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(51) Int.Cl.⁶ A61K 31/70

(30) 1996/01/18 (08/588,564) US

(54) **COMPOSITIONS ET PROCEDES S'APPLIQUANT A LA
PREVENTION ET AU TRAITEMENT DE
L'ATHEROSCLEROSE ET DES LESIONS CONSECUTIVES A
DES PERFUSIONS AU MOYEN DE SELS DE MAGNESIUM**

(54) **COMPOSITIONS AND METHODS FOR THE PREVENTION
AND TREATMENT OF ATHEROSCLEROSIS AND
REPERFUSION INJURY WITH MAGNESIUM SALTS**

(57) La présente invention se rapporte à des compositions et à des procédés s'appliquant à la prévention ou au traitement de l'athérosclérose et des lésions consécutives à des perfusions. Ce procédé consiste à administrer à un patient souffrant d'athérosclérose une solution aqueuse contenant un gluconate de magnésium dans une quantité suffisante pour inhiber la formation de radicaux libres provoquée par l'oxygène et la peroxydation lipidique. Selon une autre réalisation, l'invention se rapporte à la prévention de la resténose chez des patients subissant une intervention chirurgicale de pontage ou d'angioplastie. Selon une réalisation différente, la présente invention se rapporte à un procédé de prévention ou de traitement de lésions ischémiques consécutives à des perfusions, procédé consistant à administrer à un patient sujet aux lésions consécutives à des perfusions une quantité thérapeutiquement efficace de gluconate de magnésium afin d'inhiber la formation de radicaux libres provoquée par l'oxygène. La solution aqueuse de l'invention comprend du gluconate de magnésium dans une quantité suffisante permettant d'inhiber la formation de radicaux libres provoquée par l'oxygène dans les tissus cardiaques et endothéliaux.

(57) The present invention provides compositions and methods for the prevention or treatment of atherosclerosis and reperfusion injury. The method comprises administering to a patient having atherosclerosis an aqueous solution containing magnesium gluconate in an amount sufficient to inhibit oxygen free radical production and lipid peroxidation. In another embodiment the invention provides for the prevention of restenosis in patients undergoing bypass surgery or angioplasty. In a different embodiment, the present invention provides a method for prevention or treatment of ischemic reperfusion injury comprising the step of administering to a patient at risk of reperfusion injury a therapeutically effective amount of magnesium gluconate to inhibit oxygen free radical production. The aqueous solution of the invention comprises magnesium gluconate in an amount sufficient to inhibit oxygen free radical production in cardiac and endothelial tissues.



