Title: COMBINATION PRODUCTS OF A GUANYLATE CYCLASE INHIBITOR AND A LOCAL ANESTHETIC FOR PAIN RELIEF

The present invention is directed to a safe and effective therapy for the treatment of pain associated surgical incisions, wounds, cuts and abrasions. The present invention relates to a pharmaceutical composition comprising a combination of a guanylate cyclase inhibitor and a topical anesthetic or a combination of a redox dye and a topical anesthetic. The compound and treatment may be used to relieve pain resulting from surgical procedures, especially anal surgery.
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BACKGROUND OF THE INVENTION

The present invention relates to the field of compounds and treatments for the relief of pain, especially the treatment of postoperative pain and the pain resulting from incisions and wounds.

In the world today, there is a great deal of pain for which a panacea has not yet been discovered. As a result, many of the most important pharmaceutical compounds and medical treatments are for the relief of pain. Effective pain relieving compounds and treatments have largely met with great success. Among the many ways to measure the success of a pharmaceutical compound include its success in the marketplace and acceptance by physicians. Some of the products that have achieved such phenomenal widespread market success are pain relievers including aspirin, acetaminophen (marketed as Tylenol®), ibuprofen (marketed as Motrin IB® and Advil®), ketoprofen (marketed as Orudis KT®), and naproxen sodium (marketed as Aleve®). Therefore, there is a great need for new and improved compounds and techniques for effective pain relief.

As an example, one of the oldest and well-known pain relievers is aspirin. Aspirin is a trade name for acetylsalicylic acid, a common analgesic. The earliest known uses of the drug can be traced back to the Greek physician Hippocrates in the fifth century B.C. Hippocrates used powder extracted from the bark of willows to treat pain and reduce fever. Scientists never really understood the inner workings of this drug however. It was not until the 1970s, when British pharmacologist John Vane, Ph.D. began work on aspirin that people began to understand how aspirin really works. Vane and his colleagues found that aspirin inhibited the release of a hormone like substance called prostaglandin. This chemical regulates certain body functions, such as blood vessel elasticity and changing the functions of blood platelets. Thus can aspirin affect blood clotting and ease inflammation.
Many modern medical procedures involve surgical procedures in which incisions are made and wounds may result. Surgical procedures may be needed to address matters that are life threatening, cosmetic, therapeutic, diagnostic, corrective, or preventative, or combinations of these. In many cases, postoperative pain results from the incision or wound. The pain from the incision or wound of a surgical procedure is a serious side effect. The recovery time from the operation may be affected in part by the degree of the postoperative pain. For example, the postoperative pain may be so severe that the subject who was operated on may require bed rest for a week or longer. Furthermore, a patient may decide to forgo the operation altogether solely because the postoperative pain is known to be so severe.

A specific example of a surgical procedure where the pain is known to be extremely severe is the case of anal surgery, such as a hemorrhoidectomy. The pain is so severe that just changing the wound dressing is an agonizing experience. The pain associated with anal surgery is so renown that many people who are candidates for anal surgery opt for nonsurgical treatment because of the associated pain and discomfort. Hemorrhoids are dilated veins around the anal opening. They are a consequence of prolonged constipation, or occasionally, diarrhea. They most commonly occur at three main points equidistant around the circumference of the anus. Uncomplicated hemorrhoids are seldom painful. The main symptom is bleeding and in first degree hemorrhoids, which never appear at the anus, bleeding at the end of defecation is the only symptom. Second degree hemorrhoids protrude beyond the anus as an uncomfortable swelling, but return spontaneously. Third degree hemorrhoids remain outside the anus and frequently require surgery. In a hemorrhoidectomy, or the surgical operation to remove hemorrhoids, the hemorrhoids are tied and then excised. After the hemorrhoids have been excised, possible complications are bleeding, or anal stricture which is a narrowing of the anus. In addition, the surgical wound is quite painful, and pain medication for the alleviation of this side effect is the rule not the exception.
There have been many attempts to address the pain associated with anal surgery. For example, U.S. patent numbers 5,679,707, 5,780,487, 5,648,381, and 5,437,291 discuss various compounds and treatments for hemorrhoids. However, the prior art techniques have shortcomings, and generally do not address the pain associated with the surgical wound. Moreover, at least one of the techniques involves the use of the botulinum neurotoxin which may be deadly if not properly administered.

Therefore, a safe and effective compound and therapy is needed for the treatment of pain associated with incisions and wounds, especially those resulting from surgery. The present invention fulfills this and other needs.

SUMMARY OF THE INVENTION

The present invention provides a safe and effective compound and therapy for treating and relieving the pain associated with cuts, abrasions, incisions, and wounds. In a specific application, the compound may be used to relieve the postoperative pain from surgical incisions and wounds. For example, in the case of anal surgery, such as a hemorrhoidectomy, the pain is known to be extremely severe, and the compound and therapy of the present invention is used to relieve this pain and discomfort. The present invention is also applicable to other operations, procedures, and conditions where pain is a side effect.

In one aspect, the present invention relates to a pharmaceutical composition comprising a guanylate cyclase inhibitor; a topical anesthetic; and a pharmaceutically acceptable carrier. The pharmaceutical composition is preferably used to treat pain from surgical wounds. In some preferred aspects, the guanylate cyclase inhibitor is methylene blue, leuko methylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid, retinol or mixtures thereof. The topical anesthetics include, but are not limited to, lidocaine, procaine and bupivacaine.

In another aspect, the present invention relates to a pharmaceutical composition comprising: a redox dye; a topical anesthetic; and a
pharmaceutically acceptable carrier. The pharmaceutical composition is preferably used to treat pain from surgical wounds. In certain preferred aspects, the redox dye includes, but is not limited to, methylene blue, toluidine blue, neutral red, tetrazolium salts, chloranil and dichlorophenolindophenol.

