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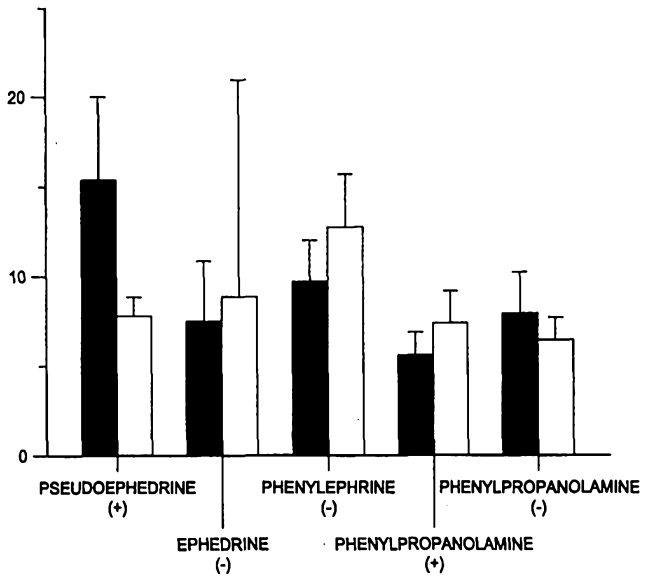

(57) Abstract

The present invention provides pharmaceutical compositions which include (-)-phenylpropanolamine and a pharmaceutically acceptable carrier, wherein the (-)-phenylpropanolamine is substantially free of (+)-phenylpropanolamine. In another embodiment, the present invention provides methods of relieving nasal and bronchial congestion and of inducing pupil dilation which include administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal. The (-)-phenylpropanolamine used in the present methods is substantially free of (+)-phenylpropanolamine, and of the adverse side effects associated with administration of (+)-phenylpropanolamine.

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(-)-PHENYLPROPANOLAMINE AS A SYMPATHOMIMETIC DRUG

FIELD OF THE INVENTION:

The present application provides pharmaceutical compositions and methods of using the sympathomimetic composition of (-)-phenylpropanolamine as a decongestant, physiological antagonist of histamine, mydriatic agent, and for treating other conditions typically treated with sympathomimetic drugs. The present compositions of (-)-phenylpropanolamine are substantially-free of (+)-phenylpropanolamine. As a result, the present compositions and methods have increased potency and reduced adverse side effects. Adverse side effects avoided by the present invention include drug interactions, central nervous system stimulation and depression. According to the present invention, the potency of the present (-)-phenylpropanolamine compositions is greater than (+)-phenylpropanolamine alone, or a racemic mixture of (+)- and (-)-phenylpropanolamine, and the (-) isomer of phenylpropanolamine can be used at lower doses, for example, to provide improved decongestion therapy.

BACKGROUND OF THE INVENTION:

Sympathomimetic drugs are structurally and pharmacologically related to amphetamine. They act by binding to or activating α- and β-adrenergic receptors, resulting in vascular constriction, reduced blood flow and/or reduced secretion of fluids into the surrounding tissues. Such receptor binding generally decreases the amount of mucous secreted into nasal passages. Sympathomimetic drugs are therefore used to treat nasal congestion, allergies and colds. In addition, because they can influence the activity of the central nervous system, sympathomimetic drugs are also used as appetite suppressants. They are known as mydriatic agents, meaning that they dilate the eye.

At the present time, sympathomimetic drugs often are sold as racemic mixtures. However, one stereoisomer may interact more selectively than the other with the receptors involved in sympathomimetic action. Isolation and use of the more selective stereoisomer may therefore reduce not only the required dosage, but many unwanted side effects.

Many organic compounds exist in optically active forms. This means that they have the ability to rotate the plane of plane-polarized light. An optically active compound is often described as a chiral compound. Such a chiral compound has at least one asymmetric carbon atom which can exist in two different, mirror-image configurations. Compounds which have the same composition but are mirror images of each other are called enantiomers. The prefixes *d* and *l*, or (+) and (-), identify the direction in which an enantiomer rotates light. The *d* or (+) enantiomer is dextrorotatory. In contrast, the *l* or (-) enantiomer is levorotatory. A mixture of (+) and (-) enantiomers is called a racemic mixture. An alternative classification system for stereoisomers uses prefixes (S) and (R).

This classification system is based on the structure of the compound rather than on the optical activity of the compound.

Alpha-(1-aminoethyl)benzenemethanol is a molecule with two chiral, or optically active, centers. As such, this compound has four stereoisomers, occurring as two pairs of enantiomers. One pair of enantiomers is called "phenylpropanolamine" or "norephedrine." The other pair is called "norpseudoephedrine," "nordefrin" or "pseudonorephrine."

The subject matter of this application relates to phenylpropanolamine (norephedrine), which has two enantiomers, (+)-phenylpropanolamine and (-)-phenylpropanolamine. The structures of the enantiomers of phenylpropanolamine and norpseudoephedrine are depicted below.

According to the present invention, and the rules of structural chemistry, (-)-phenylpropanolamine is (1R, 2S)-(-)-phenylpropanolamine.

The racemic mixture of (+)- and (-)-phenylpropanolamine, is known as a sympathomimetic amine which can be used as a decongestant or anoretic. However, the racemic mixture has undesirable side effects -- it may be contraindicated in patients having glaucoma and is known to stimulate the central nervous system. 95 AMERICAN HOSPITAL FORMULATORY SERVICE 846. Moreover, according to the present invention, the (-)-isomer of phenylpropanolamine does not interact with other drugs, for example, with antihistamines. Hence, a need exists for a composition having the beneficial decongestant and mydriatic activities of the racemic mixture of (+)- and (-)-phenylpropanolamine, without its undesirable side effects.

SUMMARY OF THE INVENTION:

According to the present invention, the (-) enantiomer of phenylpropanol-amine is a much more potent decongestant, and a better antagonist of histamine than is (+)-phenylpropanolamine but does not cause central nervous system stimulation, and does not interact with other drugs. Hence, the present compositions of (-)-phenylpropanolamine can be provided in lower dosages than the racemic mixture, and will, surprisingly, counteract the physiological effects of histamine as well as act as a decongestant, without causing the adverse side effects of the racemic mixture or the (+)-enantiomer.

The present invention is directed to a pharmaceutical composition containing (-)-phenylpropanolamine and a pharmaceutically acceptable carrier, wherein the pharmaceutical composition is substantially-free of (+)-phenylpropanolamine. The pharmaceutical composition has (-)-phenylpropanolamine in a therapeutic dosage suitable for treating nasal or bronchial congestion, counteracting the physiological effects of histamine, dilating the pupil, treating attention deficit hyperactivity disorder (ADHD), suppressing the appetite and for treating other conditions typically treated with

sympathomimetic drugs. Upon administration to a mammal in a therapeutically effective amount, the present compositions have reduced side effects relative to administration of (+)-phenylpropanolamine, or a racemic phenylephrine mixture of (+)- and (-)-phenylpropanolamine. Side effects caused by administration of (+)-phenylpropanolamine, or the racemic mixture of (+)- and (-)-phenylpropanolamine, include increased central nervous system stimulation and depression, and increased intraocular pressure and drug interactions, for example, with antihistamines.

The present invention is also directed to a method of relieving nasal and bronchial congestion which includes administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein such (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine. This method permits administration of less drug than a method which includes administration of a racemic mixture of (+)- and (+)-phenylpropanolamine, or a composition of (+)-phenylpropanolamine alone. In this embodiment, a therapeutically effective amount of (-)-phenylpropanolamine is a dosage suitable for treating nasal and/or bronchial congestion.

The present invention is also directed to a method of antagonizing the physiological effects of histamine which includes administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein such (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine. According to the present invention, (-)-phenylpropanolamine is surprisingly a more potent physiological antagonist of histamine than is (+)-phenylpropanolamine. This method has less side effects than a method which includes administration of a racemic mixture of (+)- and (+)-phenylpropanolamine, or a composition of (+)-phenylpropanolamine alone. In this embodiment, a therapeutically effective amount of (-)-phenylpropanolamine is a dosage suitable for relieving the physiological effects of histamine, for example, nasal congestion, inflammation and allergic responses.

The present invention is further directed to a method of dilating the pupil which includes administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein the (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine. The (-)-phenylpropanolamine is preferably administered topically. In this embodiment, a therapeutically effective amount of (-)-phenylpropanolamine is a dosage suitable for dilating the eye pupil. This method has less side effects than does a method which includes administration of a racemic mixture of (+)- and (-)-phenylpropanolamine, or (+)-phenylpropanolamine alone, in that it avoids substantial increases in intraocular pressure.