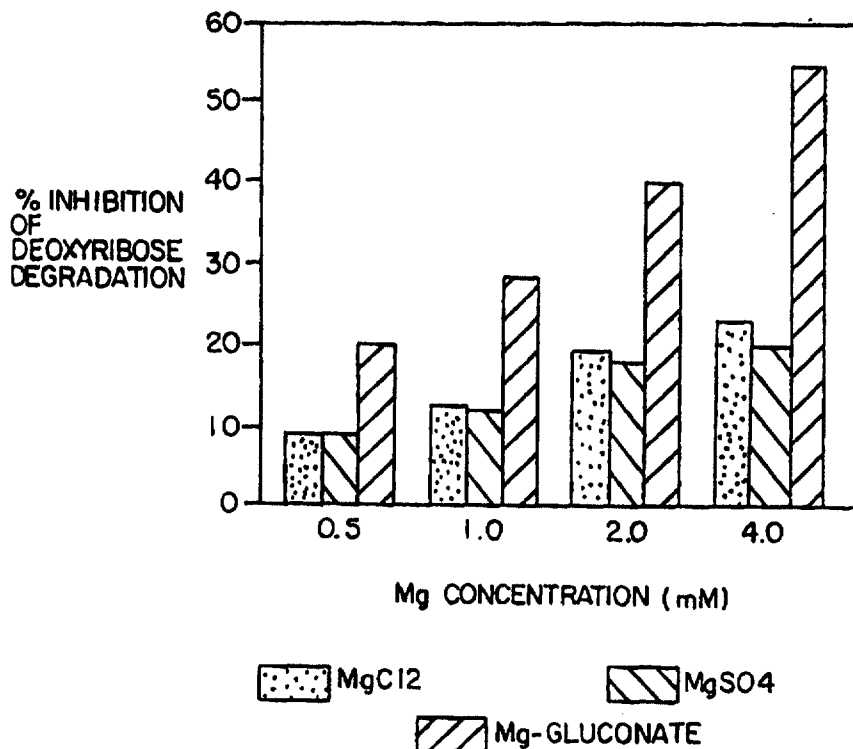
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International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁶ : A61K 31/70		A3	(11) International Publication Number: WO 97/25959
			(43) International Publication Date: 24 July 1997 (24.07.97)
(21) International Application Number: PCT/US97/00821 (22) International Filing Date: 17 January 1997 (17.01.97) (30) Priority Data: 08/588,564 18 January 1996 (18.01.96) US (71) Applicant (for all designated States except US): FLEMING & COMPANY, PHARMACEUTICALS [US/US]; 1600 Fenpark Drive, Fenton, MO 63026 (US). (72) Inventors; and (75) Inventors/Applicants (for US only): FLEMING, Thomas, E. [US/US]; 24 Bopp Lane, St. Louis, MO 63131 (US). MANSMANN, Herbert, C., Jr. [US/US]; 1004 Garrett Mill Road, Newton Square, PA 19073 (US). (74) Agent: STIEFEL, Maurice, B.; Bryan Cave L.L.P., 40th floor, 245 Park Avenue, New York, NY 10167 (US).		(81) Designated States: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, US, UZ, VN, ARIPO patent (KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i> (88) Date of publication of the international search report: 18 September 1997 (18.09.97)	
(54) Title: COMPOSITIONS AND METHODS FOR THE PREVENTION AND TREATMENT OF ATHEROSCLEROSIS AND REPERFUSION INJURY WITH MAGNESIUM SALTS			

(57) Abstract

The present invention provides compositions and methods for the prevention or treatment of atherosclerosis and reperfusion injury. The method comprises administering to a patient having atherosclerosis an aqueous solution containing magnesium gluconate in an amount sufficient to inhibit oxygen free radical production and lipid peroxidation. In another embodiment the invention provides for the prevention of restenosis in patients undergoing bypass surgery or angioplasty. In a different embodiment, the present invention provides a method for prevention or treatment of ischemic reperfusion injury comprising the step of administering to a patient at risk of reperfusion injury a therapeutically effective amount of magnesium gluconate to inhibit oxygen free radical production. The aqueous solution of the invention comprises magnesium gluconate in an amount sufficient to inhibit oxygen free radical production in cardiac and endothelial tissues.



**COMPOSITIONS AND METHODS FOR THE PREVENTION
AND TREATMENT OF ATHEROSCLEROSIS AND
REPERFUSION INJURY WITH MAGNESIUM SALTS**

1. INTRODUCTION

The present invention relates to compositions and methods for the prevention and/or treatment of atherosclerosis. The method comprises administering to a patient having atherosclerosis an aqueous solution containing magnesium gluconate in an amount sufficient to inhibit lipid peroxidation and oxidative modification of lipoproteins. The present invention also provides methods for the prevention of restenosis in a patient undergoing a surgical procedure to treat atherosclerosis. In another embodiment, the present invention provides a method for prevention and/or treatment of ischemic reperfusion injury. The method comprises administering to a patient at risk of reperfusion injury an aqueous solution containing magnesium gluconate in an effective amount to limit anticipated ischemic injury and/or after the onset of an ischemic event.

2. BACKGROUND OF THE INVENTION

The present invention relates generally to the prevention and/or treatment of cardiovascular disease. More specifically, the present invention relates to compositions and methods of prevention and/or treatment of atherosclerosis and reperfusion injury.

Cardiovascular disease is the leading cause of death in the United States. Atherosclerotic heart disease is among the most serious and costly health problems in industrialized nations. Atherosclerosis is not a single disease entity. Atherosclerotic

lesions take different forms, depending upon their anatomic site, the age, genetic and physiological status of the affected individual, and upon the risk factors to which each individual may have been exposed. The lesions of atherosclerosis occur principally within the intima and include the fatty streak, the fibrous plaque and the complicated plaques. Ross, R. and Blomset, J.A., 1976, *N. Engl. J. Med.* 295: 369.

