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(71) Applicant *(for all designated States except US)*: **YISSUM, Research Development Company of the Hebrew University of Jerusalem** [IL/IL]; Hi-Tech Park Edmond J. Safra Campus, Givat Ram, P.O.B 39135, 91390 Jerusalem (IL).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **TOUITOU, Elka** [IL/IL]; 6 Demumit Street, Canada Hill, 93893 Jerusalem (IL). **GODIN, Biana** [IL/IL]; 11/51 Pinhas Hebroni St., 96633 Jerusalem (IL). **DUCHI, Shaher** [IL/IL]; P.O.B. 249, 30055 Kfar Rama Israel (IL).

(74) Agents: **PYERNIK RUTMAN** et al.; Beit Etzion, 91 Herzl St., P.O.Box 10012, 84106 Beer-Sheva (IL).

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(54) Title: COMPOSITIONS FOR NASAL DELIVERY

(57) Abstract: A pharmaceutical composition comprising the combination of cannabidiol and glatiramer acetate, and a pharmaceutically acceptable carrier.

Compositions for Nasal Delivery

The delivery of biologically active materials to the skin and cell membranes by means of an aqueous vehicle that comprises the combination of lipid vesicles and water miscible organic solvents has been described in the art.

For example, an aqueous carrier system containing phospholipids and ethanol was described in EP 158441, with the weight ratio between the aforementioned components being from 40:1 to 1:20.

U.S. Patent No. 5,711,965 describes a solution comprising phospholipids, ethanol and water in a weight ratio of 10:16:74, respectively.

U.S. Patent Nos. 5,540,934 and 5,716,638 and WO 03/000174 describe an aqueous composition containing vesicles (ethosomes) in the presence of ethanol.

U.S. Patent No. 6,627,211 describes a carrier suitable for the administration of an anti-convulsive agent to the nasal mucous membranes. It appears that the content of organic solvents in this carrier is relatively high (30% to 60% ethanol and 30 to 60% propylene glycol).

WO 2007/043057 describes the use of phospholipid, one or more C2-C4 alcohols and water in the preparation of a vesicular composition adapted for intranasal administration of an active agent, wherein the concentrations of the phospholipid and the one or more alcohols in the composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, and the water content of the composition is not less than 30% by weight. Throughout the description, the concentrations of the ingredients are expressed relative to the total weight of the composition.

US 6,410,588 discloses the use of cannabidiol for the treatment of multiple sclerosis.

SUMMARY OF INVENTION

The present invention provides a pharmaceutical composition comprising the combination of cannabidiol and glatiramer acetate, and a pharmaceutically acceptable carrier. The carrier is preferably adapted for nasal or parenteral administration. The carrier may thus be in a liquid form (including viscous liquids), or semi-solid (e.g., a gel, cream). A carrier which comprises water, one or more C2-C4 alcohols and phospholipids has been found to be especially suitable in this regard. The cannabidiol and the glatiramer acetate are preferably present in the composition in synergistically effective amounts. More specifically, the concentration of cannabidiol and glatiramer acetate are within the ranges of 0.5-40 % and 0.5-30 %, respectively.

Most preferably the present invention provides a pharmaceutical composition adapted for intranasal administration comprising a combination of glatiramer acetate and cannabidiol and a carrier comprising not less than 30% by weight water, from 12 to 30% by weight C2-C4 alcohol(s) and from 0.2 to 10% phospholipids arranged in a vesicular structure. The C2-C4 alcohol is preferably ethanol. Preferably, the composition further comprises a polyol, and more specifically, propylene glycol, at a concentration in the range of 1 to 30% by weight. The composition comprising cannabidiol and glatiramer acetate is suitable for the treatment of multiplesclerosis.

The present invention also provides the use of cannabidiol, glatiramer acetate and a pharmaceutically acceptable carrier in the preparation of a nasally or parenterally administrable composition for the treatment of multiplesclerosis, wherein the carrier preferably comprises phospholipid, one or more C2-C4 alcohols and water, wherein the concentrations of said phospholipid and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, and the water content of said composition is not less than 30% by weight. The C2-C4 alcohol is preferably ethanol, and the composition may further comprise a polyol such as propylene glycol.

The present invention also provides a pharmaceutical composition for intranasal administration comprising a combination of diclofenac and at least one active ingredient selected from the group consisting of diazepam and fentanyl and a carrier,

wherein said carrier comprises not less than 30% by weight water, from 12 to 30% by weight C2-C4 alcohol(s) and from 0.2 to 10% phospholipids arranged in a vesicular structure. Preferably, the composition further comprises a polyol, and more specifically, propylene glycol, at a concentration in the range of 1 to 30% by weight.

The present invention also provides the use of diclofenac and at least one active ingredient selected from the group consisting of diazepam and fentanyl, and a pharmaceutically acceptable carrier in the preparation of a nasally administrable composition for the treatment of pain, wherein the carrier comprises phospholipid, one or more C2-C4 alcohols and water, wherein the concentrations of said phospholipid and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, and the water content of said composition is not less than 30% by weight. The C2-C4 alcohol is preferably ethanol, and the composition may further comprise a polyol such as propylene glycol.

In another aspect, the present invention provides a pharmaceutical composition for treating insomnia comprising a therapeutically effective amount of brotizolam or pharmaceutically acceptable salt thereof, phospholipids, one or more C2-C4 alcohols and water, wherein the concentrations of the phospholipids and the one or more alcohols in the composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of the composition being not less than 30% by weight, the phospholipids forming vesicles in the composition. The C2-C4 alcohol is preferably ethanol, and the composition may further comprise a polyol such as propylene glycol. The present invention also provides the use of brotizolam, phospholipids, one or more C2-C4 alcohols and water in the preparation of a composition adapted for intranasal administration, wherein the concentrations of said phospholipids and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of said composition being not less than 30% by weight. The C2-C4 alcohol is preferably ethanol, and the composition may further comprise a polyol such as propylene glycol.

In another aspect, the present invention provides a pharmaceutical composition comprising a therapeutically effective amount of prednisolone or pharmaceutically

acceptable derivatives or salts thereof, phospholipids, one or more C2-C4 alcohols and water, wherein the concentrations of the phospholipids and the one or more alcohols in the composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of the composition being not less than 30% by weight, the phospholipids forming vesicles in the composition. The C2-C4 alcohol is preferably ethanol, and the composition may further comprise a polyol such as propylene glycol. The present invention also provides the use of prednisolone, phospholipids, one or more C2-C4 alcohols and water in the preparation of a composition adapted for intranasal administration, wherein the concentrations of said phospholipids and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of said composition being not less than 30% by weight. The composition is useful in the treatment of multiplescelerosis.

Phospholipids suitable for use in the preparation of the composition according to the present invention include phosphatidylcholine (PC), hydrogenated phosphatidylcholine, phosphatidic acid (PA), phosphatidylserine (PS), phosphatidylethanolamine (PE), phosphatidylglycerol (PPG) and phosphatidylinositol (PL). The chemical structure of phospholipids that may be used according to the present invention is described in US 4,614,730, which is incorporated herein by reference. Preferably, the phospholipids are present in the composition of the invention at a concentration of 0.5 to 5% by weight.

The term C2-C4 alcohols, as used herein, refers to alkanols containing two, three or four carbon atoms. The alcohols to be used according to the present invention specifically include ethanol, 1-propanol, isopropyl alcohol and *tert*-butyl alcohol, with the former being especially preferred. The concentration of ethanol in the composition contemplated by the present invention for use as an intranasal drug delivery vehicle is preferably in the range of 15 to 27% by weight. The weight ratio between the alcohol(s) and the phospholipids is not less than 2:1, and more preferably not less than 5:1.