Examples of topical anesthetic include, but are not limited to, lidocaine, procaine and bupivacaine.

In still another aspect, the present invention relates to a method of treating a mammalian wound comprising: administering to a mammal a therapeutically acceptable amount of a guanylate cyclase inhibitor; and a pharmaceutically acceptable carrier. In certain embodiments, the guanylate cyclase inhibitor is methylene blue, leuko methylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid or retinol. In certain other aspects, the method further comprises applying or administering a topical anesthetic as a combination therapy with the guanylate cyclase inhibitor.

In still yet another aspect, the present invention relates to a method of treating a mammalian wound, comprising administering to a mammal a therapeutically acceptable amount of a redox dye and a pharmaceutically acceptable carrier. In certain aspects of this method, the method further comprises administering a topical anesthetic.

Other methods of the present invention include a method of relieving pain from a wound in a mammal, comprising administering to the mammal a therapeutically acceptable amount of a guanylate cyclase inhibitor; and a pharmaceutically acceptable carrier. In addition, the present invention provides another method of relieving pain from a wound in a mammal, comprising administering to the mammal a therapeutically acceptable amount of a redox dye and a pharmaceutically acceptable carrier.

A still further aspect of the present invention is the use of a compound including a first substance having properties like methylene blue and a second substance having properties like lidocaine to treat pain associated with incisions or wounds. Further, these incisions or wounds may result from surgical
procedures. Still further, the compounds with the first and second substances is used to treat postoperative pain resulting from anal surgery. The compound with first and second substances is used to treat and relieve pain associated with toothache, circumcision, or Caesarean section operations. In a specific embodiment, the first substance is about one percent in solution and the second substance is about two percent in solution. The compound with first and second substances is applied topically, transdermally, or interdermally.

In another aspect of the present invention, the compound of the present invention may be applied to the site of the cut, abrasion, incision, or wound. The compound may be applied topically, transdermally, or subdermally. For example, after a surgical procedure is completed, the compound of the present invention may be injected at the site of the incision area. The depth of the injection depends on the incision or cut, and there may be multiple injections to ensure the entire incision area is treated. This single treatment may be effective to relieve pain from the surgical incisions, and further treatment with the compound is not needed. Moreover, other pain relief drugs, such as oral or injected painkillers (e.g., Dolantin or Demerol) are not required. The compound of the present invention may be applied as needed to suit the specific application.

In other aspects, the present invention relates to a kit comprising: a guanylate cyclase inhibitor or a redox dye; a topical anesthetic; and instructions for use. Furthermore, the present invention also relates to apparatuses and techniques for effectively and hygienically dispersing the compounds of the present invention. For example, the compound may be supplied in disposable syringes, individually wrapped one-time use applicators and sponges, and individually wrapped bandages, among others. It is also important that the compound of the present invention be distributed to provide for a relatively long shelf life to ensure the compound is effective at the time of use.

Other features, objects and advantages of this invention and its preferred embodiments will become apparent from the detailed description which follows.
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BRIEF DESCRIPTION OF THE DRAWING

Figure 1 illustrates the chemical formulae of some guanylate cyclase inhibitors of this invention.

Figure 2 illustrates the chemical formulae of some redox dyes of this invention.

DEFINITIONS

The term "guanylate cyclase inhibitor" refers to a compound which inhibits the guanylate cyclase induced formation of the second messenger, cyclic GMP. Methylene blue is a preferred guanylate cyclase inhibitor.

The term "redox dye" refers to compounds which indicate by color change that either a chemical reduction or chemical oxidation has taken place. Methylene blue is a preferred redox dye.

The term "topical" or "topically" refers to a particular spot which can be in, or on any part of a mammal including, but not limited to, the epidermis, any other dermis or other body tissue. Topical administration means the direct contact of the anesthetic, redox dye or guanylate cyclase inhibitor with tissue, such as a skin or membrane.

The term "therapeutically effective amount" refers to the amount of active ingredient sufficient to produce the necessary effect. The active ingredient can be a guanylate cyclase inhibitor or a redox dye if reversible nerve inhibition is desirable. The active ingredient can also be a local anesthetic if topical anesthesia is desired.

DETAILED DESCRIPTION OF THE INVENTION

A. Pharmaceutical Compositions

In one aspect, the present invention relates to a pharmaceutical composition comprising a guanylate cyclase inhibitor; a topical anesthetic; and a pharmaceutically acceptable carrier. Guanylate cyclase inhibitors refer to compounds which inhibit guanylate cyclase induced formation of cyclic GMP. Examples of guanylate cyclase inhibitors suitable for use in the present
invention include, but are not limited to, methylene blue, leuko methylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid and retinol.

Methylene blue inhibits nitric acid stimulated guanylate cyclase activity. Nitric acid increases cyclic GMP synthesis by activating the soluble heme-containing form of guanylate cyclase. Nitric acid is one product of a reaction catalyzed by nitric oxide synthetase that uses arginine as its substrate (see, Deutsch et al., Neuropsychopharmacology, 15:37-43 (1996)).

Local anesthetics generally are esters or amides of benzoic acid derivatives, administered either as the free base or the acid addition salt. The anesthetic agents of this invention are those known, or of a type known, in the art. Examples of local or topical anesthetics of this invention include, but are not limited to, lidocaine, prilocaine mepivacaine, bupivacaine, dibuacine and etidocaine. Other suitable anesthetics include, but are not limited to, procaine, tetracaine, propoxycaine, chloroprocaine, benzocaine, cocaine, hexylcaine, piperocaine, oxyprocaine, and proparacaine. Other suitable anesthetics include, but are not limited to, cyclomethycaine, dimethisoquin, ketocaffine, diperodon, dyclonine, and pramoxine, all of which are typically administered in the form of the acid addition hydrochloride or sulfate salts.