Lipids play a central role in the pathogenesis of atherosclerosis, especially in individuals with markedly increased elevations of plasma low-density lipoproteins (LDL). The accumulation of lipid within proliferated smooth muscle cells, within macrophages in the lesions, and within the extracellular connective tissue matrix, are common findings in atherosclerotic lesions. Geer, J.C. et al., 1961, *Am. J. Pathol.* 38: 263. Injury to the endothelium that results in alterations in permeability permits plasma constituents such as lipoproteins to have more ready access to the artery wall. Oxidation of LDL is one of the deleterious effects of oxidative stress. Free radicals released from injured endothelial cells, smooth muscle cells and macrophages induce peroxidation of LDL lipids, resulting in formation of hydroperoxides of arachidonic acid and other unsaturated lipid moieties of LDL lipids. Steinbrecher, U.P. et al., 1984, *Proc. Natl. Acad. Sci. USA* 81: 3883; Morel, D.W. et al., 1984, *Arteriosclerosis* 4: 375.

A number of pharmaceutical interventions have been proposed for prevention and/or treatment of atherosclerosis. In many cases of atherosclerosis, invasive surgical procedures of bypass or angioplasty are required to reestablish an occluded lumen to proper size. At present, more than 220,000 patients

per year undergo coronary artery bypass surgery in the United States. Virtually all grafts early after implantation show loss of endothelium and deposition of fibrin and platelet thrombi to form a plaque. In some patients the circumferential intimal plaque continues to proliferate in the ensuing months and in some cases this leads to significant narrowing of the graft lumen or restenosis, by an atherosclerotic process. Forty percent of patients have restenosed arteries within 5 years of bypass grafts. Bulkley, B.H. et al., 1977, *Circulation* 55: 163.

Coronary angioplasty is an effective method of improving blood flow to the heart. However, changes in the structure of the vessel wall after angioplasty include intimal proliferation, deposition of thrombi and thrombosis. In approximately 30 percent of patients restenosis occurs within three to six months.

Cardiac tissue is susceptible to ischemia wherein the tissue suffers from a decrease or cessation of perfusion by blood due to blockage of the blood vessels by atherosclerosis, heart attack, stroke, during tissue neurosis, or organ transplant. A cardiac ischemic event can also be caused from therapeutic interventions such as bypass surgery or angioplasty.

Clinical and experimental studies have indicated that timely reperfusion of ischemic myocardium can reduce the amount of necrosis after acute coronary artery occlusion. Kloner, R.A., et al., 1983 *Circulation* 68 (Suppl I) I-8. However, reperfusion, while terminating ischemia, can also cause further damage to jeopardized cells. Frame, L.H. et al., 1983, *J. Clin. Invest.* 72: 535. This phenomenon has been termed "reperfusion injury". Several mechanisms have been proposed to explain its

occurrence after bypass surgery and angioplasty, including cell swelling due to sodium and water influx, calcium influx and formation of calcium phosphate granules within mitochondria, hemorrhage, and generation of oxygen free radicals, particularly superoxide anions ($^{\circ}\text{O}_2^-$) at the time of reoxygenation.

There is increasing evidence that hypoxia or ischemia followed by reoxygenation or reperfusion increases production of oxygen radical species. The production of oxygen radicals and lipid peroxides during reperfusion are believed to result in tissue necrosis or damage. Administration of exogenous quenchers of free radicals, such as superoxide dismutase, catalase or both, improve cardiac function and limit infarct size when administered after global ischemia. McCord, J.M., 1985, 312: 159-163; Jolly, S.R., et al., 1984, *Cir. Res.* 54: 277-285. Other examples of clinically significant ischemic reperfusion injury include intestinal ischemia, necrotic brain-tissue, damage to the organ during organ transplantation or ischemic injuries in patients receiving artificial blood substitutes.

3. SUMMARY OF THE INVENTION

The present invention relates to compositions and methods for the prevention and/or treatment of atherosclerosis and/or reperfusion injury.

More specifically, the present invention provides a method of preventing the initiation and/or progression of atherosclerotic lesions. The present invention also provides a method for preventing the restenosis of coronary vessels following angioplasty or bypass surgery. To this end, the present invention provides a method comprising administering to a patient having atherosclerosis an aqueous

solution containing magnesium gluconate in sufficient amounts to inhibit oxygen free radical production and lipid peroxidation. In different embodiments of the invention, the composition is administered enterally or parenterally.