The present invention is further directed to a method of treating attention deficit hyperactivity disorder which includes administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein the (-)-phenylpropanolamine is substantially-

free of (+)-phenylpropanolamine. In this embodiment, a therapeutically effective amount of (-)-phenylpropanolamine is a dosage suitable for relieving the symptoms of attention deficit disorder. This method has less side effects than does a method which includes administration of a racemic mixture of (+)- and (-)-phenylpropanolamine, or (+)-phenylpropanolamine alone.

The present invention is further directed to a method of suppressing the appetite which includes administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein the (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine. In this embodiment, a therapeutically effective amount of (-)-phenylpropanolamine is a dosage suitable for suppressing or depressing the appetite. This method requires a smaller dosage and has less side effects than does a method which includes administration of a racemic mixture of (+)- and (-)-phenylpropanolamine, or (+)-phenylpropanolamine alone.

The present invention is also directed to a method of treating conditions typically treated with sympathomimetic drugs, which includes administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein such (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine. This method requires administration of less drug than does a method which includes administration of a composition of (+)- and (-)-phenylpropanolamine, or of (+)-phenylpropanolamine alone. In this embodiment, a therapeutically effective amount of (-)-phenylpropanolamine is a dosage suitable for treating the condition typically treated with a sympathomimetic drug.

BRIEF DESCRIPTIONS OF THE DRAWINGS:

Figure 1 depicts the mean arterial blood pressure after administration of a "75% dose" of the indicated enantiomers, with (stippled bar) and without (solid bar) histamine treatment. The "75% dose" is 75% of the dosage required to produce a 10% increase in mean arterial blood pressure. As indicated, (-)-phenylpropanolamine causes a lesser increase in blood pressure at this 75% dose than is observed for several enantiomers which are commercially available as decongestants.

Figure 2 provides the observed percent nasal airway pressure after a 75% dose of the different enantiomers, with (stippled bar) and without (solid bar) histamine treatment. The "75% dose" is 75% of the dosage required to produce a 10% increase in mean arterial blood pressure. Even at this reduced dosage, the (-) enantiomer of phenylpropanolamine is more effective than the (+) enantiomer.

DETAILED DESCRIPTION OF THE INVENTION:

The present invention provides pharmaceutical compositions of (-)-phenylpropanolamine and a pharmaceutically acceptable carrier that are substantially free of

(+)-phenylpropanolamine. The present invention also provides methods of using such (-)-phenylpropanolamine compositions for treating colds, nasal congestion, bronchial congestion, histamine-related inflammations, allergies, attention deficit disorder, and for other conditions typically treated with sympathomimetic drugs. The present invention also provides methods for dilating the pupil. Accordingly, the present compositions of (-)-phenylpropanolamine can be used as decongestants, bronchodilators, physiological antagonists of histamine, agents for treating attention deficit disorder, mydriatric agents, and the like.

The structures of the free amines of (+)-phenylpropanolamine and (-)-phenylpropanolamine are: CAS #: 37577-28-9, [S-(R*, S*)]-alpha- CAS #: 492-41-1, (1-aminoethyl)benzenemethanol, [R-(R*, S*)]-alpha-(1-aminoethyl)benzenemethanol.

As used herein, the term "substantially free of (+)-phenylpropanolamine" means that the composition contains at least 90% (-)-phenylpropanolamine and 10% or less (+)-phenylpropanolamine. In a more preferred embodiment, "substantially free of (+)-phenylpropanolamine" means that the composition contains at least 95% (-)-phenylpropanolamine and 5% or less (+)-phenylpropanolamine: Still more preferred is an embodiment wherein the pharmaceutical composition contains 99% or more (-)-phenylpropanolamine and 1% or less (+)-phenylpropanolamine.

According to the present invention, compositions of (-)-phenylpropanolamine which are substantially free of (+)-phenylpropanolamine are also substantially free of the adverse side effects related to administration of (+)-phenylpropanolamine. Such adverse side effects include but are not limited to: central nervous system stimulation, central nervous system depression and drug interactions. As a result, administration of the present compositions of (-)-phenylpropanolamine produces reduced side effects relative to the administration of the racemic phenylpropanolamine mixture or the (+)-stereoisomer of phenylpropanolamine.

The (-)-phenylpropanolamine of this invention may be prepared by known procedures. Methods for separating the stereoisomers in a racemic mixture are well-known to the skilled artisan.

The present invention also provides pharmaceutically acceptable salts of (-)-phenylpropanolamine. For example, (-)-phenylpropanolamine can be provided as a hydrochloride, bitartrate, tannate, sulfate, stearate, citrate or other pharmaceutically acceptable salt. Methods of making such pharmaceutical salts of (-)-phenylpropanolamine are readily available to one of ordinary skill in the art.

As used herein, "pharmaceutically acceptable carrier" includes any and all solvents, dispersion media, coatings, isotonic and absorption delaying agents, sweeteners and the like. The pharmaceutically acceptable carriers may be prepared from a wide range of materials including, but not limited to, diluents, binders and adhesives, lubricants, disintegrants,

coloring agents, bulking agents, flavoring agents, sweetening agents and miscellaneous materials such as buffers and adsorbents that may be needed in order to prepare a particular therapeutic composition. The use of such media and agents for pharmaceutical active substances is well known in the art. Except insofar as any conventional media or agent is incompatible with the active ingredient, its use in the therapeutic compositions is contemplated.

According to the present invention, (-)-phenylpropanolamine does not interact with other drugs, whereas the (+) enantiomer of phenylpropanolamine does interact with drugs. For example, when (-)-phenylpropanolamine is administered with an antihistamine like triprolidine, no effect is observed. However, when (+)-phenyl-propanolamine is administered with triprolidine, significant changes in central nervous system activity (stimulation or depression) are observed.

Due to the lack of drug interaction which (-)-phenylpropanolamine exhibits, supplementary active ingredients, such as additional antihistamines and decongestants, can be incorporated into the present compositions. The amount of the added antihistamine or decongestant present in the pharmaceutical composition will depend upon the particular drug used. Typical antihistamines include: diphenhydramine; chlorpheniramine; astemizole; terfenadine; terfenadine carboxylate; brompheniramine; triprolidine; acrivastine; and loratadine

The present invention further contemplates a method of relieving nasal and bronchial congestion which comprises administering a therapeutically effective amount of (-)-phenylpropanolamine, which is substantially free of (+)-phenylpropanolamine. Administration of (-)-phenylpropanolamine avoids many of the side effects related to administering (+)-phenylpropanolamine. Side effects which can be reduced or avoided by using the present methods include central nervous system stimulation, central nervous system depression and drug interactions.

According to the present invention, (-)-phenylpropanolamine is surprisingly more effective as a decongestant and as a physiological antagonist of histamine than is (+)-phenylpropanolamine. As a physiological antagonist of histamine, (-)-phenylpropanolamine counteracts the physiological effects of histamine. Histamine can cause physiological effects like nasal congestion, bronchial congestion, inflammation and the like. This present invention contemplates (-)-phenylpropanolamine to counteract all of these histamine-related physiological responses.

The present invention also contemplates a method of treating histamine- related inflammation and/or sinus congestion which comprises administering a therapeutically effective amount of (-)-phenylpropanolamine. The pharmaceutical compositions of (-)-phenylpropanolamine used for this method are substantially-free of (+)-phenylpropanolamine and induce less side effects than does administration of a composition

containing (+)-phenylpropanolamine, or the racemic mixture of phenylpropanolamine.

The present invention further contemplates a method of dilating the pupil which comprises administering a therapeutically effective amount of (-)-phenylpropanolamine to the eye. According to the present invention, administration of (-)-phenylpropanolamine induces less intraocular pressure than does administration of (+)-phenylpropanolamine or of the racemic mixture of phenylpropanolamine. The pharmaceutical compositions of (-)-phenylpropanolamine used for this method are substantially-free of (+)-phenylpropanolamine.

According to the present invention, a therapeutically effective amount of (-)-phenylpropanolamine is an amount sufficient to relieve the symptoms of a condition which can be treated by a sympathomimetic drug. In one embodiment, an amount sufficient to reduce the symptoms of a condition which can be treated by a sympathomimetic drug is an amount of (-)-phenylpropanolamine sufficient to bind or activate an adrenergic receptor, for example, an α- or a β-adrenergic receptor. When the condition is nasal congestion, the therapeutically effective amount is the amount needed to reduce nasal congestion. When bronchial congestion is the condition, the therapeutically effective amount is the amount needed to reduce bronchial congestion or provide bronchodilation. When inflammation and/or allergic reaction is the condition, the therapeutically effective amount is the amount needed to counteract the physiological effects of histamine. When eye pupil dilation is the desired, such a therapeutically effective amount of (-)-phenylpropanolamine is an amount of (-)-phenylpropanolamine sufficient to dilate the pupil. Preferably, such a pharmaceutically effective amount also produces less side effects than are observed upon administration of (+)-phenylpropanolamine, or a racemic mixture of (+)- and (-)-phenylpropanolamine. The skilled artisan can readily determine the necessary therapeutically effective amounts for treating these conditions, particularly in light of the teachings provided herein.