According to a particularly preferred embodiment of the invention, the composition further comprises one or more water miscible polyols, and especially glycols (1,2-diols, such as ethylene glycol and propylene glycol, with the latter being especially preferred), at a concentration of 1 to 30% by weight, and preferably 5 to 20 by weight.

The compositions of the present invention may be prepared by mixing together the various components, namely, water, phospholipids, one or more C2-C4 alcohols (and possibly also one or more polyols) and the active ingredient under conditions that allow the formation of vesicles. More specifically, the compositions of the present invention may be conveniently prepared by dissolving the phospholipids in the alcohol (or in the alcohol/glycol mixture), followed by the addition of the active ingredient, either in the form of an aqueous solution thereof or in a solid form, with a subsequent addition of water. The preparation of the composition is preferably carried out under stirring, typically at room temperature or at an elevated temperature, which is preferably not higher than 50°C.

Alternatively, a dispersion of the phospholipids and the active ingredient in water is prepared, into which the alcohol, optionally together with polyol (e.g., a mixture of ethanol and propylene glycol) are added with stirring, possibly under heating.

It is also possible to first prepare freeze-dried lipid vesicles having the active ingredient encapsulated therein, and subsequently dispersing the same in a mixture of water, the C2-C4 alcohol and optionally polyol.

It should be noted that the composition according to the present invention may include additional excipients that are well known in the art, such as surfactants, preservatives, thickening agents, co-solvents, adhesives, antioxidants, buffers, viscosity and absorption enhancing agents and agents capable of adjusting the pH and osmolarity of the formulation.

Suitable surfactants that can be used in accordance with the present invention include ionic, nonionic or amphoteric surface active agents. More specifically, hydrophilic surfactants (e.g. Tweens, Tween 80, Myrj, Brjs, Labrasol etc.) or lipophilic surfactants

(eg. Span 20, Span 60, Myrj, Arlacel 83 and such) may be suitably used, preferably at a concentration in the range of 0-25% by weight.

Suitable preservatives that can be used with the present formulations include, for example, benzyl alcohol, parabens, chlorobutanol, benzalkonium salts and combinations thereof. Some examples of antioxidants include tocopherols, butyl hydroxytoluene, sodium metabisulfite, potassium metabisulfite, ascorbyl palmitate and the like. These preservatives and antioxidants may be present in the formulations in a concentration of from about 0.001% up to about 5%w/w.

Regarding buffers, the nasal delivery system may include a buffer for maintaining the formulation at a pH of about 7.0. The particular buffer, of course, can vary depending upon the particular nasal delivery system used, as well as the specific active molecule selected. Buffers that are suitable for use in the present invention include, for example, acetate, citrate, prolamine, carbonate and phosphate buffers and combinations thereof. The pharmaceutical formulations of the present invention may include a pH adjusting agent.

Regarding thickening agents, the viscosity of the formulations of the present invention can be maintained at a desired level using a pharmaceutically acceptable thickening agent. Thickening agents that can be added to the compositions of the present invention include for example, methyl cellulose, xanthan gum, tragacanth, adhesives, guar gum, carboxymethyl cellulose, hydroxypropyl cellulose, carbomer, polyvinyl alcohol, alginates, acacia, chitosans, mucoadhesive polymer-systems like poly(acrylates), cellulose derivatives, hyaluronic acid, hyaluronic acid derivatives, chitin, collagen, pectin, starch, poly(ethylene glycol), sulfated polysaccharides, carrageenan, Na-alginate, gelatin, pectin and combinations thereof. The desired concentration of the thickening agent will depend upon the agent selected and the viscosity desired.

The compositions may also comprise gel forming or bioadhesive compounds such as carbopols, alginates, scleroglucan, cellulose derivatives, starch, albumin, pluronic gels, diethyl aminoethyl (DEAE)-sephadex, polycarbophil, hyaluronic acid,

hyaluronates, starch, gelatin, cholagen and others. Compositions can also be incorporated in the w/o cream, o/w cream, hydrophilic ointment or lipophilic ointment, gels, other semi-solid bases. The compositions could be delivered to the nasal cavity as drops, mists, aerosols, instillations, by use of pipetor, special devices, evaporators, vaporizators and such.

The formulations of the present invention may also include agents such as tolerance enhancers to reduce or prevent drying of the mucus membrane and to prevent irritation thereof.

The compositions according to the present invention may be applied to the nasal cavity as liquids, sprays, aerosols, nebulizers or semi-solid preparations. Semisolid preparations may be on the base of gels, w/o or o/w creams or hydrophilic/lipophilic ointments. The compositions may contain molecularly dispersed (soluble, solubilized, etc.) active agent or the fine particles/crystals of the active agent. The compositions could be administered from nasal sprays, metered-dose sprays, squeeze bottles, liquid droppers, disposable one-dose droppers, nebulizers, cartridge systems with unit-dose ampoules, single-dose pumps, bi-dose pumps, multiple-dose pumps or any other device. For example, the compositions of the invention may be stored in/delivered from a spray or aerosol device/container as described in details in Remington's Pharmaceutical Sciences (16th edition, Chapters 83 and 92).

Regarding spray devices, it should be noted that both single (unit) dose or multiple dose systems may be used. Typically, a spray device comprises a bottle and a pump; such devices are commercially available from various sources. Typically, the volume of liquid that is dispensed in a single spray actuation is in the range of from 5 to 250 microlitters/each nostril/single administration and the concentration of the active ingredient in the formulation may be readily adjusted such that one or more spray into the nostrils will comply with the dosage regimen.

It should be noted that the composition comprising cannabidiol and glatiramer acetate as described hereinabove may be applied also parenterally (e.g., by means of intravenous injection).

Cannabidiol is 2-[3-Methyl-6-(1-methylethenyl)-2-cyclohexen-1-yl]-5-pentyl-1,3-benzenediol. A method for the synthesis of cannabidiol of (-)-form has been described, for example by T. Petrzilka et al., *Helv. Chim. Acta* 52, 1102 (1969); H. J. Kurth et. al., *Z. Naturforsch.* 36B, 275 (1981); Synthesis of cannabidiol of (±)-form has been described, for example by R. Mechoulam, Y. Gaoni, *J. Am. Chem. Soc.* 87, 3273 (1965). Cannabidiol may be administered at a daily dose of 10 – 400 mg.

Glatiramer acetate is typically administered at a daily dose of 1 to 60 mg. Glatiramer acetate is a mixture of polypeptides composed of alanine, glutamic acid, lysine, and tyrosine in a molar ratio of approximately 4.6:1.5:3.6:1.0, respectively, which is synthesized by chemically polymerizing the four amino acids, forming products with average molecular weights ranging from about 4000 to about 13,000 daltons. The corresponding molar fractions are approximately 0.427 for alanine, 0.141 for glutamic acid, 0.337 for lysine and 0.093 for tyrosine, and may vary by about +/-10 A method for synthesis of Glatiramer acetate has been described for example in US 7049399. Glatiramer acetate is typically administrated at a daily dose of 20 mg injected subcutaneously (SC).

Diclofenac is 2-(2-(2,6-dichlorophenylamino)phenyl)acetic acid. A method for synthesis of diclofenac has been described for example by A. Sallman, R. Pfister, U.S. Pat. 3,558,690, (1971). Diclofenac is typically administrated at a daily dose of 25-150 mg.

