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(54) Title: N-OXIDE ARYL SULFONES AND SULFOXIDES

(57) Abstract: N-oxide aryl sulfones and sulfoxides of Formula (I) are provided: in which the variables are as described herein. Such compounds may be used to modulate bradykinin receptor activity *in vivo*or *in vitro*, and are particularly useful in the treatment of conditions responsive to Bi modulation in humans, domesticated companion animals and livestock animals, including inflammation and pain. Pharmaceutical compositions and methods for using them to treat such disorders are provided, as are methods for using such ligands for receptor localization studies and various *in vitro* assays.

# N-OXIDE ARYL SULFONES AND SULFOXIDES

## FIELD OF THE INVENTION

This invention relates generally to N-oxide aryl sulfones and sulfoxides, and to the use of such compounds to treat conditions responsive to bradykinin receptor-1 ($B_1$) modulation. The invention further relates to the use of such compounds as reagents for the identification of other agents that bind to $B_1$, and as probes for the detection and localization of $B_1$.

## CROSS-REFERENCE TO RELATED APPLICATIONS

This application claims priority to U.S. Provisional Application 60/823,265, filed August 23, 2006, which is hereby incorporated by reference in its entirety.

## DESCRIPTION OF THE SEQUENCE LISTING

| | |
|---|---|
| SEQ ID NO:1 | $B_1$ PCR primer number 1 |
| SEQ ID NO:2 | $B_1$ PCR primer number 2 |
| SEQ ID NO:3 | $B_1$ PCR primer number 3 |
| SEQ ID NO:4 | $B_1$ PCR primer number 4 |

## BACKGROUND OF THE INVENTION

Millions of people throughout the world suffer debilitating and incapacitating pain. Current treatments for pain can be beneficial, but such treatments are not completely effective, and typically have undesirable side effects. For example, non-steroidal anti-inflammatory drugs are commonly used, but are only moderately effective against pain, and have serious renal and gastrointestinal side effects at high doses. Opiates, such as morphine, are potent analgesics, but their usefulness is limited because of adverse side effects, such as physical addictiveness and withdrawal properties, as well as respiratory depression, mood changes, and decreased intestinal motility with concomitant constipation, nausea, vomiting, and alterations in the endocrine and autonomic nervous systems. There is thus a need for effective agents for the treatment of pain.

Bradykinin (BK) is a nonapeptide that functions in cardiovascular homeostasis, contraction and relaxation of smooth muscles, inflammation and pain. The effects of BK are mediated by specific G protein-coupled BK receptors, of which there are at least two distinct subtypes termed $B_1$ and $B_2$. The $B_2$ receptor is expressed constitutively in a variety of tissues. In contrast, the $B_1$ receptor is inducibly expressed in response to pathophysiological conditions such as inflammation, pain, trauma, bacterial infection, burns and shock.

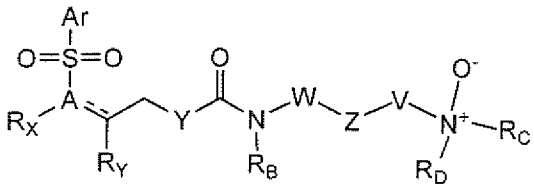
Accordingly, $B_1$ is a particularly attractive drug target for these and other conditions, and agents that act at this receptor may be targeted specifically to injured tissues, with minimal effects in

normal tissues. In addition, compounds that bind to $B_1$ and/or modulate the activity of $B_1$ also find use as research tools.

There is thus a need in the art for small molecule modulators of $B_1$ activity. The present invention fulfills this need and provides further related advantages.

## SUMMARY OF THE INVENTION

The present invention provides N-oxide aryl sulfones and sulfoxides that satisfy Formula I:



Formula I

or are a pharmaceutically acceptable salt, solvate (*e.g.*, hydrate) or ester of such a compound.

Within Formula I:

A is N, C or CH;

==== represents a single or double bond;

Ar is phenyl or a 5- or 6-membered heteroaryl, each of which is optionally substituted, and each of which is preferably substituted with from 0 to 4 substituents independently chosen from $R_1$;

$R_X$ and $R_Y$ are:

(i) independently chosen from:

(a) hydrogen; and

(b) $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl and ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, each of which is optionally substituted, and each of which is preferably substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, amino and $C_1$-$C_4$alkyl; or

(ii) taken together to form a 4- to 10-membered carbocycle or a 4- to 10-membered heterocycle, each of which is optionally substituted and each of which is preferably substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, halogen, amino and $C_1$-$C_8$alkyl;

Y is a group of the formula $(CH_2)_r$-Q-$(CH_2)_p$, which is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from (i) amino, hydroxy, cyano, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl and $C_1$-$C_6$haloalkyl; (ii) substituents of the same carbon atom or adjacent carbon atoms that are taken together to form $C_3$-$C_6$cycloalkyl, and (iii) substituents that are taken together with $R_X$ or $R_Y$ to form an optionally substituted 4- to 7-membered carbocycle or 4- to 7-membered heterocycle, wherein:

Q is absent, $CH_2$, CH=CH, O, S or $NR_{10}$, wherein $R_{10}$ is hydrogen or $C_1$-$C_6$alkyl; and

r and p are independently chosen integers ranging from 0 to 6;

W is $C_1$-$C_6$alkylene that is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

Z is absent, divalent phenyl, $NR_G$ or $CR_ER_F$ wherein:

$R_G$ is hydrogen, $C_1$-$C_6$alkyl or taken together with $R_B$ or $R_D$ to form a 4- to 7-membered heterocycloalkyl that includes W and is optionally substituted; preferably such heterocycloalkyl is substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

$R_E$ is hydrogen, $C_1$-$C_6$alkyl or taken together with $R_B$ to form a 4- to 7-membered heterocycloalkyl that includes W and is optionally substituted; preferably such heterocycloalkyl is substituted with from 0 to 4 substituents independently chosen from $R_{20}$; and

$R_F$ is hydrogen, $C_1$-$C_6$alkyl or taken together with $R_D$ to form a 4- to 7-membered heterocycloalkyl that includes V and is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

V is absent or $C_1$-$C_6$alkylene that is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from (i) $R_{20}$ and (ii) groups that are taken together with $R_D$ to form a 4- to 7-membered heterocycle that is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

Each $R_1$ is independently chosen from:

(i) halogen, hydroxy, cyano, amino, nitro, aminocarbonyl, aminosulfonyl and –COOH;

(ii) $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_1$-$C_6$alkoxy, $C_1$-$C_6$alkylthio, $C_1$-$C_6$alkylsulfinyl, $C_1$-$C_6$alkoxycarbonyl, $C_1$-$C_6$alkylsulfonyl$C_0$-$C_4$alkyl, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, mono- or di-($C_1$-$C_6$alkyl)aminosulfonyl$C_0$-$C_4$alkyl, mono- or di-($C_1$-$C_6$alkyl)aminocarbonyl$C_0$-$C_4$alkyl and (4- to 8-membered heterocycloalkyl)$C_0$-$C_4$alkyl; each of which is optionally substituted and each of which is preferably substituted with from 0 to 6 substituents independently chosen from halogen, hydroxy, cyano and amino; and

(iii) groups that are taken together to form a fused carbocyclic ring that is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl;

$R_B$ is:

(i) hydrogen, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_2$-$C_6$alkyl ether, mono- or di-($C_1$-$C_6$alkyl)amino$C_1$-$C_4$alkyl or (4- to 7-membered heterocycloalkyl)$C_0$-$C_4$alkyl, each of which is optionally substituted and each of which is preferably substituted with from 0 to 6 substituents independently chosen from:

(a) amino, halogen, hydroxy, cyano and oxo; and

(b) $C_1$-$C_6$alkyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_1$-$C_6$alkoxy, phenyl$C_0$-$C_4$alkyl and (5- or 6-membered heterocycle)$C_0$-$C_4$alkyl, each of which is optionally substituted and each of which is preferably substituted with from 0 to 4 substituents independently chosen from

amino, cyano, halogen, hydroxy, $C_1$-$C_6$alkyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, $C_1$-$C_6$alkoxycarbonyl, mono- or di-($C_1$-$C_6$alkyl)amino, phenyl$C_0$-$C_4$alkyl and phenyl$C_0$-$C_4$alkoxy; or

(ii) taken together with $R_E$ or $R_G$ to form an optionally substituted heterocycloalkyl;

$R_C$ is absent, $C_1$-$C_6$alkyl or taken together with $R_D$ to from a 4- to 7-membered heterocycle that is optionally substituted and is preferably substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

$R_D$ is $C_1$-$C_6$alkyl or taken together with $R_C$, $R_F$, $R_G$ or a substituent of V to form an optionally substituted 4- to 7-membered heterocycle; and

Each $R_{20}$ is independently selected from:

(i) hydroxy, oxo, cyano and amino; and

(ii) $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl, each of which is optionally substituted, and each of which is preferably substituted with from 0 to 2 substituents independently chosen from hydroxy, halogen, oxo, cyano, $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl.

Within certain aspects, N-oxide aryl sulfones and sulfoxides of Formula I, and other Formulas provided herein, are $B_1$ modulators and exhibit a $K_i$ of no greater than 5 micromolar, 2 micromolar, 1 micromolar, 500 nanomolar, 100 nanomolar, 50 nanomolar or 10 nanomolar in a $B_1$ binding assay and/or have an $EC_{50}$ or $IC_{50}$ value of no greater than 5 micromolar, 2 micromolar, 1 micromolar, 500 nanomolar, 100 nanomolar, 50 nanomolar or 10 nanomolar in an assay for determination of $B_1$ agonist or antagonist activity.

In certain embodiments, $B_1$ modulators provided herein are $B_1$ antagonists; preferably such antagonists exhibit no detectable $B_1$ agonist activity.

Within certain aspects, N-oxide aryl sulfones and sulfoxides of Formula I are labeled with a detectable marker (*e.g.*, radiolabeled or fluorescein conjugated).

The present invention further provides, within other aspects, pharmaceutical compositions comprising at least one N-oxide aryl sulfone or sulfoxide of Formula I in combination with a physiologically acceptable carrier or excipient.

Methods are further provided for inhibiting agonist-induced $B_1$ activity. Within certain such aspects, the inhibition takes place *in vitro*. Such methods comprise contacting a $B_1$ receptor with at least one $B_1$ antagonist as described herein, under conditions and in an amount or concentration sufficient to detectably inhibit agonist-induced $B_1$ activity. Within other such aspects, the $B_1$ receptor is in a patient. Such methods comprise contacting cells expressing a $B_1$ receptor in a patient with at least one $B_1$ antagonist as described herein in an amount or concentration that would be sufficient to detectably inhibit agonist-induced $B_1$ activity in cells expressing a cloned $B_1$ receptor *in vitro*.

The present invention further provides methods for treating a condition responsive to $B_1$ receptor modulation in a patient, comprising administering to the patient a therapeutically effective amount of at least one N-oxide aryl sulfone or sulfoxide of Formula I.

Within other aspects, methods are provided for treating pain in a patient, comprising administering to a patient suffering from (or at risk for) pain a therapeutically effective amount of at least one N-oxide aryl sulfone or sulfoxide of Formula I. Pain conditions that may be treated include, but are not limited to, inflammatory pain, acute pain, dental pain, back pain, surgical pain, headache, neuropathic pain, and pain associated with osteoarthritis or trauma.

Within further aspects, the present invention provides methods for determining the presence or absence of $B_1$ in a sample, comprising: (a) contacting a sample with an N-oxide aryl sulfone or sulfoxide of Formula I under conditions that permit binding of the compound to $B_1$; and (b) detecting a signal indicative of a level of the compound bound to $B_1$.

In yet another aspect, the present invention provides methods of preparing the compounds disclosed herein, including the intermediates.

These and other aspects of the present invention will become apparent upon reference to the following detailed description.

## DETAILED DESCRIPTION

As noted above, the present invention provides N-oxide aryl sulfones and sulfoxides, which may be used *in vitro* or *in vivo* in a variety of contexts, as described herein.

### TERMINOLOGY

Compounds are generally described herein using standard nomenclature. For compounds having asymmetric centers, it should be understood that (unless otherwise specified) all of the optical isomers and mixtures thereof are encompassed. In addition, compounds with carbon-carbon double bonds may occur in Z- and E- forms, with all isomeric forms of the compounds being included in the present invention unless otherwise specified. If a compound exists in various tautomeric forms, a recited compound is not limited to any one specific tautomer, but rather is intended to encompass all tautomeric forms. Certain compounds are described herein using a general formula that includes variables (*e.g.*, Y, Ar, $R_1$). Unless otherwise specified, each variable within such a formula is defined independently of any other variable, and any variable that occurs more than one time in a formula is defined independently at each occurrence.

The term "N-oxide aryl sulfone or sulfoxide" encompasses all compounds that satisfy Formula I, including compounds in which the Ar moiety is heteroaryl and compounds in which the Ar moiety is aryl. This term further includes pharmaceutically acceptable salts, solvates (*e.g.*, hydrates) and esters of such compounds.

A "pharmaceutically acceptable salt" of a compound recited herein is an acid or base salt that is suitable for use in contact with the tissues of human beings or animals without excessive toxicity or

carcinogenicity, and preferably without irritation, allergic response, or other problem or complication. Such salts include mineral and organic acid salts of basic residues such as amines, as well as alkali or organic salts of acidic residues such as carboxylic acids. Specific pharmaceutically acceptable anions for use in salt formation include, but are not limited to, acetate, 2-acetoxybenzoate, ascorbate, benzoate, bicarbonate, bitartrate, bromide, calcium edetate, carbonate, chloride, citrate, dihydrochloride, diphosphate, edetate, estolate (ethylsuccinate), formate, fumarate, gluceptate, gluconate, glutamate, glycolate, glycollylarsanilate, hexylresorcinate, hydrabamine, hydrobromide, hydrochloride, hydroiodide, hydroxymaleate, hydroxynaphthoate, iodide, isethionate, lactate, lactobionate, malate, maleate, mandelate, methylbromide, methylnitrate, methylsulfate, mucate, napsylate, nitrate, pamoate, pantothenate, phenylacetate, phosphate, polygalacturonate, propionate, salicylate, stearate, subacetate, succinate, sulfamate, sulfanilate, sulfate, sulfonates including besylate (benzenesulfonate), camsylate (camphorsulfonate), edisylate (ethane-1,2-disulfonate), esylate (ethanesulfonate), 2-hydroxyethylsulfonate, mesylate (methanesulfonate), triflate (trifluoromethanesulfonate) and tosylate (*p*-toluenesulfonate), tannate, tartrate, teoclate and triethiodide. Similarly, pharmaceutically acceptable cations for use in salt formation include, but are not limited to ammonium, benzathine, chloroprocaine, choline, diethanolamine, ethylenediamine, meglumine, procaine, and metals such as aluminum, calcium, lithium, magnesium, potassium, sodium and zinc. Those of ordinary skill in the art will recognize further pharmaceutically acceptable salts for the compounds provided herein. In general, a pharmaceutically acceptable acid or base salt can be synthesized from a parent compound that contains a basic or acidic moiety by any conventional chemical method. Briefly, such salts can be prepared by reacting the free acid or base forms of these compounds with a stoichiometric amount of the appropriate base or acid in water or in an organic solvent, or in a mixture of the two; generally, the use of nonaqueous media, such as ether, ethyl acetate, ethanol, methanol, isopropanol or acetonitrile, is preferred.

It will be apparent that each compound provided herein may, but need not, be formulated as a solvate (*e.g.*, a hydrate) or non-covalent complex. In addition, the various crystal forms and polymorphs are within the scope of the present invention. Also provided herein are prodrugs of the compounds provided herein. A "prodrug" is a compound that may not fully satisfy the structural requirements of a formula provided herein, but is modified *in vivo*, following administration to a patient, to produce a compound within the scope of such formula. For example, a prodrug may be an acylated derivative of a compound as provided herein. Prodrugs include compounds wherein hydroxy, amine or sulfhydryl groups are bonded to any group that, when administered to a mammalian subject, cleaves to form a free hydroxy, amino, or sulfhydryl group, respectively. Examples of prodrugs include, but are not limited to, acetate, formate, phosphate and benzoate derivatives of alcohol and amine functional groups within the compounds provided herein. Prodrugs of the compounds provided herein may be prepared by modifying functional groups present in the compounds in such a way that the modifications are cleaved *in vivo* to yield the parent compounds.

As used herein, the term "alkyl" refers to a straight or branched chain saturated aliphatic hydrocarbon. Alkyl groups include groups having from 1 to 8 carbon atoms ($C_1$-$C_8$alkyl), from 1 to 6 carbon atoms ($C_1$-$C_6$alkyl) and from 1 to 4 carbon atoms ($C_1$-$C_4$alkyl), such as methyl, ethyl, propyl, isopropyl, n-butyl, *sec*-butyl, *tert*-butyl, pentyl, 2-pentyl, isopentyl, neopentyl, hexyl, 2-hexyl, 3-hexyl or 3-methylpentyl. "$C_0$-$C_4$alkyl" refers to a single covalent bond ($C_0$) or an alkylene group having 1, 2, 3 or 4 carbon atoms; "$C_0$-$C_2$alkyl" refers to a single covalent bond or a methylene or ethylene group.

"Alkylene" refers to a divalent alkyl group, as defined above. $C_1$-$C_4$alkylene is an alkylene group having 1, 2, 3 or 4 carbon atoms.

"Alkenyl" refers to straight or branched chain alkene groups, which comprise at least one unsaturated carbon-carbon double bond. Alkenyl groups include $C_2$-$C_8$alkenyl, $C_2$-$C_6$alkenyl and $C_2$-$C_4$alkenyl groups, which have from 2 to 8, 2 to 6 or 2 to 4 carbon atoms, respectively, such as ethenyl, allyl or isopropenyl. "Alkynyl" refers to straight or branched chain alkyne groups, which have one or more unsaturated carbon-carbon bonds, at least one of which is a triple bond. Alkynyl groups include $C_2$-$C_8$alkynyl, $C_2$-$C_6$alkynyl and $C_2$-$C_4$alkynyl groups, which have from 2 to 8, 2 to 6 or 2 to 4 carbon atoms, respectively.

A "cycloalkyl" is a saturated or partially saturated cyclic group in which all ring members are carbon, such as cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl and partially saturated variants thereof. Certain cycloalkyl groups are $C_3$-$C_8$cycloalkyl, in which the ring contains from 3 to 8 ring members, all of which are carbon. A "($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl" is a $C_3$-$C_8$cycloalkyl group linked via a single covalent bond or a $C_1$-$C_4$alkylene group.

By "alkoxy," as used herein, is meant an alkyl group attached via an oxygen bridge (*i.e.*, -O-alkyl). Alkoxy groups include $C_1$-$C_6$alkoxy and $C_1$-$C_4$alkoxy groups, which have from 1 to 6 or from 1 to 4 carbon atoms, respectively. Methoxy, ethoxy, propoxy, isopropoxy, n-butoxy, *sec*-butoxy, *tert*-butoxy, n-pentoxy, 2-pentoxy, 3-pentoxy, isopentoxy, neopentoxy, hexoxy, 2-hexoxy, 3-hexoxy, and 3-methylpentoxy are representative alkoxy groups.

Similarly, "alkylthio" refers to an alkyl group as attached via a sulfur bridge (*i.e.*, –S–alkyl). Alkylthio groups include $C_1$-$C_6$alkylthio and $C_1$-$C_4$alkylthio groups, which have from 1 to 6 or from 1 to 4 carbon atoms, respectively.

"Alkylsulfinyl" refers to groups of the formula –(SO)–alkyl, in which the sulfur atom is the point of attachment. Alkylsulfinyl groups include $C_1$-$C_6$alkylsulfinyl and $C_1$-$C_4$alkylsulfinyl groups, which have from 1 to 6 or from 1 to 4 carbon atoms, respectively.

"Alkylsulfonyl" refers to groups of the formula –($SO_2$)–alkyl, in which the sulfur atom is the point of attachment. Alkylsulfonyl groups include $C_1$-$C_6$alkylsulfonyl and $C_1$-$C_4$alkylsulfonyl groups, which have from 1 to 6 or from 1 to 4 carbon atoms, respectively. The term "$C_1$-$C_6$alkylsulfonyl$C_0$-$C_4$alkyl refers to a $C_1$-$C_6$alkylsulfonyl that is linked via a single covalent bond or a $C_1$-$C_4$alkylene group (*i.e.*, -($C_0$-$C_4$alkyl)–($SO_2$)–($C_1$-$C_6$alkyl)).

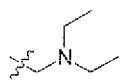
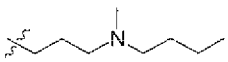
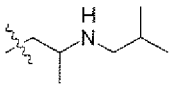
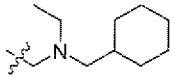
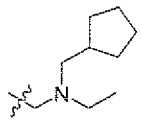
The term "alkanoyl" refers to an acyl group (*e.g.*, –(C=O)–alkyl), where attachment is through the carbon of the keto group. Alkanoyl groups include $C_2$-$C_8$alkanoyl, $C_2$-$C_6$alkanoyl and $C_2$-$C_4$alkanoyl groups, which have from 2 to 8, 2 to 6 or 2 to 4 carbon atoms, respectively. "$C_1$alkanoyl" refers to –(C=O)H, which (along with $C_2$-$C_8$alkanoyl) is encompassed by the term "$C_1$-$C_8$alkanoyl." Ethanoyl is $C_2$alkanoyl.

Similarly, "alkyl ether" refers to a linear or branched ether substituent. Alkyl ether groups include $C_2$-$C_8$alkyl ether, $C_2$-$C_6$alkyl ether and $C_2$-$C_4$alkyl ether groups, which have 2 to 8, 6 or 4 carbon atoms, respectively. A $C_2$ alkyl ether is $–CH_2–O–CH_3$.

The term "alkoxycarbonyl" refers to an alkoxy group linked via a carbonyl (*i.e.*, a group having the general structure –C(=O)–O–alkyl). Alkoxycarbonyl groups include $C_1$-$C_8$, $C_1$-$C_6$ and $C_1$-$C_4$alkoxycarbonyl groups, which have from 1 to 8, 6 or 4 carbon atoms, respectively, in the alkyl portion of the group. "$C_1$alkoxycarbonyl" refers to $–C(=O)–O–CH_3$.

"Alkylamino" refers to a secondary or tertiary amine that has the general structure –NH–alkyl or –N(alkyl)(alkyl), wherein each alkyl is selected independently from alkyl, cycloalkyl and (cycloalkyl)alkyl groups. Such groups include, for example, mono- and di-($C_1$-$C_8$alkyl)amino groups, in which each $C_1$-$C_8$alkyl may be the same or different, as well as mono- and di-($C_1$-$C_6$alkyl)amino groups and mono- and di-($C_1$-$C_4$alkyl)amino groups.

"Alkylaminoalkyl" refers to an alkylamino group linked via an alkylene moiety (*i.e.*, a group having the general structure –alkylene–NH–alkyl or –alkylene–N(alkyl)(alkyl)) in which each alkyl is selected independently from alkyl, cycloalkyl and (cycloalkyl)alkyl groups. Alkylaminoalkyl groups include, for example, mono- and di-($C_1$-$C_8$alkyl)amino$C_1$-$C_6$alkyl, and mono- and di-($C_1$-$C_6$alkyl)amino$C_1$-$C_4$alkyl. "Mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl" refers to a mono- or di-($C_1$-$C_6$alkyl)amino group linked via a single covalent bond or a $C_1$-$C_4$alkylene group. The following are representative alkylaminoalkyl groups:

It will be apparent that the definition of "alkyl" as used in the terms "alkylamino" and "alkylaminoalkyl" differs from the definition of "alkyl" used for all other alkyl-containing groups, in the inclusion of cycloalkyl and (cycloalkyl)alkyl groups (*e.g.*, ($C_3$-$C_7$cycloalkyl)$C_0$-$C_2$alkyl).

The term "aminocarbonyl" refers to an amide group (*i.e.*, $–C(=O)NH_2$). "Mono- or di-($C_1$-$C_6$alkyl)aminocarbonyl$C_0$-$C_4$alkyl" refers to an aminocarbonyl group in which one or both hydrogens are replaced with an independently selected $C_1$-$C_6$alkyl group, and which is linked via a single covalent bond or a $C_1$-$C_4$alkylene group.

Similarly, "aminosulfonyl" refers to a sulfonamide group (*i.e.*, $–SO_2NH_2$). "Mono- or di-($C_1$-$C_6$alkyl)aminosulfonyl$C_0$-$C_4$alkyl" refers to an aminosulfonyl group in which one or both hydrogens

are replaced with an independently selected $C_1$-$C_6$alkyl group, and which is linked via a single covalent bond or a $C_1$-$C_4$alkylene group.

The term "halogen" refers to fluorine, chlorine, bromine or iodine.

The term "oxo," as used herein refers to an oxygen substituent of a carbon atom that results in the formation of a carbonyl group (C=O). An oxo group that is a substituent of a nonaromatic carbon atom results in a conversion of $-CH_2-$ to $-C(=O)-$. An oxo group that is a substituent of an aromatic carbon atom results in a conversion of $-CH-$ to $-C(=O)-$ and may result in a loss of aromaticity.