The pharmaceutical compositions of the present invention contain (-)-phenylpropanolamine in a therapeutically effective amount that is sufficient to provide decongestion, bronchodilation, antagonize the effects of histamine, produce a mydriatic response or treat attention deficit hyperactivity disorder, while having less side effects than would similar doses of (+)-phenylpropanolamine or the racemic mixture of (+)- and (-)-phenylpropanolamine. Such a therapeutically effective amount would be about 0.01 micrograms (μg) to about 50 milligrams (mg) per kilogram of body weight, and preferably of about 0.1 μg to about 10 mg per kg of body weight. More preferably, the dosage can range from about 1.0 μg to about 1 mg per kg of body weight. Even more preferably, the dosage can range from about 5.0 μg to about 50 μg per kg of body weight. Dosages can be readily determined by one of ordinary skill in the art and can be readily formulated into the subject pharmaceutical compositions.

The subject (-)-phenylpropanolamine may be administered by any convenient route. For example, (-)-phenylpropanolamine may be inhaled, ingested, topically applied or parenterally injected. The subject (-)-phenylpropanolamine may be incorporated into a cream, solution or suspension for topical administration. (-)-Phenylpropanolamine is preferably inhaled or administered orally or topically. The skilled artisan can readily determine the route for a specific use.

The following examples further illustrate the invention.

EXAMPLE 1

α -Adrenergic and β-Adrenergic Receptor Binding Studies

Many physiological processes are mediated by the binding of chemical compounds to $_1$, $_2$ and receptors. For example, many compounds which reduce nasal congestion bind to $_1$ and $_2$ receptors and some reduce bronchial congestion by binding to receptors. Accordingly, a compound that binds to $_1$, $_2$ and/or $_2$ receptors may be an effective nasal or bronchial decongestant.

More specifically, $_2$ adrenergic receptors, concentrated on precapillary arterioles in the nasal mucosa, induce arteriolar vasoconstriction when activated by a sympathomimetic compound. Such vasoconstriction decrease blood flow through those vessels and reduces excess extracellular fluid associated with nasal congestion and a runny nose. On the other hand, $_1$ adrenergic receptors are concentrated on postcapillary venules in the nasal mucosa. Binding to $_1$ receptors induces venoconstriction, which also reduces nasal congestion.

Compounds that bind to $_2$ receptors may also help relieve the symptoms of nasal congestion because $_2$ receptor binding is related to increased bronchodilation and reduced airway resistance.

The binding of (-)-phenylpropanolamine to $_1$, $_2$ and $_2$ various receptors was compared to the receptor binding of (+)pseudoephedrine, (-)ephedrine, (-)-phenylephrine and (+)-phenylpropanolamine. The (+) isomer of pseudoephedrine is a known decongestant, sold under the trade name SUDAFED®. (-)-Phenylephrine (Neo-Synephrine®) and (-)-ephedrine are also known to be effective decongestants.

Methods:

Membrane Preparations.

PULMONARY ALPHA-1 AND BETA-2 RECEPTORS: The lungs of mongrel dogs were separated from cartilaginous airways and major blood vessels, weighed, chopped and placed into 10 volumes of ice-cold buffered sucrose (50mM Tris-HCl pH 7.4, 1mM EGTA, 0.32M Sucrose). The tissue was then homogenized in a Polytron tissue homogenizer. The homogenate was filtered through two layers of cheesecloth, and the filtrate was dounced three times using a Con-Turque Potter homogenizer. The dounced filtrate was centrifuged

at 1000 x g for 15 min at 4 C. The supernatant was recentrifuged at 30,000 x g for 30 min at 4 C and the resulting pellet was washed and resuspended in 10 volumes of Tris buffer (50mM Tris HCl, pH 7.4, 1mM EGTA) and incubated at 37 C for 30 min in a shaking water bath. The suspension was centrifuged at 4 C at 30,000 x g for 30 min and the resulting pellet washed in 10 volumes of Tris buffer. The final pellet was resuspended in 0.5 volume of 50mM Tris HCl, pH 7.4, 1mM EGTA, 25mM $MgCl_2$. Protein concentration was then determined by the Lowry method and the final suspension was adjusted to 10 mg of protein/ml, aliquoted and stored at 90 C. Particulates were also prepared for $_2$ receptors using the identical procedure except the final protein concentration was adjusted to 0.1 g/ml.

BRAIN ALPHA-2 RECEPTORS: Membranes of mongrel dogs were harvested from the canine frontal cortex and prepared as described for lung except that the final membrane protein concentration was adjusted to 0.5 mg/ml.

Binding Assays.

ALPHA-1 BINDING, ^{3}H-PRAZOCIN: Canine lung membrane preparations (500ug protein/100 ul) were incubated with ^{3}H-Prazocin (77.9Ci/mmol) for 60 min at 25 C in a final volume of 0.25 ml of buffer (50 mM Tris-HCl/1mM EGTA, pH 7.4). Each experimental point was determined in triplicate. Nonspecific binding was determined separately for each concentration point using 10 μM phentolamine. The final concentration of ^{3}H-Prazocin was 0.7-1.1 nM in competition studies and between 0.1 and 10nM in saturation experiments. All binding assay incubations were terminated by rapid dilution with 2 ml of ice-cold wash buffer (50nM Tris-HCl, pH 7.4) and filtration through Whatman GF/B filters using a Brandel receptor-binding harvester. The filters were washed twice more with 4 ml of wash buffer and then added to 6 ml Cytoscint (ICN, Costa Mesa CA) for liquid scintillation counting (Barnes et al., 1983). In all experiments less than 17% of the added radio ligand was bound, and specific binding was about 65-70% of total binding.

ALPHA-2 BINDING P-^{125}IODOCLONIDINE. Canine brain membranes (50 ug protein/100 ul) were incubated with p-iodoclonidine (2200 Ci/mmol) for 120 min at 25 C in a final volume of 0.25 ml. Nonspecific binding was determined in separate incubations in the presence of 10 uM phentolamine. The final concentration of p-iodoclonidine was 44-45 pM in competition studies and between 50 pM and 10 nM in saturation experiments. Bound and free were separated and the bound quantitated as described above for the ICYP assays. An average of 6% of radioligand was bound, and specific binding was about 91% of total binding.

BETA-2 BINDING, ^{125}IODOCYANOPINDOLOL (^{125}ICYP). Canine lung membranes (10

ug protein/ 100 ul) were incubated with ^{125}ICYP (2200 Ci/mmol) for 110 min at 30 C in a final volume of 0.25 ml. Nonspecific binding was determined in separate incubations in the presence of 2 μM dipropranolol. Each experimental point was determined in triplicate. The final concentration of ^{125}ICYP was 8-12 pM in competition studies and between 2 and 200 pM in saturation experiments. Incubations were terminated as described above for the $_1$ assays. Filters were placed into polyethylene tubes and the bound ligand was determined by gamma spectrometry (Sano et al., 1993). An average of 27% of radio ligand was bound, and specific binding was about 90% of total binding.

All data were analyzed with the aid of microcomputer nonlinear curve fitting programs (PRISM 2.0, Graphpad Software, San Diego CA).

Results:

The receptors resident in each of the three membrane preparations were evaluated by standard saturation analysis following the addition of increasing concentrations of the appropriate radioligand. In the case of the α_1- and β_2- assays the mathematical analysis was consistent with a one site fit. The α_2-receptor analysis was best fit as two sites, one high and one low affinity.

The radio ligand added for subsequent α_2-displacement assays was adjusted to evaluate only the high affinity receptor. Contributions from p-iodoclonidine binding to imidazoline receptors in the α_2- displacement assay were evaluated with epinephrine. Epinephrine easily displaced all bound p-iodoclonidine which indicates that at the concentrations employed, p-iodoclonidine labeled few if any imidazoline receptors. Similarly, with the β_2-assay, contributions from the binding of ICYP to β_1 sites was evaluated with the β_1-selective antagonist, atenolol. Atenolol was largely ineffective in displacing ICYP from pulmonary membranes indicating little if any β_1 binding within the assay. All subsequent analyses with displacement by individual test compounds used the Kd determined from the saturation analysis since it is generally considered a more reliable estimate of the true equilibrium dissociation constant.

Table 1 provides the binding characteristics of the α_1-receptors in the membrane preparation for prazocin. The Kd is the apparent equilibrium dissociation constant for prazocin. The BMAX is the number of α_1-receptor binding sites for prazocin in this membrane preparation expressed as femtomoles per mg protein.