Diazepam is 7-chloro-1,3-dihydro-1-methyl-5-phenyl-2H-1,4-benzo-diazepin-2-one. A method for the synthesis of diazepam has been described, for example by Sternbach LH, Reeder E, Keller O, & Metlesics W. [Quinazolines and 1,4-benzodiazepines III substituted 2-amino-5-phenyl-3H-1,4-benzodiazepine 4-oxides. J Org Chem, 26: 4488-4497, 1961]. Diazepam is typically administered at a daily dose of 0.2 to 100 mg.

Fentanyl is N-(1-phenethyl-4-piperidyl)-N-phenyl-propanamide. Fentanyl is typically administered at a daily dose of 100 to 400 mg. Its preparation is described, for example, in Gupta, P. K et al., J. Chem. Res. 2005, 7, 452-453 and in Def. Res. Dev. Establ. ,Gwalior 474 002, India; Eng.)

Brotizolam is 8-bromo-6-(o-chlorophenyl)-1-methyl-4H-s-triazolo[3,4c]thieno[2,3e]1,4-diazepine.A method for its synthesis has been described, for example in Japanese Patent Unexamined Publication No. 80899/1976 (U.S. Pat. No. 4,094,984). Brotizolam is typically administered at a daily dose of 0.01 to 1 mg.

Prednisolone is 11,17-dihydroxy-17- (2-hydroxyacetyl)- 10,13-dimethyl-6,7,8,9,10,11,12,13,14,15,16,17- dodecahydrocyclopenta [a]phenanthren-3-one. A method for synthesis of Prednisolone has been described for example by Kurosawa T, Ikegawa S, Chiba H, Ito Y, Nakagawa S, Kobayashi K, Tohma M. [Convenient synthesis of 18-hydroxylated cortisol and prednisolone. Steroids. 1992 Sep;57(9):426-9.] Prednisolone is typically administrated at a daily dose of 5-60 mg.

As used herein, nasally administering or nasal administration includes administering the compositions into norstils of the nose to the mucous membranes of the nasal passage or nasal cavity of the mammal. Such formulations can be administered, for example, as a nasal spray, nasal inhaler, nasal drop, aerosol, propellants, pressured dispersion, aqueous aerosol, nebulizer, nasal suspension, instillation, nasal gel, nasal ointment and nasal cream by aid of any new or old type device. Administration of compositions of the present invention may also take place using a nasal tampon or nasal sponge containing the compositions.

It should be noted that the active compound identified herein may be provided in the form of a pharmaceutically acceptable salt, racemic and separate enantiomers, when applicable.

In the drawings

Figures 1A-B are bar diagrams showing the latency (Figure 1A) and total sleeping time (Figure 1B) in mice following nasal administration of brotizolam (drug dose 0.25mg/kg or 2.5mg/kg), 5 minutes prior to sleep induction with pentobarbitone sodium (40 mg/kg), as compared to oral administration of an aqueous drug solution (at the sane dosage) and versus untreated control;

Figure 2 is a graph showing clinical manifestations of Experimental Allergic Encephalomyelitis (EAE) induced by Myelinoligodendrocyte glycoprotein (MOG peptide) (n=6) for prophylactic intranasal administration of prednisolone composition at a dose of 3mg/Kg animal, as compared to subcutaneous administration of aqueous drug solution (at the sane dosage) and versus untreated control;

Figure 3 is a graph showing clinical manifestations of EAE induced by MOG peptide for intranasal administration of prednisolone composition at a dose of 5.7 mg/Kg animal and 13.7 mg/Kg animal, as compared to subcutaneous administration of an aqueous drug solution (at 13.7 mg/kg) and versus untreated control;

Figure 4 is a bar diagram showing the results of a writhing test in mice following nasal administration of an amalgam composition of diazepam and/or diclofenac, versus untreated control; and

Figure 5 is a graph showing clinical manifestations of EAE following intranasal administration of CBD and GA, versus subcutaneous administration thereof and versus untreated control.

Examples

Reference Example 1

Nasal composition of the invention containing Glatiramer acetate (GA)

Component, %w/w	
Phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
GA aqueous solution for injection	to 100

The final composition contains 12mg/mL GA.

Reference Example 2

GA solution for subcutaneous (SC) administration- control solution

Component	
Copaxone® injection (20mg/mL)	180 µl
Normal saline to	3000 µl

Reference Example 3

Diazepam nasal composition (low dose)

Component	*%w/w*
Diazepam	0.3125
Phospholipid	2
Ethanol	15
Propylene Glycol	22
Vitamin E	0.2
Water (DDW)	to 100

Reference Example 4

Diazepam nasal composition (high dose)

Component	%
Diazepam	0.625
Phospholipid	2
Ethanol	15
Propylene Glycol	22
Vitamin E	0.2
Water (DDW)	to 100

The above formulation was prepared as follows: phospholipid was dissolved in ethanol and propylene glycol was added to this solution followed by addition of Vitamin E and mixing. Then, Diazepam was added to this mixture. Water was added slowly with constant stirring by a Heidolph overhead stirrer. The composition is stirred for additional 15 minutes. The final composition contains 6.25 mg/mL Diazepam.

Reference Example 5

Nasal composition of the invention containing Glatiramer acetate (GA)

Component, %w/w	
Phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
GA aqueous solution for injection	To 100

10 grams of the above formulation were prepared as follows: 0.5 g soy phospholipid was dissolved in 1.5 g ethanol and 2 g propylene glycol was added to this solution followed by addition of Vitamin E and mixing for 5 minutes. Then, GA aqueous solution containing 20mg/mL GA was added slowly with constant stirring by a

Heidolph overhead stirrer. The composition is stirred for additional 15 minutes. The final composition contains 12mg/mL GA.

Reference Example 6

GA solution for subcutaneous (SC) administration- control solution

Copaxone® injection (20mg/mL)	180 µl
Normal saline to	3000 µl

Reference Example 7

GA solution for nasal administration- control solution

Component	
Copaxone® injection (20mg/mL)	600 µl
Distilled water	400 µl

Example 1

Brotizolam (hypnotic/sleep effect) nasal composition

Component,	*mg*
Brotizolam	0.25
Phospholipid	20
Ethanol	150
Propylene Glycol	200
Vitamin E	2
Water (DDW)	To 1 ml

The formulation was prepared according to the methods described above. Final formulation contains 0.25mg/g Brotizolam.

Doses of Brotizolam in humans: 0.01 to about 1 mg preferably from about 0.05 to about 0.3 mg for one administration before sleep.

Example 2

Hypnotic (sleep) effect of Brotizolam in mice following nasal administration of the composition of the invention- Test of composition of Example 1

The efficacy of the intranasal administration of the above brotizolam-containing compositions was tested by means of the following experimental protocol using the assessment of pentobarbitone-induced sleeping in mice. In this test described below, the time in minutes, after treatment with pentobarbitone and loss of righting reflex was taken as sleep latency, whilst the time between loss of righting reflex and the regain of right reflex was taken as the duration of sleep. The model is widely used in rats and mice to evaluate the hypnotic/sedative effects of drugs [Avoka et al., J. Ethnopharmacol. 2006; 103:166-75].

Experimental protocol:

The experiments were carried out on Female C57Bl/6 mice, 6-7weeks old.