In another embodiment of the invention, a method is provided for preventing restenosis in a patient undergoing a surgical procedure for treatment of atherosclerosis, comprising administering to the patient before and after the surgical procedure a therapeutically effective amount of magnesium gluconate.

The present invention also provides a method for treating and/or reducing ischemic reperfusion injury comprising administering to a patient undergoing a surgical procedure and in danger of reperfusion injury, a therapeutically effective amount of magnesium gluconate to inhibit oxygen free radical production. In different embodiments of the method the surgical procedures are bypass surgery and angioplasty.

The compositions of the invention comprise aqueous solutions suitable for enteral or parenteral administration containing magnesium gluconate in an amount sufficient to inhibit oxygen free radical production and lipid peroxidation in cardiac and endothelial tissues.

The examples presented in Sections 5 and 6 below detail the use according to the methods of the invention of magnesium gluconate in clinical and experimental models of atherosclerosis and reperfusion injury.

4. DESCRIPTION OF THE FIGURES

Figure 1. Effects of magnesium salts formation of malondialdehyde in endothelial cell membranes.

Figure 2. Effects of magnesium salts on site-specific OH.-mediated deoxyribose oxidation.

5. DETAILED DESCRIPTION OF THE INVENTION

The present invention relates to compositions and methods for the prevention and/or treatment of atherosclerosis and reperfusion injury.

More specifically, the present invention provides a method of preventing the initiation and/or progression of atherosclerotic lesions.

The lesions of atherosclerosis occur principally within intima and include the fatty streak, the fibrous plaque and the complicated plaques. Fatty streaks are accumulations of lipid laden myointimal cells at branch points of vessels and can be identified at any age or sex. The fatty streak is ubiquitous in young people and does not inevitably lead to advanced lesions of atherosclerosis, but is of interest as a possible precursor of advanced atherosclerotic lesions.

Fibrous plaques develop around the age of 25 in those populations in which there is a high incidence of atherosclerosis. The fibrous plaques

are white and viewed as a more advanced atherosclerotic lesion with potential for lumina narrowing and degeneration. The fibrous plaques are derived from fatty streaks that continue the process of cell proliferation, lipid accumulation and connective tissue formation. The third and most advanced atheroma is the complicated plaque, composed of fibrous tissue, fibrin, calcium, intracellular and extracellular lipid, and having a necrotic lipid-rich core. The lesions may disintegrate and ulcerate and become involved with thrombotic episodes that may lead to occlusive disease.

There are three pathogenic theories of atheroma formation: the lipogenic theory, the response-to-injury theory, and the monoclonal theory. The monoclonal theory suggests that each atherosclerotic lesion is derived from a single smooth-muscle cell and this cell serves as the progenitor for all of the proliferating cells within the lesion.

The lipogenic theory implicates lipids in the initiation and progression of atherosclerosis in individuals with markedly increased elevations of plasma low-density lipoproteins (LDL). The accumulation of lipid within proliferated smooth muscle cells, within macrophages in the lesions, and within the extracellular connective tissue matrix,

are common findings in lesions of atherosclerosis.

Geer, J.C., et al., 1961, *Am. J. Pathol.* 38:263.

The response-to-injury theory suggests some form of "injury" to the endothelium which results in structural and/or functional alterations in the endothelial cells. Ross, H. and Harker, L., 1976, *Science* 193: 1094. Injury to the endothelium that results in alterations in permeability permits plasma constituents such as lipoproteins to have more ready access to the artery wall. Oxidation of LDL is one of the deleterious effects of oxidative stress. Oxygen free radicals released from injured endothelial cells, smooth muscle cells and macrophages induce peroxidation of LDL lipids, resulting in formation of hydroperoxides of arachidonic acid and other unsaturated lipid moieties of LDL lipids.

The present invention provides a method of preventing the initiation and/or progression of atherosclerosis by inhibiting oxygen free radical production by cardiac tissue, endothelial cells and cellular elements in the atherosclerotic lesions such as macrophages and neutrophil leukocytes.

