(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
14 April 2022 (14.04.2022)

WIPO | PCT

(10) International Publication Number
WO 2022/076852 A1

(51) International Patent Classification:
A61K 31/02 (2006.01) *A61K 9/00* (2006.01)

(21) International Application Number:
PCT/US2021/054221

(22) International Filing Date:
08 October 2021 (08.10.2021)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
63/089,263 08 October 2020 (08.10.2020) US
17/317,551 11 May 2021 (11.05.2021) US

(71) Applicant: **ADS THERAPEUTICS LLC** [US/US]; 15615 Alton Parkway, Suite 450, Irvine, California 92618 (US).

(72) Inventors: **NI, Jinsong**; 15615 Alton Parkway, Suite 450, Irvine, California 92618 (US). **YANG, Rong**; 15615 Alton Parkway, Suite 450, Irvine, California 92618 (US).

(74) Agent: **SHAN, Feng**; SZDC Law PC, 1629 K Street NW, Suite 300, Washington, District of Columbia 20006 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, IT, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, WS, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report (Art. 21(3))*

(54) Title: TOPICAL OPHTHALMOLOGICAL COMPOSITIONS

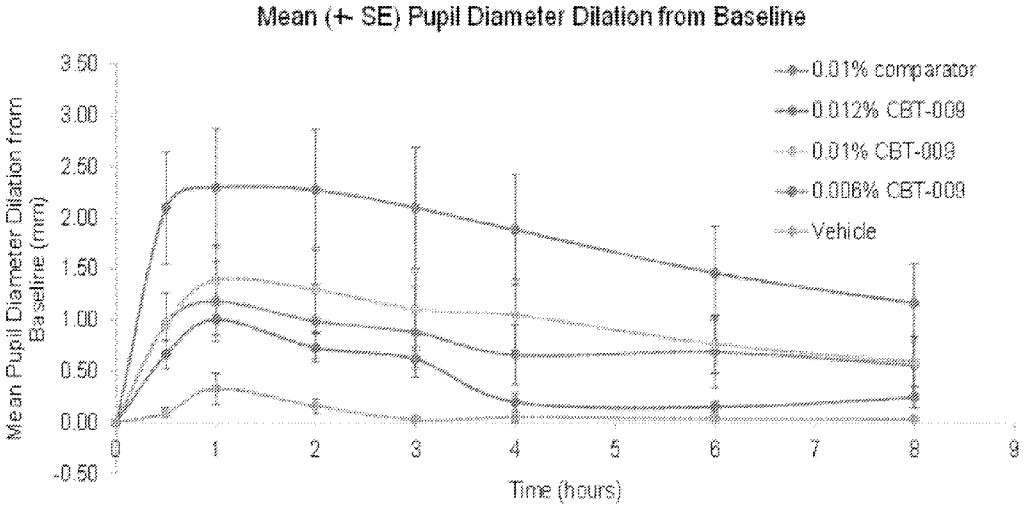


FIG. 2

(57) Abstract: A topical ophthalmological composition includes a therapeutically effective amount of a muscarinic receptor antagonist as an active pharmaceutical ingredient; and a semifluorinated alkane, as a liquid vehicle. The topical ophthalmological composition treats an ocular disease.

WO 2022/076852 A1

TOPICAL OPHTHALMOLOGICAL COMPOSITIONS

The present application claims priority to US Provisional Application No. 63/089,263, filed on October 8, 2020 and US Non-provisional Application No. 17/317,551, filed on May 11, 2021, both of which are incorporated by reference for all purposes as if fully set forth herein.

FIELD OF THE INVENTION

[0001] The present invention relates to topical ophthalmological compositions of a muscarinic receptor antagonist dissolved in semifluorinated alkane as a liquid vehicle, wherein, the formulation of atropine is used for treating myopia.

BACKGROUND OF THE INVENTION

[0002] Atropine is an anti-muscarinic compound and is a competitive antagonist of muscarinic receptors. It has anti-parasympathetic functions. It is used for several indications such as anticholinergic poisoning and bradycardia. In the eye, it is traditionally used for dilating pupil. Recently, low dose of atropine is shown be able to attenuate the progression of myopia in young adults (Li 2019). For the myopia indication, atropine is approved in only a few countries as of now.