The following groups of animals were used (n=4):

Group 1: Intranasal administration of Brotizolam composition at a dose of 0.25mg/Kg animal (18µl/mouse). 9µl of the Composition from Example 1 were administered in each nostril.

Group 2: Oral administration of Brotizolam at a dose of 0.25mg/kg animal (18µl/mouse).

Group 3: Untreated control animals given orally with water.

The animals were administered with the above compositions and 5 minutes later injected with Sodium Phenobarbital-SP (40mg/kg i.p). The onset of the loss of righting reflex and the duration of the loss of the righting reflex were recorded as latency and total sleeping time in minutes (TST), respectively.

Figure 1 presents the results of this experiment. It appears that nasal treatment with Brotizolam given only 5 minutes prior pentobarbitone administration significantly shortened the latency and significantly enhanced TST of pentobarbitone induced sleep. Oral Brotizolam had no significant effect on both parameters as compared to control.

FORMULATIONS FOR TREATMENT AND PROPHYLAXIS OF MULTIPLE SCLEROSIS

The efficiency of the nasal carrier of the invention is shown in the use of formulations for prophylaxis and treatment o neurological symptoms developed in an Experimental Allergic Encephalomyelitis (EAE) model (an animal model for multiple sclerosis)

Prophylaxis of Multiple Sclerosis

Example 3

Nasal composition of the invention containing corticosteroids.

Component,	*%w/w*
Prednisolone	0.25
Soy phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
Water (DDW)	to 100

The composition was prepared by the methods described in previous examples. The final composition contains 2.5mg/mL prednisolone.

Example 4

Prednisolone solution for subcutaneous (SC) administration- control solution

Component, %w/w	
PEG400	30
Prednisolone	0.05
Normal saline	69.95

Prednisolone was dissolved in the mixture of PEG400 and saline. A clear solution containing 0.5mg/mL prednisolone was obtained.

Example 5

Effect of corticosteroid nasal formulation on prevention of EAE

EAE in mice is an accepted model of Multiple Sclerosis in humans.

Induction of EAE:

EAE was induced in female C57Bl/6 mice, 6-7weeks old (weighting 17-18g) using a slightly modified previously published protocol [Kataoka H, et al. Cell Mol Immunol. 2005]. By using this procedure, the mice were immunized with Myelinoligodendrocyte glycoprotein (MOG_{35-55}) peptide mixed with mycobacteria in Complete Freund's adjuvant (CFA), followed by immediate and 48 hours later injections of Pertussis toxin.

Clinical assessment is performed on the basis of the following EAE score scale.

Score	Signs	Description
0	Normal behavior	No neurological signs
0.5	Distal limb tail	The distal tail is limp and droops wobbly
1	Limb tail	The tail is limp and droops wobbly
2	Wright reflex	
3	Ataxia	Walk when the hinds legs are unsteady
4	Early paralysis	The rat/mouse has difficulties standing on his legs but still has remnants of movements
5	Full paralysis	The rat/mouse can't move its legs at all, it looks thinner and emaciated. Incontinence
6	Moribund-death	

The following experimental groups were used, each containing 6 animals:

Group 1: Prophylactic intranasal administration of Prednisolone composition (Example 3) at a dose of 3mg/Kg animal (0.05mg/20µl/mouse). 10µl of the Composition were administered in each nostril twice daily, from 1st to 4th day after EAE induction, and then 1.5mg/kg once a day (0.025mg/10µl per mouse, 5µl/norise) until the end of the experiment.

Group 2: Prophylactic subcutaneous administration: Prednisolone 3mg/kg (0.05mg/100µl per mouse), administrated as a control subcutaneously in solution of Example 4. The solution was injected subcutaneously twice daily, from 1st to 4th day

after EAE induction, and then 1.5mg/kg was administered once a day SC (0.025mg/50ul per mouse) until the end of the experiment.

Group 3: Control: no treatment.

The results presented in Figure 2 show that intranasal administration of the Composition of the invention containing Prednisolone (Example 3) efficiently prevented the development of EAE as compared to SC administration of the same drug dose in solution.

Treatment of Multiple Sclerosis

Example 6

Nasal composition of the invention containing corticosteroids

Component, %w/w	
Prednisolone	0.5
Phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
Water (DDW)	59.3

The composition was prepared by the methods described in previous examples. The final composition contains 5mg/mL prednisolone.

Example 7

Nasal composition of the invention containing corticosteroids

Component	*%w/w*
Prednisolone	1-12
Phospholipid	1-5
Ethanol	12-20
Propylene Glycol	5-25

Tocopherol acetate	0.2
Water/Saline/Buffer	To 100

The composition was prepared by the methods described in previous examples. The nasal composition contains 10-120mg/mL prednisolone.

The composition could be administered as drops, nebulation, spray, device, special device for nasal delivery to brain.

Doses in humans: The initial dosage of prednisolone may vary from 2 mg to 60 mg per day. The initial dosage should be maintained or adjusted until a satisfactory response is noted.

The nasal composition is administered from 1 to 4 times every day or every other day or once a week.

Example 8

Effect of GA and prednisolone innovative formulation on treatment of EAE

EAE was induced in Female C57Bl/6 mice 6-7weeks old with body weight 16.84 ±1.23g, according to protocol described in Example 5.

EAE Treatment with nasal compositions of the invention versus controls:

Compositions were administered once daily, beginning on ~ 11^{th} day (treatment was initiated when individual mice developed a clinical score EAE ≥0.5) until the end of the study.

Four experimental groups were used, each containing 6 animals:

Group 1: Intranasal administration of prednisolone composition (Example 6) at a dose of 5.7mg/Kg animal (0.1mg/20µl/mouse). 10µl of the Composition were administered in each nostril once daily.

Group 2: Intranasal administration of GA composition (Reference Example 1) at a dose of 13.7mg/Kg animal (240mcg/20µl/mouse). 10µl of the Composition were administered in each nostril once daily.

Group 3: Subcutaneous administration of GA control solution (Reference Example 2) at a dose of 13.7 mg/kg. 200µl of the solution were administered once a day to the mice (240µg GA/animal/day).

Group 4: Control: no treatment.

Figure 3 is a plot presenting the results obtained in this experiment. The results show that intranasal administration of Prednisolone from the Compositions of the invention (Example 6) efficiently treated EAE (a model of Multiple Sclerosis). For example, from day 22 to the end of the experiment the following EAE scores were obtained at the plateau:

Group 1 (Intranasal prednisolone novel composition) ~ 0.4

Group 2 (Intranasal GA reference composition) ~from 0.5 to 0

Group 3 (Subcutaneous GA reference solution) ~ 1.5

Group 4 (Untreated control) ~3

Pain Treatment with Intranasal Administration of Diazepam and Diclofenac Mixture Composition

Example 9

Diazepam (lower dose) and diclofenac amalgam nasal composition

Component, %w/w	
Diazepam	0.3125
Diclofenac Sodium	1.25
Phospholipid	2
Ethanol	15
Propylene Glycol	22
Vitamin E	0.2
Distilled Water	To 100

The above formulation was prepared as follows: phospholipid was dissolved in ethanol and propylene glycol was added to this solution followed by addition of Vitamin E and mixing. Then, Diazepam was added to this mixture followed by addition of diclofenac sodium and stirring for another 10 minutes. Water was added slowly with constant stirring by a Heidolph overhead stirrer. The composition is stirred for additional 15 minutes. The final composition contains 6.25 mg/mL Diazepam.