In addition, the salts of the foregoing anesthetics are also useful in the present invention. Such salts include, but are not limited to, a dyclonine salt, a prilocaine salt, a tetracaine salt, a bupivacaine salt, a mepivacaine salt, a lidocaine salt, a procaine salt, an etidocaine salt, and a dibuacine salt. In a preferred embodiment, the guanylate cyclase inhibitor is methylene blue and the topical anesthetic is lidocaine. The pharmaceutical composition is preferably used to treat pain from a surgical wound.

With reference to Figure 1, methylene blue is a phenothiazine derivative and has been used as a biological stain and is known as a guanylate cyclase inhibitor (see, Huang et al., Eur. J Pharmacol, 205:209-294 (1991)). The methylene blue used for medicinal purposes has the molecular formula C_{16}H_{18}N_{3}SCl•3H_{2}O. Methylene blue is known to be an antidote for cyanide
and nitrate poisoning. Methylene blue, also known as methylthionine chloride, is a member of the class of compounds known as redox dyes. In addition to methylene blue, other redox dyes include, but are not limited to, toluidine blue, neutral red, leuko methylene blue, tetrazolium salts, chloranil, and dichlorophenolindophenol (see, Figure 2).

As such, in another embodiment, the present invention relates to a pharmaceutical composition comprising: a redox dye; a topical anesthetic; and a pharmaceutically acceptable carrier. Suitable redox dyes include, but are not limited to, methylene blue, toluidine blue, neutral red, tetrazolium salts, chloranil and dichlorophenolindophenol. The pharmaceutical composition is preferably used to treat pain from a wound.

Although it is understood that the methylene blue used for medicinal purposes has the molecular formula C₁₆H₁₈N₃SCl•3H₂O, other forms of methylene blue and derivatives thereof are also suitable for use in the present invention. These include, but are not limited to, methylene blue having various hydrate amounts, methylene blue with various counter ions (e.g., bromide, fluoride, iodide, and others), and methylene blue derivatives wherein the exocyclic ring nitrogen atoms are mono or disubstituted with C₁-C₆ alkyl.

As used herein the term alkyl includes, but is not limited to, branched or unbranched hydrocarbon chains, such as, methyl, ethyl, n-propyl, iso-propyl, n-butyl, sec-butyl, iso-butyl, tert-butyl, octa-decyl and 2-methylpentyl. These groups can be optionally substituted with one or more flinitional groups which are attached commonly to such chains, such as, hydroxyl, bromo, fluoro, chloro, iodo, mercapto or thio, cyano, alkylthio, heterocyclyl, aryl, heteroaryl, carboxyl, carbalkoyl, alkyl, alkenyl, nitro, amino, alkoxy, amido, and the like to form alkyl groups such as trifluoromethyl, 3-hydroxyhexyl, 2-carboxypropyl, 2-fluoroethyl, carboxymethyl, cyanobutyl and the like.

Without being bound to any particular theory, it is believed that methylene blue has a high affinity for components of the nervous system, and in particular the nerve endings. The administration of methylene blue to a mammal, such as a human being, reversibly inhibits the nerve ending, impairing
their function. The reversible inhibition of the nerve endings results in inactivity of the nerves or impairment of the nerve function with concomitant pain relief. In addition, the administration of the local anesthetic reduces or eliminates the pain of the temporary nerve inhibition. Typically, the body repairs the nerve impairment in about three weeks to a month.

B. **Uses, Dosages and Schedules**

The dosage of the composition used in accordance with the invention varies depending on the composition and the condition being treated. The age, weight, and clinical condition of the recipient patient; and the experience and judgment of the clinician or practitioner administering the therapy are among the factors affecting the selected dosage. Other factors include the route of administration to the patient, the patient's medical history, the severity of the disease process, and the potency of the particular compound. The dose should be sufficient to ameliorate symptoms or signs of the disease treated without producing unacceptable toxicity to the patient.

Typically, the dosage is administered once and preferably only once, but more or less frequent dosing can be recommended by the clinician. The single dose can be administered over a period of time. Thus, multiple sub-effective doses can be administered until cumulatively the effective dose is reached. The effective amount of the active ingredient is that amount which provides either subjective relief of symptoms or an objectively identifiable improvement as noted by the clinician or other qualified observer.

The compositions described in this invention can be used to treat a variety of disorders including pain relief of surgical wounds, anal itching, neurodermatitis, pain relief from the wound caused by a hemorrhoidectomy, dermatological damage, skin lesions, anal fissures and for the reversible inhibition of nerve endings.

The pain relieving abilities of the compositions of the present invention are effective for all surgical wounds and incisions. These surgical operations include those which treat disease, injury, deformities, microsurgery, and various
other conditions which require surgical treatment. In addition, other areas
where the compositions of the present invention are effective include, but are
not limited to, toothaches, such as tooth extraction, circumcision, and Caesarean
section.

As such, in another embodiment, the present invention relates to a
method of treating a mammalian wound. The method comprising:
administering to a mammal a therapeutically acceptable amount of a guanylate
cyclase inhibitor and a pharmaceutically acceptable carrier. In a particularly
preferred embodiment, the method further comprises administering a topical
anesthetic, such as lidocaine. The topical anesthetic is used to soothe the
sensation caused by the action of the guanylate cyclase inhibitor. Preferably,
the guanylate cyclase inhibitor is methylene blue, but other guanylate cyclase
inhibitors can be used.

In yet another embodiment, the present invention relates to a method of
treating a mammalian wound. The method comprising: administering to a
mammal a therapeutically acceptable amount of a redox dye and a
pharmaceutically acceptable carrier. In a preferred embodiment, the method
further comprises administering a topical anesthetic.

In still other aspects, the present invention relates to a method of
relieving pain from a surgical wound or incision in a mammal. The method
comprising: administering to a mammal a therapeutically acceptable amount of
a guanylate cyclase inhibitor and a pharmaceutically acceptable carrier. In a
particularly preferred embodiment, the method further comprises administering
a topical anesthetic, such as lidocaine. The topical anesthetic is used to soothe
the sensation caused by the action of the guanylate cyclase inhibitor.
Preferably, the guanylate cyclase is methylene blue.