[0003] Myopia, or nearsightedness, is a condition in which people can see close objects clearly, but objects farther away appear blurred. Myopia occurs if the eyeball is too long or the cornea (the clear front cover of the eye) is too curved so that distant objects can't be focused correctly on retina. Myopia is the most common eye disorder worldwide. About 30 percent of the U.S. population has myopia. The etiology of myopia is unknown. Genetics is believed to have a role in myopia. Myopia development may be affected by how a person uses the eyes. It may occur in school-age children and progresses until about age 20. However, myopia may also develop in adults due to visual stress or health conditions such as diabetes. Myopia may increase the risk of other ocular diseases (Wu 2019).

[0004] Atropine solution (water-based) formulations have been tested in multiple clinical trials and is proven to be able to slow down the progression of myopia (Cooper 2018, Li 2019, Yam 2020). In the water-based formulation, atropine is prone to degradation at neutral pH solution once the container is open to the air, therefore, the shelf life of the product at neutral pH is often less than 1 year. Low pH of 3-6 in the formulation is used to

increase the stability of atropine in solution (Berton 2020; Saito 2019). However, low pH is also known to cause irritation and discomfort in the eye.

[0005] This invention uses an organic liquid carrier to create a more stable and less irritating formulation of atropine for ocular, in particular myopia, indications.

SUMMARY OF THE INVENTION

[0006] In one embodiment, a topical ophthalmological composition includes: a therapeutically effective amount of a muscarinic receptor antagonist as an active pharmaceutical ingredient; and a semifluorinated alkane, as a liquid vehicle. The topical ophthalmological composition treats an ocular disease.

[0007] In another embodiment, the muscarinic receptor antagonist is selected from the group consisting of atropine, pirenzepine, aclidinium bromide, benztropine, cyclopentolate, diphenhydramine, doxylamine, dimenhydrinate, dicyclomine, darifenacin, flavoxate, hydroxyzine, ipratropium, mebeverine, oxybutynin, procyclidine, scopolamine, solifenacin, tropicamide, tiotropium, trihexyphenidyl, and tolterodine.

[0008] In another embodiment, the muscarinic receptor antagonist is atropine.

[0009] In another embodiment, the atropine is in a free base form or a salt form.

[0010] In another embodiment, the concentration of the atropine in a free base form is from about 0.0001% to about 1.0% (w/w), preferably, from about 0.001% to about 0.1% (w/w), more preferably, from about 0.01% to about 0.1% (w/w).

[0011] In another embodiment, the semifluorinated alkane is a compound of formula RFRH or of formula RFRHRF, wherein RF is a perfluorinated hydrocarbon with 15 or less carbon atoms, and wherein RH is a non-fluorinated hydrocarbon with 15 or less carbon atoms.

[0012] In another embodiment, the semifluorinated alkane is selected from F4H5, F4H6, F6H4, F6H6, F6H8 and F6H10.

[0013] In another embodiment, the semifluorinated alkane is F6H8 (perfluorohexyloctane).

[0014] In another embodiment, the topical ophthalmological composition further includes an organic cosolvent selected from the group consisting of phenylethyl alcohol, ethanol, isopropanol, glycerol, propylene glycol, and polyethylene glycol.

[0015] In another embodiment, the organic cosolvent is ethanol or phenylethyl alcohol.

[0016] In another embodiment, the concentration of ethanol is about 1% (w/w) or less, for example, 0.001% to 1% (w/w); or the concentration of phenylethyl alcohol is about 1% (w/w) or less, for example, 0.001% to 1% (w/w).

[0017] In another embodiment, the topical ophthalmological composition is a non-aqueous solution, a suspension, or an emulsion.

[0018] In another embodiment, the atropine in the topical ophthalmological composition is chemically stable for at least 0.5 year, for at least 1 year, or for at least 2 years.

[0019] In another embodiment, the topical ophthalmological composition is adapted for topically administering as eye drops to an eye of a patient.

[0020] In another embodiment, the topical ophthalmological composition causes minimal irritation in the eye.

[0021] In another embodiment, the ocular disease is myopia.

[0022] In another embodiment, the topical ophthalmological composition slows a myopia progression.

[0023] It is to be understood that both the foregoing general description and the following detailed description are exemplary and explanatory and are intended to provide further explanation of the invention as claimed.

BRIEF DESCRIPTION OF THE DRAWINGS

[0024] The accompanying drawings, which are included to provide a further understanding of the invention and are incorporated in and constitute a part of this

specification, illustrate embodiments of the invention and together with the description serve to explain the principles of the invention.

[0025] In the drawings:

[0026] Figure 1 shows the chromatogram of Atropine (tR: 12.947) standard solution.

[0027] Figure 2 shows pupil dilation effect in rabbit: CBT-009 = atropine F6H8 formulation; Comparator = atropine water formulation.