Example 10

Diazepam and diclofenac amalgam nasal composition

Component, %w/w	
Diazepam	1
Diclofenac Sodium	7.5
Phospholipid	2.5
Ethanol	16
Propylene Glycol	25
Vitamin E	0.2
Buffer	To 100

Example 11

Composition containing opiates and anti-inflammatory agents

Component, %w/w	
Fentanyl	0.01-1
Diclofenac Sodium	0.5-10
Phospholipid	3
Ethanol	17
Propylene Glycol	17
Vitamin E	0.2
Distilled Water	To 100

Suggested dose of Fentanyl is in the range of 10-300 mcg per each administration.

Suggested dose of diclofenac 0.05-100mg per each administration.

Example 12

Test of analgesic effect of Diazepam and Diclofenac nasal compositions in mice

The analgesic efficacy of the intranasal administration of the diclofenac-diazepam and diazepam containing composition (compositions detailed in Examples 9 and reference examples 3-4 respectively) was tested.

The experiments were carried out on female C75/BL mice (8-9 weeks).
The following experimental groups of 4 animals, each, were used:

Group 1: Intranasal administration of Diazepam low dose composition as given in Reference Example 3.
Group 2: Intranasal administration of Diazepam high dose composition as given in Reference Example 4.
Group 3: Intranasal administration of Diazepam and Diclofenac composition containing a low dose of Diazepam and Diclofenac as given in Example 9.
The results were compared to the untreated control animals.

Intraperitoneal injection of a weak solution of acetic acid induces a nociceptive stereotyped behavior (writhing) that mimics acute pain. This model is widely used to evaluate the analgesic (antinociceptive) effects of anti pain drugs.

The mice received treatment under Isoflurane® anesthesia. Half an hour after the treatment, all the mice in the 3 groups were intraperitoneally administered with acetic acid 0.6% (10 ml/kg) and individually housed in cages with a smooth flat floor.
Antinociception effect was recorded by counting the number of writhes 5 minutes after the injection of acetic acid for a period of 10 minutes. A writhe is indicated by abdominal constriction and stretching of at least one hind limb.
From results presented in Figure 4 it is obvious that administration of the nasal composition of Diclofenac and Diazepam, as given in Example 9, totally diminished the pain induced by injection of acetic acid solution. The composition has much higher effect than Diazepam administered at the same low dose and even Diazepam administered at a twice higher dose. The implication of these results could be in reducing the drug doses when using the combined composition.

Multiple Sclerosis Treatment with Intranasal Administration of Glatiramer Acetate (GA) and Cannabidiol (CBD) Mixture Compositions

Example 13

Nasal composition of the invention containing GA and CBD (cannabidiol)

Component, %w/w	
CBD	4
Phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
GA aqueous solution for injection	To 100

The composition was prepared by the methods described above.

The final composition contains 11.2mg/mL GA and 40mg/mL CBD.

Example 14

Nasal composition of the invention containing GA and CBD

Component, %w/w	
CBD	2
Phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
GA aqueous solution for injection	To 100

The composition was prepared by the methods described above.

The final composition contains 11.2mg/mL GA and 20mg/mL CBD.

Example 15

Nasal composition of the invention containing GA and CBD

Component, %w/w	
CBD	1.2
Phospholipid	5
Ethanol	15
Propylene Glycol	20
Vitamin E	0.2
GA aqueous solution for injection	To 100

The final composition contains 11.8mg/mL GA and 12mg/mL CDB.

Example 16

Nasal composition of the invention containing GA and CBD

Component, %w/w	
GA	0.5-12
CBD	0.1-10
Phospholipid	0.2-10
Ethanol	12-18
Propylene Glycol	0-25
Tocopherol acetate or BHT	0.02-0.5
Water/Saline/Buffer	To 100

The composition was prepared by the methods described in previous examples. CBD is mixed/ dissolved in Phospholipid/ Ethanol/ Propylene glycol phase. GA is added as an aqueous solution.

The GA solution could contain buffers, glycols, polyglycols, solubilyzers such as citrate esters, carbohydrates, mannitol, sugar alcohols, salts, stabilizers, antioxidants (BHA, BHT, sulfides, tocopherols, ascorbic acid derivatives).

The nasal composition contains 5-100mg/mL GA and 1-100mg/mL CBD.

The compositions could be administered as drops, nebulation, spray, device, special device for nasal delivery to brain.

Dosage of GA in humans: 0.5-80mg GA administered as needed.

Dosage of GA in humans: 0.5-80mg GA administered as needed.

Dosage of CBD: 0.1-500mg administered as needed.

Administration of the nasal composition could be made every day or every other day or once a week.

Example 17

Treatment of Experimental Allergic Encephalomyelitis (EAE) by means of intranasal GA-CBD combination- evidence of neurons regeneration

EAE in mice is an accepted model of Multiple Sclerosis in humans.

Induction of EAE:

EAE was induced in female C57Bl/6 mice, 6-7weeks old (weighting 17-18g) using a slightly modified previously published protocol [Kataoka H, et al. Cell Mol Immunol. 2005]. By using this procedure, the mice were immunized with Myelinoligodendrocyte glycoprotein (MOG_{35-55}) peptide mixed with mycobacteria in Complete Freund's adjuvant (CFA), followed by immediate and 48 hours later injections of Pertussis toxin.

Clinical assessment is performed on the basis of the EAE score scale (see Example 5 for details).

The following experimental groups were used, each containing 6 animals:

Compositions were administered to mice in all treatment groups once daily, beginning on ~ 11^{th} day (treatment was initiated when individual mice developed a clinical score EAE $\geq$0.5) until the end of the study.

Four experimental groups were used:

Group 1: Intranasal administration of GA-CBD composition.

Composition of Example 13 at a dose of 6.8 mg GA/Kg animal and 24.45 mg CBD/Kg animal (10µl/mouse) was administered for the first 3 days from the development of EAE.

Further, until the end of the experiment, composition of Example 14 at a dose of 6.8 mg GA/Kg animal and 12.2 mg CBD/Kg animal (10µl/mouse) was administered

Composition was administered once daily.

At the end of the experiment, the mice were sacrificed, the brain was removed for neurogenesis analysis.

Group 2: Intranasal administration of GA composition (Reference Example 5) at a GA dose of 6.8 mg/Kg animal (120mcg/10µl/mouse). Composition was administered once daily.

Group 3: Subcutaneous administration of GA control solution (Reference Example 6) at a GA dose of 6.8 mg/kg.

100µl of the solution were administered once a day to the mice (120 µg GA/animal/day).

Group 4: Control- no treatment.

Two mice were injected intraperitoneally daily from day 20 to end of experiment 50 mg/Kg BrDU (5-Br-2'-deoxyuridine), a thymidine analog that incorporates into the DNA of dividing brain neurons.

At the end of the experiment, the mice were sacrificed, the brain was removed for neurogenesis analysis.

The results presented in Figure 5 show that intranasal administration of the combination composition containing a low dose GA and CBD was much more efficient in treating EAE in mice than intranasal administration of the same dose GA alone or the same dose of GA administrated subcutaneously.

At the end of the experiment (day 26) the following EAE scores were obtained:

Group 1 (Intranasal GA-CBD compositions) ~ 0.5

Group 2 (Intranasal GA composition) ~ 2

Group 3 (Subcutaneous GA control solution) ~ 3.4

Group 4 (Untreated control) ~3

Hystological sections show a high regeneration of active neurons only on mice treated intranasaly with GA-CBD (from group 1).