A "haloalkyl" is an alkyl group that is substituted with 1 or more independently chosen halogens (*e.g.*, "$C_1$-$C_8$haloalkyl" groups have from 1 to 8 carbon atoms; "$C_1$-$C_6$haloalkyl" groups have from 1 to 6 carbon atoms). Examples of haloalkyl groups include, but are not limited to, mono-, di- or tri-fluoromethyl; mono-, di- or tri-chloromethyl; mono-, di-, tri-, tetra- or penta-fluoroethyl; mono-, di-, tri-, tetra- or penta-chloroethyl; and 1,2,2,2-tetrafluoro-1-trifluoromethyl-ethyl. Typical haloalkyl groups are trifluoromethyl and difluoromethyl.

A dash ("–") that is not between two letters or numbers is used to indicate a point of attachment for a substituent. For example, $-C(=O)NH_2$ is attached through the carbon atom.

A "carbocycle" has from 1 to 3 fused, pendant or spiro rings, each of which has only carbon ring members and each of which may, but need not, be bridged by an alkylene moiety. Typically, a carbocycle that has a single ring contains from 3 to 8 ring members (*i.e.*, $C_3$-$C_8$carbocycles); rings having from 4 or 5 to 7 ring members (*i.e.*, $C_4$-$C_7$carbocycles or $C_5$-$C_7$carbocycles) are recited in certain embodiments. Carbocycles comprising fused, pendant or spiro rings typically contain from 9 to 14 ring members. Carbocycles may be optionally substituted with a variety of substituents, as indicated. Unless otherwise specified, a carbocycle may be a cycloalkyl group (*i.e.*, each ring is saturated or partially saturated as described above) or an aryl group (*i.e.*, at least one ring within the group is aromatic). Representative aromatic carbocycles are phenyl, naphthyl, tetrahydronaphthyl and biphenyl. In certain embodiments preferred carbocycles have a single ring, such as phenyl and $C_3$-$C_8$cycloalkyl groups.

Certain carbocycles recited herein are ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl groups (*i.e.*, groups in which a 3- to 10-membered carbocyclic group (which may be cycloalkyl or aryl) is linked via a single covalent bond or a $C_1$-$C_4$alkylene group). Phenyl groups linked via a single covalent bond or $C_1$-$C_2$alkylene group are designated phenyl$C_0$-$C_4$alkyl (*e.g.*, benzyl, 1-phenyl-ethyl, 1-phenyl-propyl and 2-phenyl-ethyl). When substituted, it will be apparent that such groups may be substituted on the ring portion and/or on the alkylene portion of the group. A "phenyl$C_0$-$C_4$alkoxy" group is a phenyl ring that is linked via an oxygen atom (*i.e.*, phenoxy) or via a $C_1$-$C_4$alkoxy group (*e.g.*, benzyloxy).

A "heterocycle" (also referred to herein as a "heterocyclic group") has from 1 to 3 fused, pendant or spiro rings, at least one of which is a heterocyclic ring (*i.e.*, one or more ring atoms is a heteroatom independently chosen from oxygen, sulfur and nitrogen, with the remaining ring atoms being carbon). Typically, a heterocyclic ring comprises 1, 2, 3 or 4 heteroatoms; within certain

embodiments each heterocyclic ring has 1 or 2 heteroatoms per ring. Each heterocyclic ring generally contains from 3 to 8 ring members (rings having from 4 or 5 to 7 ring members are recited in certain embodiments) and heterocycles comprising fused, pendant or spiro rings typically contain from 9 to 14 ring members. Certain heterocycles comprise a sulfur atom as a ring member; in certain embodiments, the sulfur atom is oxidized to SO or $SO_2$. Heterocycles may be optionally substituted with a variety of substituents, as indicated. Certain heterocycles are 4- to 10-membered and comprise one or two rings; in certain embodiments, such heterocycles are monocyclic (*e.g.*, 4- to 8-membered, 5- to 8-membered, 4- to 7-membered, or 5- or 6-membered).

Certain heterocycles are heteroaryl groups (*i.e.*, at least one heterocyclic ring within the group is aromatic), such as a 5- to 10-membered heteroaryl (which may be monocyclic or bicyclic) or a 5- or 6-membered heteroaryl (*e.g.*, thienyl, imidazolyl, pyridyl or pyrimidyl). Other heterocycles are heterocycloalkyl groups (*i.e.*, do not comprise an aromatic heterocyclic ring). Certain heterocycles may be linked by a single covalent bond or via an alkylene group, as indicated, for example, by the term "(4- to 8-membered heterocycloalkyl)$C_0$-$C_4$alkyl."

A "substituent," as used herein, refers to a molecular moiety that is covalently bonded to an atom within a molecule of interest. For example, a "ring substituent" may be a moiety such as a halogen, alkyl group, haloalkyl group or other group discussed herein that is covalently bonded to an atom (such as a carbon or nitrogen atom) that is a ring member. The term "substitution" refers to replacing a hydrogen atom in a molecular structure with a substituent as described above, such that the valence on the designated atom is not exceeded, and such that a chemically stable compound *(i.e.*, a compound that can be isolated, characterized, and tested for biological activity) results from the substitution.

Groups that are "optionally substituted" are unsubstituted or are substituted by other than hydrogen at one or more available positions, typically 1, 2, 3, 4 or 5 positions, by one or more suitable groups (which may be the same or different). Optional substitution is also indicated by the phrase "substituted with from 0 to X substituents," where X is the maximum number of possible substituents. Certain optionally substituted groups are substituted with from 0 to 2, 3 or 4 independently selected substituents (*i.e.*, are unsubstituted or substituted with up to the recited maximum number of substituents).

"$B_1$," as used herein, refers to the human $B_1$ bradykinin receptor reported by Menke et al. (1994) *J. Biol. Chem. 269*:21583-21586, as well as allelic variants thereof and homologues thereof found in other species (*e.g.*, GenBank Accession Number AAX14712 (Macaca fascicularis)).

The term "$B_1$ agonist" refers to a compound that binds $B_1$ and induces signal transduction mediated by $B_1$. $B_1$ agonists include, for example, bradykinin and kallidin (lysyl-bradykinin), as well as peptide portions or variants of bradykinin or kallidin that bind $B_1$ and retain activity. Representative $B_1$ agonists include, but are not limited to, desArg$^9$bradykinin and desArg$^{10}$kallidin.

A "$B_1$ antagonist" is a compound that detectably inhibits signal transduction mediated by $B_1$. Such inhibition may be determined using the representative calcium mobilization assay provided in Example 7. Preferred $B_1$ antagonists have an $IC_{50}$ of 5 μM or less in this assay, more preferably 2 μM or less, and still more preferably 1 μM or less, 500 nM or less, 100 nM or less or 10 nM or less. In certain embodiments, the $B_1$ antagonist is specific for $B_1$ (*i.e.*, the $IC_{50}$ value in a similar assay performed using the $B_2$ receptor is greater than 2 μM and/or the $IC_{50}$ ratio ($B_2/B_1$) is at least 10, preferably 100, and more preferably at least 1000). $B_1$ antagonists preferably have minimal agonist activity (*i.e.*, induce an increase in the basal activity of $B_1$ that is less than 5% of the increase that would be induced by one $EC_{50}$ of the peptide agonist desArg$^{10}$kallidin, and more preferably have no detectable agonist activity within the assay described in Example 7). $B_1$ antagonists for use as described herein are generally non-toxic. $B_1$ antagonists include neutral antagonists and inverse agonists.

A "neutral antagonist" of $B_1$ is a compound that inhibits the activity of $B_1$ agonist (*e.g.*, desArg$^{10}$kallidin) at $B_1$, but does not significantly change the basal activity of the receptor (*i.e.*, within a calcium mobilization assay as described in Example 7 performed in the absence of agonist, $B_1$ activity is reduced by no more than 10%, more preferably by no more than 5%, and even more preferably by no more than 2%; most preferably, there is no detectable reduction in activity). Neutral antagonists may, but need not, also inhibit the binding of agonist to $B_1$.

An "inverse agonist" of $B_1$ is a compound that reduces the activity of $B_1$ below its basal activity level in the absence of activating concentrations of agonist. Inverse agonists may also inhibit the activity of agonist at $B_1$, and/or may inhibit binding of $B_1$ agonist to $B_1$. The reduction in basal activity of $B_1$ produced by an inverse agonist may be determined from a calcium mobilization assay, such as the assay of Example 7.

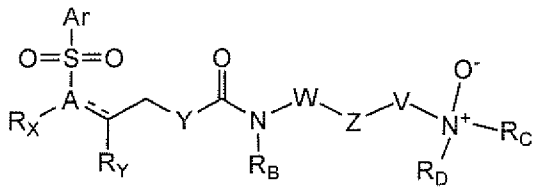
A "therapeutically effective amount" (or dose) is an amount that, upon administration to a patient, results in a discernible patient benefit (*e.g.*, provides detectable relief from a condition being treated). Such relief may be detected using any appropriate criteria. A therapeutically effective amount or dose generally results in a concentration of compound in a body fluid (such as blood, plasma, serum, CSF, synovial fluid, lymph, cellular interstitial fluid, tears or urine) that is sufficient to result in detectable alteration in $B_1$-mediated signal transduction (using an assay provided herein). The discernible patient benefit may be apparent after administration of a single dose, or may become apparent following repeated administration of the therapeutically effective dose according to a predetermined regimen, depending upon the indication for which the compound is administered. For the treatment of pain, a discernible patient benefit is generally apparent after administration of a single therapeutically effective dose, although further benefit may become apparent following repeated administrations.

A "patient" is any individual treated with an N-oxide aryl sulfone or sulfoxide as provided herein. Patients include humans, as well as other animals such as companion animals (*e.g.*, dogs and

cats) and livestock. Patients may be experiencing one or more symptoms of a condition responsive to $B_1$ modulation or may be free of such symptom(s) (*i.e.*, treatment may be prophylactic in a patient considered to be at risk for the development of such symptoms).

N-OXIDE ARYL SULFONES AND SULFOXIDES

As noted above, the present invention provides N-oxide aryl sulfones and sulfoxides of Formula I that may be used in a variety of contexts, including in the treatment of conditions responsive to $B_1$ modulation, as described herein.

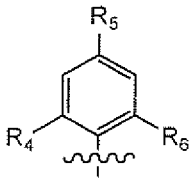


Formula I

Such compounds may also be used within *in vitro* assays (*e.g.*, assays for $B_1$ activity), as probes for detection and localization of $B_1$ and within assays to identify other $B_1$ antagonists.

Within certain N-oxide aryl sulfones and sulfoxides of Formula I, Ar is phenyl, naphthyl, pyridyl or pyrimidinyl, each of which is substituted with from 0 to 4 substituents independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl. Within other N-oxide aryl sulfones and sulfoxides of Formula I, Ar is phenyl that is substituted with from 0 to 4 substituents independently chosen from $R_1$ (*e.g.*, substituted with 1, 2 or 3 substituents independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl). Within further N-oxide aryl sulfones and sulfoxides of Formula I, each substituent (*e.g.*, 1, 2 or 3 substituents) represented by $R_1$ is independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, $C_1$-$C_6$haloalkoxy and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl.

Within certain N-oxide aryl sulfones and sulfoxides, Ar is a group of the formula:



wherein $R_4$, $R_5$ and $R_6$ are independently chosen from hydrogen, halogen, hydroxy, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy and $C_1$-$C_6$haloalkoxy. Within certain such compounds, at least one of $R_4$, $R_5$ and $R_6$ is not hydrogen; within further such compounds, at least two of $R_4$, $R_5$ and $R_6$ are not hydrogen; within still further such compounds, $R_4$, $R_5$ and $R_6$ are independently chosen from halogen, hydroxy, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy and $C_1$-$C_6$haloalkoxy (*e.g.*, $R_5$ is methyl or methoxy and $R_4$ and $R_6$ are each methyl).

Within the variable designated "Y," the sum of r and p ranges from 1 to 6 for certain N-oxide aryl sulfones and sulfoxides. Within certain such compounds, p is not zero. Certain Y groups satisfy

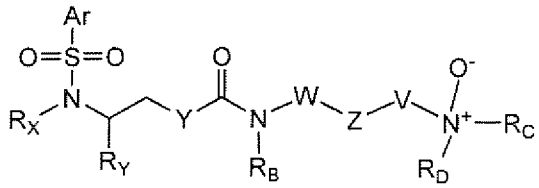
the formula $(CH_2)_r$-O-$(CH_2)_p$, which is substituted with from 0 to 4 substituents independently chosen from amino, hydroxy, cyano, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl and substituents of the same carbon atom or adjacent carbon atoms that are taken together to form $C_3$-$C_6$cycloalkyl. Representative such Y groups include, for example, –O-$CH_2$–, –$CH_2$-O-$CH_2$–, –O-$CH_2$-$CH_2$–, –$CH_2$-O-$CH_2$-$CH_2$– and –$CH_2$-$CH_2$-O-$CH_2$–. Other Y groups satisfy the formula –N($R_{10}$)-$CH_2$–. Yet another Y group is –CH=CH–.

Within other N-oxide aryl sulfones and sulfoxides of Formula I, Y is a group of the formula $(CH_2)_r$-$(CH_2)_p$ that is substituted with from 0 to 4 substituents independently chosen from amino, hydroxy, cyano, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl and substituents of the same carbon atom or adjacent carbon atoms that are taken together to form $C_3$-$C_6$cycloalkyl. Representative such Y groups include, for example, –$CH_2$–, –$CH_2$-$CH_2$–, and –$CH_2$-$CH_2$-$CH_2$–, each of which is optionally substituted as indicated above.

The variable A is N, C, or CH. It will be apparent that if A is N or CH, then ==== represents a single bond. If ==== represents a double bond, then A is carbon.

The variables $R_X$ and $R_Y$ of Formula I are, within certain embodiments, independently chosen from hydrogen and $C_1$-$C_6$alkyl. Within other embodiments, $R_X$ and $R_Y$ are taken together to form an aromatic group. Representative such aromatic groups include, for example, naphthyl, quinolinyl, phenyl and pyridyl, each of which is optionally substituted as described above. Within further embodiments, $R_X$ and $R_Y$ are taken together to form an optionally substituted partially or fully saturated ring. Representative such rings include, for example, piperazine, morpholine, thiomorpholine, piperidine and pyrrolidine.

Certain compounds of Formula I further satisfy Formula II:



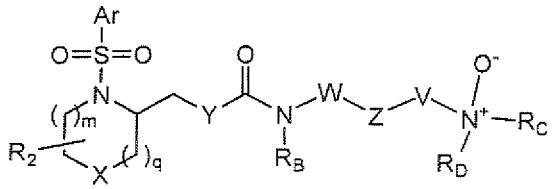

Formula II

wherein $R_X$ and $R_Y$ are:

(i) independently chosen from: (a) hydrogen; and (b) $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl, and ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, each of which is substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, amino and $C_1$-$C_4$alkyl; or

(ii) taken together to form a 4- to 10-membered heterocycle that is substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, halogen, amino and $C_1$-$C_8$alkyl;

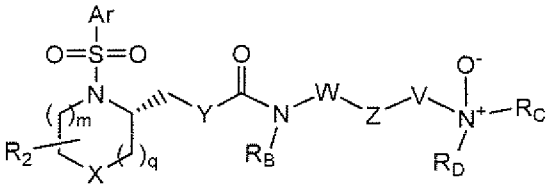
and the remaining variables are as described for Formula I. Within certain such compounds, $R_X$ and $R_Y$ are taken together to form an optionally substituted 4- to 8-membered heterocycloalkyl. Representative such compounds further satisfy Formula III:



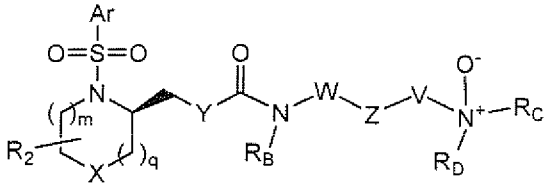
Formula III

wherein: X is $CH_2$, $NR_3$, O, S, SO or $SO_2$; m and q are independently 0, 1 or 2; $R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_6$alkyl; $R_3$ is hydrogen, $C_1$-$C_6$alkyl or $C_1$-$C_6$alkanoyl; and the remaining variables are as described for Formula I.

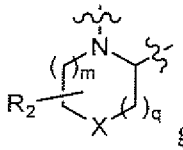
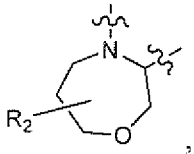
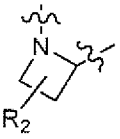
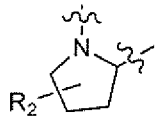
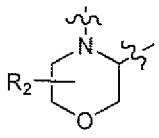
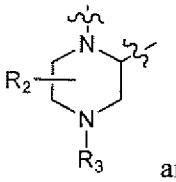
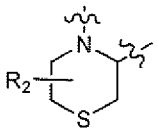
Certain N-oxide aryl sulfones and sulfoxides of Formula III further satisfy Formula IV or Formula V:



Formula IV

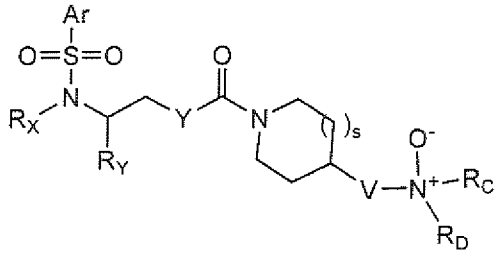


Formula V

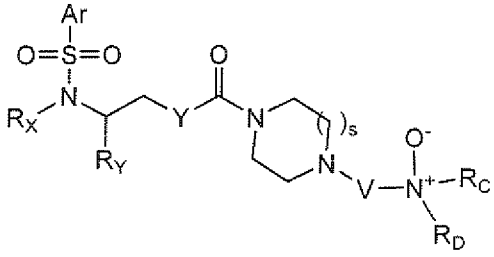
Within Formulas III-V, X is $CH_2$ in certain compounds; for other compounds X is O. Representative  groups include, for example, , , , ,  and . Within certain compounds, $R_2$ represents from 0 to 4 substituents independently chosen from $C_1$-$C_2$alkyl (*e.g.*, gem-dimethyl).

The variable $R_C$ is absent in certain embodiments. It will be apparent that $R_C$ may only be absent if $R_D$ forms a heteroaryl with $R_F$ or a substituent of V (and any heteroaryl so formed comprises V and the tetravalent nitrogen atom as ring members).

The variable "Z" is $NR_G$ or $CR_ER_F$ within certain embodiments of Formulas I-V. It will be apparent that $R_E$, $R_F$ and $R_G$ are intended to be substituents of the base carbon or nitrogen atom (*i.e.*, $NR_G$ is  and $CR_ER_F$ is ). Within certain such compounds, $R_E$ or $R_G$ is taken together with $R_B$ to form a 4- to 7-membered heterocycloalkyl that is substituted with from 0 to 4 substituents independently chosen from: (i) hydroxy, oxo, cyano and amino; and (ii) $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl; each of which is substituted with from 0 to 2 substituents independently chosen from hydroxy, halogen, oxo, cyano, $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl. Representative such compounds satisfy Formula VI or Formula VII, in which variables are as described above:

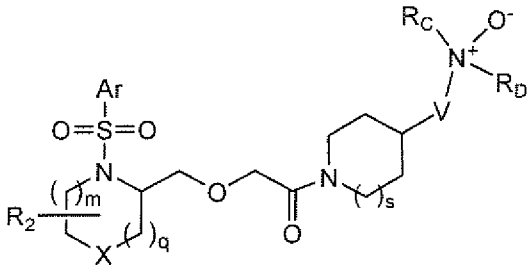


Formula VI

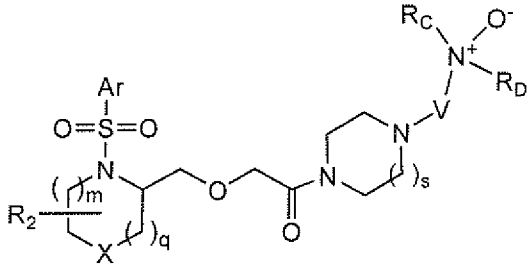


Formula VII

Within certain compounds of Formula VI, s is 0, 1 or 2; $R_D$ is $C_1$-$C_6$alkyl; V is absent or $C_1$-$C_4$alkylene; and the remaining variables are as described above. Within certain compounds of Formula VII, s is 0, 1 or 2; $R_D$ is $C_1$-$C_6$alkyl; V is $C_1$-$C_4$alkylene; and the remaining variables are as described above. Within certain compounds of Formula VI or Formula VII, $R_X$ and $R_Y$ are independently hydrogen or $C_1$-$C_6$alkyl. Other compounds of Formula VI or Formula VII further satisfy Formula VIII or Formula IX, respectively:



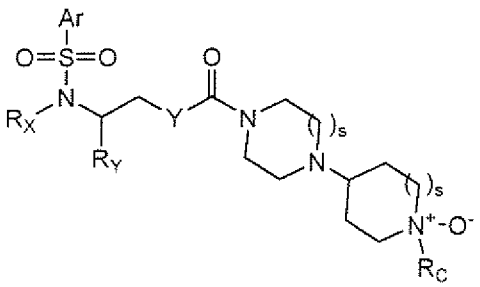
Formula VIII



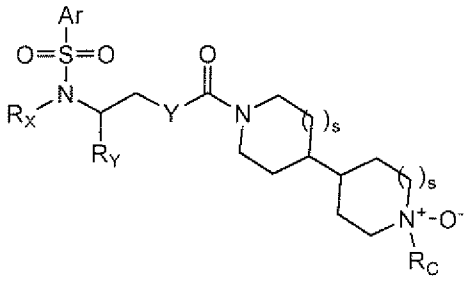
Formula IX

in which m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3; X is $NR_3$, O, S, SO or $SO_2$; $R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_6$alkyl; and the remaining variables are as described for Formula VI.

Still further N-oxide aryl sulfones and sulfoxides provided herein satisfy Formula X or Formula XI:

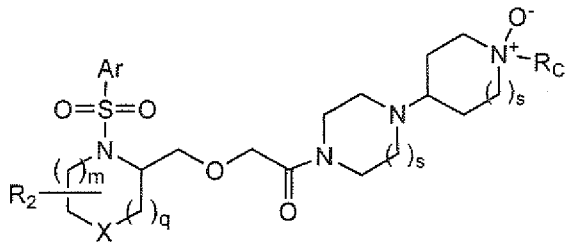


Formula X

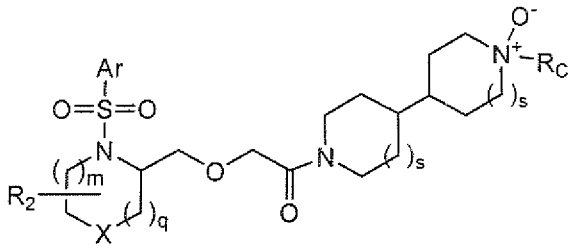


Formula XI

in which each s is independently 0, 1 or 2. Within certain compounds of Formula X or Formula XI, $R_X$ and $R_Y$ are independently hydrogen or $C_1$-$C_6$alkyl. Other compounds of Formula X or Formula XI further satisfy Formula XII or Formula XIII, respectively:



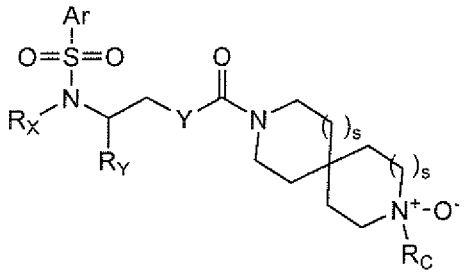
Formula XII



Formula XIII

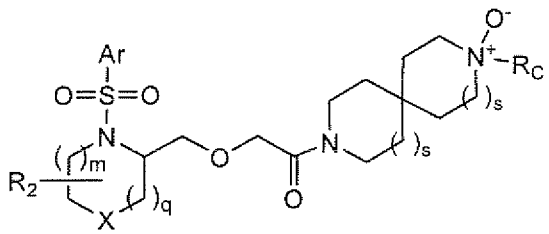
in which m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3; X is $NR_3$, O, S, SO or $SO_2$; $R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_6$alkyl; and the remaining variables are as described above.

Within further embodiments of Formulas I-V, the variable "Z" is $CR_ER_F$, and $R_F$ is taken together with $R_D$ to form a 4- to 7-membered heterocycloalkyl that is substituted with from 0 to 4 substituents independently chosen from: (i) hydroxy, oxo, cyano and amino; and (ii) $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl; each of which is substituted with from 0 to 2 substituents independently chosen from hydroxy, halogen, oxo, cyano, $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl. Representative such compounds satisfy Formula XIV:



Formula XIV

in which s is 0, 1 or 2, and the remaining variables are as described above. Within certain compounds of Formula XIV, $R_X$ and $R_Y$ are independently hydrogen or $C_1$-$C_6$alkyl. Other compounds of Formula XIV further satisfy Formula XV:



Formula XV

in which m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3; X is $NR_3$, O, S, SO or $SO_2$; $R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_8$alkyl; and the remaining variables are as described above.

Within still further embodiments of Formulas I-V, the variable "Z" is $NR_G$ and $R_G$ is hydrogen or $C_1$-$C_6$alkyl.