DETAILED DESCRIPTION OF THE ILLUSTRATED EMBODIMENTS

[0028] Reference will now be made in detail to embodiments of the present invention, example of which is illustrated in the accompanying drawings.

[0029] A muscarinic receptor antagonist is an anticholinergic agent that blocks the activities of a muscarinic acetylcholine receptor. The muscarinic receptor antagonist may be atropine, pirenzepine, aclidinium bromide, benztropine, cyclopentolate, diphenhydramine, doxylamine, dimenhydrinate, dicyclomine, darifenacin, flavoxate, hydroxyzine, ipratropium, mebeverine, oxybutynin, procyclidine, scopolamine, solifenacin, tropicamide, tiotropium, trihexyphenidyl, or tolterodine. Preferably, the muscarinic receptor antagonist is atropine or pirenzepine. More preferably, the muscarinic receptor antagonist is atropine.

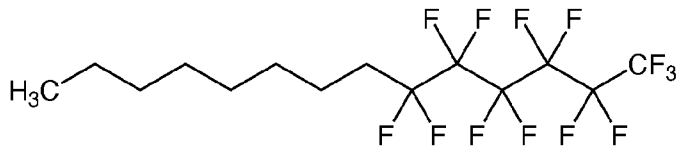
[0030] The topical ophthalmological composition of the present invention includes a therapeutically effective amount of the muscarinic receptor antagonist, e.g., atropine. A therapeutically effective amount means an amount effective to prevent development of or to alleviate the existing symptoms of the subject being treated. Determination of the effective amount is well within the capability of those skilled in the art, especially in light of the detailed disclosure provided herein.

[0031] Atropine solution (water) formulations had been previously proven effective in treating myopia, specially reducing myopia progression (Li 2019, Wu 2019). The atropine sulfate monohydrate aqueous formulation had been tested in the clinic at concentrations ranging from 0.01% to 1% and all these doses have demonstrated efficacy in myopia treatment. The solution formulation had two drawbacks. The first is that once the container opens to air, the atropine at neutral pH in the solution is prone to degradation, therefore, the shelf life of the product at neutral pH is often less than 1 year. Furthermore, this instability of

the atropine in the solution requires that the formulation is used within about a month. The second shortcoming is that the low pH, such as in the pH range of 3.5 to 6.0, used to reduce atropine degradation to increase product shelf life, can cause irritation or discomfort to the human eye as reported of adverse events in the patients.

[0032] This disclosure provides compositions using a semifluorinated alkane, in particular F6H8 (perfluorohexyloctane), as the liquid vehicle to dissolve atropine to eliminate the two shortcomings of the solution formulation. F6H8 is an amphiphilic liquid with two mutually immiscible moieties (hydrocarbon segment as RH and perfluorinated segment as RF) bound covalently. Other related analogies used in the compositions of the present inventions may be F4H5 (perfluorobutylpentane), F4H6 (perfluorobutylhexane), F6H4 (perfluorohexylbutane), F6H6 (perfluorohexylhexane), and F6H10 (perfluorohexyldecane).

[0033] The structure of F6H8 is shown below.



F6H8 ($CF_3(CF_2)_5(CH_2)_7CH_3$)

[0034] In some embodiments, the disclosure is based on the studies described in the examples that show atropine can be dissolve in F6H8 at sufficient concentration to have biological efficacy. The formulation of atropine in F6H8 is stable for prolonged times at room temperature and can be made into a product with sufficient self-life for regulatory approval. This formulation is not irritating in the eye in animal model studies when dosed at a concentration higher than what is needed for some indications.

[0035] Examples 1 and 2 demonstrated that stable atropine formulations ranging from about 0.0001 to 0.15% can be achieved in F6H8 with the addition of three co-solvents. Example 7 showed that equivalent doses of the atropine free base F6H8 organic formulation achieved similar efficacy to that of the atropine sulfate monohydrate aqueous formulation in a rabbit model. Our studies, combined with the known efficacy of the aqueous formulation in the range of 0.01-1%, indicated that the achievable range of concentration, from about 0.0001% to about 1.0% or from 0.0001% to 1.0% (free base, w/w), preferably, from about

0.001% to about 0.1% or from 0.001% to 0.1%, more preferably, about 0.01% to about 0.1% or 0.01% to 0.1%, in the F6H8 formulation will also be efficacious.

[0036] Examples

[0037] Example 1: Dissolution of atropine in F6H8.

[0038] Methods: Formulations of atropine free base were investigated according to the following procedure:

[0039] 1. Dissolving Atropine

[0040] Added more than 4 mg of atropine powder in 4 mL of F6H8 or F6H8 with 0.1% ethanol, yielding about 1 mg/mL. Stirred the formulation for 2 days.