In still other aspects, the present invention relates to a method of
relieving pain from a surgical wound in a mammal. The method comprising:
administering to the mammal a therapeutically acceptable amount of a redox
dye and a pharmaceutically acceptable carrier. In a particularly preferred
embodiment, the method further comprises administering a topical anesthetic, such as lidocaine. Preferably, the redox dye is methylene blue.

As discussed above, it is believed that the guanylate cyclase inhibitor or redox dye, such as methylene blue, has a high affinity to components of the nervous system, and in particular the nerve endings. For instance, the administration of methylene blue to a mammal, such as a human being, reversibly inhibit the nerve function. The reversible inhibition of the nerve endings results in inactivity of the nerves with concomitant pain relief. The administration of the local anesthetic either simultaneously or sequentially, reduces or eliminates the pain of the temporary nerve inhibition. Typically, the body repairs the nerve inhibition in about three weeks to a month.

As such, the present invention relates to reversible inhibition of the nerve endings in a mammal. The method comprising: administering to a mammal a guanylate cyclase inhibitor, or a redox dye in a therapeutically effective amount sufficient to reversibly inhibit the nerve and alleviate pain. In a particularly preferred embodiment, the method further comprises administering a topical anesthetic, such as lidocaine. Preferably, the guanylate cyclase inhibitor or redox dye is methylene blue.

In still yet another aspect, the present invention relates to a method of relieving pain from a wound in a mammal. The method comprising: reversibly inhibiting the nerve endings. In a preferred embodiment, the reversible inhibition is cause by administering to a mammal a guanylate cyclase inhibitor, or a redox dye in a therapeutically effective amount sufficient to reversibly inhibit the nerve and thereby alleviate pain. In a particularly preferred embodiment, the method further comprises administering a topical anesthetic, such as lidocaine. Preferably, the guanylate cyclase inhibitor or redox dye is methylene blue.

The combination therapy of the guanylate cyclase inhibitor, or a redox dye and the topical anesthetic can be done sequentially or concomitantly. If done sequentially, a preferred embodiment comprises administering the topical anesthetic first, followed by the guanylate cyclase inhibitor, or a redox dye.
In another aspect, the present invention relates to a kit. Generally, the kit comprises a guanylate cyclase inhibitor; a topical anesthetic; and instructions for use. Preferably, the guanylate cyclase inhibitors suitable for use in the kits of this invention include, but are not limited to, methylene blue, leukomethylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid, and retinol. More preferably, the guanylate cyclase is methylene blue.

In certain preferred aspects, the kit according to the present invention further comprises a topical anesthetic including, but not limited to, lidocaine, procaine and bupivacaine. Preferably, the topical anesthetic is lidocaine.

In other aspects, the kit comprises a redox dye; a topical anesthetic; and instructions for use. In certain preferred aspects, the kits of this invention are compartmentalized kits. A compartmentalized kit of this invention has a first container providing an active ingredient, such as a guanylate cyclase inhibitor or a redox dye; a second container providing an aqueous solution of a topical anesthetic, such as lidocaine; and instructions for use. Optionally, the kit contains hypodermic needles and syringes for use.

Although the invention has been described with human patients as the preferred mammalian recipient, veterinary use is also contemplated.

C. Formulations

The guanylate cyclase inhibitors or redox dyes of this invention can be formulated in a variety of carriers and delivery systems. Means of preparation, formulation and administration are known to those of skill. See generally, Remington's Pharmaceutical Science 15th ed., Mack Publishing Co., Easton, Pa. (1980).

For instance, to prepare a long-acting depot formulation, a therapeutically effective concentration of the active ingredient is placed in water, an oil, resin, biopolymer, or other suitable delivery device as is known in the art. The amount of the guanylate cyclase inhibitor or redox dye to be administered and the composition's concentration in depot formulations depends
upon the vehicle or device selected, the clinical condition of the patient, the side
effects, and the stability of the composition of the formulation. Thus, the
physician employs the appropriate preparation containing the appropriate
concentration of the guanylate cyclase inhibitor or redox dye and selects the
amount of formulation administered, depending upon clinical experience with
the patient in question or with similar patients.

In addition to the guanylate cyclase inhibitor or redox dye, and a topical
anesthetic as described above, the compositions can include, depending on the
formulation desired, pharmaceutically acceptable nontoxic carriers, or diluents
which include vehicles commonly used to form pharmaceutical compositions
for animal or human administration. The diluent is selected so as not to unduly
affect the biological activity of the combination. Examples of diluents which
are especially useful for injectable formulations are water, the various saline
solutions, Ringer's solution, dextrose solution, and Hank's solution. In addition,
the pharmaceutical composition or formulation may include additives such as
other carriers; adjuvants; or nontoxic, nontherapeutic, nonimmunogenic
stabilizers and the like.

Furthermore, excipients can be included in the formulation. Examples
include co-solvents, surfactants, oils, humectants, emollients, preservatives,
stabilizers, and antioxidants. Any pharmacologically acceptable buffer may be
used, e.g., Tris or phosphate buffers. Effective amounts of diluents, additives
and excipients are those amounts which are effective to obtain a
pharmaceutically acceptable formulation in terms of solubility, biological
activity, and others.

Thus, a composition of the invention includes a guanylate cyclase
inhibitor or redox dye, a topical anesthetic, both of which can be formulated
with a conventional, pharmaceutically acceptable carrier or vehicles for
parenteral or topical administration. Formulations can also include small
amounts of adjuvants such as buffers and preservatives to maintain isotonicity,
physiological, and pH stability. Means of preparation, formulation and

The compositions of this invention are typically administered to human patients parenterally or topically, or both. Preferably, the compositions are administered in unit dosage forms suitable for single administration of precise dosage amounts. For example, long-acting depot compositions are administered subcutaneously or intra-muscularly as precise unit doses with each dose lasting weeks to a month. Typically, one dose of the therapeutically effective amount parenterally administered cutaneous, subcutaneously, or intramuscularly will be effective.