TABLE 1

α_1-Receptor Binding Characteristics (canine lung membranes)	
Measure	Summary
Scatchard Analysis	
Kd	0.84 nM
BMAX	55
Saturation Analysis	
Kd	0.73 nM
BMAX	53

Table 2 provides the binding characteristics of the α_2-receptors in the membrane preparation for p-iodoclonidine. The Kd is the apparent equilibrium dissociation constant for p-iodoclonidine. The BMAX is the number of α_2-receptor binding sites for p-iodoclonidine in this membrane preparation expressed as femtomoles per mg protein. Note that the two site data from the Saturation Analysis is more reliable than the Scatchard Analysis because the Scatchard Analysis assumes only one site. In order to obtain both values from the Scatchard plots, the points in the transition zone were arbitrarily divided and assigned to high and low affinity plots.

TABLE 2

α_2-Receptor Binding Characteristics (canine cerebral cortex membranes)	
Measure	Summary
Scatchard Analysis	
Kd_1 (high affinity)	0.15 nM
Kd_2 (low affinity)	0.87 nM
$BMAX_1$ (high affinity)	67
$BMAX_2$ (low affinity)	120
Saturation Analysis	
Kd_1 (high affinity)	0.15 nM
Kd_2 (low affinity)	3.01 nM
$BMAX_1$ (high affinity)	57
$BMAX_2$ (low affinity)	73

Table 3 provides the binding characteristics of the β_2-receptors in the membrane preparation for ^{125}iodocyanopindolol (ICYP). The Kd is the apparent equilibrium dissociation constant for ICYP. The BMAX is the number of β_2-receptor binding sites for ICYP in this membrane preparation expressed as femtomoles per mg protein.

TABLE 3

β_2-Receptor Binding characteristics (canine lung membranes)			
Measure	Run 1	Run 2	Summary
Scatchard Analysis			
Kd	9.9 pM	7.8 pM	8.9 pM
BMAX	150	139	145
Saturation Analysis			
Kd	9.6 pM	9.3 pM	9.5 pM
BMAX	149	142	146

The concentration of test drug required to inhibit 50% of specific prazocin, p-iodoiclonidine or ICYP binding (IC_{50}) is provided in Table 4. The Ki values of α_1, α_2 and

β_2-receptors for each drug are also provided in Table 4, where the Ki is $IC_{50} \div (1 + I/Kd)$. The variable, I, is the concentration of tracer added and the variable, Kd, is the equilibrium dissociation constant empirically determined for this receptor population. In general, lower IC_{50} and/or K_i values mean that less drug need be administered to effectively bind to these α_1, α_2 and β_2-receptors.

TABLE 4

	Alpha-1		Alpha-2		Beta-2		Ki-Ratio		
Drugs	IC_{50}	K_i	IC_{50}	K_i	IC_{50}	K_i	$\alpha 1/\alpha 2$	$\alpha 1/\beta 2$	$\beta 2/\alpha 2$
(+)-Pseudoephedrine	691	299	28	21	502	220	14.23	1.35	10.48
(-)-Ephedrine	109	47	0.77	0.59	12	5	79.67	9.40	8.47
(-)-Phenylephrine	7	3	0.02	0.015	10	5	200.00	0.60	333.33
(+)-phenylpropanola-mine	1464	612	15	12	486	215	51.00	2.85	17.92
(-)-phenylpropanol-amine	116	48	0.31	0.24	50	22	200.00	2.18	91.67

These data show that (-)-phenylpropanolamine has significantly lower IC_{50} and/or K_I values, than does (+)-phenylpropanolamine and several known decongestants.

EXAMPLE 2

(-)-Phenylpropanolamine Induces Pupil Dilation Without Increasing Intraocular Pressure

The induction of pupil dilation or mydriasis by (-)-phenylpropanolamine was compared to the pupil dilation caused by (+)pseudoephedrine, (-)ephedrine, (-)-phenylephrine and (+)-phenylpropanolamine. The (-) enantiomer of phenylephrine is known to be a mydriatic agent.

Methods:

Enantiomers (-)-phenylpropanolamine, (+)-pseudoephedrine, (-)-ephedrine, (-)-phenylephrine and (+)-phenylpropanolamine were evaluated for their efficacy in

producing mydriasis and for their effects on intraocular pressure (IOP). These agents were administered topically as either 1 and 2% solutions in buffered saline. Pupillary diameter and IOP were measured in all animals over a six hour time period during the day to minimize diurnal variations in IOP and pupil diameter.

The experiments were performed on adult male New Zealand white rabbits weighing 3.0-6.0 kg. All rabbits were caged individually and maintained on a 12hr/12hr light/dark schedule with free access to food and water. All animal procedures were in conformity with the ARVO Resolution on the Use and Care of Animals in Research. All treated rabbits had served as controls by having received a saline treatment on a different day.

Drug or saline-control solutions were applied to the superior aspect of the globe in a volume of 25 µl and allowed to spread over the cornea and sclera, while a conjunctival trough was formed by retracting the lower eyelid for approximately 30 seconds. Only one eye received drug treatment. The contralateral eye served as a control. Saline (or PBS) and drug treated rabbits were treated and observed simultaneously. A single dose was given at 0 time and IOP and pupil diameter measured at -1.0, -0.5, 0.5, 1, 3 and 5 hrs post-treatment.

IOP measurements were recorded with an Alcon Applanation Pneumotonograph (Surgical Products Division, Alcon Laboratories, Inc., Ft. Worth, TX) in rabbits placed in Lucite restraining cages. Initial topical application of a two drop 0.5% proparacaine HCl (Ophthetic®, Allergan Pharmaceuticals, Inc.) was performed on each rabbit.

Pupil diameter was measured visually at the point of the greatest horizontal diameter with a transparent millimeter ruler. All measurements were made under the identical ambient lighting conditions.

Mean and Standard Error values were used to construct time-response and dose-response curves for the treated and contralateral eye of research rabbits. The data were analyzed statistically by an analysis of variance and a Bonferoni's test for significance. $P<0.05$ was the accepted level of significance.

Results:

Although some variation in baseline IOP was noted among the total rabbits tested, there were no significant changes in IOP or pupil diameter (PD) in the saline control groups (Tables 5-7) during the six hour time period selected for drug testing.

The adrenergic agonist (+)-pseudoephedrine is known to be an active sympathomimetic amine which has both α- and β- agonist activity. In this study, (+)-pseudoephedrine produced mydriasis in only the treated eye. A slight acute elevation in IOP in the treated eye was observed following 1% and 2% topical application of (+)-pseudoephedrine. A delayed elevation in IOP was also observed in the contralateral eye.

(-)-Ephedrine increased IOP but had no effect on pupil diameter.

(-)-Phenylephrine, which is clinically available as a mydriatic agent, produces both

an elevation in IOP and an increase in pupil diameter.

At a 1% topical dose, (-)-phenylpropanolamine caused significant mydriasis and only mild IOP. Even higher doses (-)-phenylpropanolamine resulted in only mild IOP elevation, while causing marked mydriasis. (+)-Phenylpropanolamine was less effective than (-)-phenylpropanolamine. (Tables 5-7).

TABLE 5 - IOP in mmHg

Time in Hr.	-1	-0.5	0.5	1	3	5
SALINE U	27±0.9	25±0.4	26±1.5	25±1.4	27±0.9	26±1.2
(15) T	26±0.9	25±1.1	26±1.1	25±1.3	27±0.8	26±0.8
SALINE U	20±0.7	19±0.9	20±1.0	19±1.0	20±1.0	19±1.1
(15) T	19±0.8	18±0.9	18±0.8	17±0.7	19±0.9	18±1.1
DRUG (1%)						
(+)-Pseudoephedrine	19±1.0	18±1.6	18±2.0	19±1.2	20±2.0	21±0.9
U	20±0.2	20±1.7	19±2.3	22±1.4	21±2.0	23±2.3
T						
(-)-Ephedrine U	19±1.7	17±1.0	20±1.9	19±1.2	16±0.9	15±1.0
T	22±2.0	23±0.9	23±2.5	26±1.0	24±0.6	23±0.9
(-)-Phenylephrine	16±1.0	15±2.1	18±1.6	19±1.3	20±1.9	19±1.7
U	20±1.6	16±1.5	24±0.6	24±1.0	23±1.5	21±1.9
T						
(+)-	23±0.9	22±1.3	21±1.2	22±1.1	23±1.5	22±1.0
Phenylpropanolamine	25±1.2	22±1.7	25±0.7	24±1.2	22±1.2	21±0.7
U						
T						
(-)-	23±2.1	21±2.3	27±2.0	25±0.9	25±1.9	22±1.3
Phenylpropanolamine	25±0.2	22±1.6	25±2.7	25±2.2	23±2.6	24±3.4
U						
T						
DRUG (2%)						
(+)-Pseudoephedrine	20±1.0	16±10	18±2.3	17±2.1	18±1.2	22±1.4
U	24±1.1	18±1.4	26±0.8	23±1.3	22±2.1	23±2.4
T						
(-)-Ephedrine U	17±1.2	19±0.9	19±1.4	19±1.9	18±0.7	19±1.5
T	16±2.0	16±0.7	16±1.0	15±0.4	17±0.7	19±0.8
(-)-Phenylephrine	17±1.9	17±1.9	20±2.0	20±2.4	14±1.7	14±2.0
U	18±1.6	15±1.5	12±0.8	14±2.0	16±0.8	14±1.3
T						
(+)-	21±1.0	20±1.4	19±1.5	17±0.3	17±1.0	15±0.7
Phenylpropanolamine	24±1.2	23±1.4	24±0.9	27±0.9	20±1.8	18±1.0
U						
T						
(-)-	17±2.1	17±1.4	19±2.9	20±1.7	22±1.9	19±1.2
Phenylpropanolamine	18±2.0	16±1.3	21±1.9	21±1.9	21±1.7	20±2.1
U						
T						