Representative N-oxide aryl sulfones and sulfoxides provided herein include, but are not limited to, those specifically described in the Examples below. It will be apparent that the specific

compounds recited herein are representative only, and are not intended to limit the scope of the present invention. Further, as noted above, all compounds of the present invention may be present as a free acid or base or as a pharmaceutically acceptable salt, hydrate or ester.

As noted above, within certain aspects, compounds provided herein are $B_1$ modulators. In addition, certain compounds provided herein are specific for $B_1$. $B_1$ modulator activity may be confirmed using a calcium mobilization assay, such as the assay described in Example 7, herein.

If desired, binding activity of the compounds provided herein to $B_1$ may be confirmed using the representative assay described in Example 6, herein, or using an assay described by Fox et al. (2005) *Br. J. Pharmacol. 144*:889-99. Preferred $B_1$ modulators exhibit a $K_i$ within such an assay of 5 micromolar or less, more preferably 2 micromolar or less, 1 micromolar or less, 500 nanomolar or less, 100 nanomolar or less or 10 nanomolar or less.

*In vivo* activity of $B_1$ modulators provided herein may be confirmed using any of a variety of animal models including, but not limited to, those described in the following documents (each of which is hereby incorporated by reference for its disclosure of the recited animal model):

Wood et al. (2003) *J. Med. Chem. 46*:1803-06 – carrageenan-induced mechanical pressure hyperalgesia;

Conley et al. (2005) *Eur. J. Pharmacol. 527*:44-51 – thermal antinociception and carrageenan-induced mechanical pressure hypersensitivity;

Gougat et al. (2004) *J. Pharmacol. Exper. Therap. 309*:661-669 – UV irradiation-induced thermal hyperalgesia and chronic constriction of the sciatic nerve-induced neuropathy and in ischemia-induced injury;

Fox et al. (2005) *Br. J. Pharmacol. 144*:889-99 – complete Freund's adjuvant-induced mechanical pressure hypersensitivity;

Gabra et al. (2005) *J. Neuropathol. Exp. Neurol.* 64:782-89 – diabetes-induced peripheral neuropathic pain;

Pesquero et al. (2000) *Proc. Natl. Acad. Sci. USA 97*:8140-45 – carrageenan-induced pleurisy (inflammation around the lung);

Lawson et al. (2005) *Eur. J. Pharmacol. 514*:69-78 – diabetes-induced vascular disease;

Hirata et a (2003) *Eur. J. Pharmacol. 474*:255-60 – ACE inhibitor-induced cough;

Mazzuferi et al. (2005) *Neuroscience. 135*:979-86 – neuronal hyperexcitability in epilepsy.

If desired, compounds provided herein may be evaluated for certain pharmacological properties including, but not limited to, oral bioavailability (preferred compounds are orally bioavailable to an extent allowing for therapeutically effective doses of less than 140 mg/kg, preferably less than 50 mg/kg, more preferably less than 30 mg/kg, even more preferably less than 10 mg/kg, still more preferably less than 1 mg/kg and most preferably less than 0.1 mg/kg), toxicity (a preferred compound is nontoxic when a therapeutically effective amount is administered to a subject), side effects (a preferred compound produces side effects comparable to placebo when a

therapeutically effective amount of the compound is administered to a subject), serum protein binding and *in vitro* and *in vivo* half-life (a preferred compound exhibits an *in vivo* half-life allowing for Q.I.D. dosing, preferably T.I.D. dosing, more preferably B.I.D. dosing, and most preferably once-a-day dosing). In addition, differential penetration of the blood brain barrier may be desirable. Routine assays that are well known in the art may be used to assess these properties, and identify superior compounds for a particular use. For example, assays used to predict bioavailability include transport across human intestinal cell monolayers, including Caco-2 cell monolayers. Penetration of the blood brain barrier of a compound in humans may be predicted from the brain levels of the compound in laboratory animals given the compound (*e.g.*, intravenously). Serum protein binding may be predicted from albumin binding assays. Compound half-life is inversely proportional to the frequency of dosage of a compound. *In vitro* half-lives of compounds may be predicted from assays of microsomal half-life as described herein.

As noted above, preferred compounds provided herein are nontoxic. In general, the term "nontoxic" as used herein shall be understood in a relative sense and is intended to refer to any substance that has been approved by the United States Food and Drug Administration ("FDA") for administration to mammals (preferably humans) or, in keeping with established criteria, is susceptible to approval by the FDA for administration to mammals (preferably humans). In addition, a highly preferred nontoxic compound generally satisfies one or more of the following criteria: (1) does not substantially inhibit cellular ATP production; (2) does not significantly prolong heart QT intervals; (3) does not cause substantial liver enlargement, or (4) does not cause substantial release of liver enzymes.

As used herein, a compound that does not substantially inhibit cellular ATP production is a compound that satisfies the criteria set forth in Example 8, herein. In other words, cells treated as described in Example 8 with 100 μM of such a compound exhibit ATP levels that are at least 50% of the ATP levels detected in untreated cells. In more highly preferred embodiments, such cells exhibit ATP levels that are at least 80% of the ATP levels detected in untreated cells.

A compound that does not significantly prolong heart QT intervals is a compound that does not result in a statistically significant prolongation of heart QT intervals (as determined by electrocardiography) in guinea pigs, minipigs or dogs upon administration of a dose that yields a serum concentration equal to the $EC_{50}$ or $IC_{50}$ for the compound. In certain preferred embodiments, a dose of 0.01, 0.05, 0.1, 0.5, 1, 5, 10, 40 or 50 mg/kg administered parenterally or orally does not result in a statistically significant prolongation of heart QT intervals. By "statistically significant" is meant results varying from control at the $p<0.1$ level or more preferably at the $p<0.05$ level of significance as measured using a standard parametric assay of statistical significance such as a student's T test.

A compound does not cause substantial liver enlargement if daily treatment of laboratory rodents (*e.g.*, mice or rats) for 5-10 days with a dose that yields a serum concentration equal to the $EC_{50}$ or $IC_{50}$ for the compound results in an increase in liver to body weight ratio that is no more than

100% over matched controls. In more highly preferred embodiments, such doses do not cause liver enlargement of more than 75% or 50% over matched controls. If non-rodent mammals (*e.g.*, dogs) are used, such doses should not result in an increase of liver to body weight ratio of more than 50%, preferably not more than 25%, and more preferably not more than 10% over matched untreated controls. Preferred doses within such assays include 0.01, 0.05. 0.1, 0.5, 1, 5, 10, 40 or 50 mg/kg administered parenterally or orally.

Similarly, a compound does not promote substantial release of liver enzymes if administration of twice the minimum dose that yields a serum concentration equal to the $EC_{50}$ or $IC_{50}$ for the compound does not elevate serum levels of ALT, LDH or AST in laboratory rodents by more than 100% over matched mock-treated controls. In more highly preferred embodiments, such doses do not elevate such serum levels by more than 75% or 50% over matched controls. Alternatively, a compound does not promote substantial release of liver enzymes if, in an *in vitro* hepatocyte assay, concentrations (in culture media or other such solutions that are contacted and incubated with hepatocytes *in vitro*) that are equal to the $EC_{50}$ or $IC_{50}$ for the compound do not cause detectable release of any of such liver enzymes into culture medium above baseline levels seen in media from matched mock-treated control cells. In more highly preferred embodiments, there is no detectable release of any of such liver enzymes into culture medium above baseline levels when such compound concentrations are five-fold, and preferably ten-fold the $EC_{50}$ or $IC_{50}$ for the compound.

In other embodiments, certain preferred compounds do not inhibit or induce microsomal cytochrome P450 enzyme activities, such as CYP1A2 activity, CYP2A6 activity, CYP2C9 activity, CYP2C19 activity, CYP2D6 activity, CYP2E1 activity or CYP3A4 activity at a concentration equal to the $EC_{50}$ or $IC_{50}$ for the compound.

Certain preferred compounds are not clastogenic (*e.g.*, as determined using a mouse erythrocyte precursor cell micronucleus assay, an Ames micronucleus assay, a spiral micronucleus assay or the like) at a concentration equal the $EC_{50}$ or $IC_{50}$ for the compound. In other embodiments, certain preferred compounds do not induce sister chromatid exchange (*e.g.*, in Chinese hamster ovary cells) at such concentrations.

For detection purposes, as discussed in more detail below, compounds provided herein may be isotopically-labeled or radiolabeled. For example, such compounds may have one or more atoms replaced by an atom of the same element having an atomic mass or mass number different from the atomic mass or mass number usually found in nature. Examples of isotopes that can be present in the compounds provided herein include isotopes of hydrogen, carbon, nitrogen, oxygen, phosphorous, fluorine and chlorine, such as $^{2}H$, $^{3}H$, $^{11}C$, $^{13}C$, $^{14}C$, $^{15}N$, $^{18}O$, $^{17}O$, $^{31}P$, $^{32}P$, $^{35}S$, $^{18}F$ and $^{36}Cl$. In addition, substitution with heavy isotopes such as deuterium (*i.e.*, $^{2}H$) can afford certain therapeutic advantages resulting from greater metabolic stability, for example increased in vivo half-life or reduced dosage requirements and, hence, may be preferred in some circumstances.

PREPARATION OF N-OXIDE ARYL SULFONES AND SULFOXIDES

Compounds provided herein may generally be prepared using standard synthetic methods. In general, starting materials are commercially available from suppliers such as Sigma-Aldrich Corp. (St. Louis, MO), or may be synthesized from commercially available precursors using established protocols. By way of example, a synthetic route similar to that shown in any of the following Schemes may be used, together with synthetic methods known in the art of synthetic organic chemistry, or variations thereon appreciated by those skilled in the art. It will be apparent that the reagents and synthetic transformations in the following Schemes and in the art can be readily modified to produce additional compounds of Formula I. Each variable in the following Schemes refers to any group consistent with the description of the compounds provided herein.

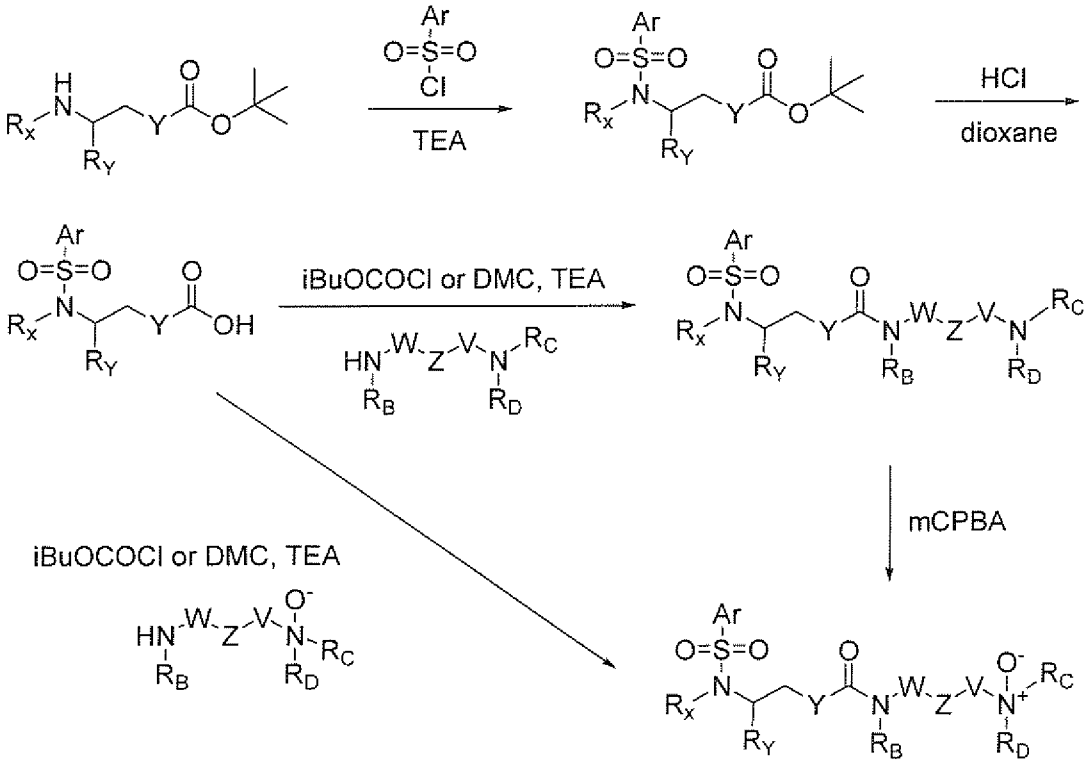
When a protecting group is required, an optional deprotection step may be employed. Suitable protecting groups and methodology for protection and deprotection, such as those described in *Protecting Groups in Organic Synthesis* by T. Greene, are well known. Compounds and intermediates requiring protection/deprotection will be readily apparent.

Certain abbreviations used in the following Schemes and in the Examples include:

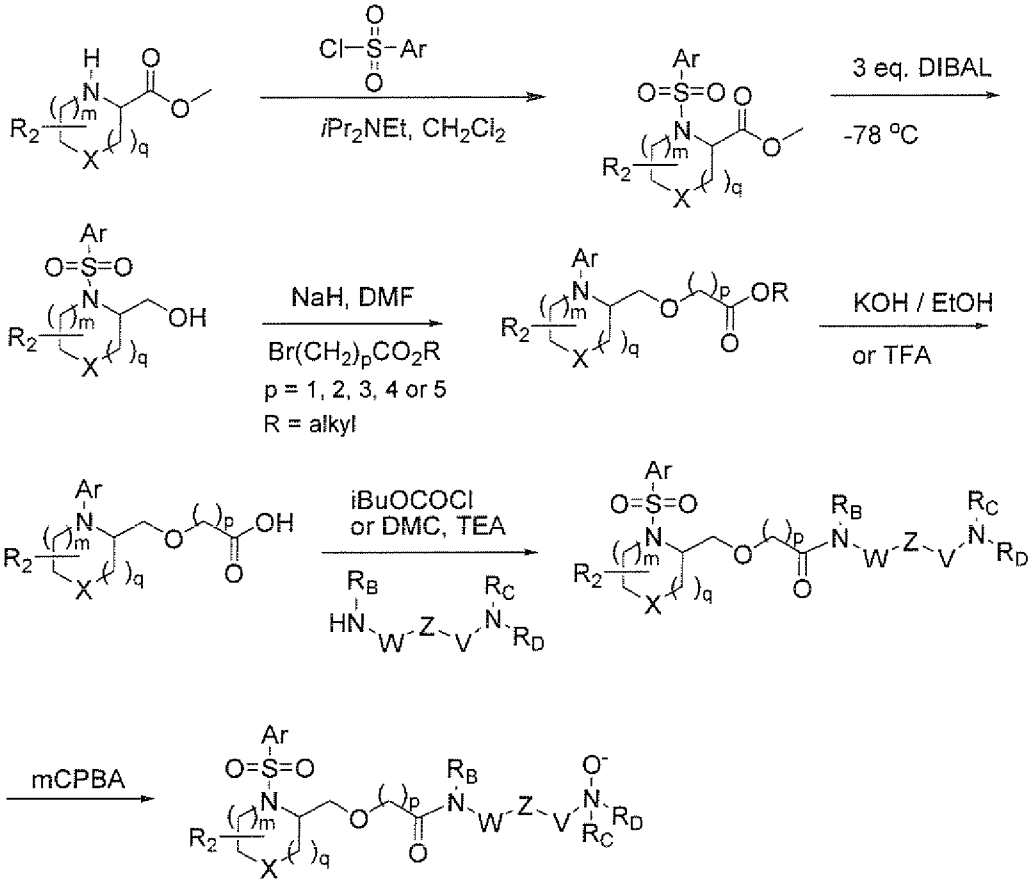
| | |
|---|---|
| AIBN | 2,2'-Azobis(2-methylpropionitrile) |
| DIBAL | diisobutylaluminium hydride |
| DMA | *N,N*-dimethylacetamide |
| DMAP | *N,N*-dimethyl-4-aminopyridine |
| DMC | 2-chloro-1,3-dimethylimidazolidinium chloride |
| DMF | dimethylformamide |
| DMSO | dimethylsulfoxide |
| Eq. | equivalent(s) |
| Et | ethyl |
| EtOH | ethanol |
| EtOAc | ethyl acetate |
| h | hour(s) |
| $^1$H NMR | proton nuclear magnetic resonance |
| LC-MS | liquid chromatography/mass spectrometry |
| mCPBA | m-chloroperoxybenzoic acid |
| MeOH | methanol |
| MHz | megahertz |
| M+1 | mass + 1 |
| min | minute(s) |
| MS | mass spectrometry |
| NMO | N-methylmorpholine N-oxide |
| δ | chemical shift |

| | |
|---|---|
| rt | room temperature |
| Ph | phenyl |
| SCX | strong cation exchange |
| TEA | triethylamine |
| TFA | trifluoroacetic acid |
| THF | tetrahydrofuran |
| TPAP | tetra-n-propylammonium perruthenate |

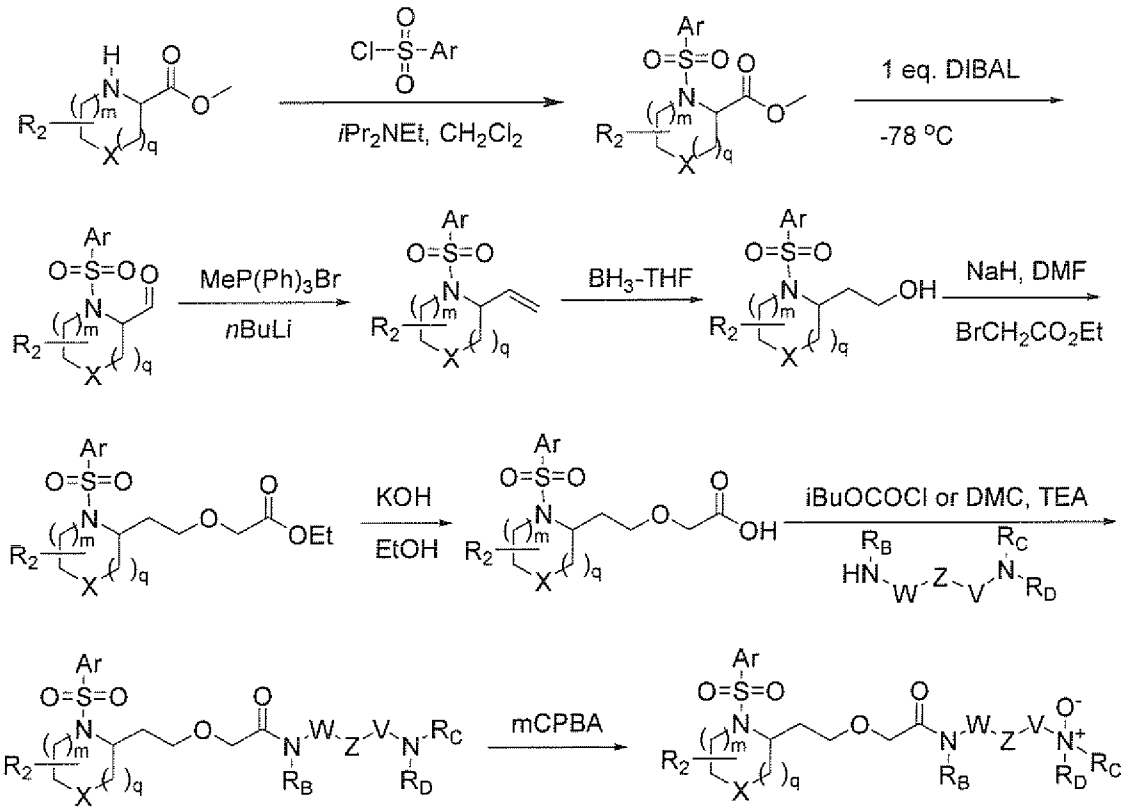
Scheme 1



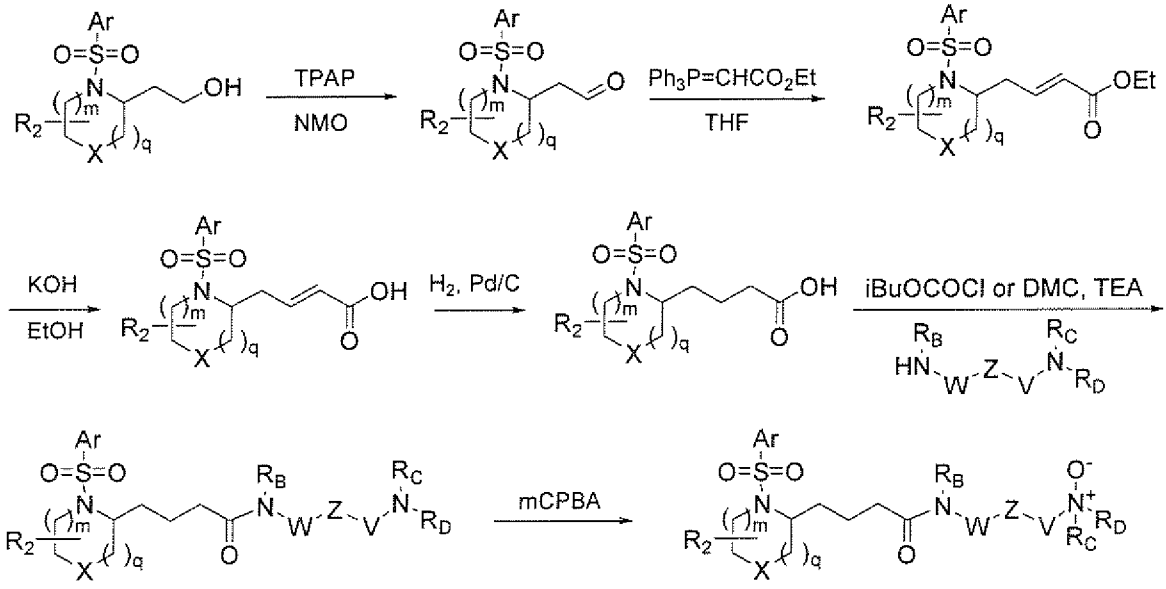
Scheme 2


Cl—S—Ar
iPr2NEt, CH2Cl2
3 eq. DIBAL
-78 °C
NaH, DMF
Br(CH2)pCO2R
p = 1, 2, 3, 4 or 5
R = alkyl
KOH / EtOH
or TFA
iBuOCOCl
or DMC, TEA
mCPBA