For injection, the compositions can be formulated into preparations by dissolving, suspending or emulsifying them in an aqueous or nonaqueous solvent, such as vegetable or other similar oils, synthetic aliphatic acid glycerides, esters of higher aliphatic acids or propylene glycol; and, if desired, with conventional additives such as solubilizers, isotonic agents, suspending agents, emulsifying agents, stabilizers, and preservatives. Preferably, the compositions of the invention may be formulated in aqueous solutions, preferably in physiologically compatible buffers such as Hank’s solution, Ringer’s solution, physiological saline buffer, or distilled water. For transmucosal administration, penetrants appropriate to the barrier to be permeated are used in the formulation. Such penetrants are generally known in the art.

Appropriate formulations for parenteral use are apparent to the practitioner of ordinary skill. Usually, the therapeutic compound, *i.e.*, the guanylate cyclase inhibitor or the redox dye, is prepared in an aqueous solution in a concentration of from about one milligram per milliliter to about fifty milligrams per milliliter. More typically, the concentration is from about one milligram per milliliter to about twenty milligrams per milliliter or about one milligram per milliliter to about five milligrams per milliliter. Most preferably, the concentration is about one milligram per milliliter. Concentrations below one milligram per milliliter may be necessary in some cases depending on the
solubility and potency of the active ingredient selected for use. The formulation, which is sterile, is suitable for various parenteral routes including dermal, intradermal, subdermal, intramuscular, cutaneous, and subcutaneous.

The topical anesthetic solution, such as lidocaine hydrochloride, is used in about a one percent solution to about a twenty percent solution, more preferably about a one percent solution to about a ten percent solution, and most preferably about a two percent solution to about a five percent solution.

The guanylate cyclase inhibitor or redox dye, such as methylene blue solution, is about a one percent solution to about a twenty percent solution, more preferably about a one percent solution to about a ten percent solution and most preferably about a one percent solution to about a five percent solution.

The ratio of lidocaine hydrochloride solution to methylene blue solution depends in part on the weight percentages used. If for example, a two percent lidocaine hydrochloride solution is used, and a one percent methylene chloride solution is used, a ratio of about eighteen-to-one to about one-to-nine can be used, more preferably, a ratio of about ten-to-one to about one-to-five can be used, and most preferably, a ratio of eight-to-one to about one-to-two is used.

The compositions of this invention can also be delivered topically. To prepare a topical formulation for the treatment of wounds, anal fissures, hemorrhoids or dermatological disorders, a therapeutically effective concentration of a guanylate cyclase inhibitors or redox dye is placed in a dermatological vehicle as is known in the art. The amount of the therapeutic guanylate cyclase inhibitor or redox dye to be administered and the compound's concentration in the topical formulations depend upon the vehicle selected, the clinical condition of the patient, the side effects and the stability of the composition in the formulation. Thus, the physician employs the appropriate preparation containing the appropriate concentration of the guanylate cyclase inhibitor or redox dye and selects the amount of formulation administered, depending upon clinical experience with the patient in question or with similar patients.
The concentration of the therapeutic compound for topical formulations, *i.e.*, the guanylate cyclase inhibitor or the redox dye, ranges from about one milligram per milliliter to about one hundred milligrams per milliliter, more preferably, from about one milligram per milliliter to about fifty milligrams per milliliter and, more preferably from about two milligrams per milliliter to about twenty milligrams per milliliter. Most preferably, a concentration of about two milligrams per milliliter is employed. Solid dispersions of the therapeutic compound as well as solubilized preparations can be used. Thus, the precise concentration is subject to modest experimental manipulation in order to optimize the therapeutic response. About 2500 milligrams of therapeutic compound per 100 grams of vehicle is useful in the treatment of skin lesions to provide a two and a half percent weight/weight (w/w) formulation. Suitable vehicles include oil-in-water or water-in-oil emulsions using mineral oils, petrolatum and the like as well as gels such as hydrogels.

The therapeutic composition is optionally administered topically by the use of a transdermal therapeutic system (*see*, Barry, *Dermatological Formulations*, (1983) p. 181 and literature cited therein). A preferred way to practice the invention is to apply the guanylate cyclase inhibitor or redox dye, together with the topical anesthetic, in a cream, lotion, ointment, or oil based carrier, directly to the skin lesions.

The therapeutic composition is alternatively administered by aerosol. This is accomplished by preparing an aqueous aerosol, liposomal preparation or solid particles containing the composition. A nonaqueous (*e.g.*, fluorocarbon propellant) suspension can be used. Sonic nebulizers are preferred because they minimize exposing the therapeutic compound to shear, which can result in degradation of the composition.

Ordinarily, an aqueous aerosol is made by formulating an aqueous solution or suspension of the therapeutic composition together with conventional pharmaceutically acceptable carriers and stabilizers. The carriers and stabilizers vary with the requirements of the particular compound, but typically include nonionic surfactants (*Tweens, Pluronics, or polyethylene*
glycol); innocuous proteins such as serum albumin, sorbitan esters, oleic acid, lecithin; amino acids such as glycine; buffers; salts; sugars or sugar alcohols. Aerosols generally are prepared from isotonic solutions.

Slow or extended-release delivery systems, including any of a number of biopolymers (biological-based systems), systems employing liposomes, and polymeric delivery systems, can be utilized with the compositions described herein to provide a continuous or long term source of therapeutic composition. Such slow release systems are applicable to formulations for topical and parenteral use.