Results are as mean ± S.E. of five-six rabbits per drug.
U = Untreated contralateral eye
T = Drug treated eye

TABLE 6 - Pupil Diameter in mm						
Time in Hr.	**-1**	**-0.5**	**0.5**	**1**	**3**	**5**
SALINE U (15) T	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2
SALINE U (15) T	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2	5±0.2 5±0.2
DRUG (1%)						
(+)-Pseudoephedrine U T	6±0.4 7±0.2	6±0.4 7±0.2	6±0.4 8±0.2	6±0.4 8±0.2	6±0.4 8±0.2	6±0.4 8±0.2
(-)-Ephedrine U T	6±0.4 6±0.4	6±0.4 6±0.4	6±0.4 6±0.4	6±0.4 6±0.4	6±0.4 6±0.4	6±0.4 6±0.4
(-)-Phenylephrine U T	6±0.4 6±0.4	6±0.4 6±0.4	6±0.4 7±0.4	6±0.4 9±0.4	6±0.4 9±0.4	6±0.4 9±0.4
(+)-Phenylpropanolamine U T	7±0.3 7±0.3	7±0.3 7±0.3	7±0.3 8±0.3	7±0.4 8±0.4	7±0.4 8±0.4	7±0.3 7±0.5
(-)-Phenylpropanolamine U T	7±0.4 7±0.5	7±0.4 7±0.5	7±1.2 10±0.2	7±1.2 10±0.2	7±1.2 10±0.2	7±1.2 10±0.2
DRUG (2%)						
(+)-Pseudoephedrine U T	6±0.4 6±0.4	6±0.4 6±0.4	6±0.4 7±0.4	6±0.4 7±0.4	6±0.4 7±0.4	6±0.4 7±0.4
(-)-Ephedrine U T	5±0.2 5±0.2	5±0.2 5±0.2	9±0.2 9±0.2	9±0.2 9±0.2	9±0.2 9±0.2	9±0.2 9±0.2
(-)-Phenylephrine U T	6±0.2 6±0.2	6±0.2 6±0.2	12±0.2 12±0.2	12±0.2 12±0.2	12±0.2 12±0.2	12±0.2 12±0.2
(+)-Phenylpropanolamine U T	6±0.4 6±0.4	6±0.4 6±0.4	7±0.7 8±0.4	7±0.5 8±0.4	7±0.5 8±0.4	7±0.7 8±0.4
(-)-Phenylpropanolamine U T	5±0.5 5±0.5	5±0.5 5±0.5	5±0.5 6±0.5	5±0.5 8±0.5	5±0.5 8±0.5	5±0.5 8±0.5

Results are as mean ± S.E. of five-six rabbits per drug.
U = Untreated contralateral eye
T = Drug treated eye

TABLE 7 - Mydriatic Responses				
DRUGS	**TREATED EYE**		**UNTREATED EYE**	
SALINE	0		0	
(+)-Pseudoephedrine	1%	+	1%	0
	2%	+	2%	0
(-)-Ephedrine	1%	0	1%	0
	2%	+++	2%	+++
(+)-Phenylephrine	1%	++	1%	0
	2%	+++	2%	+++
(+)-Phenylpropanolamine	1%	+	1%	0
	2%	++	2%	+
(-)-Phenylpropanolamine	1%	++	1%	+
	2%	++	2%	0

SCALE: 0 = No change; + = 0-2 mm change; ++ = 2-4 mm change; +++ = >4 mm change

EXAMPLE 3

(-)-Phenylpropanolamine Does Not Stimulate the Central Nervous System

Many sympathomimetic compounds stimulate the central nervous system. This is one reason that decongestants have side effects like insomnia. These tests compare the degree of central nervous system stimulation, in the presence and absence of an antihistamine, of (-)-phenylpropanolamine, (+)-pseudoephedrine, (-)-ephedrine, (-)-phenylephrine and (+)-phenylpropanolamine. Unlike many sympathomimetic compounds, (-)-phenylpropanolamine does not stimulate the central nervous system.

Moreover, decongestants are often sold in combination with other active ingredients (e.g. Claritin-D® and Seldane-D®). In products containing two or more active ingredients, interactions between the active ingredients are undesirable. In these tests, the extent of (-)-phenylpropanolamine interaction with a known antihistamine triprolidine, was observed and compared to any such interaction between triprolidine and (+)-pseudoepdedrine, (-)-ephedrine and (-)-phenylephrine.

Methods:

Animals

Male Swiss-Webster mice (HSD ND4, Harlan Sprague Dawley, Houston, TX) aged 2-3 months were used in these studies. Each dose group consisted of 8 mice.

The mice were housed 2 to 4 per cage in 30.4 x 22.9 x 15.2 cm clear polycarbonate cages with food and water available *ad libitum* for at least one week prior to locomotor activity testing. The colony was maintained at 23±1 C, on a normal light-dark cycle beginning at 0700 hr. All testing took place during the light portion of the light-dark cycle.

Apparatus

Horizontal (forward movement) locomotor activity was measured using a standardized, optical activity monitoring system [Model KXYZCM (16), Omnitech Electronics, Columbus, OH]. Activity was monitored in forty 40.5 x 40.5 x 30.5 cm clear acrylic chambers that where housed in sets of two within larger sound-attenuating chambers. A panel of 16 infrared beams and corresponding photodetectors were spaced 2 cm apart along the sides and 2.4 cm above the floor of each activity chamber. A 7.5-W incandescent light above each chamber provided dim illumination via a rheostat set to 20% of full scale. Fans provided an 80-dB ambient noise level within the chamber.

Drugs.

(+)-Pseudoephedrine, (-)-ephedrine, (-)-phenylephrine, (+)-phenylpropanolamine, (-)-phenylpropanolamine and (+)-amphetamine were obtained from Sigma Chemical Co. Triprolidine HCl was obtained from Research Biochemicals International, (Natick, MA). All compounds were dissolved in 0.9% saline and injected i.p. in a volume of 10 ml/kg body weight.

Procedure.

Locomotor stimulant effects. In these studies, mice were placed in the activity testing chambers immediately following injection of saline or a dose of one of the test compounds ranging from 0.1 mg/kg to 250 mg/kg. (+)-Amphetamine was used as a positive control. The total horizontal distance traversed (cm) was recorded at 10 minute intervals for a 2-hour session. Separate groups of 8 mice were assigned to each dose or saline group, and dose-effect testing continued for each compound until maximal stimulant or depressant effects could be estimated. A separate control group was tested along with each compound.

For compounds with significant stimulant effects, the potency and efficacy were estimated for the 30-minute time period in which maximal stimulant effects were observed at the lowest dose. Using TableCurve 2D v2.03 (Jandel Scientific), the mean average total distance traversed (cm/10 min) for that period was fit to a 3-parameter logistic peak function of $\log_{10}$ dose (with the constant set to the mean of the saline group), and the maximum effect estimated from the resulting curve. The ID_{50} (dose producing ½ maximal stimulant activity) was estimated from a linear regression against $\log_{10}$ dose of the ascending portion of the dose-effect curve. The stimulant efficacy was the peak effect of the compound (cm/10 min) as estimated from the logistic peak function, minus the mean control distance traveled (cm/10 min), and was expressed for each stimulant compound as a ratio to the stimulant efficacy determined for (+)-amphetamine.