A free radical is a molecule with an odd, unpaired electron which makes the molecule unstable and highly reactive. Tribble, D.L., et al., 1987, *Hepatology* 7: 377-386. Small amounts of these

compounds are produced endogenously by the mitochondrial electron transport system and the endoplasmic reticulum in microsomes and peroxisomes. Oxygen free radicals, the superoxide anion (O_2^-), the hydroxyl radical ($^{\circ}OH$), and their intermediary, hydrogen peroxide (H_2O_2), are believed to be generated during ischemia and at the time of reperfusion. These free radicals interact with other cellular constituents such as deoxyribonucleic acid (DNA) and lipids with subsequent formation of multiple degradation products. Lipid peroxidation forms lipid peroxides and aldehydes that interact with protein sulfhydryl groups perpetuating cellular damage. Del Maestro, R.F., 1980, *Acta. Physiol. Scand.* 492 (Suppl.): 153-168.

Normally, protective mechanisms are present in the cell to prevent damage by free radicals. For example, the primary mechanism of clearance of O_2^- from biologic systems is superoxide dismutase, which catalyzes the dismutation of O_2^- to H_2O_2 and O_2 . The cytoplasmic enzymes glutathione peroxidase and catalase provide the final detoxification steps with the reduction of H_2O_2 to O_2 . Glutathione peroxidase seems to be a more active enzyme than catalase in protecting myocardial cells from H_2O_2 -mediated damage. Fridovich, I., 1983, *Annu. Rev. Pharmacol. Toxicol.*

23: 239-257; Fantone, J.C., et al., 1982, *Am. J. Pathol.* 107: 395-418.

The present invention provides a method comprising administering to a patient having atherosclerosis a composition of the invention containing magnesium gluconate in sufficient amounts to inhibit oxygen free radical production and lipid peroxidation associated with it.

The present invention also provides a method for preventing and treating restenosis of coronary vessel following angioplasty or bypass surgery. To that end, the present invention provides methods for preventing or treating restenosis in a patient undergoing a surgical procedure for treatment of atherosclerosis, comprising administering to the patient prior to and/or after the surgical procedure, a therapeutically effective amount of magnesium gluconate.

In different embodiments of the invention, the composition of the invention is administered enterally or parenterally in the prevention or treatment of atherosclerosis.

The compositions of the invention comprise aqueous solutions containing magnesium gluconate in an amount sufficient to inhibit oxygen free radical production and lipid peroxidation in cardiac and endothelial tissues.

The present invention also provides compositions and methods for preventing or treating ischemic reperfusion injury. By definition, reperfusion injury refers to cell death or damage caused by reperfusion, in contrast to cell death or damage caused by the preceding ischemic episode. The mechanism by which reperfusion injury occurs involves the generation of oxygen free radicals when oxygen and blood elements are reintroduced into the ischemic bed at reperfusion. For example, during perfusion of the isolated heart, malondialdehyde, which is a byproduct of the lipid peroxidation, remains unchanged during hypoxia, but increases significantly during reoxygenation. Kloner, R.A., et al., 1983, *Circulation* 68:(Suppl. I): I-8 - I-15.

In a different embodiment, the present invention also provides a method for preventing and treating ischemic reperfusion injury comprising administering to a patient undergoing a surgical procedure and at risk of reperfusion injury a therapeutically effective amount of magnesium gluconate to inhibit oxygen free radical production.

In yet further embodiments, application of the methods of the invention include such procedures as bypass surgery and angioplasty.

Administration of magnesium gluconate results in cellular resistance to oxygen free radical

production and free radical stress. The preferred composition of the invention contains magnesium gluconate, desirably at a concentration of about 0.2mM to about 0.4M.

The present invention can be used as an adjunct with other cardiac therapy, e.g. with a thrombolytic drug administered to a patient who has had a myocardial infarction.

Magnesium is an important metallocoenzyme for many enzyme reactions. In the method of the present invention, magnesium gluconate inhibits the production of oxygen free radicals thereby reducing the cellular damage due to oxidative stress.