In one preferred delivery system, the compositions of the present invention are present on a patch, such as a transdermal patch, a dressing, a bandage, a gauze, a membrane or some other carrier which will allow the delivery system to stay adhered to the site of interest. As used herein, the site of interest can be a surgical wound, an anal fissure, a hemorrhoid, a lesion, a surgical incision or some other area wherein pain relief is desired. In this way, the composition is in direct contact with the site of interest. This delivery system optionally contains an adhesive which attaches and preferably strongly attaches to the site of interest. Optionally, the patch or bandage will be affixed to the site of interest with additional adhesive, such as surgical tape or wound dressing tape. (See, U.S. patent numbers 5,419,913, 5,156,847, and 4,806,614 the teachings of which are incorporated herein by reference).

As such, in another aspect, the present invention provides a wound dressing comprising a sterile bandage including an effective amount of a guanylate cyclase inhibitor or a redox dye and a topical anesthetic sufficient to alleviate pain. The guanylate cyclase inhibitors, redox dyes and topical anesthetics have been described above. Preferably, the guanylate cyclase inhibitor or redox dye is methylene blue and the topical anesthetic is lidocaine.

The material for bandage manufacture is well known to those of skill in the art. Suitable bandage materials include, but are not limited to, nontoxic polymers, particularly those types used to carry drugs for transdermal delivery, natural or synthetic elastomers, polyisobutylene, styrene, butadiene, acrylic acid
polyacrylates, cellulose and cellulose derivatives and polymers typically used for bandages and wound dressings.

In another embodiment, the compositions of the present invention are useful for coating medical devices. These medical devices include, but are not limited to, catheters, needles, sutures, clips, staples and other instruments wherein a mammal is subject to pain during insertion of the device or instrument.

In other aspects, the compositions of the present invention are useful for coating medical devices which are implanted for use. These devices include, but are not limited to, insulin pumps, dialysis needles and transfusion needles. Those skilled in the art will be aware of other medical devices wherein the present invention may be delivered.

D. Examples

Example 1

This example illustrates a clinical trial and therapy using a composition of the present invention.

i. Participants

A total of 5000 subjects were selected for this trial. Each individual had a clinically established case of third degree hemorrhoids. Subjects were male or female, between the age of 20 years to 65 years, with various body weights, and no diagnosed complications. The 5000 subjects represented a cross section of the population. All subjects underwent hemorrhoidectomies. All subjects were of Chinese descent. Physical parameters such as body weight, body mass index, height, medications, and diet were measured and recorded for each subject.

ii. Dosage and Duration

The subjects were initially screened and randomly assigned into one of two groups. Group I consisted of those taking the traditional pain relief medication only. Group II were those using the compositions as set forth herein
as well as in some instances other pain relief medication. The composition of
the present invention was provided to each subject in at least one dose and in
some instances multiple subeffective doses, which cumulatively equaled the
effective dose. The total dosage of four-to-one lidocaine (two percent)
methylene blue (two percent) was provided to each subject.
With reference to Table 1, it can be seen that about twenty-five percent
of the subjects who took the compositions as set forth herein also took oral pain
relief medication. The composition set out in example 3 was the composition
used in this trial. The reason some patients took oral medication is due to the
fact that these subjects took the pain medication prophylactically.

iii. Study Protocol
Each participant was on a light diet at least twelve hours prior to
surgery. Sterile surgical hemorrhoidectomies were performed on each patient,
and the surgeries were performed without complications. The compositions of
the present invention were generally administered when the patients were in an
unconscious or semiconscious state, after the hemorrhoidectomy was
performed. The administration was typically performed right after surgery.
In most instances, the composition was injected with an effective dose,
or alternatively, multiple injections with subeffective doses that cumulatively
equaled an effective dose into the area where the patient felt pain.

iv. Analysis
When asked about the local feeling after the operation, most patients
reported a local sensation, presumably due to the action of the composition on
the local nerve endings rather than pain. As shown in the tabulated data below,
the percentage of Dolantin (Demerol) injection for pain relief was drastically
reduced when the composition of the present invention was used.
v. **Statistical Analyses of the Data**

All the parameters were statistically analyzed and the data from each group compared.

vi. **Results**

Table 1 and Table 2 summarizes the study, with Table 1 containing the data from participants who were administered the compositions of the present invention. Table 2 contains the data from participants taking traditional medication.

Typically, pain in the anal area will induce spasm of the bladder sphincter which will cause concomitant urine retention. About seven percent of the clinical participants needed a urine induction tube. By using the compositions of this invention, the extent of the above problem was reduced and the urine tube usage dropped to less than one percent.

The pain relieving action of compositions of this invention lasted over twenty days and the local nerve endings were completely restored in about thirty days. No side effects were observed in any of the cases.

**Table 1**

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<th>oral pain relief medicine</th>
<th>injection of Dolantin</th>
<th>urine tube required</th>
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<tr>
<td>No. cases</td>
<td>1860</td>
<td>640</td>
<td>12</td>
<td>8</td>
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<td>percent</td>
<td>74.4%</td>
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**Table 2**

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<th>injection of Dolantin</th>
<th>urine tube required</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. cases</td>
<td>2500</td>
<td>464</td>
<td>186</td>
</tr>
<tr>
<td>percent</td>
<td>100%</td>
<td>18.56%</td>
<td>7.44%</td>
</tr>
</tbody>
</table>

Note: Some of the patients who took oral pain medicine also took the injected Dolantin. Therefore, the total case number is over 2500.
vii. **Conclusion**

This example illustrates that using the compositions of the present invention, clinical subject participants can return to their usual activities with minimal or no discomfort on the second day after surgery. For most patients, hospitalization is usually unnecessary. The compositions of this invention also make it possible to perform hemorrhoidectomies on older and weaker patients. It makes the surgical therapy a much more easy experience for patients.