For compounds with significant depressant effects, the potency and efficacy were estimated for the 30-minute time period in which maximal depression occurred at the lowest dose. The mean average total distance traversed (cm/10 min) for that period were fit to a linear function of $\log_{10}$ dose of the descending portion of the dose-effect curve. The ID_{50} was the dose producing ½ maximal depressant activity, where maximal depression = 0 cm/30 min. Efficacy was the ratio of maximal depressant effect to maximum possible depression for each compound (mean average total distance of the control group minus the lowest mean total distance, expressed as a ratio to the control group total distance).

H_1 receptor antagonist interaction studies. The potential for each compound to interact with an H_1 antihistamine was determined by testing whether a known antihistamine, triprolidine, produced a dosage shift in the observed stimulant or depressant effects of each sympathomimetic compound. Triprolidine was used as an example of the class of H_1 receptor antagonists that are typically used as antihistaminic drugs. Twenty minutes prior to administering each test sympathomimetic compound, either triprolidine (at 0.01, 0.1, 1.0, or 25 mg/kg) or saline was injected. The mice were then immediately placed in the apparatus for a two hour session. Doses of the test compound were selected from the ascending or descending time of the dose-effect curve determined from the compound-alone studies. Eight mice were tested for each triprolidine/ sympathomimetic combination.

Statistical analysis. Time course data for each compound were considered in 2-way analyses of variance with dose as a between-group and time as a within-group factor. The dose-effect data were considered in 1-way analyses of variance, and planned individual comparisons were conduced between each dose and the saline control group. Interaction studies were considered in 2-way analyses of variance, with Pretreatment and Test dose as the factors.

Two-way analyses of variance were conducted on horizontal distance traveled using dose as a between-subject factor and time as a within-subject factor. Only (+)-amphetamine exhibited a significant dose- and time-effect, with an interaction of dose and time (all $Fs>2.7$; all p values <0.01).

Results:

The effects of sympathomimetic enantiomers on locomotor activity are summarized in Table 8.

Locomotor stimulant effects

Time course.

Mice injected with (+)amphetamine showed a dose- and time-dependent increase in the distance traversed within 10 minutes following injection. The peak stimulant effects occurred during the first 30 minutes following 2.5 mg/kg and continued for at least 60

minutes.

(-)-Ephedrine resulted in increased locomotion within 40 minutes following 50 to 100 mg/kg, with peak effects occurring 60 to 90 minutes following injection and diminishing thereafter.

Little or no stimulant effects were evident within two hours following treatment with (+)-phenylpropanolamine, (+)-pseudoephedrine or (-)-phenylpropanolamine (Table 8).

Locomotor depressant effects

Time course. (+)-Amphetamine and (-)-ephedrine treatment did not cause locomotor depression. However, treatment with (+)-pseudoephedrine, (+)-phenylpropanolamine and (-)-phenylpropanolamine did result in some locomotor depression within 10 to 20 minutes following injection. These effects lasted from 20 minutes to $\geq$ 2 hours, depending upon dose and compound.

Depressant efficacy/potency.

Dose-response relationships for locomotor depressant effects of the sympathomimetics are provided in Table 8, for the time period in which the maximal depressant effects were first observed as a function of dose. The maximal depressant effect was estimated as the difference between the control group mean and the mean of the dose group with lowest locomotor activity. The maximum possible effect was assumed to be equivalent to the mean of the control group. Depressant efficacy was the ratio of maximal depressant effect to the maximum possible effect, and was highest for (+)-pseudoephedrine (0.58). The ID_{50} for depressant effects was estimated from a linear regression through the descending portion of the dose-effect curve, assuming zero locomotor activity (horizontal distance) as the maximal effect. The order of potency for the depression was:

(+)-phenylpropanolamine≈(-)-phenylephrine > (-)-phenylpropanolamine >> (+)-pseudoephedrine

TABLE 8

		Stimulation			Depression		
Compound	Range[1]	Efficacy[2]	Potency[3]	Time[4]	Efficacy[5]	Potency[6]	Time[7]
(+)-Pseudoephedrine	1-100	0.21	12.6	40-70	0.58	72.4	10-40
(-)-Ephedrine	0.5-250	0.80	38.2	50-80	0.45	7.4	10-40
	0.1-10	0	-	60-90	0.77	2.3	0-30
(-)-Phenylephrine	2.5-25	0.19	7.9	50-80	0.76	2.6	90-120
(+)-Phenylpropanolamine	1-25	0	-	60-90	0.70	5.8	10-40
(-)-Phenylpropanolamine							

1 Dose range tested in mg/kg.
2 The ratio of the maximal stimulant effect of the test compound to the maximal effect of (+)amphetamine.
3 The dose resulting in ½ the maximal stimulant effect (ID_{50}) in mg/kg i.p.
4 The 30-min period following injection in which the maximal stimulant effect occurred.
5 The ratio of the maximal depressant effect to the maximum possible effect (zero locomotor activity)
6 The dose resulting in ½ the maximal depressant effect (ID_{50}) in mg/kg i.p.
7 The 30-min minute period following injection in which the maximal depression occurred.
8 "-" denotes absence of significant effect

Triprolidine interactions.

Triprolidine alone. When injected immediately prior to testing, doses of triprolidine from 0.25 to 25 mg/kg failed to affect horizontal distance during the 2-hour test period. Dose-dependent depression of locomotion was observed following 50 and 100 mg/kg, beginning within 10-minutes following injection and lasting for 30 to 40 minutes. A separate one-way analysis of variance on average distance/ 10 min for the period 0-30 minutes following injection suggested a significant dose main effect where $F(8,102) = 7.7$ and $p<0.00$ 1, although individual comparisons of dose groups with control in that analysis verified that significant effects of triprolidine were restricted to the 50 and 100 mg/kg doses (ps<.01).

Triprolidine interactions. When tested for dose-response in mice pretreated with 0.01, 0.1, or 1.0 mg/kg triprolidine, (-)-phenylpropanolamine and (-)-phenylephrine did not show significant modification of stimulant or depressant effects. Significant effects for pretreatment with triprolidine were observed for (+)-pseudoephedrine, (-)-ephedrine and (+)-phenylpropanolamine.

Conclusion.

(-)-Phenylpropanolamine causes no central nervous system stimulation and has less potency for depression than does (+)-phenylpropanolamine. Moreover, combining triprolidine and (-)-phenylpropanolamine did not give rise to any negative drug interactions such as central nervous system stimulation or depression. Hence, (-)-phenylpropanolamine does not appear to interact with H_1 histamine receptors, and may be used in combination with other drugs, such as H_1 antihistamines, particularly, triprolidine.

EXAMPLE 4

Decongestant Activity of (-)-phenylpropanolamine

The decongestant activity of (-)-phenylpropanolamine was compared to that of (+)pseudoephedrine, (-)ephedrine, (-)-phenylephrine and (+)-phenylpropanol-amine, in normal and histamine-challenged rats.

Experimental Protocol:

The method was based on one reported by Lung for the measurement of nasal airway resistance. Sprague Dawley rats (weight range 247-365 gram) were anesthetized with sodium pentobarbital intra-peritoneally (50 mg/ka). Rats were placed on a heating pad, in a V trough, dorsal side down. A tracheotomy was performed and a tracheal cannula was positioned, tied and left open to room air. A cannula was placed into the superior part of the trachea and was advanced till lodged in the posterior nasal opening. Normal saline (0.5 ml) was injected into the nasal cannula to confirm position as well as to moisten the nasal mucosa. After nasal cannulation was confirmed the cannula was tied in place with a suture placed around the trachea. Excess fluid was expelled from the nasal airway with a short (2-3 second) air flow via the nasal cannula. Additionally, in studies correlating blood pressure changes to those in the nasal airway pressure, a cannula was positioned in the internal carotid artery (PE. 50) and connected to a multipen (Grass) recorder using pressure transducer (Isotec).

Nasal airway pressure was measured using a validyne pressure transducer (with a 2.25 cm H_2O range membrane) connected to a multipen recorder (Grass). Air was passed through an in-line direct measure flow meter (Gilmont instruments) connected to the nasal opening cannula. Pressure was measured in the line with a constant flow rate (150 ml/min) of air. Enantiomeric drugs were directly injected into the jugular vein using a 30 gauge needle. All injections were of a constant 0.1 ml volume. In the congestion challenged groups congestion was achieved by an intranasal administration of histamine (50 mM,

0.02ml/nostril). The histamine was expelled after 2 min with a short nasal cannula airflow and subsequent enantiomeric drug doses were directly injected into the jugular vein. The doses of injection for each of the enantiomers tested were determined from a previous study in our laboratory in which each of the dose of drug we chose resulted in an increase in mean arterial pressure (MAP) of 10% (Table 9). The dose causing a 10% increase in MAP served as our "100%" dose for the initial nasal airway studies.