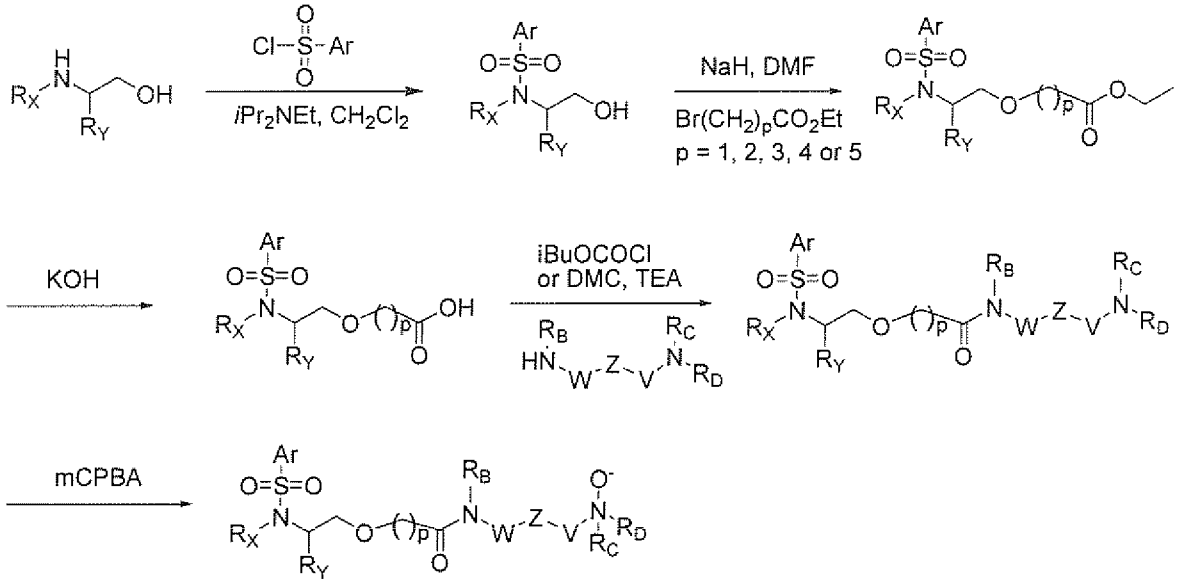
Scheme 3



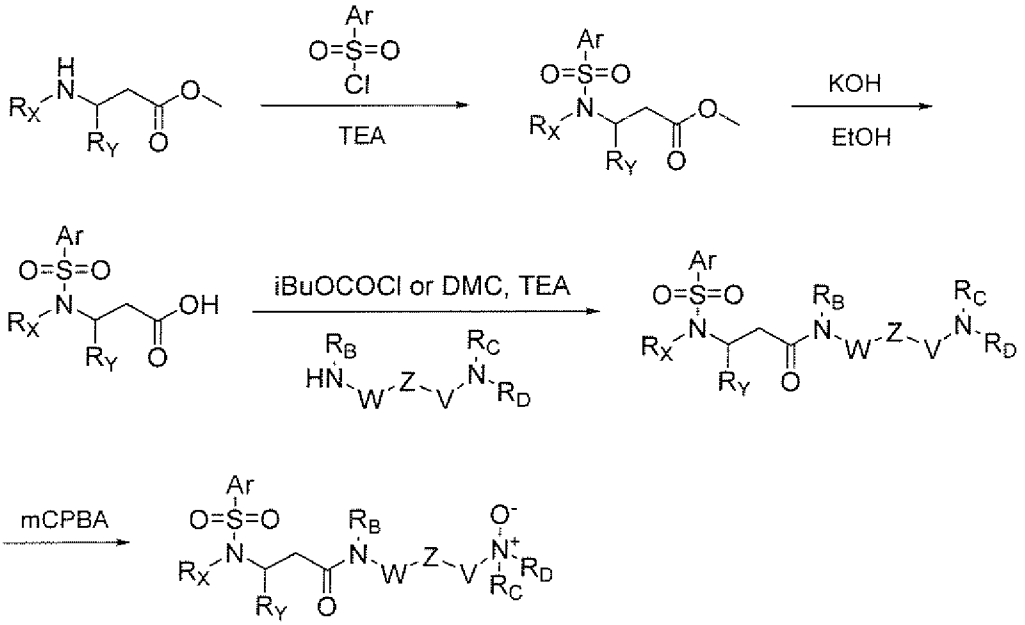

Scheme 4



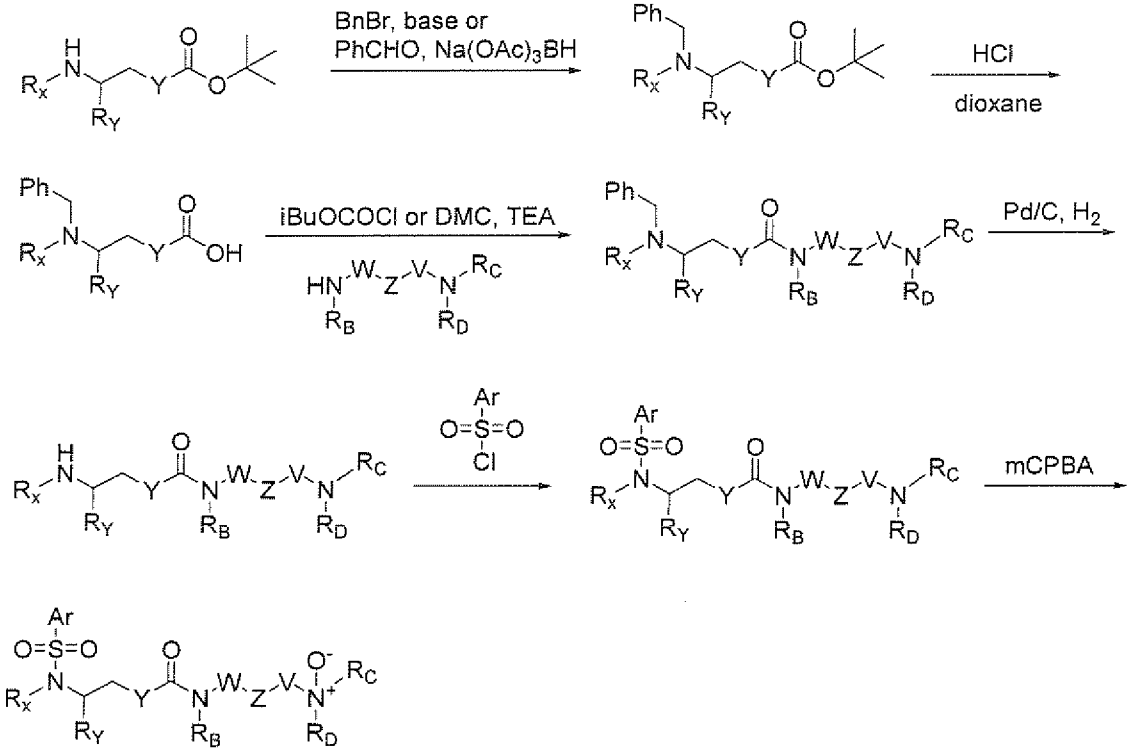
Scheme 5




Scheme 6



Scheme 7




In certain embodiments, a compound provided herein may contain one or more asymmetric carbon atoms, so that the compound can exist in different stereoisomeric forms. Such forms can be, for example, racemates or optically active forms. As noted above, all stereoisomers are encompassed by the present invention. Nonetheless, it may be desirable to obtain single enantiomers (*i.e.*, optically active forms). Standard methods for preparing single enantiomers include asymmetric synthesis and resolution of the racemates. Resolution of the racemates can be accomplished, for example, by conventional methods such as crystallization in the presence of a resolving agent, or chromatography using, for example a chiral HPLC column.

Compounds may be radiolabeled by carrying out their synthesis using precursors comprising at least one atom that is a radioisotope. Each radioisotope is preferably carbon (*e.g.*, $^{14}C$), hydrogen (*e.g.*, $^{3}H$), sulfur (*e.g.*, $^{35}S$) or iodine (*e.g.*, $^{125}I$). Tritium labeled compounds may also be prepared catalytically via platinum-catalyzed exchange in tritiated acetic acid, acid-catalyzed exchange in tritiated trifluoroacetic acid, or heterogeneous-catalyzed exchange with tritium gas using the compound as substrate. In addition, certain precursors may be subjected to tritium-halogen exchange with tritium gas, tritium gas reduction of unsaturated bonds, or reduction using sodium borotritide, as appropriate. Preparation of radiolabeled compounds may be conveniently performed by a radioisotope supplier specializing in custom synthesis of radiolabeled probe compounds.

## PHARMACEUTICAL COMPOSITIONS

The present invention also provides pharmaceutical compositions comprising one or more N-oxide aryl sulfones and sulfoxides provided herein, together with at least one physiologically

acceptable carrier or excipient. Pharmaceutical compositions may comprise, for example, one or more of water, buffers (*e.g.,* neutral buffered saline or phosphate buffered saline), ethanol, mineral oil, vegetable oil, dimethylsulfoxide, carbohydrates (*e.g.,* glucose, mannose, sucrose or dextrans), mannitol, proteins, adjuvants, polypeptides or amino acids such as glycine, antioxidants, chelating agents such as EDTA or glutathione and/or preservatives. In addition, other active ingredients may (but need not) be included in the pharmaceutical compositions provided herein.

Pharmaceutical compositions may be formulated for any appropriate manner of administration, including, for example, topical, oral (including, but not limited to, sublingual), nasal, rectal or parenteral administration. The term parenteral as used herein includes subcutaneous, intradermal, intravascular (*e.g.,* intravenous), intramuscular, spinal, intracranial, intrathecal and intraperitoneal injection, as well as any similar injection or infusion technique. In certain embodiments, compositions suitable for oral use are preferred. Such compositions include, for example, tablets, troches, lozenges, aqueous or oily suspensions, dispersible powders or granules, emulsion, hard or soft capsules, or syrups or elixirs. Within yet other embodiments, compositions of the present invention may be formulated as a lyophilizate.

Compositions intended for oral use may further comprise one or more components such as sweetening agents, flavoring agents, coloring agents and/or preserving agents in order to provide appealing and palatable preparations. Tablets contain the active ingredient in admixture with physiologically acceptable excipients that are suitable for the manufacture of tablets. Such excipients include, for example, inert diluents (*e.g.,* calcium carbonate, sodium carbonate, lactose, calcium phosphate or sodium phosphate), granulating and disintegrating agents (*e.g.,* corn starch or alginic acid), binding agents (*e.g.,* starch, gelatin or acacia) and lubricating agents (*e.g.,* magnesium stearate, stearic acid or talc). The tablets may be uncoated or they may be coated by known techniques to delay disintegration and absorption in the gastrointestinal tract and thereby provide a sustained action over a longer period. For example, a time delay material such as glyceryl monosterate or glyceryl distearate may be employed.

Formulations for oral use may also be presented as hard gelatin capsules wherein the active ingredient is mixed with an inert solid diluent (*e.g.,* calcium carbonate, calcium phosphate or kaolin), or as soft gelatin capsules wherein the active ingredient is mixed with water or an oil medium (*e.g.,* peanut oil, liquid paraffin or olive oil).

Aqueous suspensions contain the active material(s) in admixture with excipients suitable for the manufacture of aqueous suspensions. Such excipients include suspending agents (*e.g.,* sodium carboxymethylcellulose, methylcellulose, hydropropylmethylcellulose, sodium alginate, polyvinylpyrrolidone, gum tragacanth and gum acacia); and dispersing or wetting agents (*e.g.,* naturally-occurring phosphatides such as lecithin, condensation products of an alkylene oxide with fatty acids such as polyoxyethylene stearate, condensation products of ethylene oxide with long chain aliphatic alcohols such as heptadecaethyleneoxycetanol, condensation products of ethylene oxide with

partial esters derived from fatty acids and a hexitol such as polyoxyethylene sorbitol monooleate, or condensation products of ethylene oxide with partial esters derived from fatty acids and hexitol anhydrides such as polyethylene sorbitan monooleate). Aqueous suspensions may also comprise one or more preservatives, such as ethyl or n-propyl p-hydroxybenzoate, one or more coloring agents, one or more flavoring agents, and one or more sweetening agents, such as sucrose or saccharin.

Oily suspensions may be formulated by suspending the active ingredient(s) in a vegetable oil (*e.g.*, arachis oil, olive oil, sesame oil or coconut oil) or in a mineral oil such as liquid paraffin. The oily suspensions may contain a thickening agent such as beeswax, hard paraffin or cetyl alcohol. Sweetening agents such as those set forth above, and/or flavoring agents may be added to provide palatable oral preparations. Such suspensions may be preserved by the addition of an anti-oxidant such as ascorbic acid.

Dispersible powders and granules suitable for preparation of an aqueous suspension by the addition of water provide the active ingredient in admixture with a dispersing or wetting agent, suspending agent and one or more preservatives. Suitable dispersing or wetting agents and suspending agents are exemplified by those already mentioned above. Additional excipients, such as sweetening, flavoring and coloring agents, may also be present.

Pharmaceutical compositions may also be formulated as oil-in-water emulsions. The oily phase may be a vegetable oil (*e.g.*, olive oil or arachis oil), a mineral oil (*e.g.*, liquid paraffin) or a mixture thereof. Suitable emulsifying agents include naturally-occurring gums (*e.g.*, gum acacia or gum tragacanth), naturally-occurring phosphatides (*e.g.*, soy bean lecithin, and esters or partial esters derived from fatty acids and hexitol), anhydrides (*e.g.*, sorbitan monoleate) and condensation products of partial esters derived from fatty acids and hexitol with ethylene oxide (*e.g.*, polyoxyethylene sorbitan monoleate). An emulsion may also comprise one or more sweetening and/or flavoring agents.

Syrups and elixirs may be formulated with sweetening agents, such as glycerol, propylene glycol, sorbitol or sucrose. Such formulations may also comprise one or more demulcents, preservatives, flavoring agents and/or coloring agents.

Formulations for topical administration typically comprise a topical vehicle combined with active agent(s), with or without additional optional components. Suitable topical vehicles and additional components are well known in the art, and it will be apparent that the choice of a vehicle will depend on the particular physical form and mode of delivery. Topical vehicles include water; organic solvents such as alcohols (*e.g.*, ethanol or isopropyl alcohol) or glycerin; glycols (*e.g.*, butylene, isoprene or propylene glycol); aliphatic alcohols (*e.g.*, lanolin); mixtures of water and organic solvents and mixtures of organic solvents such as alcohol and glycerin; lipid-based materials such as fatty acids, acylglycerols (including oils, such as mineral oil, and fats of natural or synthetic origin), phosphoglycerides, sphingolipids and waxes; protein-based materials such as collagen and gelatin; silicone-based materials (both non-volatile and volatile); and hydrocarbon-based materials

such as microsponges and polymer matrices. A composition may further include one or more components adapted to improve the stability or effectiveness of the applied formulation, such as stabilizing agents, suspending agents, emulsifying agents, viscosity adjusters, gelling agents, preservatives, antioxidants, skin penetration enhancers, moisturizers and sustained release materials. Examples of such components are described in Martindale--The Extra Pharmacopoeia (Pharmaceutical Press, London 1993) and Martin (ed.), Remington's Pharmaceutical Sciences. Formulations may comprise microcapsules, such as hydroxymethylcellulose or gelatin-microcapsules, liposomes, albumin microspheres, microemulsions, nanoparticles or nanocapsules.

A topical formulation may be prepared in a variety of physical forms including, for example, solids, pastes, creams, foams, lotions, gels, powders, aqueous liquids and emulsions. Typical modes of delivery for topical compositions include application using the fingers; application using a physical applicator such as a cloth, tissue, swab, stick or brush; spraying (including mist, aerosol or foam spraying); dropper application; sprinkling; soaking; and rinsing. Controlled release vehicles can also be used.

A pharmaceutical composition may be prepared as a sterile injectible aqueous or oleaginous suspension. The compound(s) provided herein, depending on the vehicle and concentration used, can either be suspended or dissolved in the vehicle. Such a composition may be formulated according to the known art using suitable dispersing, wetting and/or suspending agents such as those mentioned above. Among the acceptable vehicles and solvents that may be employed are water, 1,3-butanediol, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils may be employed as a solvent or suspending medium. For this purpose any bland fixed oil may be employed, including synthetic mono- or diglycerides. In addition, fatty acids such as oleic acid find use in the preparation of injectible compositions, and adjuvants such as local anesthetics, preservatives and/or buffering agents can be dissolved in the vehicle.

Compounds may also be formulated as suppositories (*e.g.*, for rectal administration). Such compositions can be prepared by mixing the drug with a suitable non-irritating excipient that is solid at ordinary temperatures but liquid at the rectal temperature and will therefore melt in the rectum to release the drug. Suitable excipients include, for example, cocoa butter and polyethylene glycols.

Pharmaceutical compositions may be formulated for release at a pre-determined rate. Instantaneous release may be achieved, for example, via sublingual administration (*i.e.*, administration by mouth in such a way that the active ingredient(s) are rapidly absorbed via the blood vessels under the tongue rather than via the digestive tract). Controlled release formulations (*i.e.*, formulations such as a capsule, tablet or coated tablet that slows and/or delays release of active ingredient(s) following administration) may be administered by, for example, oral, rectal or subcutaneous implantation, or by implantation at a target site. In general, a controlled release formulation comprises a matrix and/or coating that delays disintegration and absorption in the gastrointestinal tract (or implantation site) and thereby provides a delayed action or a sustained action

over a longer period. One type of controlled-release formulation is a sustained-release formulation, in which at least one active ingredient is continuously released over a period of time at a constant rate. Preferably, the therapeutic agent is released at such a rate that blood (*e.g.*, plasma) concentrations are maintained within the therapeutic range, but below toxic levels, over a period of time that is at least 4 hours, preferably at least 8 hours, and more preferably at least 12 hours. Such formulations may generally be prepared using well known technology and administered by, for example, oral, rectal or subcutaneous implantation, or by implantation at the desired target site. Carriers for use within such formulations are biocompatible, and may also be biodegradable; preferably the formulation provides a relatively constant level of modulator release. The amount of modulator contained within a sustained release formulation depends upon, for example, the site of implantation, the rate and expected duration of release and the nature of the condition to be treated or prevented.

Controlled release may be achieved by combining the active ingredient(s) with a matrix material that itself alters release rate and/or through the use of a controlled-release coating. The release rate can be varied using methods well known in the art, including (a) varying the thickness or composition of coating, (b) altering the amount or manner of addition of plasticizer in a coating, (c) including additional ingredients, such as release-modifying agents, (d) altering the composition, particle size or particle shape of the matrix, and (e) providing one or more passageways through the coating. The amount of modulator contained within a sustained release formulation depends upon, for example, the method of administration (*e.g.*, the site of implantation), the rate and expected duration of release and the nature of the condition to be treated or prevented.

The matrix material, which itself may or may not serve a controlled-release function, is generally any material that supports the active ingredient(s). For example, a time delay material such as glyceryl monosterate or glyceryl distearate may be employed. Active ingredient(s) may be combined with matrix material prior to formation of the dosage form (*e.g.*, a tablet). Alternatively, or in addition, active ingredient(s) may be coated on the surface of a particle, granule, sphere, microsphere, bead or pellet that comprises the matrix material. Such coating may be achieved by conventional means, such as by dissolving the active ingredient(s) in water or other suitable solvent and spraying. Optionally, additional ingredients are added prior to coating (*e.g.*, to assist binding of the active ingredient(s) to the matrix material or to color the solution). The matrix may then be coated with a barrier agent prior to application of controlled-release coating. Multiple coated matrix units may, if desired, be encapsulated to generate the final dosage form.

In certain embodiments, a controlled release is achieved through the use of a controlled release coating (*i.e.*, a coating that permits release of active ingredient(s) at a controlled rate in aqueous medium). The controlled release coating should be a strong, continuous film that is smooth, capable of supporting pigments and other additives, non-toxic, inert and tack-free. Coatings that regulate release of the modulator include pH-independent coatings, pH-dependent coatings (which may be used to release modulator in the stomach) and enteric coatings (which allow the formulation

to pass intact through the stomach and into the small intestine, where the coating dissolves and the contents are absorbed by the body). It will be apparent that multiple coatings may be employed (*e.g.*, to allow release of a portion of the dose in the stomach and a portion further along the gastrointestinal tract). For example, a portion of active ingredient(s) may be coated over an enteric coating, and thereby released in the stomach, while the remainder of active ingredient(s) in the matrix core is protected by the enteric coating and released further down the GI tract. pH dependent coatings include, for example, shellac, cellulose acetate phthalate, polyvinyl acetate phthalate, hydroxypropylmethylcellulose phthalate, methacrylic acid ester copolymers and zein.

In certain embodiments, the coating is a hydrophobic material, preferably used in an amount effective to slow the hydration of the gelling agent following administration. Suitable hydrophobic materials include alkyl celluloses (*e.g.*, ethylcellulose or carboxymethylcellulose), cellulose ethers, cellulose esters, acrylic polymers (*e.g.*, poly(acrylic acid), poly(methacrylic acid), acrylic acid and methacrylic acid copolymers, methyl methacrylate copolymers, ethoxy ethyl methacrylates, cyanoethyl methacrylate, methacrylic acid alkamide copolymer, poly(methyl methacrylate), polyacrylamide, ammonio methacrylate copolymers, aminoalkyl methacrylate copolymer, poly(methacrylic acid anhydride) and glycidyl methacrylate copolymers) and mixtures of the foregoing. Representative aqueous dispersions of ethylcellulose include, for example, AQUACOAT® (FMC Corp., Philadelphia, PA) and SURELEASE® (Colorcon, Inc., West Point, PA), both of which can be applied to the substrate according to the manufacturer's instructions. Representative acrylic polymers include, for example, the various EUDRAGIT® (Rohm America, Piscataway, NJ) polymers, which may be used singly or in combination depending on the desired release profile, according to the manufacturer's instructions.

The physical properties of coatings that comprise an aqueous dispersion of a hydrophobic material may be improved by the addition or one or more plasticizers. Suitable plasticizers for alkyl celluloses include, for example, dibutyl sebacate, diethyl phthalate, triethyl citrate, tributyl citrate and triacetin. Suitable plasticizers for acrylic polymers include, for example, citric acid esters such as triethyl citrate and tributyl citrate, dibutyl phthalate, polyethylene glycols, propylene glycol, diethyl phthalate, castor oil and triacetin.

Controlled-release coatings are generally applied using conventional techniques, such as by spraying in the form of an aqueous dispersion. If desired, the coating may comprise pores or channels to facilitate release of active ingredient. Pores and channels may be generated by well known methods, including the addition of organic or inorganic material that is dissolved, extracted or leached from the coating in the environment of use. Certain such pore-forming materials include hydrophilic polymers, such as hydroxyalkylcelluloses (*e.g.*, hydroxypropylmethylcellulose), cellulose ethers, synthetic water-soluble polymers (*e.g.*, polyvinylpyrrolidone, cross-linked polyvinylpyrrolidone and polyethylene oxide), water-soluble polydextrose, saccharides and polysaccharides and alkali metal salts. Alternatively, or in addition, a controlled release coating may include one or more orifices,

which may be formed my methods such as those described in US Patent Nos. 3,845,770; 4,034,758; 4,077,407; 4,088,864; 4,783,337 and 5,071,607. Controlled-release may also be achieved through the use of transdermal patches, using conventional technology (*see, e.g.,* US Patent No. 4,668,232).

Further examples of controlled release formulations, and components thereof, may be found, for example, in US Patent Nos. 4,572,833; 4,587,117; 4,606,909; 4,610,870; 4,684,516; 4,777,049; 4,994,276; 4,996,058; 5,128,143; 5,202,128; 5,376,384; 5,384,133; 5,445,829; 5,510,119; 5,618,560; 5,643,604; 5,891,474; 5,958,456; 6,039,980; 6,143,353; 6,126,969; 6,156,342; 6,197,347; 6,387,394; 6,399,096; 6,437,000; 6,447,796; 6,475,493; 6,491,950; 6,524,615; 6,838,094; 6,905,709; 6,923,984; 6,923,988; and 6,911,217; each of which is hereby incorporated by reference for its teaching of the preparation of controlled release dosage forms.

In addition to or together with the above modes of administration, a compound provided herein may be conveniently added to food or drinking water (*e.g.,* for administration to non-human animals including companion animals (such as dogs and cats) and livestock). Animal feed and drinking water compositions may be formulated so that the animal takes in an appropriate quantity of the composition along with its diet. It may also be convenient to present the composition as a premix for addition to feed or drinking water.

N-oxide aryl sulfones and sulfoxides provided herein are generally administered in a therapeutically effective amount. Preferred systemic doses are no higher than 50 mg per kilogram of body weight per day (*e.g.,* ranging from about 0.001 mg to about 50 mg per kilogram of body weight per day), with oral doses generally being about 5-20 fold higher than intravenous doses (*e.g.,* ranging from 0.01 to 40 mg per kilogram of body weight per day). The amount of active ingredient that may be combined with the carrier materials to produce a single dosage unit will vary depending, for example, upon the patient being treated and the particular mode of administration. Dosage units will generally contain from about 10 μg to about 500 mg of an active ingredient. Optimal dosages may be established using routine testing, and procedures that are well known in the art.

Pharmaceutical compositions provided herein may, but need not, further comprise one or more additional pharmaceutical agents, such as an anti-inflammatory agent or analgesic.

Anti-inflammatory agents include, for example, non-steroidal anti-inflammatory drugs (NSAIDs), non-specific and cyclooxygenase-2 (COX-2) specific cyclooxgenase enzyme inhibitors, gold compounds, corticosteroids, methotrexate, leflunomide, cyclosporine A, IM gold, minocycline, azathioprine, tumor necrosis factor (TNF) receptor antagonists, soluble TNF alpha receptor (etanercept), anti-TNF alpha antibodies (*e.g.,* infliximab and adalimumab), anti-C5 antibodies, interleukin-1 (IL-1) receptor antagonists (*e.g.,* anakinra or IL-1 trap), IL-18 binding protein, CTLA4-Ig (*e.g.,* abatacept), anti-human IL-6 receptor monoclonal antibody (*e.g.,* tocilizumab), LFA-3-Ig fusion proteins (*e.g.,* alefacept), LFA-1 antagonists, anti-VLA4 monoantibody (*e.g.,* natalizumab), anti-CD11a monoclonal antibody, anti-CD20 monoclonal antibody (*e.g.,* rituximab), anti-IL-12 monoclonal antibody, anti-IL-15 monoclonal antibody, CDP 484, CDP 870, chemokine receptor

antagonists, selective iNOS inhibitors, p38 kinase inhibitors, integrin antagonists, angiogenesis inhibitors, and TMI-1 dual inhibitors. Further anti-inflammatory agents include meloxicam, rofecoxib, celecoxib, etoricoxib, parecoxib, valdecoxib and tilicoxib.

NSAIDs include, but are not limited to, ibuprofen, flurbiprofen, naproxen or naproxen sodium, diclofenac, combinations of diclofenac sodium and misoprostol, sulindac, oxaprozin, diflunisal, piroxicam, indomethacin, etodolac, fenoprofen calcium, ketoprofen, sodium nabumetone, sulfasalazine, tolmetin sodium, and hydroxychloroquine. One class of NSAIDs consists of compounds that inhibit cyclooxygenase (COX) enzymes; such compounds include celecoxib and rofecoxib. NSAIDs further include salicylates such as acetylsalicylic acid or aspirin, sodium salicylate, choline and magnesium salicylates, and salsalate, as well as corticosteroids such as cortisone, dexamethasone, methylprednisolone, prednisolone, prednisolone sodium phosphate, and prednisone.

