT Rule 13.1 states that the international application shall relate to one invention only or to a group of inventions so linked as to form a single inventive concept ("requirement of unity of invention").

PCT Rule 13.2 states that unity of invention referred to in Rule 13.1 shall be fulfilled only when there is a technical relationship among those inventions involving one or more of the same or corresponding special technical features.

Annex B, Part 1(c), further defines independent and dependent claims. Unity of invention only is concerned in relation to independent claims. Dependent claims are defined as a claim which contains all the features of another claim and is the same category as the other claim. The category of a claim refers to the classification of claims according to subject matter, e.g. product, process, use, apparatus, means, etc.

Annex B, Part 1(e) indicates the permissible combinations of different categories of claims. Part 1(e(i)) states that inclusion of an independent claim for a given product, an independent claim for a process specially adapted for the manufacture of the said product, and an independent claim for a use of the said product is permissible.

Herein, two additional independent claims, i.e. 22 & 23, are included in the application, and appear contrary to PCT Rule 13.