**Example 2**

This example illustrates a clinical trial and therapy by topical application. A patient having anal fissures is selected for therapy using the present invention. A composition of methylene blue and lidocaine is prepared in a cream vehicle at a concentration of one percent to five percent (weight/volume), typically two and a half percent and is applied to the affected skin. After the first day of treatment, no pain is associated with the anal fissure.

**Example 3**

This example illustrates a formulation of the composition.

In the clinical trial as set forth in example 1, the formulation known as composition 1 had the following specification: one milliliter of two percent methylene blue (ten milligrams); four milliliters of two percent lidocaine; five milliliters of distilled water.

All publications, patents and patent applications mentioned in this specification are herein incorporated by reference into the specification in their entirety for all purposes. Although the invention has been described with reference to preferred embodiments and examples thereof, the scope of the present invention is not limited only to those described embodiments. As will be apparent to persons skilled in the art, modifications and adaptations to the above-described invention can be made without departing from the spirit and scope of the invention, which is defined and circumscribed by the appended claims.
WHAT IS CLAIMED IS:

1. A pharmaceutical composition comprising:
   a) a guanylate cyclase inhibitor;
   b) a topical anesthetic; and
   c) a pharmaceutically acceptable carrier therefor.

2. A pharmaceutical composition according to claim 1,
   wherein said guanylate cyclase inhibitor is a member selected from the group
   consisting of methylene blue, leuko methylene blue, 6-amininoquinoline-5,8-
   quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid and retinol.

3. A pharmaceutical composition according to claim 1,
   wherein said guanylate cyclase inhibitor is methylene blue.

4. A pharmaceutical composition according to claim 1,
   wherein said topical anesthetic is a member selected from the group consisting
   of lidocaine, procaine and bupivacaine.

5. A pharmaceutical composition according to claim 4,
   wherein said topical anesthetic is lidocaine.

6. A pharmaceutical composition according to claim 3,
   wherein said topical anesthetic is lidocaine.

7. A pharmaceutical composition according to claim 6,
   wherein said topical anesthetic is two percent lidocaine hydrochloride solution
   and said guanylate cyclase inhibitor is a one percent methylene blue solution in
   a ratio of about eighteen-to-one to about one-to-nine.

8. A pharmaceutical composition comprising:
   a) a redox dye;
   b) a topical anesthetic; and
   c) a pharmaceutically acceptable carrier therefor.

9. A pharmaceutical composition according to claim 8,
   wherein said redox dye is a member selected from the group consisting of
   methylene blue, toluidine blue, neutral red, tetrazolium salts, chloranil and
   dichlorophenolindophenol.
10. A pharmaceutical composition according to claim 8, wherein said redox dye is methylene blue.

11. A pharmaceutical composition according to claim 8, wherein said topical anesthetic is a member selected from the group consisting of lidocaine, procaine and bupivacaine.

12. A pharmaceutical composition according to claim 11, wherein said topical anesthetic is lidocaine.

13. A method of treating a mammalian wound, said method comprising administering to said mammal a therapeutically acceptable amount of a guanylate cyclase inhibitor; and a pharmaceutically acceptable carrier therefor.

14. A method according to claim 13, further comprising administering a topical anesthetic.

15. A method according to claim 14, wherein said guanylate cyclase inhibitor is a member selected from the group consisting of methylene blue, leuko methylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid and retinol.

16. A method according to claim 14, wherein said guanylate cyclase is methylene blue.

17. A method of treating a mammalian wound, said method comprising administering to said mammal a therapeutically acceptable amount of a redox dye; and a pharmaceutically acceptable carrier therefor.

18. A method according to claim 17, further comprising administering a topical anesthetic.

19. A method according to claim 18, wherein said redox dye is a member selected from the group consisting of methylene blue, toluidine blue, neutral red, tetrazolium salts, chloranil and dichlorphenolindophenol.

20. A method according to cl 18, wherein said redox dye is methylene blue.

21. A method of relieving pain in a mammal, said method comprising administering to said mammal a therapeutically acceptable amount
of a guanylate cyclase inhibitor; and a pharmaceutically acceptable carrier therefor.

22. A method according to claim 21, further comprising administering a topical anesthetic.

23. A method according to claim 22, wherein said guanylate cyclase inhibitor is a member selected from the group consisting of methylene blue, leuko methylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid and retinol.

24. A method according to claim 22, wherein said guanylate cyclase is methylene blue.

25. A method of relieving pain in a mammal, said method comprising administering to said mammal a therapeutically acceptable amount of a redox dye; and a pharmaceutically acceptable carrier therefor.

26. A method according to claim 25, further comprising administering a topical anesthetic.

27. A method according to claim 26, wherein said redox dye is a member selected from the group consisting of methylene blue, toluidine blue, neutral red, tetrazolium salts, chloranil and dichlorophenolindophenol.

28. A method according to claim 26, wherein said redox dye is methylene blue.

29. A method of reversibly inhibiting a nerve ending in a mammal, said method comprising: administering to said mammal a therapeutically acceptable amount of a guanylate cyclase inhibitor and a pharmaceutically acceptable carrier therefor.

30. A method according to claim 29, further comprising administering a topical anesthetic.

31. A method according to claim 29, wherein said guanylate cyclase is methylene blue.

32. A kit comprising:
   a) a guanylate cyclase inhibitor;
   b) a topical anesthetic; and
c) instructions for use.

33. A kit according to claim 32, wherein said guanylate cyclase inhibitor is a member selected from the group consisting of methylene blue, leuko methylene blue, 6-anilinoquinoline-5,8-quinone, N-methylhydroxylamine, hydroxylamine, ethacrynic acid and retinol.

34. A kit according to claim 32, wherein said guanylate cyclase is methylene blue.

35. A kit according to claim 32, wherein said topical anesthetic is a member selected from the group consisting of lidocaine, procaine and bupivacaine.