TABLE 9

Dosage of Enantiomer which raised mean arterial pressure 10%

Drug Name	Dog (ug/kg)	Rat (ug)
(+)-Pseudoephedrine	200	60
(-)-Ephedrine	100	30
(-)-Phenylephrine	10	3-5
(+)-Phenylpropanolamine	400	120
(-)-Phenylpropanolamine	20	6

Three investigations were performed as follows:

Investigation 1: A comparison was made of the effect of the different enantiomers on nasal airway resistance prior to and following histamine congestion. The amount of drug required to raise the mean arterial pressure by 10% was chosen as the " 100% dose" for these decongestant studies (see Table 9). Control changes in nasal airway resistance were obtained by recording nasal airway resistance prior to and following this dose 100% dose. In a test group of rats the 100% dose was injected into the jugular vein two minutes after nasal airway congestion was produced by introduction of 0.02 ml/nostril of 50 mM histamine into the nasal airway. Nasal airway resistance was thus increased by histamine challenge. To assess the effect of administering an enantiomer changes in airway resistance were then observed.

Investigation 2: A comparison of the effect on nasal airway resistance of various doses of each enantiomer was made to determine an effective dose of the enantiomer. Dosages tested were 50%, 25%, 10% and 5% of the "100%" enantiomer dosage required to increase the mean arterial pressure 10%. Changes in nasal airway resistance were observed by comparing pre-enantiomer injection nasal airway resistance with decreases in nasal airway resistance following jugular vein injection of the enantiomer. Five rats were tested at each dose for each of the enantiomers.

Investigation 3: A 75% dose of enantiomer was tested following 0.02 ml/nostril of 50mM histamine. As before, this "75% dose" was 75% of the dose required to increase the mean arterial pressure 10%. The 75% dosages employed were as provided in Table 10.

Table 10

Drug	75% Dosage (μg/kg i.v.)
(+)-Pseudoephedrine	150
(-)-Ephedrine	75
(-)-Phenylephrine	7.5
(+)-Phenylpropanolamine	300
(-)-Phenylpropanolamine	15

Blood pressure was monitored. Effects on airway resistance and blood pressure of each of the eight enantiomeric were evaluated at the 75% dose prior to and following histamine in five rats for each enantiomer and each histamine condition.

Results

Investigation 1:

Each drug gave rise to a significant decrease in nasal airway pressure, relative to control, in non-histamine-challenged rats (Table 11). While the control for the (-)-phenylephrine was significantly different from the other controls, this difference in control level did not translate into a difference caused by administration of the drug.

TABLE 11

Drug	Control (mm H_2O)	Post Drug (mm H_2O)	% Change	Paired t test pValue
(+)-Pseudoephedrine	9 ± 0.5	7 ± 0.9	-21.3 ± 7.6	0.015
(-)-Ephedrine	7 ± 0.8	6 ± 0.5	-14 ± 3.4	0.034
(-)-Phenylephrine	5 ± 0.2	4 ± 0.2	-20.3 ± 4.3	0.010
(+)-Phenylpropanol-amine	7 ± 0.2	6 ± 0.2	-10.5 ± 1.4	0.001
(-)-Phenylpropanol-amine	8 ± 0.6	7 ± 0.7	-8.81 ± 2.6	0.028

* mm water.

In the histamine-challenged rats, administration of each drug again showed a significant decrease in nasal passage pressure (Table 12). However, after treatment with histamine, rats treated with (-)-phenylpropanolamine showed a significantly greater reduction in airway passage resistance than did rats treated with (+)-phenylpropanolamine (Table 12). These data indicate that (-)-phenylpropanolamine is a stronger antagonist of the physiological effects of histamine than is (+)-phenylpropanolamine.

TABLE 12

Drug	Control*	t test p Value	Post* Histamine	t test pValue	Post* Drug	%Change
(+)-Pseudo-ephedrine	6.6 ±0.6	0.1	9.3 ±1.8	0.001	6.1 ±1.5	-36.7±2.7
(-)-Ephedrine	6.7 ±0.4	0.06	8.2 ±0.7	0.007	6.4 ±0.6	-21.8±3.5
(-)-Phenyl-ephrine	7.5 ±0.5	0.04	13.2 ±2.1	0.05	10.5 ±2.1	-22.3±8.5
(+)-Phenyl-propanolamine	0.5 ±0.9	0.004	12.2 ±0.8	0.04	10.6 ±1.0	-13.6 ±4.4
(-)-Phenyl-propanolamine	7.8 ±0.2	0.07	10.5 ±1.0	0.04	7.6 ±0.4	-25.2±5.6

* mm water.

The results indicate that the antihistamine and/or decongestant activity of (-)-phenylpropanolamine was superior to (+)-phenylpropanolamine.

Investigation #2:

Table 13 summarizes the mean nasal airway pressure of different enantiomer dosages ranging from 5%, 10%, 25% and 50% of the dose that produced a 10% change in resting mean arterial pressure (the "100%" dose). The standard error of the mean is also provided. In general, the 50% dose was an approximate threshold dose at which nasal airway pressure was reduced.

TABLE 13
Mean Decrease in Nasal Airway Pressure With Variable Enantiomer Dosages*

Drug	5%	10%	25%	50%
(+)-Pseudoephedrine	-3.8 ± 2.8	-6.8 ± 3.6	-13.5 ± 4.3	-16.1 ± 2.4
(-)-Ephedrine	-1.0 ± 0.8	-2.5 ± 1.5	-3.6 ± 1.9	-1.9 ± 2.0
(-)-Phenylephrine	-1.6 ± 0.9	-4.8 ± 0.8	-12.1 ± 2.3	-5.2 ± 1.6
(+)-Phenyl-propanolamine	-0.03 ± 0.3	-0.7 ± 1.2	-0.8 ± 1.0	-0.07 ± 1.5
(-)-Phenyl-propanolamine	0.1 ± 0.7	-1.4 ± 0.5	-1.4 ± 2.1	-1.9 ± 0.6

*percent enantiomer dose that increased the dog resting mean arterial pressure 10%.

Investigation #3:

Figure 1 summarizes the percent change in mean arterial blood pressure after a 75% dose of various enantiomers. As indicated, (-)-phenylpropanolamine causes a lesser increase in blood pressure than is observed for several enantiomers which are commercially available as decongestants.

Figure 2 provides the observed percent nasal airway pressure after a 75% dose of the different enantiomers.

WHAT IS CLAIMED:

1. A pharmaceutical composition comprising (-)-phenylpropanolamine in a therapeutic dosage suitable for treating nasal congestion and a pharmaceutically acceptable carrier, wherein said (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine.

2. The pharmaceutical composition of Claim 1 wherein said composition is substantially-free of a side effect related to administration of (+)-phenylpropanolamine.

3. The pharmaceutical composition of Claim 2 wherein said side effect is central nervous system stimulation.

4. The pharmaceutical composition of Claim 2 wherein said side effect is central nervous system depression.

5. The pharmaceutical composition of Claim 2 wherein said side effect is a drug interaction.

6. The pharmaceutical composition of Claim 5 wherein said drug interaction is with an antihistamine.

7. The pharmaceutical composition of Claim 1 wherein said therapeutic dosage is sufficient to reduce nasal or bronchial congestion.

8. The pharmaceutical composition of Claim 1 wherein said therapeutic dosage is sufficient to activate an α -adrenergic receptor.

9. The pharmaceutical composition of Claim 1 wherein said therapeutic dosage is sufficient to counteract the physiological effects of histamine.

10. The pharmaceutical composition of Claim 1 wherein said therapeutic dosage is about 0.01 μg to about 50 mg per kg of body weight.

11. A method of relieving nasal and bronchial congestion which comprises administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein said (-)-phenylpropanolamine is substantially free of (+)-phenylpropanolamine.

12. The method of Claim 11 wherein said method has less side effects than administration of a racemic mixture of (+)- and (-)-phenylpropanolamine.

13. The method of Claim 11 wherein said method reduces a side effect related to administering (+)-phenylpropanolamine.

14. The method of Claim 13 said side effect is central nervous system depression.

15. The method of Claim 13 said side effect is a drug interaction.

16. The pharmaceutical composition of Claim 15 wherein said drug interaction is with an antihistamine.

17. The method of Claim 11 wherein said therapeutically effective amount is sufficient to reduce nasal and bronchial congestion.

18. The method of Claim 11 wherein said therapeutically effective amount is an

amount of (-)-phenylpropanolamine sufficient to activate an α -adrenergic receptor.

19. The method of Claim 11 wherein said therapeutically effective amount is an amount of (-)-phenylpropanolamine sufficient to act as an antihistamine.