Certain analgesics for use in combination with $B_1$ modulators provided herein are also anti-inflammatory agents, and are listed above. Other such medications are analgesic agents, including narcotic agents which typically act at one or more opioid receptor subtypes (*e.g.*, μ, κ and/or δ), preferably as agonists or partial agonists. Such agents include opiates, opiate derivatives and opioids, as well as pharmaceutically acceptable salts and hydrates thereof. Specific examples of narcotic analgesics include, within preferred embodiments, alfentanil, alphaprodine, anileridine, bezitramide, buprenorphine, butorphanol, codeine, diacetyldihydromorphine, diacetylmorphine, dihydrocodeine, diphenoxylate, ethylmorphine, fentanyl, heroin, hydrocodone, hydromorphone, isomethadone, levomethorphan, levorphane, levorphanol, meperidine, metazocine, methadone, methorphan, metopon, morphine, nalbuphine, opium extracts, opium fluid extracts, powdered opium, granulated opium, raw opium, tincture of opium, oxycodone, oxymorphone, paregoric, pentazocine, pethidine, phenazocine, piminodine, propoxyphene, racemethorphan, racemorphan, sulfentanyl, thebaine and pharmaceutically acceptable salts and hydrates of the foregoing agents.

Other examples of narcotic analgesic agents include acetorphine, acetyldihydrocodeine, acetylmethadol, allylprodine, alphracetylmethadol, alphameprodine, alphamethadol, benzethidine, benzylmorphine, betacetylmethadol, betameprodine, betamethadol, betaprodine, clonitazene, codeine methylbromide, codeine-N-oxide, cyprenorphine, desomorphine, dextromoramide, diampromide, diethylthiambutene, dihydromorphine, dimenoxadol, dimepheptanol, dimethylthiamubutene, dioxaphetyl butyrate, dipipanone, drotebanol, ethanol, ethylmethylthiambutene, etonitazene, etorphine, etoxeridine, furethidine, hydromorphinol, hydroxypethidine, ketobemidone, levomoramide, levophenacylmorphan, methyldesorphine, methyldihydromorphine, morpheridine, morphine, methylpromide, morphine methylsulfonate, morphine-N-oxide, myrophin, naloxone, naltyhexone, nicocodeine, nicomorphine, noracymethadol, norlevorphanol, normethadone, normorphine, norpipanone, pentazocaine, phenadoxone, phenampromide, phenomorphan, phenoperidine,

piritramide, pholcodine, proheptazoine, properidine, propiran, racemoramide, thebacon, trimeperidine and the pharmaceutically acceptable salts and hydrates thereof.

Further specific representative analgesic agents include, for example acetaminophen (paracetamol); aspirin and other NSAIDs described above; NR2B antagonists; capsaicin receptor antagonists; anti-migraine agents; anticonvulsants such as oxcarbazepine and carbamazepine; antidepressants (such as TCAs, SSRIs, SNRIs, substance P antagonists, etc.); spinal blocks; pentazocine/naloxone; meperidine; levorphanol; buprenorphine; hydromorphone; fentanyl; sufentanyl; oxycodone; oxycodone/acetaminophen, nalbuphine and oxymorphone. Still further analgesic agents include CB2-receptor agonists, such as AM1241, capsaicin receptor antagonists and compounds that bind to the $\alpha 2\delta$ subunit of voltage-gated calcium channels, such as gabapentin and pregabalin.

Representative anti-migraine agents for use in combination with a $B_1$ modulator provided herein include CGRP antagonists, ergotamines and 5-$HT_1$ agonists, such as sumatripan, naratriptan, zolmatriptan and rizatriptan.

Pharmaceutical compositions may be packaged for treating conditions responsive to $B_1$ modulation (*e.g.*, treatment of pain, inflammation or other disorder(s) recited herein). Packaged pharmaceutical preparations generally comprise a container holding a therapeutically effective amount of a pharmaceutical composition as described above and instructions (*e.g.*, labeling) indicating that the composition is to be used for treating a condition responsive to $B_1$ modulation in a patient (*e.g.*, pain or other disorder as indicated herein). In certain embodiments, a packaged pharmaceutical preparation comprises one or more N-oxide aryl sulfone or sulfoxide provided herein and one or more additional agents in the same package, either in separate containers within the package or in the same container (*i.e.*, as a mixture). Preferred mixtures are formulated for oral administration (*e.g.*, as pills, capsules, tablets or the like). In certain embodiments, the package comprises a label bearing indicia indicating that the components are to be taken together for the treatment of pain.

METHODS OF USE

Within certain aspects, the present invention provides methods for treating a condition responsive to $B_1$ modulation in a patient. The patient may be afflicted with such a condition, or may be free of symptoms but considered at risk for developing such a condition. A condition is "responsive to $B_1$ modulation" if the condition or symptom(s) thereof are alleviated, attenuated, delayed or otherwise improved by modulation of $B_1$ activity. In general, such methods comprise administering to the patient a therapeutically effective amount of at least one N-oxide aryl sulfone or sulfoxide as provided herein.

Conditions responsive to $B_1$ modulation include, for example pain; inflammation including neuroinflammation (such as atherosclerosis), inflammation associated with airway diseases (*e.g.*, asthma, including allergic asthma, exercise-induced bronchoconstriction, occupational asthma, and other non-allergic asthmas), and inflammatory skin disorders (*e.g.*, psoriasis and eczema)); respiratory

disorders including bronchoconstriction, asthma, chronic obstructive pulmonary disease (*e.g.*, emphysema), chronic cough (including ACE-inhibitor cough), adult respiratory distress syndrome, bronchitis, pneumonia, allergic rhinitis and vasomotor rhinitis; vascular edema (including diabetes-related vascular disease); and epilepsy.

Other conditions responsive to $B_1$ modulation include diabetes (*e.g.*, type II or non insulin dependent, as well as diabetic vasculopathy, diabetic neuropathy, diabetic retinopathy, post capillary resistance and symptoms associated with insulitis), seizure disorders (*e.g.*, epilepsy), multiple sclerosis, liver disease, cardiovascular disorders (*e.g.*, atherosclerosis, congestive heart failure and myocardial infarction), neurodegenerative diseases (*e.g.*, Alzheimer's disease and Parkinson's disease) rheumatoid arthritis, infection, cancer, cranial trauma, rhinitis, septic shock, endotoxic and pancreatic shock, anaphylaxis, inflammatory bowel disease, irritable bowel syndrome, pancreatitis, cystitis, uveitis, vascular permeability, gingivitis, osteoporosis, benign prostatic hyperplasia, hyperactive bladder, cerebral edema, vasodilation, hypotension associated with sepsis, edema resulting from trauma associated with burns, sprains or fracture, cerebral edema, angiodema, Crohn's disease and ulcerative colitis. $B_1$ modulators may also be used as smooth muscle relaxants for treating spasms of the gastrointestinal tract of uterus. In certain embodiments, the condition responsive to $B_1$ modulation is pain or inflammation.

Pain that may be treated using the $B_1$ modulators provided herein may be chronic or acute and includes, but is not limited to, peripheral nerve-mediated pain (especially neuropathic pain, such as pain due to diabetes, postherpetic neuralgia, nerve injury, vulvodynia, root avulsions, painful traumatic neuropathy and painful polyneuropathy). Compounds provided herein may also be used in the treatment of, for example, visceral pain (*e.g.*, pancreatitis, interstitial cystitis and renal colic), persistent hyperalgesia, inflammatory pain, repetitive motion pain, carpel tunnel syndrome, perioperative pain, algesia, oral neuropathic pain, toothache (dental pain), denture pain, ocular pain, postherpetic neuralgia, diabetic neuropathy, chemotherapy-induced neuropathy, reflex sympathetic dystrophy, trigeminal neuralgia, bone and joint pain (*e.g.*, pain associated with osteoarthritis), rheumatoid arthritis, myofascial pain (*e.g.*, muscular injury and fibromyalgia), Guillain-Barre syndrome, meralgia paresthetica, burning-mouth syndrome and/or pain associated with nerve and root damage, including as pain associated with peripheral nerve disorders (*e.g.*, nerve entrapment and brachial plexus avulsions, amputation, peripheral neuropathies including bilateral peripheral neuropathy, tic douloureux, atypical facial pain, nerve root damage, and arachnoiditis). Additional neuropathic pain conditions include causalgia (reflex sympathetic dystrophy - RSD, secondary to injury of a peripheral nerve), neuritis (including, for example, sciatic neuritis, peripheral neuritis, polyneuritis, optic neuritis, postfebrile neuritis, migrating neuritis, segmental neuritis and Gombault's neuritis), neuronitis, neuralgias (*e.g.*, those mentioned above, cervicobrachial neuralgia, cranial neuralgia, geniculate neuralgia, glossopharyngial neuralgia, migranous neuralgia, idiopathic neuralgia, intercostals neuralgia, mammary neuralgia, mandibular joint neuralgia, Morton's neuralgia,

nasociliary neuralgia, occipital neuralgia, red neuralgia, Sluder's neuralgia, splenopalatine neuralgia, supraorbital neuralgia and vidian neuralgia), musculoskeletal pain, AIDS-related neuropathy, MS-related neuropathy, central nervous system pain (*e.g.,* pain due to brain stem damage, sciatica, and ankylosing spondylitis), and spinal pain, including spinal cord injury-related pain.

Headache, including headaches involving peripheral nerve activity may also be treated as described herein. Such headache pain includes, for example, sinus, cluster (*i.e.,* migranous neuralgia) and tension headaches, migraine, temporomandibular pain and maxillary sinus pain. For example, migraine headaches may be prevented by administration of a compound provided herein as soon as a pre-migrainous aura is experienced by the patient.

Further conditions that can be treated as described herein include Charcot's pains, intestinal gas pains, ear pain, heart pain, muscle pain, eye pain, orofacial pain (*e.g.,* odontalgia), abdominal pain, gynaecological pain (*e.g.,* menstrual pain, dysmenorrhoea, pain associated with cystitis, labor pain, chronic pelvic pain, chronic prostitis and endometriosis), acute and chronic back pain (*e.g.,* lower back pain), gout, scar pain, hemorrhoidal pain, dyspeptic pains, pain associated with angina, nerve root pain, "non-painful" neuropathies, complex regional pain syndrome, homotopic pain and heterotopic pain – including pain associated with carcinoma, often referred to as cancer pain (*e.g.,* in patients with bone cancer), pain (and inflammation) associated with venom exposure (*e.g.,* due to snake bite, spider bite, or insect sting) and trauma associated pain (*e.g.,* post-surgical pain such as postmastectomy, postthoracotomy and stump pain; phantom limb pain, episiotomy pain, pain from cuts, musculoskeletal pain, bruises and broken bones, and burn pain, especially primary hyperalgesia associated therewith). Additional pain conditions that may be treated as described herein include pain associated with respiratory disorders as described above, autoimmune diseases, immunodeficiency disorders, hot flashes, inflammatory bowel disease, gastroesophageal reflux disease (GERD), irritable bowel syndrome and/or inflammatory bowel disease.

In certain embodiments, pain treated with $B_1$ modulators provided herein is inflammatory pain, acute pain, dental pain, back pain, surgical pain, headache, neuropathic pain or pain from osteoarthritis or trauma.

It will be apparent that compounds provided herein may be administered alone or in combination with one or more additional agents that are suitable for treating the disorder of interest. Within such combination therapy, the compound(s) and additional agent(s) may be present in the same pharmaceutical composition, or may be administered separately in either order. Representative anti-inflammatory agents and analgesics for use in combination therapy include those indicated above.

Within other aspects, $B_1$ modulators provided herein may be used within combination therapy for the treatment of conditions involving pain and/or inflammatory components. Such conditions include, for example, autoimmune disorders and pathologic autoimmune responses known to have an inflammatory component including, but not limited to, arthritis (especially rheumatoid arthritis), psoriasis, Crohn's disease, lupus erythematosus, irritable bowel syndrome, tissue graft rejection, and

hyperacute rejection of transplanted organs. Other such conditions include trauma (*e.g.,* injury to the head or spinal cord), cardio- and cerebro-vascular disease and certain infectious diseases. Within such combination therapy, a $B_1$ modulator is administered to a patient along with an additional analgesic and/or anti-inflammatory agent. The $B_1$ modulator and additional analgesic and/or anti-inflammatory agent may be present in the same pharmaceutical composition, or may be administered separately in either order.

Administration to the patient can be by way of any means discussed above, including oral, topical, nasal or transdermal administration, or intravenous, intramuscular, subcutaneous, intrathecal, epidural, intracerebroventricular or like injection. Oral administration is preferred in certain embodiments (*e.g.,* formulated as pills, capsules, tablets or the like).

Treatment regimens may vary depending on the compound used and the particular condition to be treated. In general, a dosage regimen of 4 times daily or less is preferred, with 1 or 2 times daily particularly preferred. It will be understood, however, that the specific dose and treatment regimen for any particular patient will depend upon a variety of factors including the activity of the specific compound employed, the age, body weight, general health, sex and diet of the patient, the time of administration, the route of administration, the rate of excretion, any drug combination and the severity of the particular disease undergoing therapy. Dosages are generally as described above; in general, the use of the minimum dose sufficient to provide effective therapy is preferred. Patients may generally be monitored for therapeutic effectiveness using medical or veterinary criteria suitable for the condition being treated or prevented.

Suitable dosages for $B_1$ modulators (either alone or within such combination therapy) are generally as described above. Dosages and methods of administration of any additional agent(s) (e.g., anti-inflammatory and/or analgesic agents) can be found, for example, in the manufacturer's instructions or in the *Physician's Desk Reference.* In certain embodiments, combination administration results in a reduction of the dosage of the additional agent required to produce a therapeutic effect (*i.e.,* a decrease in the minimum therapeutically effective amount). Thus, preferably, the dosage of additional agent in a combination or combination treatment method of the invention is less than the maximum dose advised by the manufacturer for administration of the agent without combination with a compound of Formula I. More preferably this dose is less than ¾, even more preferably less than ½, and highly preferably less than ¼ of the maximum dose, while most preferably the dose is less than 10% of the maximum dose advised by the manufacturer for administration of the agent(s) when administered without combination administration as described herein. It will be apparent that the dose of compound as provided herein needed to achieve the desired effect may similarly be affected by the dose and potency of the additional agent.

Within separate aspects, the present invention provides a variety of non-pharmaceutical *in vitro* and *in vivo* uses for the compounds provided herein. For example, such compounds may be labeled and used as probes for the detection and localization of $B_1$ (in samples such as cell

preparations or tissue sections, preparations or fractions thereof). In addition, compounds provided herein that comprise a suitable reactive group (such as an aryl carbonyl, nitro or azide group) may be used in photoaffinity labeling studies of receptor binding sites. In addition, compounds provided herein may be used as positive controls in assays for receptor activity, as standards for determining the ability of a candidate agent to bind to $_{B1}$, or as radiotracers for positron emission tomography (PET) imaging or for single photon emission computerized tomography (SPECT). Such methods can be used to characterize $B_1$ receptors in living subjects. For example, a compound may be labeled using any of a variety of well known techniques (*e.g.*, radiolabeled with a radionuclide such as tritium, as described herein), and incubated with a sample for a suitable incubation time (*e.g.*, determined by first assaying a time course of binding). Following incubation, unbound compound is removed (*e.g.*, by washing), and bound compound detected using any method suitable for the label employed (*e.g.*, autoradiography or scintillation counting for radiolabeled compounds; spectroscopic methods may be used to detect luminescent groups and fluorescent groups). As a control, a matched sample containing labeled compound and a greater (*e.g.*, 10-fold greater) amount of unlabeled compound may be processed in the same manner. A greater amount of detectable label remaining in the test sample than in the control indicates the presence of $B_1$ in the sample. Detection assays, including receptor autoradiography (receptor mapping) of $B_1$ in cultured cells or tissue samples may be performed as described by Kuhar in sections 8.1.1 to 8.1.9 of Current Protocols in Pharmacology (1998) John Wiley & Sons, New York.

The following Examples are offered by way of illustration and not by way of limitation. Unless otherwise specified all reagents and solvent are of standard commercial grade and are used without further purification. Using routine modifications, the starting materials may be varied and additional steps employed to produce other compounds provided herein.

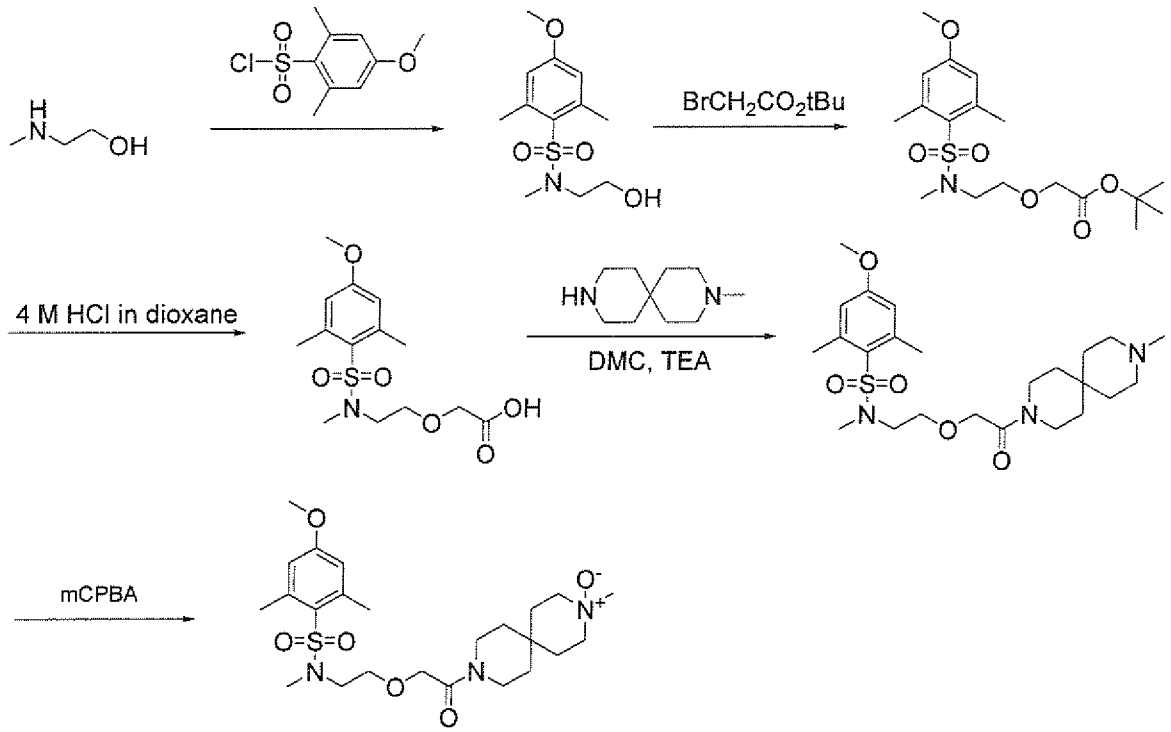
EXAMPLES

Mass spectroscopy data in the following Examples is Electrospray MS, obtained in positive ion mode using a Micromass Time-of-Flight LCT (Micromass, Beverly MA), equipped with a Waters 600 pump (Waters Corp.; Milford, MA), Waters 996 photodiode array detector, and a Gilson 215 autosampler (Gilson, Inc.; Middleton, WI). MassLynx (Advanced Chemistry Development, Inc; Toronto, Canada) version 4.0 software with OpenLynx Global Server™, OpenLynx™ and AutoLynx™ processing is used for data collection and analysis. MS conditions are as follows: capillary voltage = 3.5 kV; cone voltage = 30 V, desolvation and source temperature = 350°C and 120°C, respectively; mass range = 181-750 with a scan time of 0.22 seconds and an interscan delay of 0.05 seconds.

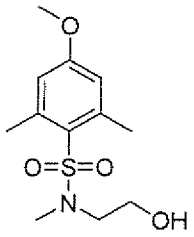
Sample volume of 1 microliter is injected onto a 50x4.6mm Chromolith SpeedROD RP-18e column (Merck KGaA, Darmstadt, Germany), and eluted using a 2-phase linear gradient at a flow rate of 6 ml/min. Sample is detected using total absorbance count over the 220-340nm UV range. The

elution conditions are: Mobile Phase A - 95% water, 5% MeOH with 0.05% TFA; Mobile Phase B - 5% water, 95% MeOH with 0.025% TFA. The following gradient is used: 0-0.5 min 10-100%B, hold at 100%B to 1.2 min, return to 10%B at 1.21 min. Inject to inject cycle is 2.15 min.

EXAMPLE 1. SYNTHESIS OF 4-METHOXY-N,2,6-TRIMETHYL-N-{2-[2-(9-METHYL-9-OXIDO-3,9-DIAZASPIRO[5.5]UNDEC-3-YL)-2-OXOETHOXT]ETHYL}BENZENESULFONAMIDE (COMPOUND 1)

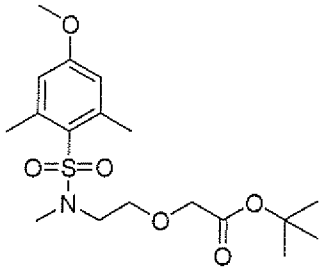



Step 1. N-(2-Hydroxyethyl)-4-methoxy-2,6,N-trimethylbenzenesulfonamide



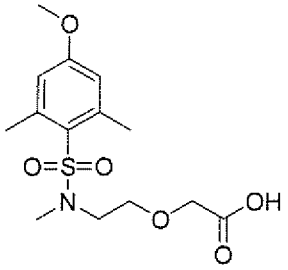
4-Methoxy-2,6-dimethylbenzenesulfonyl chloride (12 g, 51 mmol) is added to a solution of 2-(methylamino)ethanol (4.6 mL, 61 mmol), diisopropylethylamine (22 mL, 130 mmol), and $CH_2Cl_2$ (130 mL) under $N_2$. The reaction vessel is sealed and left to stir for 21 h. The solution is washed with 1:1 sat. aq. $NH_4Cl$: $H_2O$ (2 x 200 mL) and brine (200 mL). The aqueous layers are extracted with $CH_2Cl_2$ (200 mL). The combined organic layers are dried over $Na_2SO_4$, filtered, and concentrated to afford the title compound as a pale yellow oil.

Step 2. {2-[(4-Methoxy-2,6-dimethylbenzenesulfonyl)methylamino]ethoxy}acetic acid tert-butyl ester



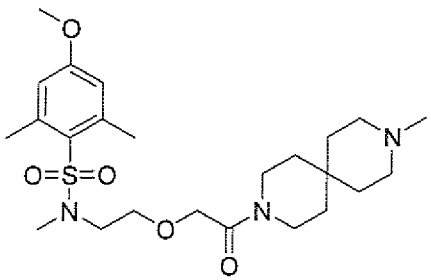
Sodium hydride, 60% dispersion in mineral oil, (3.7 g, 90 mmol) is added to a solution of (2-hydroxyethyl)-4-methoxy-2,6,N-trimethylbenzenesulfonamide (14.5 g) and DMF (300 mL) at 0 °C under $N_2$. After 15 min, t-butyl bromoacetate (11.5 mL, 78 mmol) is added and the cold bath is removed. After 16 h, the reaction mixture is poured into 50% sat. aqueous $NH_4Cl$ (400 mL). The solution is extracted with EtOAc (2 x 250 mL). The combined organic layers are washed with 50% sat. aqueous $NH_4Cl$ (400 mL), dried over $Na_2SO_4$, filtered, and concentrated. Purification by flash column chromatography (5:1 hexanes:EtOAc, 250 g $SiO_2$) affords the title compound as a pale yellow oil.

Step 3. {2-[(4-Methoxy-2,6-dimethylbenzenesulfonyl)methylamino]ethoxy}acetic acid



{2-[(4-Methoxy-2,6-dimethylbenzenesulfonyl)methylamino]ethoxy}acetic acid *tert*-butyl ester (18.9 g, 48.8 mmol) is treated with 4 M HCl in dioxane (60 mL) at rt overnight. The volatiles are removed under reduced pressure to afford a tan solid. $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$: 6.65 (s, 2H), 4.13 (s, 2H), 3.82 (s, 3H), 3.72 (t, 2H), 3.53 (t, 2H), 2.70 (s, 3H), 2.60 (s, 6H).

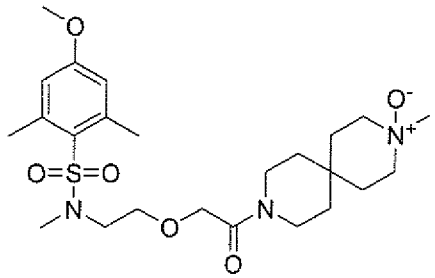
Step 4. 4-Methoxy-2,6,N-trimethyl-N-{2-[2-(9-methyl-3,9-diaza-spiro[5.5]undec-3-yl)-2-oxo-ethoxy]ethyl}benzenesulfonamide



A 1.0 M solution of TEA in toluene (0.15 mL) is added to a solution of {2-[(4-methoxy-2,6-dimethylbenzenesulfonyl)methylamino]ethoxy}acetic acid (24 mg, 72 μmol), 3-methyl-3,9-diaza-

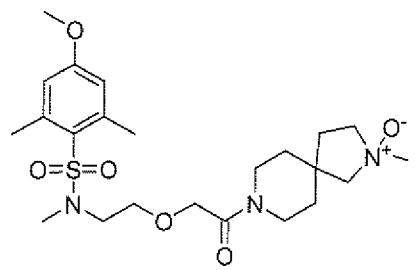
spiro[5.5]undecane (10 mg, 60 μmol), and DMA (0.3 mL) under $N_2$. A 0.2 M solution of 2-chloro-1,3-dimethylimidazolidinium chloride in acetonitrile (0.6 mL) is added. The reaction vessel is sealed and the mixture is warmed to 50 °C for 2.5 h. After cooling to ambient temperature, 1 M aqueous NaOH (1.5 mL) is added and then the volatiles are removed under reduced pressure. The aqueous residue is extracted with EtOAc (2 x 1.5 mL). The organics are loaded onto two 0.5 g SCX columns. Each column is washed with MeOH (3 x 3 mL) and then eluded with 5:5:2 EtOAc : MeOH : TEA (4.5 mL). Concentration of the organics under reduced pressure afforded the title compound as a pale yellow film. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 6.62 (s, 2H), 4.10 (s, 2H), 3.81 (s, 3H), 3.67 (t, 2H), 3.52 (dd, 2H), 3.37 (t, 2H), 3.34 (dd, 2H), 2.78 (s, 3H), 2.60 (s, 6H), 2.35-2.44 (m, 4H), 2.29 (s, 3H), 1.42-1.60 (m, 8H).