36. A kit according to claim 32, wherein said topical anesthetic is lidocaine.

37. A kit comprising:
   a) a redox dye;
   b) a topical anesthetic; and
   c) instructions for use.

38. A kit according to claim 37, wherein said redox dye is a member selected from the group consisting of methylene blue, toluidine blue, neutral red, tetrazolium salts, chloranil and dichlorophenolindophenol.

39. A kit according to claim 37, wherein said redox dye is methylene blue.

40. A kit according to claim 37, wherein said topical anesthetic is a member selected from the group consisting of lidocaine, procaine and bupivacaine.

41. A kit according to claim 37, wherein said topical anesthetic is lidocaine.

42. A wound dressing comprising:
   a) a sterile bandage;
   b) an effective amount of a member selected from the group consisting of a guanylate cyclase inhibitor or a redox dye; and
   c) a topical anesthetic sufficient to alleviate pain.
43. A wound dressing according to claim 42, wherein said guanylate cyclase inhibitor is methylene blue.

44. A wound dressing according to claim 42, wherein said topical anesthetic is lidocaine.

45. A method of relieving pain caused by a wound in a mammal, said method comprising: reversibly inhibiting a nerve ending in said mammal thereby relieving pain.

46. A method according to claim 45, wherein said inhibition is chemically induced.

47. A method according to claim 46, wherein said chemical is methylene blue.
FIGURE 1

Methylene Blue

Leuko Methylene Blue

6-anilinoquinoline-5,8-quinone

CH$_3$-NH$^-$OH · HCl
N-hydroxylamine hydrochloride

H$_2$NOH · HCl
hydroxylamine hydrochloride

Ethacrynic acid

Retinol
FIGURE 2

Toluidine Blue

Neutral Red

Tetrazolium Violet

Chloranil
### INTERNATIONAL SEARCH REPORT

**Classifications of Subject Matter**

|-------|-----------|-----------|-----------|------------|------------|-----------|

According to the International Patent Classification (IPC) or to both national classifications and IPC

**Fields Searched**

- **Minimum documentation searched (classification system followed by classification symbols)**
  - IPC 7 A61K

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

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

### DOCUMENTS CONSIDERED TO BE RELEVANT

<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
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| X        | DATABASE WI 
  Section Ch, Week 199721 
  Derwent Publications Ltd., London, GB; 
  Class B05, AN 1997-234164 
  XP002134044 
  & RO 111 151 A (OHANOVICI M), 
  abstract | 1-6, 8-41, 45-47 |
| X        | DATABASE WI 
  Section Ch, Week 199741 
  Derwent Publications Ltd., London, GB; 
  Class B02, AN 1997-436147 
  XP002134045 
  & CN 1 116 093 A (XINCHENG GOLD MINE SHANDONG PROV), 
  7 February 1996 (1996-02-07) 
  abstract | 1-4, 8-11, 32-35, 37-40 |

Further documents are listed in the continuation of box C. Patent family members are listed in annex.

- **A** document defining the general state of the art which is not considered to be of particular relevance
- **E** earlier document but published on or after the international filing date
- **L** document which may throw doubts on the priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- **O** document relating to an oral discussion, use, exhibition or other means
- **P** document published prior to the international filing date but later than the priority date claimed
- **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
- **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
- **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
- **A** document member of the same patent family

**Date of the actual completion of the international search**

29 March 2000

**Date of mailing of the international search report**

06/04/2000

Name and address of the ISA

European Patent Office, P.B. 5818 Patentfair 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl
Fax (+31-70) 540-3016

Authorized officer

Siouatou, E
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| X        | WO 95 31195 A (J. OLESEN)  
page 3, line 21 | 21, 25, 29 |
| X        | DUARTE I D G ET AL: "PERIPHERAL ANALGESIA AND ACTIVATION OF THE NITRIC OXIDE-CYCLIC GMP PATHWAY"  
EUROPEAN JOURNAL OF PHARMACOLOGY, NL, AMSTERDAM,  
vol. 186, no. 2/03,  
21 September 1990 (1990-09-21), pages 289-293, XP002038072  
ISSN: 0014-2999  
page 292, left-hand column, paragraph 4. right-hand column | 21-31, 45-47 |
| X        | DUARTE I D G ET AL: "ANALGESIA BY DIRECT ANTAGONISM OF NOCICEPTOR SENSITIZATION INVOLVES THE ARGinine-NITRIC OXIDE-CGMP PATHWAY"  
EUROPEAN JOURNAL OF PHARMACOLOGY, NL, AMSTERDAM,  
vol. 217, no. 2/03,  
7 July 1992 (1992-07-07), pages 225-227, XP000609168  
ISSN: 0014-2999  
abstract  
page 226, right-hand column, last paragraph | 21-31, 45-47 |
| A        | SCHAFFER M R ET AL: "NITRIC OXIDE REGULATES WOUND HEALING"  
JOURNAL OF SURGICAL RESEARCH, US, ACADEMIC PRESS INC., SAN DIEGO, CA,  
vol. 63, no. 1, 1 June 1996 (1996-06-01), pages 237-240, XP000610179  
ISSN: 0022-4804  
abstract | 1-47 |
| A        | BENRATH J ET AL: "SUBSTANCE P AND NITRIC OXIDE MEDIATE WOUND HEALING OF ULTRAVIOLET PHOTODAMAGED RAT SKIN: EVIDENCE FOR AN EFFECT OF NITRIC OXIDE ON KERATINOCYTE PROLIFERATION"  
NEUROSCIENCE LETTERS, IE, LIMERICK,  
vol. 200, no. 1,  
10 November 1995 (1995-11-10), pages 17-20, XP000610409  
ISSN: 0304-3940  
abstract | 1-47 |
INTERNATIONAL SEARCH REPORT

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

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

   because they relate to subject matter not required to be searched by this Authority, namely:
   Remark: Although claims 13–31 AND 45–47 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

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 8.4(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:

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.
<table>
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<th>Publication date</th>
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<td>RO 111151</td>
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