Example 18

Effect of intranasal treatment with GA-CBD composition on EAE in mice: regeneration in untreated control animals and animals administrated with GA aqueous solution

Treatment:

EAE was induced in Female C57Bl/6 as previously described in Example 5.

This experiment was carried out to test if nasal administration of GA solution not in the carrier of the invention is effective in EAE model.

Compositions were administered once daily, beginning on ~ 11^{th} day (treatment was initiated when individual mice developed a clinical score EAE $\geq$0.5) until the end of the study.

The following experimental groups were used, each containing 6 animals:

Group 1: Intranasal administration of GA aqueous control (Reference Example 7) at a high dose of 13.7mg/Kg animal (240mcg/20µl/mouse). 10µl of the Composition were administered in each nostril once daily.

Group 2: Control: no treatment.

The results show that intranasal administration of GA from aqueous solution had no effect on EAE (a model of Multiple Sclerosis) and were not different from untreated mice.

Starting from day 16 after EAE induction, animals were re-divided into four new groups according to the following treatment schedule scheme.

Group 1A: Two mice from the group previously treated with intranasal GA aqueous solution now were treated intranasally with composition of Example 15.

Group 1B: Two mice from the group previously treated with intranasal GA aqueous solution now were untreated.

Group 2A: Three previously untreated mice – remained untreated

Group 2B: One previously untreated mouse now was treated intranasally with composition of Example 15.

Table VI presents the results of experiment in Example 18. The results show the reduction in EAE scores when the mice from day 16 where treated with the combination of GA and CBD, pointing toward regeneration of the condition. In Group 1A, on day 22 the mean score was 2 showing a reduction of 1 score (from 3 to 2) and

1A, on day 22 the mean score was 2 showing a reduction of 1 score (from 3 to 2) and in Group 2B the reduction was by 2 scores, from score 4 to score 2. In both untreated mice groups, 1B and 2A, the scores remained essentially the same.

Table VI

Day	Mean scores			
	Group 1A	Group 1B	Group 2A	Group 2B
16	3	3	3.5	4
17	2.5	3	4	2
18	2	3.35	4	2
19	2.5	3.8	4	2
20	3	3	3.65	2
21	2	3	3.65	2
22	2	3	3.65	2

Example 19

Therapeutic efficacy of a new drug combination glatiramer acetate and cannabidiol (GA & CBD) administrated nasally and subcutaneous in EAE mice model

Experiments were conducted on female C57Bl/6 mice, 6-7weeks old.

Mice were immunized with 300μg of MOG_{35-55} peptide mixed with 0.1 ml (5 mg mycobacterium) in Complete Freund's adjuvant (CFA). A volume of 0.2 ml of the mixture was injected at the base of the tail for each mouse. Pertussis toxin (200 ng/ mouse) was injected immediately and 48 hours later in volume of 0.1 ml. Clinical assessment was performed on the basis of the following scale (EAE score, see Example 5).

The following formulations were prepared:

Formulation A

Composition	% w/w
GA aqueous solution	58.6
CBD	1.2
Ethanol	15
Propylene glycol	21
Phospholipon 90 G	4
Vitamin E acetate	0.2

The composition is stirred for additional 15 minutes. The final composition contains 12 mg/mL GA and 12 mg/ml CBD.

Formulation B

Composition	% w/w
GA aqueous sol. For injection	12
CBD	0.24
Ethanol	15
Propylene glycol	21
Phospholipon 90 G	4
Vitamin E acetate	0.2
Distilled water	47.6

The final composition contains 2.4 mg/mL GA and 2.4 mg/ml CBD.

Experimental groups were prepared as follows, each group containing 5 animals:
Group A- Intranasal – Glatiramer acetate and CBD -Formulation A: 5 mice received each, the combination (GA & CBD Glatiramer acetate 6.8 mg/kg and CBD 6.7 mg/kg) as 10 microliters formulation A once a day. Treatment was initiated when individual mice developed a clinical score EAE $\geq$0.5 until the end of the study.

Group B- Subcutaneous– Glatiramer acetate and CBD -Formulation B: 5 mice received each, the combination (GA & CBD Glatiramer acetate 6.8 mg/kg and CBD 6.7 mg/kg) as 50 microliters formulation B once a day. Treatment was initiated when individual mice developed a clinical score EAE ≥0.5 until the end of the study.

Group C. Control: no treatment: Untreated mice

Results:

Group	Incidence	Mortality	Mean (±SE) Duration (days)	Mean (± SE) Onset (days)	Mean (± SE) Group Score	Mean (± SE) Maximal Score
Formulation A IN-GA/CBD Therapeutic 6.7mg/6.7mg/kg	5/5	0	16 ± 1.79	14 ± 1.8	0.39 ± 0.29	1.2 ± 0.33
Formulation B SC-GA/CBD Therapeutic 6.7mg/6.7mg/kg	4/5	1	18.5 ± 0.87	11.5 ± 0.9	1.25 ± 0.48	2.25 ± 0.48
C-No-treatment	4/5	1	19 ± 1.78	11 ± 1.78	2.67 ± 0.62	4.5 ± 0.5

The above table and Figure 5 presents clinical manifestations of EAE following intranasal administration of CBD and GA, following subcutaneous administration thereof versus untreated control and show that both the nasal administration and the subcutan administration of the drug combination were statistically significant efficient versus control ill mice in reducing the illness . * $P<0.05$ Formulation A vs. Control, **$P<0.05$ Formulation B vs. Control with the nasal administration being the most efficient treatment .

Claims

1) A pharmaceutical composition comprising the combination of cannabidiol and glatiramer acetate, and a pharmaceutically acceptable carrier.

2) A pharmaceutical composition according to claim 1, wherein the carrier is adapted for nasal administration.

3) A pharmaceutical composition according to claim 2, wherein the carrier is a liquid carrier.

4) A pharmaceutical composition according to claim 2, wherein the carrier comprises water, one or more C2-C4 alcohols and phospholipids.

5) A pharmaceutical composition according to claim 4 adapted for intranasal administration, which composition comprises the combination of glatiramer acetate and cannabidiol in a carrier comprising not less than 30% by weight water, from 12 to 30% by weight C2-C4 alcohol(s) and from 0.2 to 10% phospholipids arranged in a vesicular structure.

6) A pharmaceutical composition according to claim 5, wherein the C2-C4 alcohol is ethanol.

7) A pharmaceutical composition according to claim 5, which further comprises one or more water-miscible polyols.

8) Use of cannabidiol in combination with glatiramer acetate and a pharmaceutically acceptable carrier in the preparation of a nasally or parenterally administrable composition for the treatment of multiplesclerosis.

9) The use according to claim 8, wherein the carrier comprises phospholipid, one or more C2-C4 alcohols and water.

10) The use according to claim 9, wherein the concentrations of the phospholipid and the one or more C2-C4 alcohols in the composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, and the water content of said composition is not less than 30% by weight.

11) The use according to claim 10, wherein the C2-C4 alcohol is ethanol.

12) The use according to claim 10, wherein the composition further comprises one or more water-miscible polyols.

13) A pharmaceutical composition for intranasal administration comprising a combination of diclofenac and at least one active ingredient selected from the group consisting of diazepam and fentanyl, and a carrier, wherein said carrier comprises not less than 30% by weight water, from 12 to 30% by weight C2-C4 alcohol(s) and from 0.2 to 10% phospholipids arranged in a vesicular structure.