Our aqueous magnesium gluconate composition for enteral administration is obtained by mixing a suitable magnesium salt, for example, magnesium carbonate, with citric acid and glucono-delta-lactone. Desirably the following concentration ranges are utilized: magnesium carbonate in the range of 2mg per liter to 44g per liter; citric acid in the range of 2.3mg per liter to 46.2g per liter; and glucono-delta-lactone in the range of 6mg per liter to 25mg per liter. There results an aqueous solution of magnesium gluconate/citrate, i.e. a solution containing magnesium gluconate and magnesium citrate.

Our other aqueous composition for parenteral administration comprises an aqueous solution of magnesium gluconate.

5.1 PHARMACEUTICAL PREPARATIONS AND METHODS OF ADMINISTRATION

The identified compositions that prevent or treat atherosclerosis or reperfusion injury can be administered to a patient at therapeutically effective doses. A therapeutically effective dose refers to that amount of the magnesium compound sufficient to result in the amelioration of symptoms of atherosclerosis or reperfusion injury.

Toxicity and therapeutic efficacy of the magnesium compounds can be determined by standard pharmaceutical procedures in cell cultures or experimental animals, e.g., for determining the LD₅₀ (the dose lethal to 50% of the population) and the ED₅₀ (the dose therapeutically effective in 50% of the population). The dose ratio between toxic and therapeutic effects is the therapeutic index and it can be expressed as the ratio LD₅₀/ED₅₀. Compounds which exhibit large therapeutic indices are preferred. The data obtained from the cell culture assays and animal studies can be used in formulating a range of dosages for use in humans.

5.2 FORMULATIONS

Pharmaceutical compositions for use in accordance with the present invention can be formulated in conventional manner using one or more physiologically acceptable carriers, excipients or buffers.

Thus, the compounds and their physiologically acceptable salts and solvates can be formulated for administration by insufflation (either through the mouth or the nose) or oral, buccal, parenteral or rectal administration.

For oral administration, the pharmaceutical compositions can take the form of, for example, tablets or capsules prepared by conventional means with pharmaceutically acceptable excipients such as binding agents (e.g., pregelatinised maize starch, polyvinylpyrrolidone or hydroxypropyl methylcellulose); fillers (e.g., lactose, microcrystalline cellulose or calcium hydrogen phosphate); lubricants (e.g., magnesium stearate or talc); disintegrants (e.g., potato starch or sodium starch glycolate); or wetting agents (e.g., sodium lauryl sulphate). The tablets can be coated by methods well known in the art. Liquid preparations for oral administration can take the form of, for example, aqueous solutions, syrups or suspensions, or they can be presented as a dry product for

constitution with water or other suitable vehicle before use. Such liquid preparations can be prepared by conventional means with pharmaceutically acceptable additives such as suspending agents (e.g., sorbitol syrup, cellulose derivatives or hydrogenated edible fats); emulsifying agents (e.g., lecithin or acacia); non-aqueous vehicles (e.g., almond oil, oily esters, ethyl alcohol or fractionated vegetable oils); and preservatives (e.g., methyl or propyl-p-hydrobenzoates or sorbic acid). The preparations can also contain buffer salts, flavoring, coloring and sweetening agents as appropriate.

Preparations for oral administration can be formulated to give controlled release of the active compound.

For buccal administration the compositions can take the form of tablets or lozenges formulated in the conventional manner.

The compounds can be formulated for parenteral administration by injection, e.g., by bolus injection or continuous infusion. Formulations for injection can be presented in unit dosage form, e.g., in ampoules or in multi-dose containers, with an added preservative. The compositions can take such forms as suspensions, solutions or emulsions in oily or aqueous vehicles, and can contain formulatory agents such as suspending, stabilizing and/or

dispersing agents. Alternatively, the active ingredient can be in powder form for constitution with a suitable vehicle, e.g., sterile pyrogen-free water, before use.

The compounds can also be formulated in rectal compositions such as suppositories or retention enemas, e.g., containing conventional suppository bases such as cocoa butter or other glycerides.

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

The compositions can, if desired, be presented in a pack or dispenser device which can contain one or more unit dosage forms containing the active ingredient. The pack can for example comprise metal or plastic foil, such as a blister pack. The pack or dispenser device can be accompanied by instructions for administration.