Step 5. 4-Methoxy-2,6,N-trimethyl-N-{2-[2-(9-methyl-3,9-diaza-spiro[5.5]undec-3-yl)-2-oxo-ethoxy]ethyl}benzenesulfonamide



A 0.25 M solution of 3-chloroperbenzoic acid in $CH_2Cl_2$ (500 μL, 125 μmol) is added to a 0 °C solution of 4-methoxy-2,6,N-trimethyl-N-{2-[2-(9-methyl-3,9-diaza-spiro[5.5]undec-3-yl)-2-oxo-ethoxy]ethyl}benzenesulfonamide (50 mg, 104 μmol) in $CH_2Cl_2$ (500 μL). After 1.5 h, the volatiles are removed under reduced pressure. The resulting residue is purified by thin layer chromatography (eluting with 2% $NH_4OH$ in 5 : 1 $CH_2Cl_2$ : MeOH) to afford the title compound. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 6.62 (s, 2H), 4.11 (s, 2H), 3.81 (s, 3H), 3.68 (t, 2H), 3.22-3.60 (m, 12H), 3.28 (s, 3H), 2.77 (s, 3H), 2.59 (s, 6H), 2.16-2.34 (m, 2H), 1.47-1.68 (m, 4H). LC-MS *m/z* (M + $H^+$): 498.3; retention time: 0.9 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

EXAMPLE 2. SYNTHESIS OF 4-METHOXY-N,2,6-TRIMETHYL-N-{2-[2-(2-METHYL-2-OXIDO-2,8-DIAZASPIRO[4.5]DEC-8-YL)-2-OXOETHOXY]ETHYL}BENZENESULFONAMIDE (COMPOUND 2)

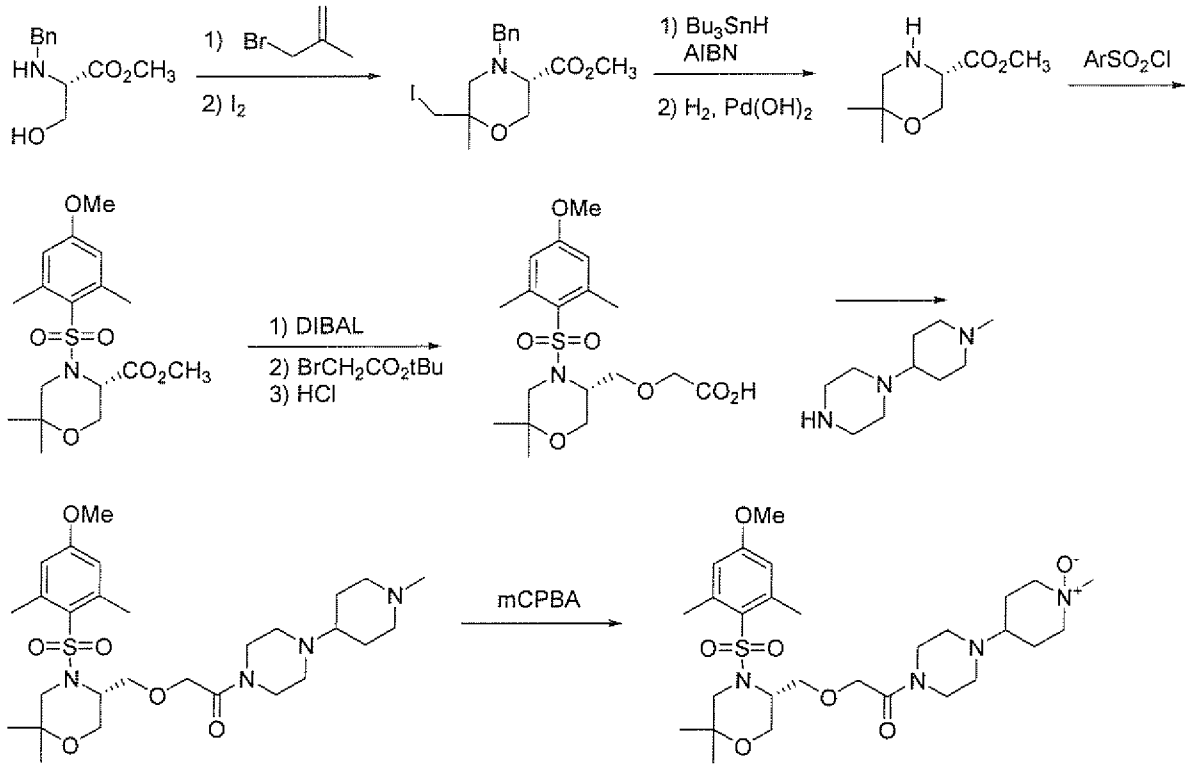


The title compound is prepared as a yellow oil using the procedure illustrated above. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 6.63 (s, 2H), 4.11 (s, 2H), 3.82 (s, 3H), 3.24-3.72 (m, 15H), 2.78 (s, 3H), 2.60

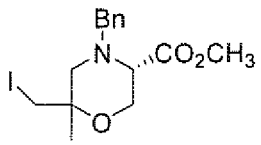
(s, 6H), 1.58-2.58 (m, 6H). LC-MS *m/z* (M + $H^+$): 484.2; retention time: 1.1 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

## EXAMPLE 3. SYNTHESIS OF ADDITIONAL REPRESENTATIVE N-OXIDE ARYL SULFONES AND SULFOXIDES

### A. (5R)-4-[(4-METHOXY-2,6-DIMETHYLPHENYL)SULFONYL]-2,2-DIMETHYL-5-({2-[4-(1-METHYL-1-OXIDOPIPERIDIN-4-YL)PIPERAZIN-1-YL]-2-OXOETHOXY}METHYL)MORPHOLINE (COMPOUND 3)

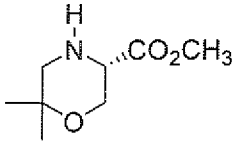



Step 1. (3S)-Methyl 4-benzyl-6-(iodomethyl)-6-methylmorpholine-3-carboxylate



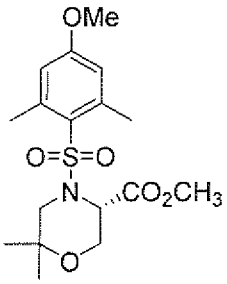

(S)-Methyl 2-(benzylamino)-3-hydroxypropanoate (27.4 g, 131 mmol) is combined with 3-bromo-2-methylpropene (25 mL, 248 mmol), $K_2CO_3$ (45 g, 327 mmol), and KI (4.3 g, 26 mmol) in 300 mL acetonitrile. The reaction mixture is stirred at rt for 3 days. Iodine (66 g, 262 mmol) is added and the reaction mixture is stirred an additional 2 h at rt. The reaction mixture is diluted with 300 mL ether and transferred to a separatory funnel. The solution is washed twice successively with 200 mL 1N $Na_2SO_3$, once with 200 mL saturated $NaHCO_3$, and once with 200 mL brine. The organic phase is dried over $MgSO_4$, filtered and concentrated *in vacuo*. The product is purified by flash chromatography, eluting with 9/1 hexanes/EtOAc to afford the title compound as a 5/1 mixture of diastereomers.

Step 2. (S)-Methyl 6,6-dimethylmorpholine-3-carboxylate



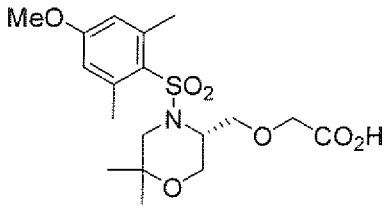
The mixture of diastereomers obtained in step 1 is combined with tributyltin hydride (73 mL, 272 mmol) in 400 mL refluxing toluene. AIBN (1 g, 6 mmol) dissolved in 40 mL toluene is added dropwise. After 2 h reflux, the reaction mixture is cooled to rt. Saturated aqueous KF solution (800 mL) is added and the resulting heterogeneous mixture is filtered through Celite and the solid washed with EtOAc. The combined solutions are transferred to a separatory funnel and washed twice with 200 mL saturated KF, and then once with 20 mL brine. The organic phase is dried over $MgSO_4$, filtered and concentrated *in vacuo*. The product is purified by flash chromatography eluting with 95/5 hexanes/EtOAc followed by EtOAc to afford (S)-methyl 4-benzyl-6,6-dimethylmorpholine-3-carboxylate as a clear oil. This oil is dissolved in a mixture of 200 mL MeOH and 20 mL acetic acid. Palladium hydroxide (5 g, 20 wt. % on carbon) is added and the reaction mixture hydrogenated on a Paar apparatus at 50 psi for 10 h. An additional 5 g of palladium hydroxide is added and the hydrogenation is continued at 50 psi for another 25 h. The reaction mixture is filtered through Celite and concentrated *in vacuo* to afford the title compound. $^1$H NMR (400 MHz, d-6-DMSO) $\delta$: 3.71-3.74 (m, 1H), 3.61-3.65 (m, 1H), 3.61 (s, 3H), 3.36 (m, 1H), 2.65 (d, 1H), 2.46 (d, 1H), 1.11 (s, 3H), 1.09 (s, 3H).

Step 3. (S)-Methyl 4-(4-methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholine-3-carboxylate



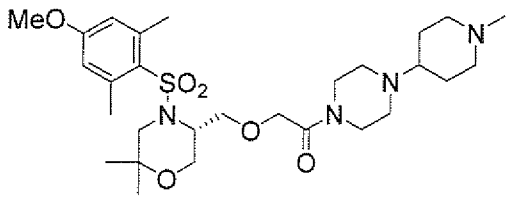
(S)-Methyl 6,6-dimethylmorpholine-3-carboxylate (2.3 g, 13 mmol) is dissolved in 50 mL pyridine. 2,6-Dimethyl-4-methoxyphenylsulfonyl chloride (3.1 g, 13 mmol) and DMAP (15 mg) are added. The reaction mixture is stirred at 50 °C for 5 h. The reaction mixture is cooled to rt, diluted with 100 mL EtOAc, and transferred to a separatory funnel. The reaction mixture is washed three times with 50 mL water and once with 50 mL brine. The organic phase is dried over $MgSO_4$, filtered, then concentrated *in vacuo*. The product is purified by flash chromatography eluting with 8/2 hexanes/EtOAc followed by 1/1 hexanes/EtOAc to afford the title compound.

Step 4. (R)-2-((4-(4-Methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholin-3-yl)methoxy)acetic acid



(S)-Methyl 4-(4-methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholine-3-carboxylate is dissolved in 8 mL of dry THF at 0 °C. DIBAL (8 mL of a 1M solution in THF) is added and the reaction mixture stirred for 30 min at 0 °C. An additional 1 mL of DIBAL solution is added and stirring is continued for 30 min. The reaction mixture is quenched by addition of solid $Na_2SO_4 \cdot 10H_2O$. The reaction mixture is filtered through Celite and concentrated *in vacuo* to afford (R)-(4-(4-methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholin-3-yl)methanol. This compound, without further purification, is dissolved in 10 mL of DMA at rt. Potassium iodide (20 mg) and sodium hydride (140 mg of a 60% oil dispersion) are added, followed by bromo *tert*-butyl acetate (0.48 mL, 3.2 mmol). The reaction mixture is stirred for 2 days, and then diluted with 50 mL EtOAc, transferred to a separatory funnel, and washed once with 20 mL water and once with 20 mL brine. The organic phase is dried over $MgSO_4$, filtered and concentrated *in vacuo*. The product is purified by flash chromatography eluting with 9/1 hexanes/EtOAc to afford (R)-*tert*-butyl 2-((4-(4-methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholin-3-yl)methoxy)acetate. This compound is dissolved in 3 mL of 4 M HCl in dioxane, and stirred at 50 °C for 3 h. The reaction mixture is cooled to rt and concentrated *in vacuo*. The product is purified by flash chromatography, eluting with 1/1 hexanes/EtOAc to afford the title compound.

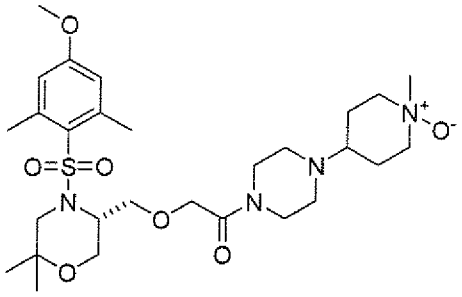
Step 5. (R)-2-((4-(4-Methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholin-3-yl)methoxy)-1-(4-(1-methylpiperidin-4-yl)piperazin-1-yl)ethanone



To a solution of (R)-2-((4-(4-methoxy-2,6-dimethylphenylsulfonyl)-6,6-dimethylmorpholin-3-yl)methoxy)acetic acid (1 mL of a 0.2 M solution in 95/5 EtOAc/TEA) is added isobutylchloroformate (1 mL of a 0.2 M solution in toluene). The reaction mixture is stirred at rt for 5 min. 1-(1-Methylpiperidin-4-yl)piperazine (1.2mL of a 0.2M solution in toluene) is added and the reaction mixture is stirred for 90 min at rt. The reaction mixture is diluted with 5 mL of EtOAc and washed with 1 mL of 1N NaOH. The organic phase is applied to a 10 g silica SPE cartridge and eluted with 10 mL of EtOAc to remove non-polar impurities, followed by 10 mL of 10/1/1 EtOAc/MeOH/TEA to elute the title compound. The solvent is removed *in vacuo* to afford the title

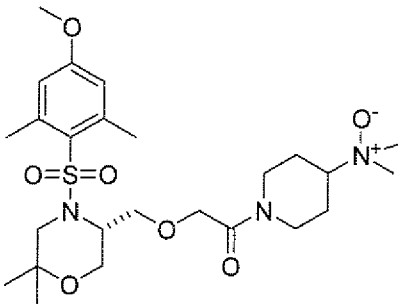
compound. $^1$H NMR (400 MHz, $CDCl_3$) δ: 6.62 (s, 2H), 4.04 (s, 2H), 3.93 (dd, 1H), 3.84-3.86 (m, 1H), 3.81 (s, 3H), 3.76 (d, 1H), 3.70 (m, 1H), 3.56 (m, 3H), 3.47 (m, 1H), 3.39 (m, 2H), 2.99 (dd, 2H), 2.90 (d, 2H), 2.60 (s, 6H), 2.52 (m, 4H), 2.25 (s, 3H), 1.95 (t, 2H), 1.75 (d, 2H), 1.56 (m, 2H), 1.15 (s, 3H), 1.11 (s, 3H).

Step 6. (5R)-4-[(4-methoxy-2,6-dimethylphenyl)sulfonyl]-2,2-dimethyl-5-({2-[4-(1-methyl-1-oxidopiperidin-4-yl)piperazin-1-yl]-2-oxoethoxy}methyl)morpholine



A 0.25 M solution of 3-chloroperbenzoic acid in $CH_2Cl_2$ (500 μL, 125 μmol) is added to a 0 °C solution of 4-[(4-methoxy-2,6-dimethylphenyl)sulfonyl]-2,2-dimethyl-5-({2-[4-(1-methylpiperidin-4-yl)piperazin-1-yl]-2-oxoethoxy}methyl)morpholine (58.7 mg, 104 μmol) in $CH_2Cl_2$ (500 μL). After 1.5 h, the volatiles are removed under reduced pressure. The resulting residue is purified by thin layer chromatography (2% $NH_4OH$ in 4 : 1 $CH_2Cl_2$ : MeOH) to afford the title compound as a tan solid. $^1$H-NMR (400 MHz, $CDCl_3$) *δ*: 6.62 (s, 2H), 4.06 (s, 2H), 3.69-3.96 (m, 4H), 3.82 (s, 3H), 3.56-3.62 (m, 3H), 3.40-3.45 (m, 2H), 3.29-3.36 (m, 2H), 3.22 (s, 3H), 3.12-3.22 (m, 2H), 2.93-3.02 (m, 2H), 2.60 s, 6H), 2.38-2.60 (m, 7H), 1.64-1.71 (m, 2H), 1.15 (s, 3H), 1.10 (s, 3H). LC-MS *m/z* ($M + H^+$): 582.3. Retention time: 1.1 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

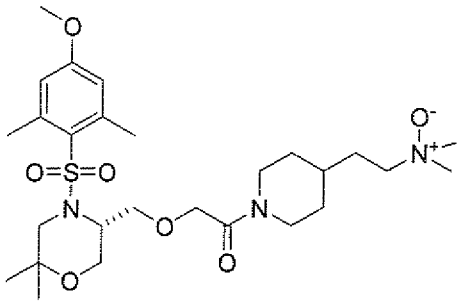
B. {1-[({(3R)-4-[(4-METHOXY-2,6-DIMETHYLPHENYL)SULFONYL]-6,6-DIMETHYLMORPHOLIN-3-YL}METHOXY)ACETYL]PIPERIDIN-4-YL}DIMETHYLAMINE OXIDE (COMPOUND 4)



The title compound as a pale yellow film is prepared as indicated above. $^1$H-NMR (400 MHz, $CDCl_3$) *δ*: 6.62 (s, 2H), 4.70-4.82 (m, 1H), 3.95-4.33 (m. 3H), 3.53-3.94 (m, 4H), 3.80 (s, 3H), 3.27-3.45 (m, 1H), 3.16 (s, 3H), 3.13 (s, 1.5H), 3.11 (s, 1.5H), 2.86-3.08 (m, 3H), 2.24-2.67 (m, 4H), 2.58

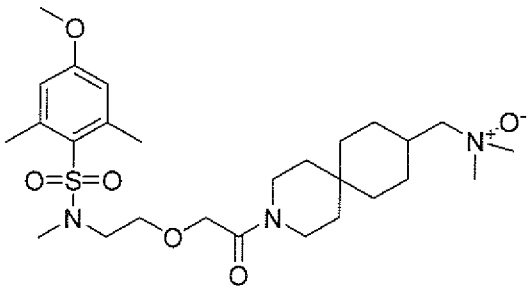
(s, 6H), 1.52-1.82 (m, 2H), 1.12 (s, 3H), 1.05 (s, 1.5H), 0.97 (s, 1.5H). LC-MS *m/z* ($M + H^+$): 527.3. Retention time: 1.1 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

C. (2-{1-[({(3R)-4-[(4-METHOXY-2,6-DIMETHYLPHENYL)SULFONYL]-6,6-DIMETHYLMORPHOLIN-3-YL}METHOXY)ACETYL]PIPERIDIN-4-YL}ETHYL)DIMETHYLAMINE OXIDE (COMPOUND 5)



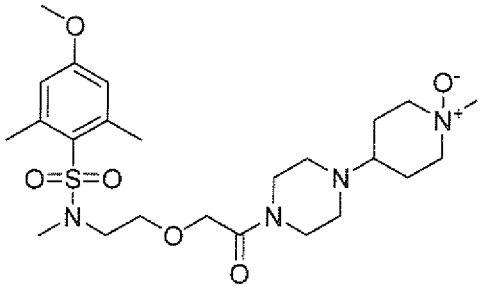
The title compound as a pale yellow film is prepared as indicated above. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 6.62 (s, 2H), 4.52 (dd, 1H), 3.52-4.19 (m, 8H), 3.63 (s, 3H), 3.23-3.32 (m, 2H), 3.18 (s, 1.5H), 3.16 (s, 3H), 3.14 (s, 1.5H), 2.81-3.05 (m, 2H), 2.59 (s, 6H), 2.48-2.61 (m, 2H), 1.52-2.01 (m, 5H), 1.10-1.35 (m, 2H), 1.14 (s, 3H), 1.10 (s, 1.5H), 1.04 (s, 1.5H). LC-MS *m/z* ($M + H^+$): 556.2. Retention time: 1.1 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

D. N-[2-(2-{9-[(DIMETHYLNITRORYL)METHYL]-3-AZASPIRO[5.5]UNDEC-3-YL}-2-OXOETHOXY)ETHYL]-4-METHOXY-N,2,6-TRIMETHYLBENZENESULFONAMIDE (COMPOUND 6)



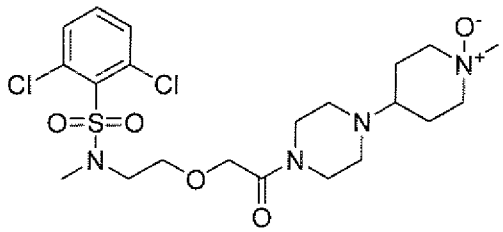
The title compound as a pale yellow film is prepared as indicated above. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 6.61 (s, 2H), 4.09 (d, 2H), 3.80 (s, 3H), 3.66 (t, 2H), 3.46-3.55 (m, 2H), 3.28-3.40 (m, 4H), 3.20 (s, 6H), 3.14 (t, 2H), 2.77 (s, 3H), 2.59 (s, 6H), 2.00-2.14 (m, 1H), 1.79-1.88 (m, 2H), 1.64-1.72 (m, 2H), 1.44-1.51 (m, 2H), 1.20-1.36 (m, 6H). LC-MS *m/z* ($M + H^+$): 540.3; retention time: 1.1 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

E. 4-METHOXY_N,2,6-TRIMETHYL-N-(2-{2-[4-(1-METHYL-1-OXIDOPIPERIDIN-4-YL)PIPERAZIN-1-YL]-2-OXOETHOXY}ETHYL)BENZENESULFONAMIDE (COMPOUND 7)



The title compound as a colorless solid is prepared as indicated above. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 6.61 (s, 2H), 4.09 (d, 2H), 3.79 (s, 3H), 3.66 (t, 2H), 3.58 (brs, 2H), 3.39-3.43 (m, 6H), 3.16-3.22 (m, 4H), 2.76 (s, 3H), 2.74 (m, 3H), 2.57 (s, 6H), 2.42 (brs, 2H), 1.68 (brs, 2H). LC-MS *m/z* ($M + H^+$): 513.3; retention time: 1.1 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

F. 2,6-DICHLORO-N-METHYL-N-(2-{2-[4-(1-METHYL-1-OXIDOPIPERIDIN-4-YL)PIPERAZIN-1-YL]-2-OXOETHOXY}ETHYL)BENZENESULFONAMIDE (COMPOUND 8)



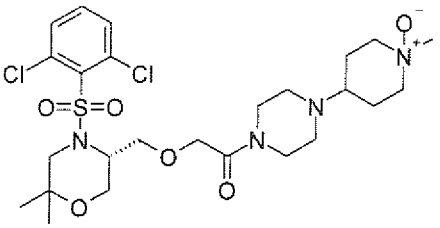
The title compound as a colorless solid is prepared as indicated above. $^1$H-NMR (400 MHz, $CDCl_3$) $\delta$: 7.44 (d, 2H), 7.30 (t, 1H), 4.15 (s, 2H), 3.72-3.75 (m, 2H), 3.60 (brs, 2H), 3.53-3.55 (m, 2H), 3.46 (brs, 3H), 3.36 (d, 2H), 3.15-3.21 (m, 4H), 3.01 (s, 2H), 2.58-2.61 (m, 4H), 2.46-2.50 (m, 4H), 1.69 (d, 2H). LC-MS *m/z* ($M + H^+$): 523.2; retention time: 1.0 min. The $IC_{50}$ (determined as described in Example 7) is 1 micromolar or less.

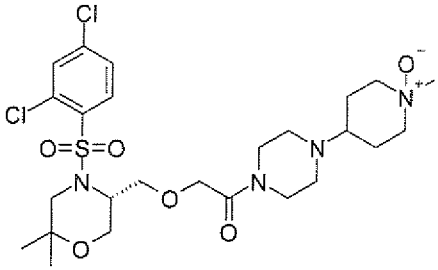
EXAMPLE 4. ADDITIONAL REPRESENTATIVE N-OXIDE ARYL SULFONES AND SULFOXIDES

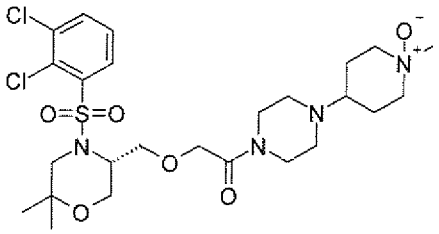
Using routine modifications, the starting materials may be varied and additional steps employed to produce other compounds provided herein. Compounds listed in Table I are prepared using such methods.