14) A pharmaceutical composition according to claim 13, wherein the C2-C4 alcohol is ethanol.

15) A pharmaceutical composition according to claim 13, which further comprises one or more water-miscible polyols.

16) The use of diclofenac and at least one active ingredient selected from the group consisting of diazepam and fentanyl, and a pharmaceutically acceptable carrier in the preparation of a nasally administrable composition for the treatment of pain, wherein the carrier comprises phospholipid, one or more C2-C4 alcohols and water, wherein the concentrations of said phospholipid and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, and the water content of said composition is not less than 30% by weight.

17) The use according to claim 16, wherein the C2-C4 alcohol present in the composition is ethanol.

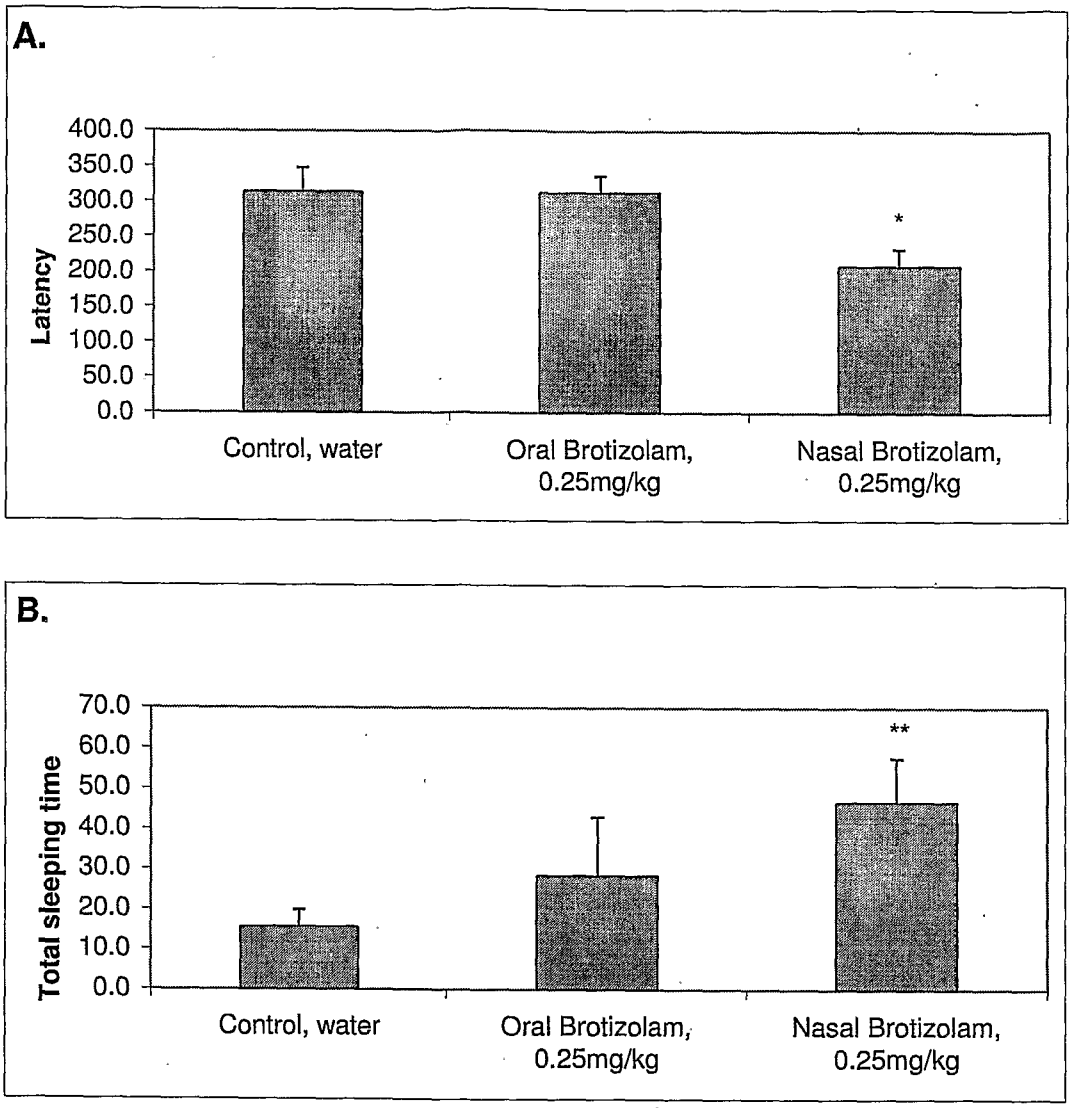
18) The use according to claim 17, wherein the composition further comprises one or more water-miscible polyols.

19) A pharmaceutical composition for treating insomnia comprising a therapeutically effective amount of brotizolam or pharmaceutically acceptable salt thereof, phospholipids, one or more C2-C4 alcohols and water, wherein the concentrations of the phospholipids and the one or more alcohols in the composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of the composition being not less than 30% by weight, the phospholipids forming vesicles in the composition.

20) Use of brotizolam, phospholipids, one or more C2-C4 alcohols and water in the preparation of a nasally administrable composition for treating insomnia, wherein the concentrations of said phospholipids and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of said composition being not less than 30% by weight.

21) A pharmaceutical composition comprising a therapeutically effective amount of prednisolone or pharmaceutically acceptable derivatives or salts thereof, phospholipids, one or more C2-C4 alcohols and water, wherein the concentrations of the phospholipids and the one or more alcohols in the composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of the composition being not less than 30% by weight, the phospholipids forming vesicles in the composition.

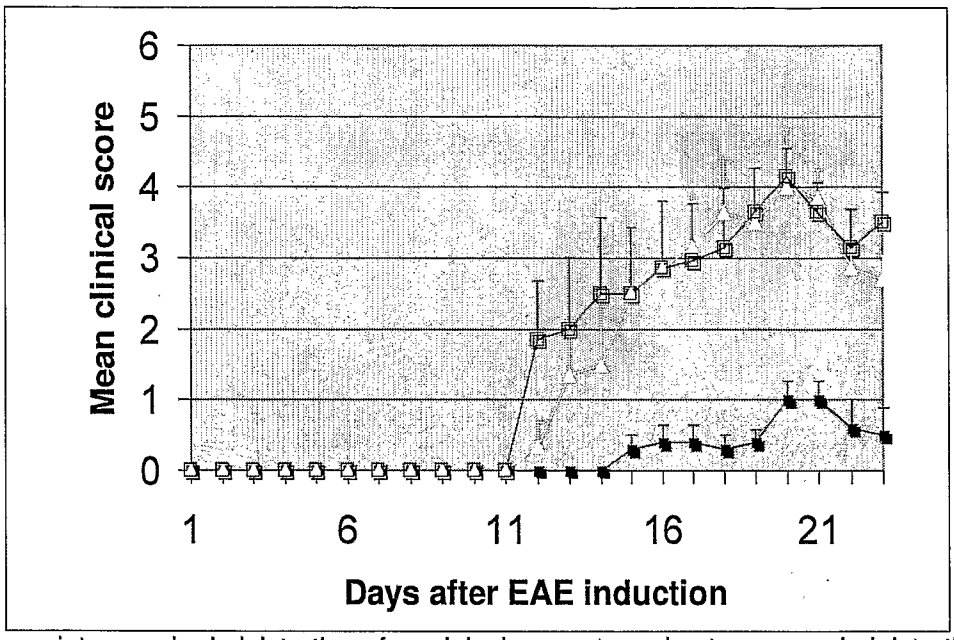
22) Use of prednisolone, phospholipids, one or more C2-C4 alcohols and water in the preparation of a composition adapted for intranasal administration, wherein the concentrations of said phospholipids and said one or more alcohols in said composition are in the ranges of 0.2 to 10% and 12 to 30% by weight, respectively, with the water content of said composition being not less than 30% by weight, the phospholipids forming vesicles in the composition.