**6. EXAMPLE: ADMINISTRATION OF MAGNESIUM
GLUCONATE IN THE TREATMENT OF
ATHEROSCLEROSIS**

Patients with hypercholesterolemia, complaining of shortness of breath and chest pain, are administered 88meq/100ml of magnesium gluconate 48 hours prior to the surgical procedure, and a divided dose up until time of surgery, i.e., bypass surgery or angioplasty. The total amount of magnesium gluconate given in the bolus injection depends on facts such as body weight, age, etc. Plasma lipid peroxide level is measured as malondialdehyde prior to and after intervention with magnesium. During the surgical procedure, the occluded artery is perfused with the 4.4% magnesium gluconate solution. Intravenous magnesium gluconate solution is administered at the dose of 4.4% depending on the clinical profile of the patient. Follow-up arteriography is carried out to monitor the recovery. Depending on the results obtained, the therapeutic regimen is developed with the ultimate goal of preventing and treating reperfusion injury and atherosclerosis.

**7. EXAMPLE: ADMINISTRATION OF MAGNESIUM
GLUCONATE IN EXPERIMENTAL MODELS OF
REPERFUSION INJURY**

Cultured Endothelial Cell Membranes

Endothelial cells were cultured in vitro in the presence of 0.25, 0.5, 1.0 and 2.0mM of magnesium

salts including magnesium gluconate, magnesium chloride and magnesium sulfate. Membrane malondialdehyde and site specific OH.- mediated deoxyribose oxidate were measured according to methods described by Mak, I.T. & Weglicki, N.B., 1994, *Method Enzymol.* 234: 620-630; and Mak, I.T., et al., 1990, *Biochem Pharm.* 40: 2169-2175. Results demonstrate that magnesium gluconate is more effective than magnesium chloride or magnesium sulfate in inhibiting free radical production (malondialdehyde) (Figure 1) and in inhibiting free radical mediated deoxyribose oxidation in a dose dependent manner. (Figure 2).

The publications cited herein are incorporated by reference in their entireties.

The present invention is not to be construed as limited in scope to the specific embodiments described herein. Indeed, various modifications of the invention in addition to those described herein will become apparent to those skilled in the art from the foregoing description and accompanying figures. Such modifications are intended to fall within the scope of the appended claims.

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WHAT IS CLAIMED IS:

1. A method for treating atherosclerosis comprising administering to a patient having atherosclerosis a therapeutically effective amount of magnesium gluconate to inhibit oxygen free radical production in cardiac and endothelial tissue.
2. The method of claim 1, wherein the magnesium gluconate is administered in an aqueous solution.
3. The method of claim 2, wherein the solution administered contains magnesium gluconate at a dose range of approximately 0.2mM to 0.4M.
4. The method of claim 2, wherein the solution is administered parenterally.
5. The method of claim 2, wherein the solution is administered enterally.
6. A method for treating atherosclerosis or restenosis comprising the step of administering to a patient prior to or after a coronary artery bypass or angioplasty therapeutically effective amount of magnesium gluconate.
7. The method of claim 6, wherein the magnesium gluconate is administered in an aqueous solution.
8. The method of claim 7, wherein the solution administered contains magnesium gluconate at a dose range of approximately 0.2M to 0.4M.
9. The method of claim 7, wherein the solution is administered enterally.

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10. The method of claim 7, wherein the solution is administered parenterally.
11. A method for treating reperfusion injury comprising the step of administering to a patient at risk of a reperfusion injury a therapeutically effective amount of magnesium gluconate.
12. The method of claim 11, wherein the magnesium gluconate is administered in an aqueous solution.
13. The method of claim 12, wherein the solution administered contains magnesium gluconate at a dose range of approximately 0.2M to 0.4M.
14. The method of claim 12, wherein the solution is administered enterally.
15. The method of claim 12, wherein the solution is administered parenterally.
16. A method for treating reperfusion injury comprising the step of administering to a patient prior to an expected ischemic event or after onset of an ischemic event a therapeutically effective amount of magnesium gluconate.
17. The method of claim 16, wherein the magnesium gluconate is administered in an aqueous solution.
18. The method of claim 17, wherein the solution administered contains magnesium gluconate at a dose range of approximately 0.2M to 0.4M.
19. A method for preventing atherosclerosis or restenosis in cardiac and endothelial tissue comprising administering a therapeutically effective amount of magnesium gluconate to inhibit oxygen free radical production.