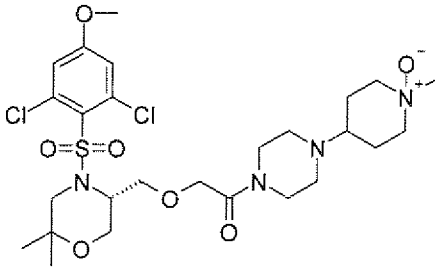
Table I

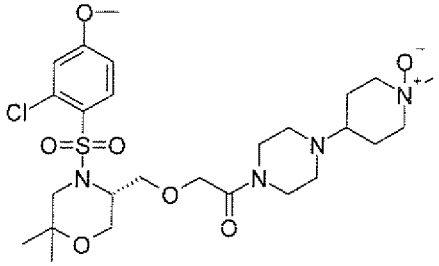
Representative N-Oxide Aryl Sulfones and Sulfoxides

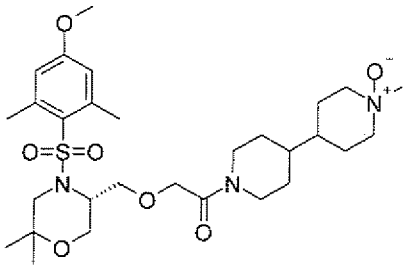
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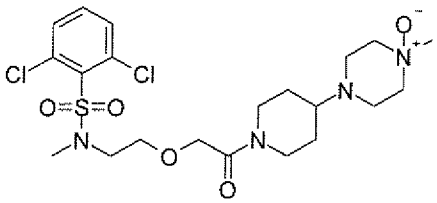
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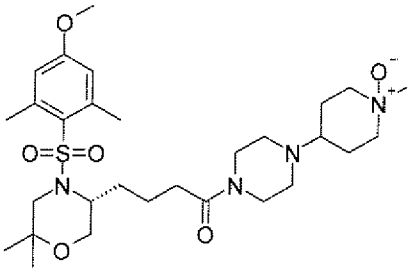
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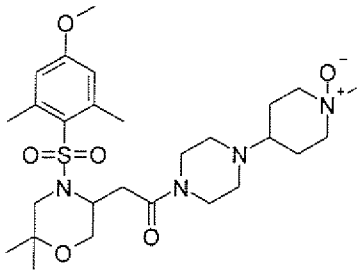
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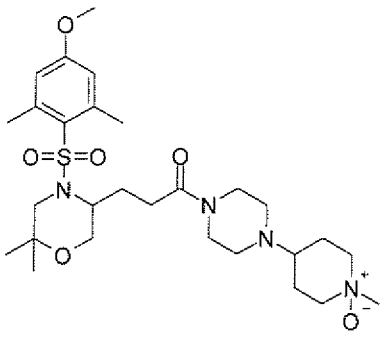
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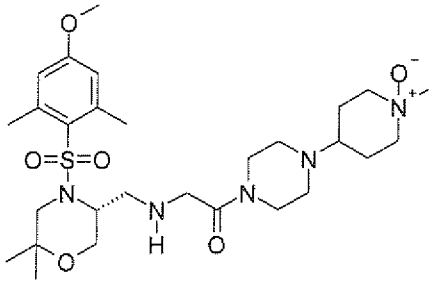
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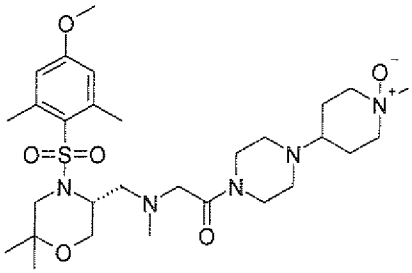
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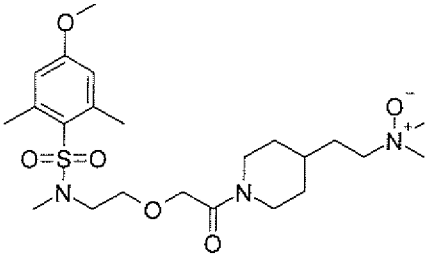
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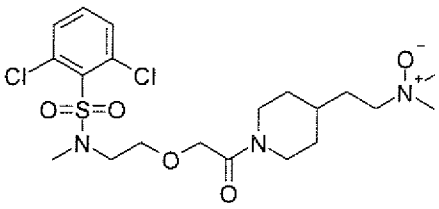
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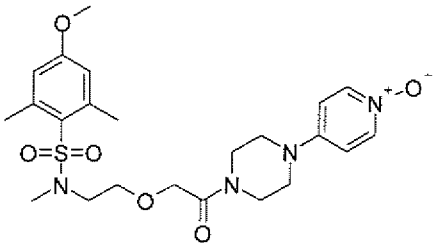
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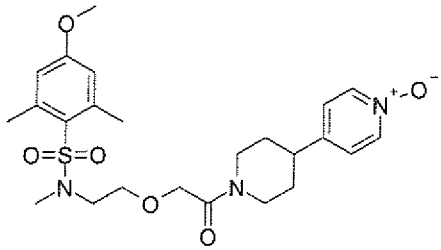
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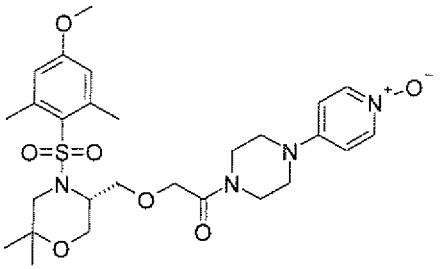
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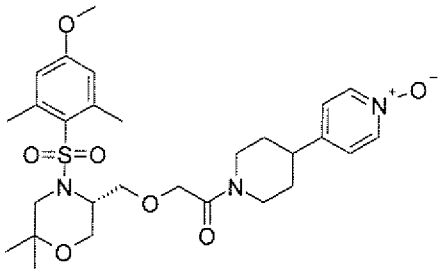
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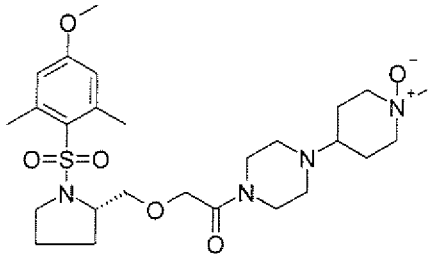
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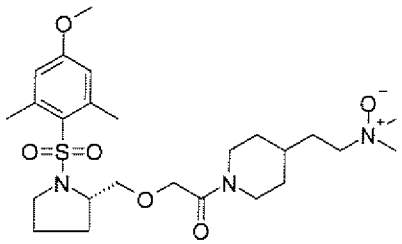
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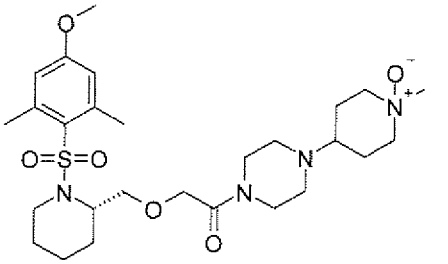
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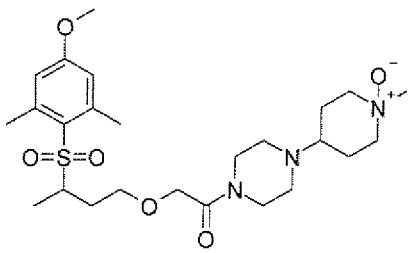
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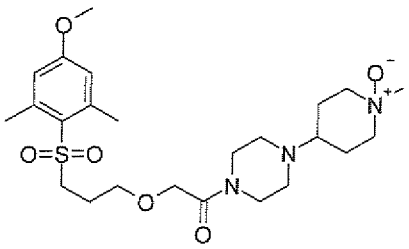
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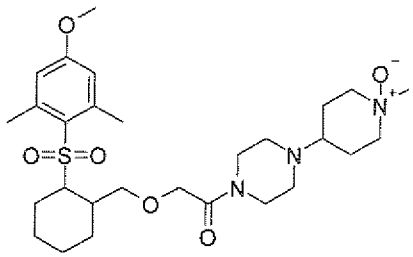
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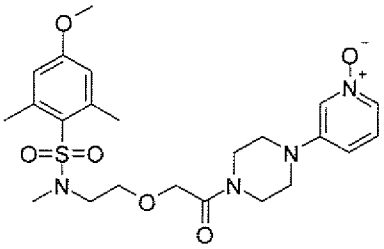
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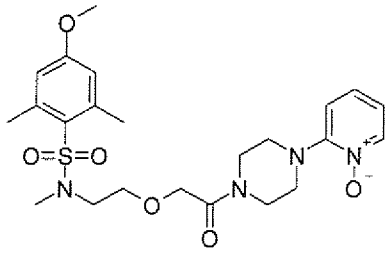
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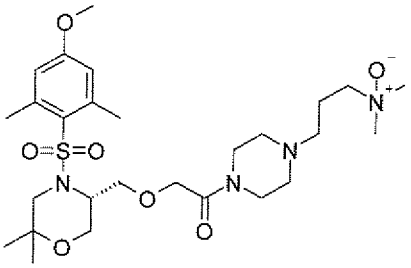
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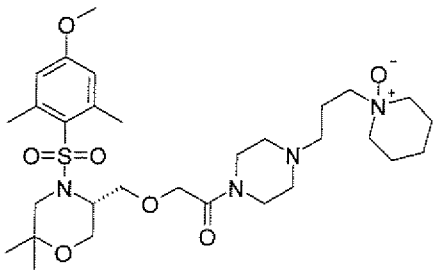
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EXAMPLE 5. PREPARATION OF $B_1$-TRANSFECTED CELLS

This Example illustrates the preparation of $B_1$-transfected cells for use in $B_1$ binding and modulation assays (Examples 6 and 7).

Cynomolgus macaque lung total RNA is isolated as described by Chomzynski et al. (1987) *Anal. Biochem. 162*:156-159. A cDNA encoding $B_1$ is cloned from the total RNA by reverse transcriptase-polymerase chain reaction (RT-PCR) with the following oligonucleotides:

Primer 1: GGCGCTAGCCACCATGGCATCCTGGCCCCCTC (SEQ ID NO:1)

Primer 2: AGCCGTCCCAGATCTGAAC (SEQ ID NO:2)

Primer 3: GATCTGGGACGGCTTGGATG (SEQ ID NO:3)

Primer 4: CGGAGCTCTTAATTCCGCCAGAAAAGTTGGA (SEQ ID NO:4)

Primer pairs 1 & 2 and 3 & 4 are used to generate overlapping cDNA fragments corresponding to the entire protein coding sequence of cynomolgus macaque $B_1$ cDNA are isolated and linked to form the full-length coding sequence (GenBank Accession Number AY788905). The construct is cloned into pcDNA 3.1 (Invitrogen, Carlsbad, CA) and transfected into Chinese hamster ovary (CHO) cells using Lipofectamine (Invitrogen), resulting in cynomolgus macaque $B_1$-expressing CHO cells. Alternatively, the construct is cloned into pBAKPAK9 (Clontech, Mountain View, CA) and transfected into Sf9 cells to generate clonal baculovirus stocks. Clonal cell lines stably expressing the cynomolgus macaque $B_1$ receptor are selected in G418. A single clonal line that exhibits high levels of receptor expression is chosen for use in binding and calcium mobilization assays (Examples 6 and

7). Clonal baculovirus stocks are used to infect Sf9 cells such that the infected cells express high levels of recombinant $B_1$ receptors. These cells are used in radioligand binding assays (Example 6).

## EXAMPLE 6. $B_1$ RECEPTOR BINDING ASSAYS

This Example illustrates a representative $B_1$ receptor binding assay that may be used to determine the binding affinity of compounds for $B_1$.

### A. [3H]-DESARG$^{10}$KALLIDIN BINDING TO INTACT IMR-90 CELLS OR CHO CELLS STABLY EXPRESSING RAT $B_1$

IMR-90 cells, which endogenously express human $B_1$, are seeded into 24 well plates at 65,000 cells per well, cultured overnight, and then treated for 3 h with 0.2 ng/mL interleukin-1 beta to induce $B_1$ expression (Menke, et al. (1994) *J. Biol. Chem.* 269:21583-86). CHO cells stably expressing rat $B_1$ are seeded into 24 well plates at 200,000 cells per well and cultured overnight. The cells are then washed 3 times with phosphate buffered saline (PBS). One hundred fifty microliters of binding buffer (50 mM Tris 7.4, 0.14 mg/mL, bacitracin, and 1 mg/mL BSA) is added to each well. Various concentrations of test compound are added to each well from DMSO solutions such the final DMSO concentration is 1% by volume; some wells receive DMSO only, and some wells receive DMSO plus 10 µM desArg$^{10}$Kallidin to define non-specific binding. All wells then receive 0.3 nM (final concentration) [3H]-desArg$^{10}$Kallidin. The plates are allowed to sit for 2 h at room temperature. Cells are then washed three times, and lysed with 400 µl Ultima Gold scintillation fluid (PerkinElmer; Boston, MA; 20 min incubation). The fluid is then transferred to counting vials counted in a Packard liquid scintillation counter (PerkinElmer). The number of counts present in the scintillation fluid is plotted as a function of antagonist compound concentration and fitted to a logistical equation using SigmaPlot (Systat Software, Point Richmond, CA) to determine each compound's $IC_{50}$ and $K_i$ (*e.g.*, as described by Szallasi, et al. (1993) *J. Pharmacol. Exp. Ther. 266*:678-83).

### B. [3H]-DESARD$^{10}$KALLIDIN BINDING TO MEMBRANE HOMOGENATES OF SF9 CELLS EXPRESSING CYNOMOLGUS MACAQUE $B_1$

Sf9 cells infected with a baculovirus carrying the coding sequence for cynomolgus macaque $B_1$ are harvested by centrifugation and frozen at -80 °C. Pellets are subsequently resuspended on ice in Tris buffered saline (TBS; 50 mM Tris (pH 7.4), 120 mM NaCl), and cells are homogenized using a polytron for 30 seconds. The crude membrane fraction is collected by centrifugation at 20,000 rpm. Membranes are washed two times with TBS and collected by centrifugation each time. Protein content of the membranes is determined after the last wash and the concentration is adjusted to 0.7 µg/uL with binding buffer (50 mM Tris 7.4, 0.14 mg/mL bacitracin, and 1.0 mg/mL BSA). To perform the binding assay, 150 microliters of membrane fraction is added to each well of a 96-well plate along with 50 µl [$^3$H]-desArg$^{10}$Kallidin (0.3 nM final) and test compound in DMSO (final

DMSO concentration = 1%). Some wells receive DMSO only, and some wells receive DMSO plus 10 μM desArg$^{10}$Kallidin to define non-specific binding. The 96 well plates are allowed to sit for 2 h at room temperature. Membrane proteins are then harvested by filtration onto GF/C filtermats (PerkinElmer) pre-soaked for 1 hr in 0.5 % polyethylenimine. After filtration, filters are dried and then counted in a Beta plate counter. The number of counts present in the scintillation fluid is plotted as a function of antagonist compound concentration and fitted to a logistical equation using SigmaPlot (Systat Software, Point Richmond, CA) to determine each compound's $IC_{50}$ and $K_i$ (*e.g.*, as described by Szallasi, *et al.* (1993) *J. Pharmacol. Exp. Ther.* 266:678-83).

## EXAMPLE 7. CALCIUM MOBILIZATION ASSAY

This Example illustrates representative calcium mobilization assays for use in evaluating test compounds for agonist and antagonist activity.

Cynomolgus macaque $B_1$-expressing CHO cells (Example 5) are plated in a 96 well plate. The cells are cultured for 1 day, after which culture media is emptied from the plate and replaced with 50 μl of KRH (Krebs-Ringer HEPES buffer: 25 mM HEPES, 5 mM KCl, 0.96 mM $NaH_2PO_4$, 1 mM $MgSO_4$, 2 mM $CaCl_2$, 5 mM glucose, 1 mM probenecid, pH 7.4) supplemented with the calcium-sensitive fluorescent dye Fluo4-AM (5 μg/ml; Teflabs, Austin, TX). The cells are then incubated at 37 °C in an environment containing 5% $CO_2$. After the 1 hour incubation, the dye solution is removed from the plate, the plate is washed once with KRH, and 100 μL KRH is added.

### DETERMINATION OF $B_1$ AGONIST $EC_{50}$

100 μL KRH + 2% DMSO is added to each well of cells, such that the final volume in each well is 200 microliters and the final DMSO concentration is 1%. Various concentrations of the $B_1$ agonist desArg$^{10}$Kallidin are added. Addition of desArg$^{10}$Kallidin elicits a fluorescent response as consequence of increased intracellular calcium. This response is measured with a FLIPR instrument (Molecular Devices, Sunnyvale, CA) and determined to be desArg$^{10}$Kallidin concentration dependent. A plot of maximum fluorescent response as a function of desArg$^{10}$Kalligin is generated and an $EC_{50}$ (concentration required to elicit a 50% of maximal response) for the response is determined using the equation:

$$y=a*(1/(1+(b/x)^c))$$

In this equation, y is the maximum fluorescence signal, x is the concentration of the $B_1$ agonist, a is the $E_{max}$, b corresponds to the $EC_{50}$ value and c is the Hill coefficient.

By replacing the desArg$^{10}$Kallidin with a test compound, this assay is also used to assess $B_1$ agonist activity of the test compound.

DETERMINATION OF ANTAGONIST ACTIVITY

Various concentrations of test compounds are added to the cell plate prepared as described above in 100 μL KRH + 2% DMSO, such that the final volume in each well is 200 microliters and the final DMSO concentration is 1%. The $EC_{50}$ concentration of desArg$^{10}$Kallidin is then added to each well of plates containing test compound to determine the extent to which each test compound inhibits an agonist-induced $B_1$ response. The maximum fluorescent response is plotted as a function of test compound concentration in order to determine the $IC_{50}$ (concentration required to inhibit 50% of the effect of agonist) for each compound at $B_1$. Antagonists of $B_1$ decrease this response by at least about 20%, preferably by at least about 50%, and most preferably by at least 80%, as compared to matched control (*i.e.,* cells treated with desArg$^{10}$Kallidin at the $EC_{50}$ concentration in the absence of test compound), at a concentration of 10 micromolar or less, preferably 1 micromolar or less.

Alternatively, the data is analyzed as follows. First, the average maximum relative fluorescent unit (RFU) response from negative control wells (no agonist) is subtracted from the maximum response detected for each of the other experimental wells. Second, average maximum RFU response is calculated for the positive control wells (agonist wells). Then, percent inhibition for each compound tested is calculated using the equation:

$$\text{Percent Inhibition} = 100 - 100 \times \left[ \frac{\text{Peak Signal in Test Wells}}{\text{Peak signal in Agonist Wells}} \right]$$

The % inhibition data is plotted as a function of test compound concentration and test compound $IC_{50}$ is determined using a linear regression in which x is ln(concentration of test compound) and y is ln(percent inhibition/(100 - percent inhibition). Data with a percent inhibition that is greater than 90% or less than 15% are rejected and are not used in the regression. The $IC_{50}$ is $e^{(-\text{intercept/slope})}$.

EXAMPLE 8. MDCK CYTOTOXICITY ASSAY

This Example illustrates the evaluation of compound toxicity using a Madin Darby canine kidney (MDCK) cell cytotoxicity assay.

1 μL of test compound is added to each well of a clear bottom 96-well plate (Packard, Meriden, CT) to give final concentration of compound in the assay of 10 μM, 100 μM or 200 μM. Solvent without test compound is added to control wells.

MDCK cells, ATCC no. CCL-34 (American Type Culture Collection, Manassas, VA), are maintained in sterile conditions following the instructions in the ATCC production information sheet. Confluent MDCK cells are trypsinized, harvested, and diluted to a concentration of 0.1 x $10^6$ cells/mL with warm (37°C) medium (VITACELL Minimum Essential Medium Eagle, ATCC catalog # 30-2003). 100 μL of diluted cells is added to each well, except for five standard curve control wells that contain 100 μL of warm medium without cells. The plate is then incubated at 37°C under 95% $O_2$,

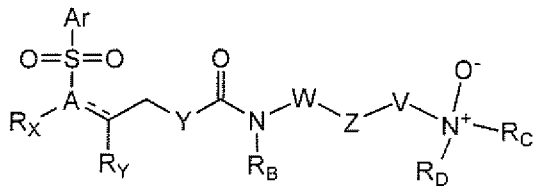
5% $CO_2$ for 2 hours with constant shaking. After incubation, 50 µL of mammalian cell lysis solution (from the Packard (Meriden, CT) ATP-LITE-M Luminescent ATP detection kit) is added per well, the wells are covered with PACKARD TOPSEAL stickers, and plates are shaken at approximately 700 rpm on a suitable shaker for 2 min.

Compounds causing toxicity will decrease ATP production, relative to untreated cells. The ATP-LITE-M Luminescent ATP detection kit is generally used according to the manufacturer's instructions to measure ATP production in treated and untreated MDCK cells. PACKARD ATP LITE-M reagents are allowed to equilibrate to room temperature. Once equilibrated, the lyophilized substrate solution is reconstituted in 5.5 mL of substrate buffer solution (from kit). Lyophilized ATP standard solution is reconstituted in deionized water to give a 10 mM stock. For the five control wells, 10 µL of serially diluted PACKARD standard is added to each of the standard curve control wells to yield a final concentration in each subsequent well of 200 nM, 100 nM, 50 nM, 25 nM, and 12.5 nM. PACKARD substrate solution (50 µL) is added to all wells, which are then covered, and the plates are shaken at approximately 700 rpm on a suitable shaker for 2 min. A white PACKARD sticker is attached to the bottom of each plate and samples are dark adapted by wrapping plates in foil and placing in the dark for 10 min. Luminescence is then measured at 22°C using a luminescence counter (*e.g.,* PACKARD TOPCOUNT Microplate Scintillation and Luminescence Counter or TECAN SPECTRAFLUOR PLUS), and ATP levels calculated from the standard curve. ATP levels in cells treated with test compound(s) are compared to the levels determined for untreated cells. Cells treated with 10 µM of a preferred test compound exhibit ATP levels that are at least 80%, preferably at least 90%, of the untreated cells. When a 100 µM concentration of the test compound is used, cells treated with preferred test compounds exhibit ATP levels that are at least 50%, preferably at least 80%, of the ATP levels detected in untreated cells.

CLAIMS

What is claimed is:

1. A compound of the formula:



or a pharmaceutically acceptable salt or hydrate thereof, wherein:

A is N, C or CH;

==== represents a single or double bond;

Ar is phenyl or a 5- or 6-membered heteroaryl, each of which is substituted with from 0 to 4 substituents independently chosen from $R_1$;

$R_X$ and $R_Y$ are:

(i) independently chosen from:

(a) hydrogen; and

(b) $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl, and ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, each of which is substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, amino and $C_1$-$C_4$alkyl; or

(ii) taken together to form a 4- to 10-membered carbocycle or heterocycle that is substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, halogen, amino and $C_1$-$C_8$alkyl;

Y is a group of the formula $(CH_2)_r$-Q-$(CH_2)_p$, which is substituted with from 0 to 4 substituents independently chosen from (i) amino, hydroxy, cyano, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl and $C_1$-$C_6$haloalkyl; (ii) substituents of the same carbon atom or adjacent carbon atoms that are taken together to form $C_3$-$C_6$cycloalkyl, and (iii) substituents that are taken together with $R_X$ or $R_Y$ to form a 4- to 7-membered carbocycle or heterocycle, wherein:

Q is absent, $CH_2$, CH=CH, O, S or $NR_{10}$, wherein $R_{10}$ is hydrogen or $C_1$-$C_6$alkyl; and

r and p are independently chosen integers ranging from 0 to 6;

W is $C_1$-$C_6$alkylene that is substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

Z is absent, divalent phenyl, $NR_G$ or $CR_ER_F$ wherein:

$R_G$ is hydrogen, $C_1$-$C_6$alkyl or taken together with $R_B$ or $R_D$ to form a 4- to 7-membered heterocycloalkyl that includes W and is substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

$R_E$ is hydrogen, $C_1$-$C_6$alkyl or taken together with $R_B$ to form a 4- to 7-membered heterocycloalkyl that includes W and is substituted with from 0 to 4 substituents independently chosen from $R_{20}$; and

$R_F$ is hydrogen, $C_1$-$C_6$alkyl or taken together with $R_D$ to form a 4- to 7-membered heterocycloalkyl that includes V and is substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

V is absent or $C_1$-$C_6$alkylene that is substituted with from 0 to 4 substituents independently chosen from (i) $R_{20}$ and (ii) groups that are taken together with $R_D$ to form a 4- to 7-membered heterocycle that is substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

Each $R_I$ is independently chosen from:

(i) halogen, hydroxy, cyano, amino, nitro, aminocarbonyl, aminosulfonyl and –COOH;

(ii) $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_1$-$C_6$alkoxy, $C_1$-$C_6$alkylthio, $C_1$-$C_6$alkylsulfinyl, $C_1$-$C_6$alkoxycarbonyl, $C_1$-$C_6$alkylsulfonyl$C_0$-$C_4$alkyl, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, mono- or di-($C_1$-$C_6$alkyl)aminosulfonyl$C_0$-$C_4$alkyl, mono- or di-($C_1$-$C_6$alkyl)aminocarbonyl$C_0$-$C_4$alkyl, and (4- to 8-membered heterocycloalkyl)$C_0$-$C_4$alkyl; each of which is substituted with from 0 to 6 substituents independently chosen from halogen, hydroxy, cyano and amino; and

(iii) groups that are taken together to form a fused carbocyclic ring that is substituted with from 0 to 4 substituents independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl;

$R_B$ is:

(i) hydrogen, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_2$-$C_6$alkyl ether, mono- or di-($C_1$-$C_6$alkyl)amino$C_1$-$C_4$alkyl, or (4- to 7-membered heterocycloalkyl)$C_0$-$C_4$alkyl, each of which is substituted with from 0 to 6 substituents independently chosen from:

(a) amino, halogen, hydroxy, cyano and oxo; and

(b) $C_1$-$C_6$alkyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_1$-$C_6$alkoxy, phenyl$C_0$-$C_4$alkyl and (5- or 6-membered heterocycle)$C_0$-$C_4$alkyl, each of which is substituted with from 0 to 4 substituents independently chosen from amino, cyano, halogen, hydroxy, $C_1$-$C_6$alkyl, ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, $C_1$-$C_6$alkoxycarbonyl, mono- or di-($C_1$-$C_6$alkyl)amino, phenyl$C_0$-$C_4$alkyl and phenyl$C_0$-$C_4$alkoxy; or

(ii) taken together with $R_E$ or $R_G$ to form an optionally substituted heterocycloalkyl;

$R_C$ is absent, $C_1$-$C_6$alkyl or taken together with $R_D$ to from a 4- to 7-membered heterocycle that is substituted with from 0 to 4 substituents independently chosen from $R_{20}$;

$R_D$ is $C_1$-$C_6$alkyl or taken together with $R_C$, $R_F$, $R_G$ or a substituent of V to form an optionally substituted 4- to 7-membered heterocycle; and

Each $R_{20}$ is independently selected from:

(i) hydroxy, oxo, cyano and amino; and

(ii) $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl, each of which is substituted with from 0 to 2 substituents independently chosen from hydroxy, halogen, oxo, cyano, $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl.