* p<0.05 Nasal Brotizolam vs. Oral Brotizolam and vs. untreated control

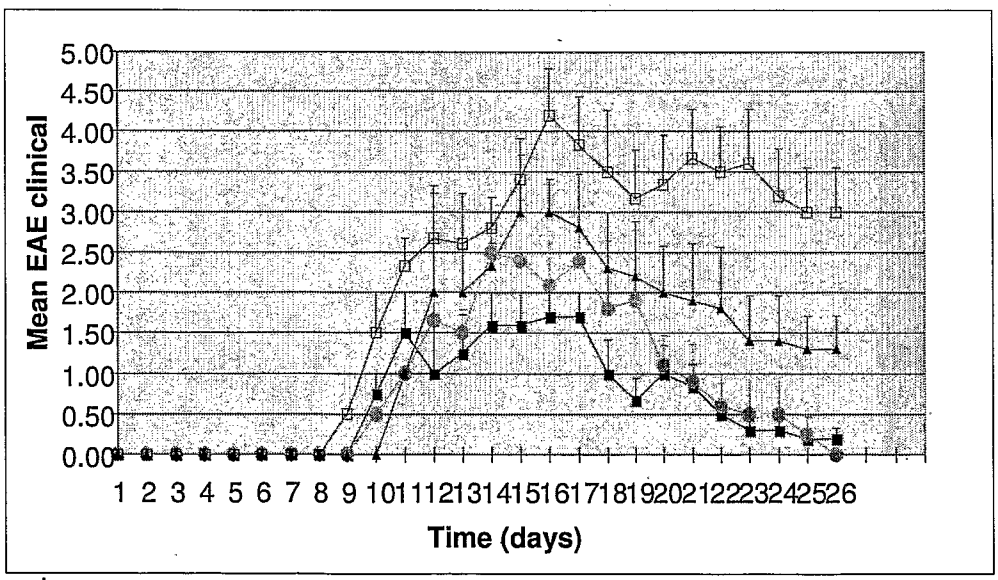
** p<0.05 Nasal Brotizolam vs. untreated control

Fig. 1



-■- intranasal administration of prednisolone; -▲- subcutaneous administration of prednisolone
-□- Untreated control animals

Fig. 2



-■ intranasal administration of prednisolone at 5.7mg/Kg animal;
-● intranasal administration of GA composition at a dose of 13.7mg/Kg animal;
-▲ subcutaneous administration of GA (13.7 mg/kg);
-□ control- no treatment

Fig. 3

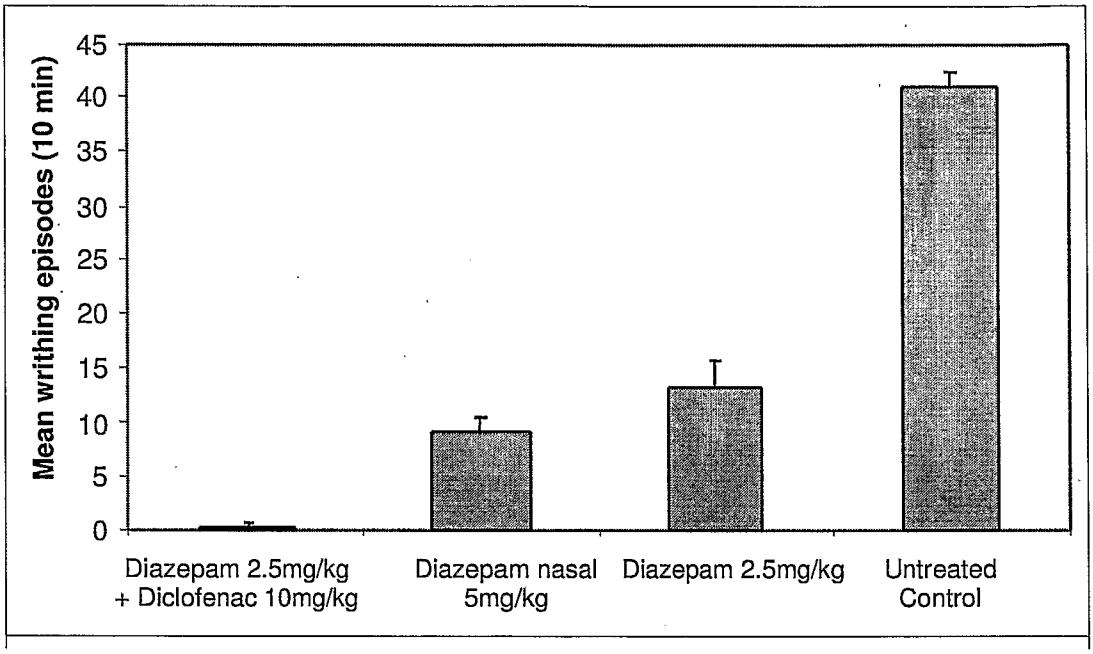
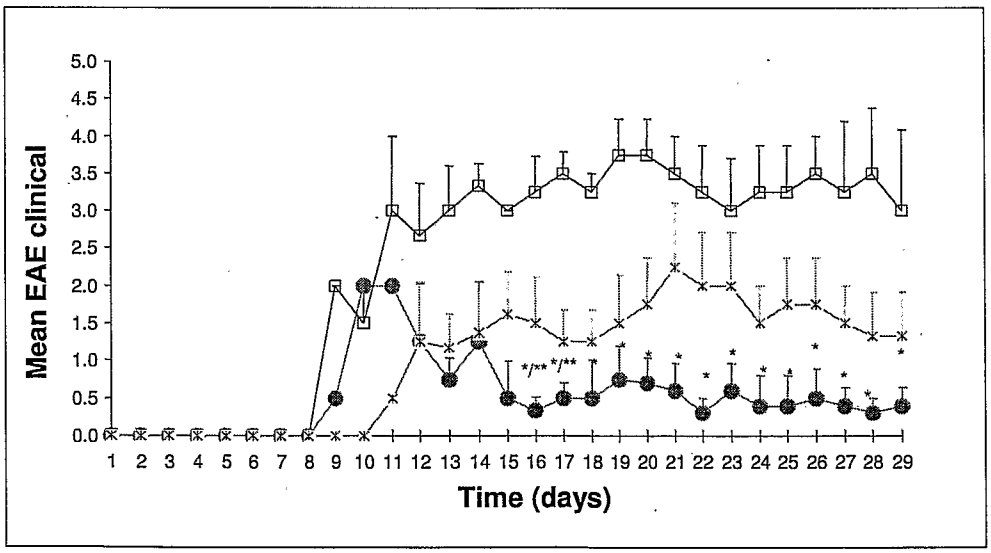


Fig. 4



^ ± SE (n=5). * P<0.05 Formulation A vs. Control, **P<0.05 Formulation B vs. Control

-□- Control- no treatment

● Intranasal GA-CBD -Formulation A

Fig. 5

✻ Subcutaneous GA- CBD -Formulation B

^Treatment was initiated when individual mouse developed a clinical score EAE ≥ 0.5