20. The method of Claim 11 wherein said therapeutic dosage is about 0.01 μg to about 50 mg per kg of body weight.

21. A method of dilating the pupil which comprises administering a therapeutically effective amount of (-)-phenylpropanolamine to a mammal, wherein said (-)-phenylpropanolamine is substantially-free of (+)-phenylpropanolamine.

22. The method of Claim 21 wherein said therapeutically effective amount is an amount of (-)-phenylpropanolamine sufficient to activate an α -adrenergic receptor.

23. The method of Claim 21 wherein said method has less side effects than administration of (+)-phenylpropanolamine.

24. The method of Claim 21 wherein said side effect is an increase in intraocular pressure.

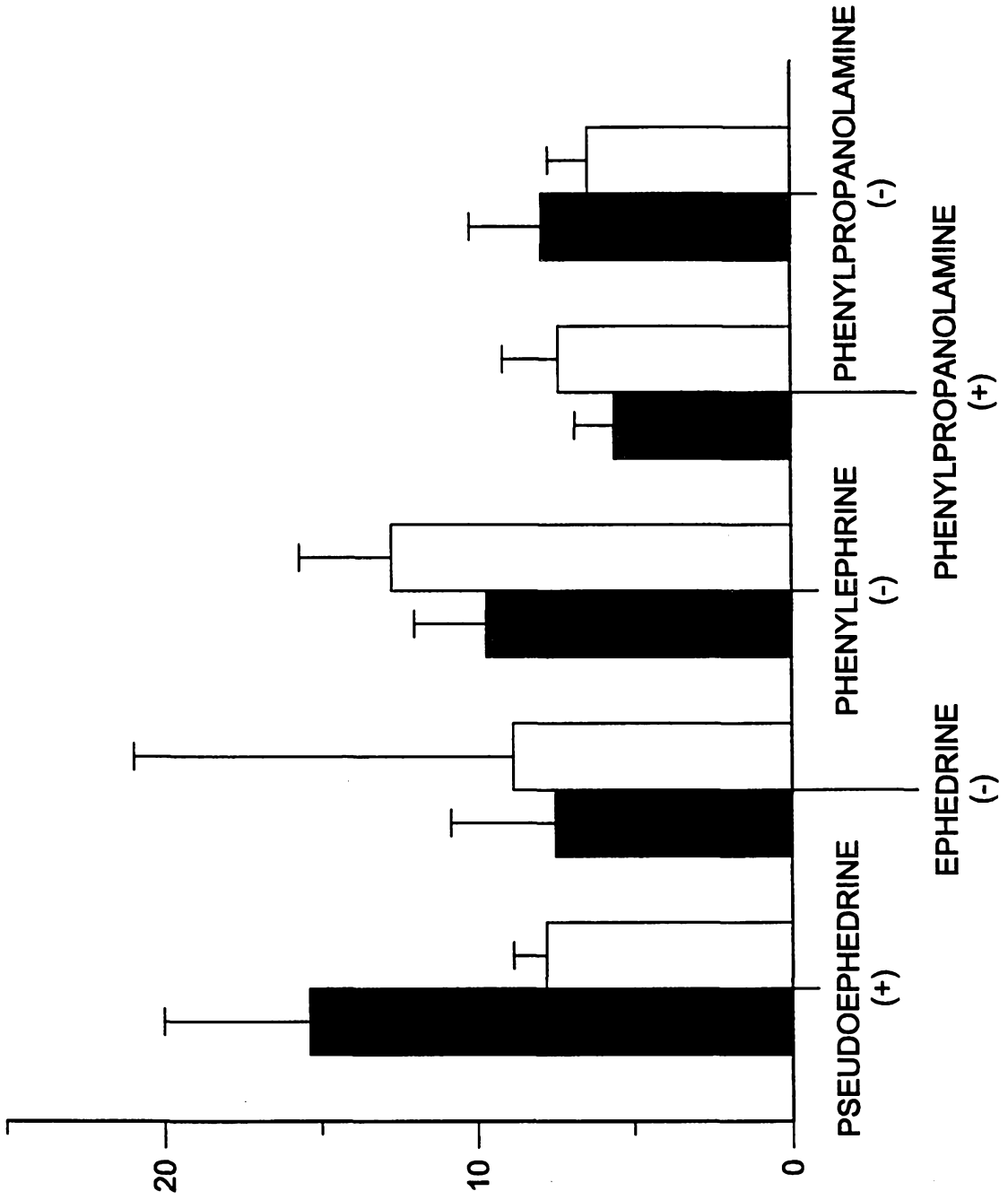
20
10
0
PSEUDOEPHEDRINE (+)
EPHEDRINE (-)
PHENYLEPHRINE (-)
PHENYLPROPANOLAMINE (+)
PHENYLPROPANOLAMINE (-)

FIG-1

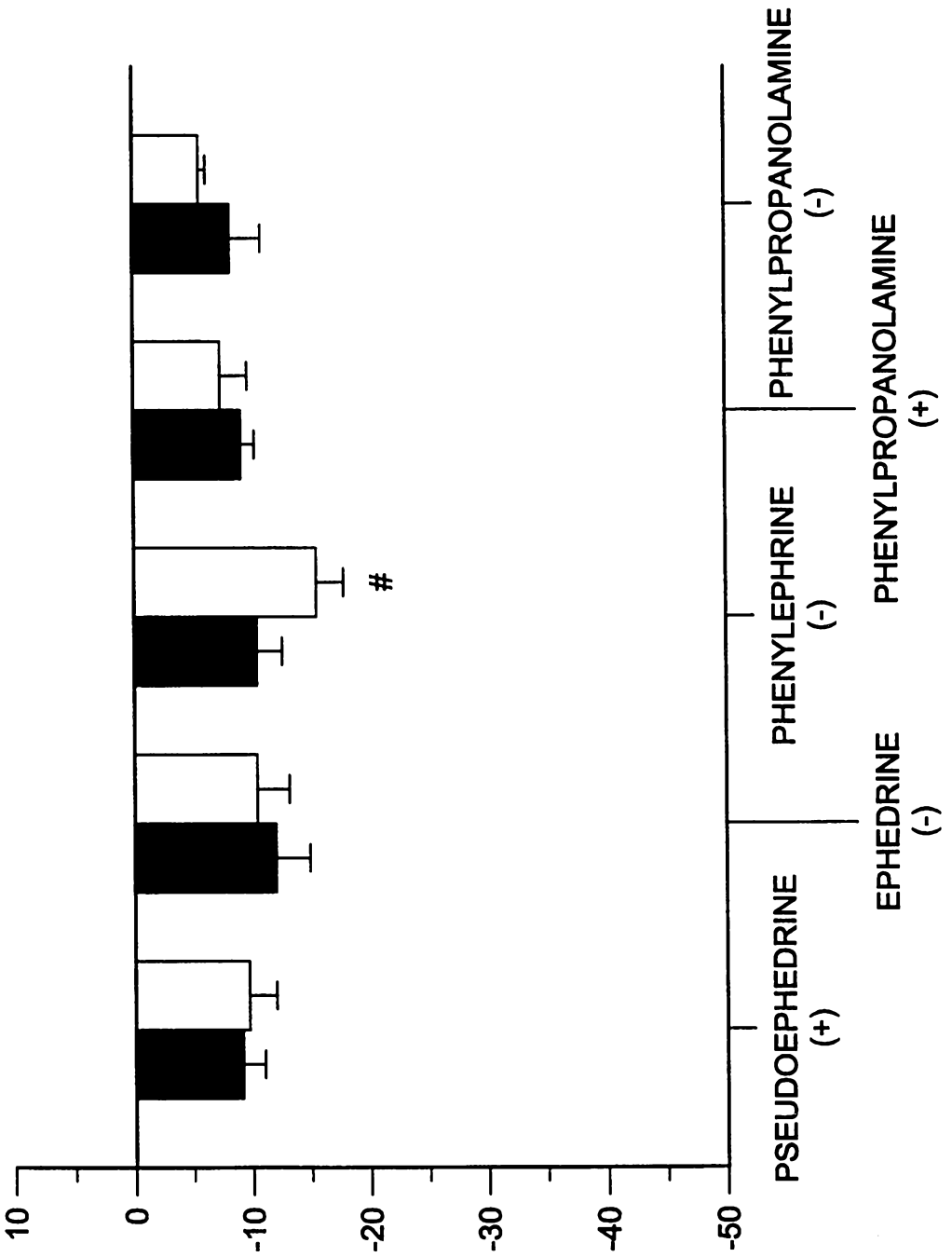
10
0
-10
-20
-30
-40
-50
#
PSEUDOEPHEDRINE (+)
EPHEDRINE (-)
PHENYLEPHRINE (-)
PHENYLPROPANOLAMINE (+)
PHENYLPROPANOLAMINE (-)

FIG-2