AMENDED SHEET

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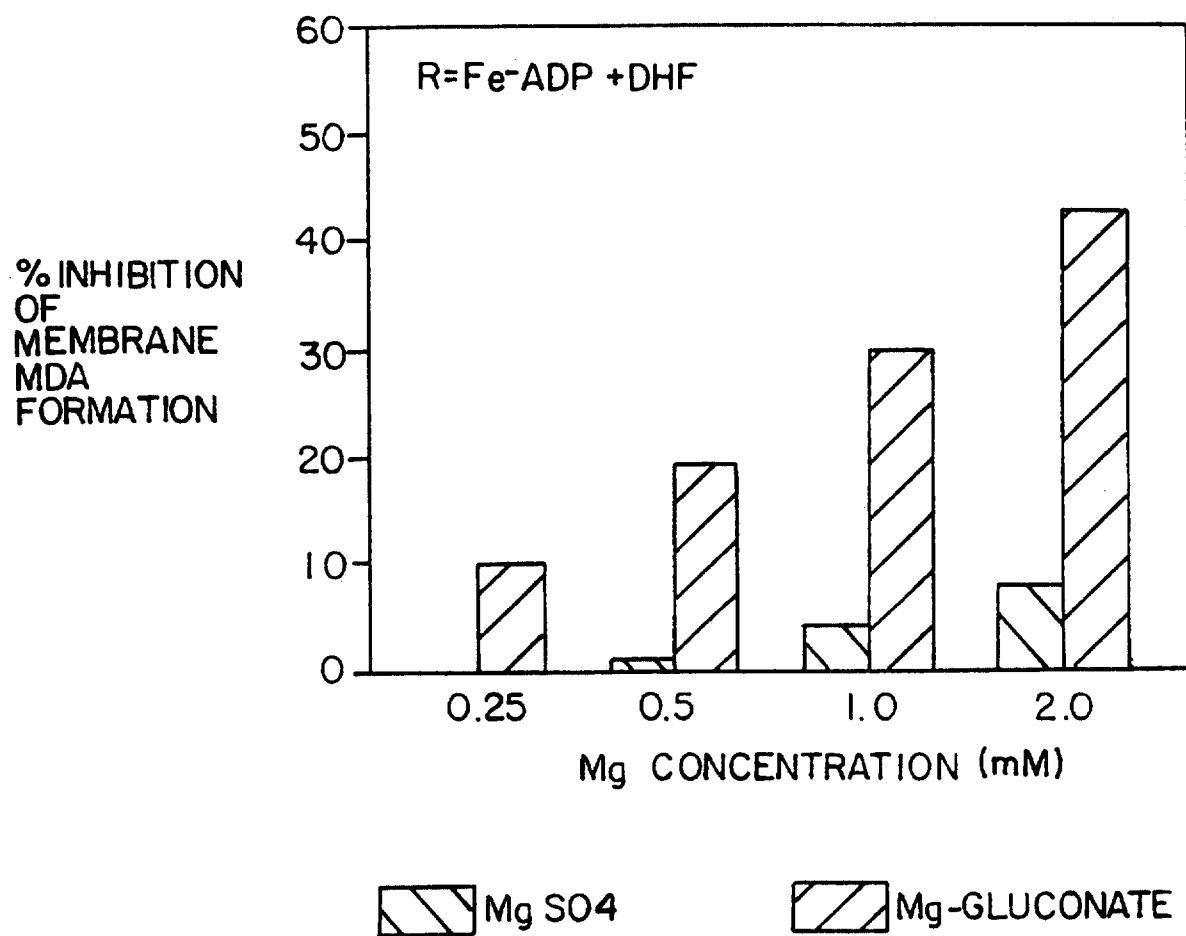
20. The method of claim 19, in which the magnesium gluconate is administered to a patient prior to or after coronary artery bypass or angioplasty.

21. A method for preventing reperfusion injury comprising the step of administering a therapeutically effective amount of magnesium gluconate to a patient at risk of a reperfusion injury.

22. The method of claim 21, wherein the magnesium gluconate is administered to a patient prior to an expected ischemic event or after onset of an ischemic event.

AMENDED SHEET

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FIG. 1

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FIG. 2