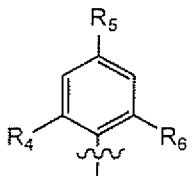
2. A compound or salt or hydrate thereof according to claim 1, wherein Ar is phenyl, naphthyl, pyridyl or pyrimidinyl, each of which is substituted with from 0 to 4 substituents independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl.

3. A compound or salt or hydrate thereof according to claim 1, wherein Ar is phenyl that is substituted with from 0 to 4 substituents independently chosen from $R_1$.

4. A compound or salt or hydrate thereof according to claim 2, wherein Ar is phenyl that is substituted with 1, 2 or 3 substituents independently chosen from halogen, hydroxy, cyano, amino, nitro, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, $C_1$-$C_6$haloalkoxy and mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl.

5. A compound or salt or hydrate thereof according to claim 1, wherein each substituent represented by $R_1$ is independently chosen from halogen, hydroxy, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy and $C_1$-$C_6$haloalkoxy.

6. A compound or salt or hydrate thereof according to claim 1, wherein Ar is




;

wherein $R_4$, $R_5$ and $R_6$ are independently chosen from halogen, hydroxy, $C_1$-$C_6$alkyl, $C_1$-$C_6$haloalkyl, $C_1$-$C_6$alkoxy, and $C_1$-$C_6$haloalkoxy.

7. A compound or salt or hydrate thereof according to claim 6, wherein $R_5$ is methyl or methoxy and $R_4$ and $R_6$ are each methyl.

8. A compound or salt or hydrate thereof according to any one of claims 1-7, wherein the sum of r and p ranges from 1 to 6.

9. A compound or salt or hydrate thereof according to claim 8, wherein p is not zero.

10. A compound or salt or hydrate thereof according to any one of claims 1-9, wherein Y is a group of the formula $(CH_2)_r$-O-$(CH_2)_p$ that is substituted with from 0 to 4 substituents independently chosen from amino, hydroxy, cyano, $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl and substituents of the same carbon atom or adjacent carbon atoms that are taken together to form $C_3$-$C_6$cycloalkyl.

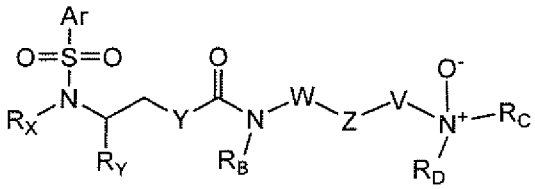
11. A compound or salt or hydrate thereof according to claim 10, wherein Y is –O-$CH_2$–, –$CH_2$-O-$CH_2$–, –O-$CH_2$-$CH_2$–, –$CH_2$-O-$CH_2$-$CH_2$– or –$CH_2$-$CH_2$-O-$CH_2$–.

12. A compound or salt or hydrate thereof according to claim 11, wherein Y is –O-$CH_2$–.

13. A compound or salt or hydrate thereof according to any one of claims 1-9, wherein Y is–N($R_{10}$)-$CH_2$–, –$CH_2$-$CH_2$–, –CH=CH–, or –$CH_2$-$CH_2$-$CH_2$–.

14. A compound or salt or hydrate thereof according to any one of claims 1-13, wherein $R_X$ and $R_Y$ are independently chosen from hydrogen and $C_1$-$C_6$alkyl.

15. A compound or salt or hydrate thereof according to anyone of claims 1-14, wherein the compound has the formula:




wherein $R_X$ and $R_Y$ are:

(i) independently chosen from:

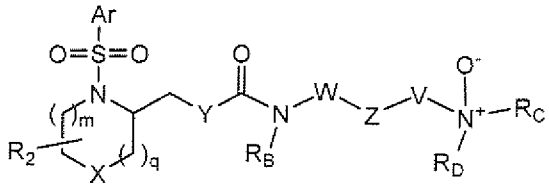
(a) hydrogen; and

(b) $C_1$-$C_6$alkyl, $C_2$-$C_6$alkenyl, $C_2$-$C_6$alkynyl, $C_1$-$C_6$haloalkyl, and ($C_3$-$C_8$cycloalkyl)$C_0$-$C_4$alkyl, each of which is substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, amino and $C_1$-$C_4$alkyl; or

(ii) taken together to form a 4- to 10-membered heterocycle that is substituted with from 0 to 4 substituents independently chosen from oxo, hydroxy, halogen, amino and $C_1$-$C_8$alkyl.

16. A compound or salt or hydrate thereof according to claim 15, wherein $R_X$ and $R_Y$ are taken together to form an optionally substituted 4- to 8-membered heterocycloalkyl.

17. A compound or salt or hydrate thereof according to claim 16, wherein the compound satisfies the formula:



wherein:

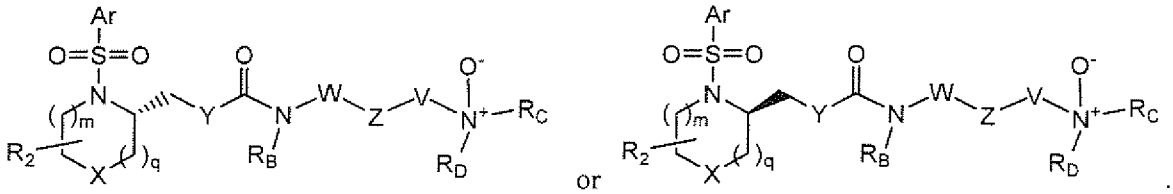
X is $CH_2$, $NR_3$, O, S, SO or $SO_2$;

m and q are independently 0, 1 or 2;

$R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_6$alkyl; and

$R_3$ is hydrogen, $C_1$-$C_6$alkyl or $C_1$-$C_6$alkanoyl.

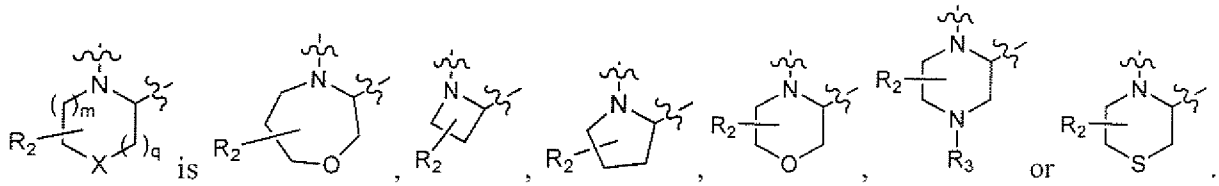
18. A compound or salt or hydrate thereof according to claim 17, wherein the compound satisfies the formula:



19. A compound or salt or hydrate thereof according to claim 17 or claim 18, wherein X is $CH_2$.

20. A compound or salt or hydrate thereof according to claim 17 or claim 18, wherein X is O.

21. A compound or salt or hydrate thereof according to claim 17 or claim 18, wherein:



22. A compound or salt or hydrate thereof according to any one of claims 17-21, wherein $R_2$ represents from 0 to 4 substituents independently chosen from $C_1$-$C_2$alkyl.

23. A compound or salt or hydrate thereof according to claim 22, wherein $R_2$ represents gem-dimethyl.

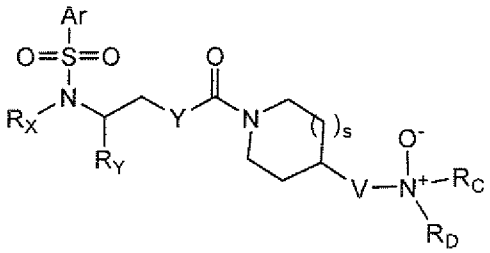
24. A compound or salt or hydrate thereof according to any one of claims 1-23, wherein Z is $NR_G$ or $CR_ER_F$.

25. A compound or salt or hydrate thereof according to claim 24, wherein $R_E$ is taken together with $R_B$ to form a 4- to 7-membered heterocycloalkyl that is substituted with from 0 to 4 substituents independently chosen from:

(i) hydroxy, oxo, cyano and amino; and

(ii) $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl; each of which is substituted with from 0 to 2 substituents independently chosen from hydroxy, halogen, oxo, cyano, $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl.

26. A compound or salt or hydrate thereof according to claim 25, wherein the compound satisfies the formula:



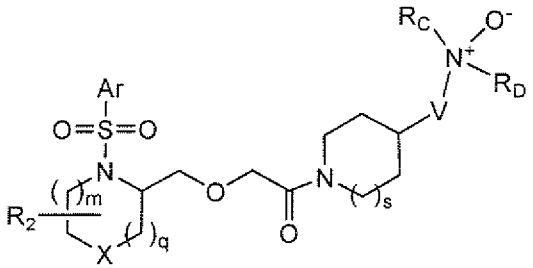
wherein:

s is 0, 1 or 2;

$R_D$ is $C_1$-$C_6$alkyl; and

V is absent or $C_1$-$C_4$alkylene.

27. A compound or salt or hydrate thereof according to claim 26, wherein the compound satisfies the formula:



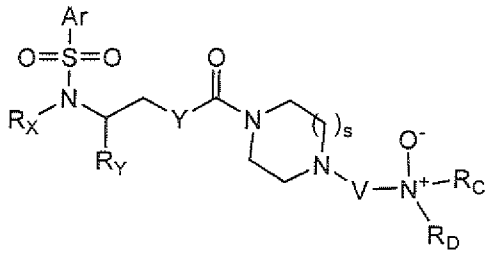
wherein:

m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3;

X is $NR_3$, O, S, SO or $SO_2$; and

$R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_6$alkyl.

28. A compound or salt or hydrate thereof according to claim 25, wherein the compound satisfies the formula:



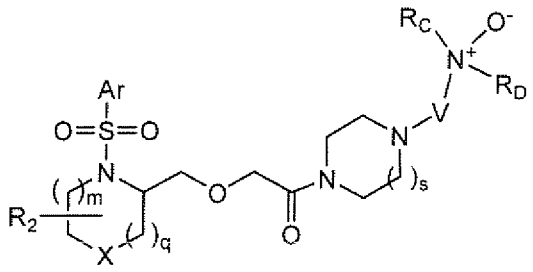
wherein:

s is 0, 1 or 2;

$R_D$ is $C_1$-$C_6$alkyl; and

V is $C_1$-$C_4$alkylene.

29. A compound or salt or hydrate thereof according to claim 28, wherein the compound satisfies the formula:



wherein:

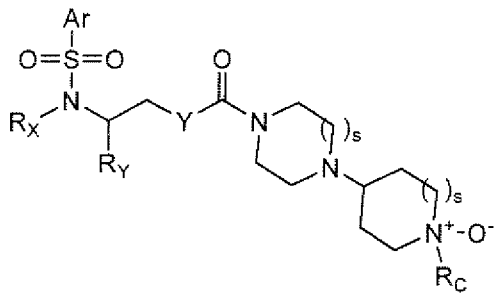
m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3;

X is $NR_3$, O, S, SO or $SO_2$; and

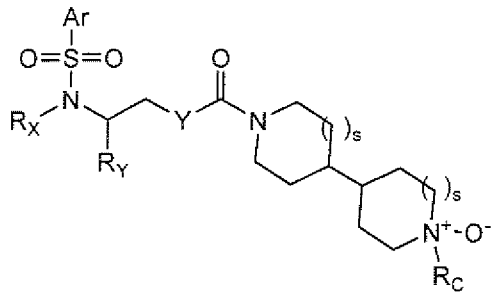
$R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_8$alkyl.

30. A compound or salt or hydrate thereof according to claim 26 or claim 28, wherein $R_X$ and $R_Y$ are independently hydrogen or $C_1$-$C_6$alkyl.

31. A compound or salt or hydrate thereof according to claim 25, wherein the compound satisfies the formula:

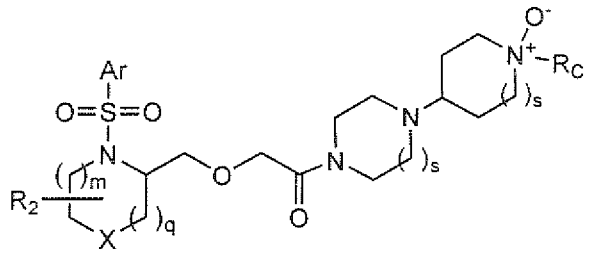
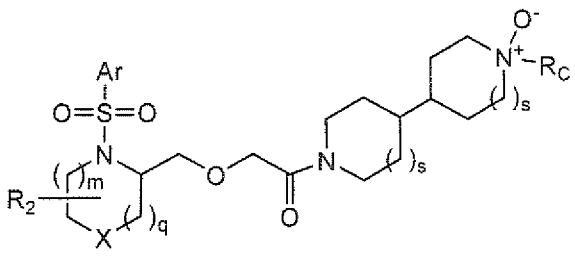


or



wherein each s is independently 0, 1 or 2.

32. A compound or salt or hydrate thereof according to claim 31, wherein the compound satisfies the formula:


or


wherein:

m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3;

X is $NR_3$, O, S, SO or $SO_2$; and

$R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_8$alkyl.

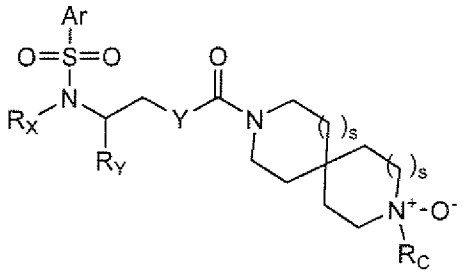
33. A compound or salt or hydrate thereof according to claim 31, wherein $R_X$ and $R_Y$ are independently hydrogen or $C_1$-$C_6$alkyl.

34. A compound or salt or hydrate thereof according to claim 24, wherein Z is $CR_ER_F$ and $R_F$ is taken together with $R_D$ to form a 4- to 7-membered heterocycloalkyl that is substituted with from 0 to 4 substituents independently chosen from:

(i) hydroxy, oxo, cyano and amino; and

(ii) $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, mono- or di-($C_1$-$C_6$alkyl)amino$C_0$-$C_4$alkyl, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl; each of which is substituted with from 0 to 2 substituents independently chosen from hydroxy, halogen, oxo, cyano, $C_1$-$C_6$alkyl, $C_1$-$C_6$alkoxy, ($C_3$-$C_{10}$carbocycle)$C_0$-$C_4$alkyl and (4- to 10-membered heterocycle)$C_0$-$C_4$alkyl.

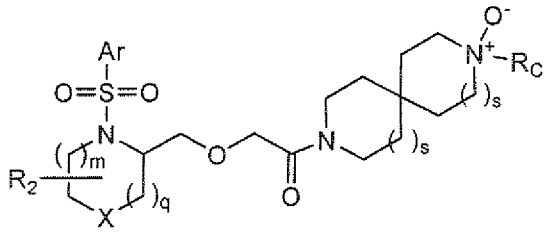
35. A compound or salt or hydrate thereof according to claim 34, wherein the compound satisfies the formula:



wherein s is 0, 1 or 2.

36. A compound or salt or hydrate thereof according to claim 35, wherein $R_X$ and $R_Y$ are independently hydrogen or $C_1$-$C_6$alkyl.

37. A compound or salt or hydrate thereof according to claim 35, wherein the compound satisfies the formula:



wherein:

m and q are independently 0, 1 or 2, such that the sum of m and q is 1, 2 or 3;

X is $NR_3$, O, S, SO or $SO_2$; and

$R_2$ represents from 0 to 4 substituents independently chosen from oxo, hydroxy and $C_1$-$C_8$alkyl.

38. A compound or salt or hydrate thereof according to claim 24, wherein Z is $NR_G$ and $R_G$ is hydrogen or $C_1$-$C_6$alkyl.

39. A compound or salt or hydrate thereof according to claim 1, wherein the compound is:

4-Methoxy-N,2,6-trimethyl-N-{2-[2-(9-methyl-9-oxido-3,9-diazaspiro[5.5]undec-3-yl)-2-oxoethoxt]ethyl}benzenesulfonamide;

4-Methoxy-N,2,6-trimethyl-N-{2-[2-(2-methyl-2-oxido-2,8-diazaspiro[4.5]dec-8-yl)-2-oxoethoxy]ethyl}benzenesulfonamide;

(5R)-4-[(4-Methoxy-2,6-dimethylphenyl)sulfonyl]-2,2-dimethyl-5-({2-[4-(1-methyl-1-oxidopiperidin-4-yl)piperazin-1-yl]-2-oxoethoxy}methyl)morpholine;

{1-[({(3R)-4-[(4-methoxy-2,6-dimethylphenyl)sulfonyl]-6,6-dimethylmorpholin-3-yl}methoxy)acetyl]piperidin-4-yl}dimethylamine oxide;

(2-{1-[({(3R)-4-[(4-methoxy-2,6-dimethylphenyl)sulfonyl]-6,6-dimethylmorpholin-3-yl}methoxy)acetyl]piperidin-4-yl}ethyl)dimethylamine oxide;

N-[2-(2-{9-[(Dimethylnitroryl)methyl]-3-azaspiro[5.5]undec-3-yl}-2-oxoethoxy)ethyl]-4-methoxy-N,2,6-trimethylbenzenesulfonamide;

4-Methoxy_N,2,6-trimethyl-N-(2-{2-[4-(1-methyl-1-oxidopiperidin-4-yl)piperazin-1-yl]-2-oxoethoxy}ethyl)benzenesulfonamide; or

2,6-Dichloro-N-methyl-N-(2-{2-[4-(1-methyl-1-oxidopiperidin-4-yl)piperazin-1-yl]-2-oxoethoxy}ethyl)benzenesulfonamide.

40. A compound or salt or hydrate thereof according to any one of claims 1-39wherein the compound exhibits no detectable agonist activity an *in vitro* assay of $B_1$ agonism.

41. A compound or salt or hydrate thereof according to any one of claims 1-40wherein the compound has an $IC_{50}$ value of 1 micromolar or less in an *in vitro* assay of $B_1$ antagonism.

42. A pharmaceutical composition, comprising at least one compound or salt or hydrate thereof according to any one of claims 1-41 in combination with a physiologically acceptable carrier or excipient.

43. A pharmaceutical composition according to claim 42 wherein the composition is formulated as an injectible fluid, an aerosol, a cream, a gel, a pill, a capsule, a syrup or a transdermal patch.

44. A method for inhibiting induction of agonist-induced $B_1$ activity *in vitro*, the method comprising contacting $B_1$ receptor with at least one compound or salt or hydrate thereof according to any one of claims 1-41 under conditions and in an amount sufficient to detectably inhibit agonist-induced $B_1$ activity.

45. A method for inhibiting induction of agonist-induced $B_1$ activity in a patient, comprising contacting cells expressing $B_1$ receptor with at least one compound or salt or hydrate thereof according to any one of claims 1-41, in an amount sufficient to detectably inhibit agonist-induced $B_1$ activity in cells expressing a cloned $B_1$ receptor *in vitro*, and thereby inhibiting agonist-induced $B_1$ activity in the patient.

46. A method according to claim 45, wherein the patient is a human.

47. A method for treating a condition responsive to $B_1$ receptor modulation in a patient, comprising administering to the patient a therapeutically effective amount of at least one compound or salt or hydrate thereof according to any one of claims 1-41, and thereby alleviating the condition in the patient.

48. A method according to claim 47, wherein the condition is inflammation or pain.

49. A method according to claim 47, wherein the condition is cough, asthma, vascular edema or epilepsy.

50. A method for treating pain in a patient, comprising administering to a patient suffering from pain a therapeutically effective amount of at least one compound or salt or hydrate thereof according to any one of claims 1-41, and thereby alleviating pain in the patient.

51. A method according to claim 50, wherein the patient is suffering from inflammatory pain, acute pain, dental pain, back pain, surgical pain, headache, neuropathic pain or pain from osteoarthritis or trauma.

52. A method according to claim 50, wherein the patient is a human.

53. A compound or salt or hydrate thereof according to claim 1, wherein the compound is radiolabeled.

54. A method for determining the presence or absence of $B_1$ receptor in a sample, comprising the steps of:

(a) contacting a sample with a compound or salt or hydrate thereof according to any one of claims 1-41, under conditions that permit binding of the compound to $B_1$ receptor; and

(b) detecting a signal indicative of a level of the compound bound to $B_1$ receptor, and therefrom determining the presence or absence of $B_1$ receptor in the sample.

55. A method according to claim 54, wherein the compound is radiolabeled, and wherein the step of detection comprises the steps of:

(i) separating unbound compound from bound compound; and

(ii) detecting the presence or absence of bound radiolabel in the sample.

56. A packaged pharmaceutical preparation, comprising:

(a) a pharmaceutical composition according to claim 42 in a container; and

(b) instructions for using the composition to treat pain.

57. The use of a compound or salt or hydrate thereof according to any one of claims 1-41 for the manufacture of a medicament for the treatment of a condition responsive to $B_1$ receptor modulation.

58. A use according to claim 57, wherein the condition is an inflammatory condition or pain.

# INTERNATIONAL SEARCH REPORT

International application No.
PCT/US07/76266

A. CLASSIFICATION OF SUBJECT MATTER
IPC: **C07D 471/10**( 2006.01)

USPC: 544/70
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. : 544/70

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 practicable, search terms used)
CAS ONLINE, STN structure search

C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category * | Citation of document, with indication, where appropriate, of the relevant passages | Relevant to claim No. |
|---|---|---|
| A, P | Database CAS ONLINE on STN, Chem. Abstr., Accession No. 2006:1124909, WO 2006113140 A2 (ELAN PHARMACEUTICALS, INC., USA) 26 October 2006 (26.10.2006), abstract. | 39 |

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

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"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)

"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

"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

"&" document member of the same patent family

| Date of the actual completion of the international search | Date of mailing of the international search report |
|---|---|
| 06 November 2007 (06.11.2007) | 20 NOV 2007 |
| Name and mailing address of the ISA/US<br>Mail Stop PCT, Attn: ISA/US<br>Commissioner for Patents<br>P.O. Box 1450<br>Alexandria, Virginia 22313-1450<br>Facsimile No. (571) 273-3201 | Authorized officer<br>Rebecca L. Anderson<br>Telephone No. (571) 272-0696 |

Form PCT/ISA/210 (second sheet) (April 2005)

# INTERNATIONAL SEARCH REPORT

International application No.
PCT/US07/76266

**Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 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:

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

2. ☒ Claims Nos.: 1-38 and 40-58
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:
Please See Continuation Sheet

3. ☒ Claims Nos.: 10-38,40-52 and 54-58
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

**Box No. III Observations where unity of invention is lacking (Continuation of item 3 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 additional fees, this Authority did not invite payment of any additional fees.
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 and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

Continuation of Box II Reason 2:
The numerous variables, e.g. A, Ar, Rx, Ry, Y, W, Z, R3, Rf, V, R1, Rb, Rc, Rd, R20, Q, R10, r, p, etc. and their voluminous, complex meanings and their virtual incomprehensible permutations and combinations make it impossible to determine the full scope and complete meaning of the claimed subject matter. As presented the claimed subject matter cannot be regarded as being a clear and concise description for which protection is sought and as such the listed claims do not comply with the requirements of PCT Article 6. Thus it is impossible to carry out a meaningful search on same. A search will be carried out on the first discernable invention of claim 39 which is the first listed compound.