[0041] 2. Preparing HPLC Samples

[0042] Centrifuged the formulations above and filtered the supernatants through 0.45 micron filters without further dilution. One sample was prepared from each solvent for HPLC analysis.

[0043] 3. Analyzing the HPLC Samples

[0044] The samples were analyzed using a RP-HPLC method with an Agilent Eclipse Plus C18 HPLC column (150 mm X 2.1 mm I.D.) connected with a guard column (12.5 mm X 2.1 mm I.D.) and a gradient elution from 100% water to 100% acetonitrile at a flow rate of 0.2 ml/min. The chromatograms were monitored at UV at 220 nm. The atropine peak is at retention time 12.947 as shown in the chromatograph in Figure 1.

[0045] Results

[0046] The solubility of atropine was determined to be 129 μg/ml (0.0129% w/w) in F6H8 alone. When 0.1% ethanol was added, the solubility was 171.5 μg/ml (0.0173% w/w). In this particular study, the free base form of atropine was used, while the mono sulfate salt was previously used in the solution formulation approved for myopia usage. The MW of the free base is 83% equivalent to the mono sulfate salt form of atropine solution formulation. The 0.01% atropine mono sulfate salt solution was previously shown effective for myopia treatment in the clinic and was approved in several countries. This 0.01% atropine salt

concentration was equivalent to 0.0083% of the free base concentration. Since the maximum atropine free base that we observed in F6H8 was 0.0129%, we concluded that the F6H8 formulation can deliver sufficient amount of atropine for the treatment of myopia. The 0.0129% concentration we observed was about 55% higher than the 0.0083% needed for efficacy. In addition, we observed that the concentration of atropine can be increased further by adding ethanol to the formulation. The addition of just 0.1% ethanol increased the solubility by 33%. Higher levels of ethanol would likely further increase the solubility of atropine in F6H8. The concentrations of atropine in the F6H8 formulations are show in Table 1.

Table 1: Concentrations of Atropine in F6H8 formulations

Sample Descriptions	Atropine in F6H8	Atropine in F6H8 with 0.1% (v/v) Ethanol
Solubility (concentrations)	129.0 μg/mL	171.5 μg/mL

[0047] Using slightly different preparation method, excess amount of atropine free base was added to 100% F6H8. The mixture was heated to 40°C and stirred for 15 minutes. After centrifugation and sit still for 16 hours, the sample (Sample ID 1379308) was aliquoted and measured at the concentration of 214.55 μg/mL. Adding additional atropine free base and continuously stirring for 66 hours, the sample (Sample ID 140933) was centrifuged, aliquoted and measured at the concentration of 219.47 μg/mL. Therefore, the saturated solubility of atropine free base in 100% F6H8 was determined at 217.01 μg/mL by averaging 214.55 μg/mL and 219.47 μg/mL.

[0048] Increased atropine solubility by addition of co-solvents:

[0049] Additional studies were carried out to determine the stable concentration of atropine when a co-solvent was added. As shown in Table 2, the addition of 0.25%, 0.50% or 0.75% phenylethyl alcohol increased the atropine solubility to 0.043%, 0.055% or 0.085%, respectively; the addition of 0.25%, 0.5% or 0.75% ethanol increased the atropine solubility to 0.031%, 0.089% or 0.108%, respectively; the addition of 0.25%, 0.5%, 0.75% or 2% isopropanol increased the atropine solubility to 0.046%, 0.055%, 0.082% or 0.153%, respectively. After serial 10x dilution of each sample of 0.75% co-solvent in F6H8, the atropine formulations remained stable at levels down to about 0.00008%.

Table 2 Concentration of atropine in F6H8 with different types of co-solvents at various concentrations

Compositions with phenylethyl alcohol as co-solvent

Topical ophthalmological composition		Atropine concentraion (w/w %)
Semifluorinated alkane (F6H8)	Co-solvent (phenylethyl alcohol)	Measured saturating concentraion by HPLC/UV
99.75%	0.25%	0.043
99.50%	0.50%	0.055
99.25%	0.75%	0.085
Semifluorinated alkane (F6H8)	Co-solvent (phenylethyl alcohol)	Observed stable concentration after 10X serial dilution of the saturating concentration
99.25%	0.75%	0.0085
99.25%	0.75%	0.00085
99.25%	0.75%	0.000085

Compositions with ethanol as co-solvent

Topical ophthalmological composition		Atropine concentraion (w/w %)
Semifluorinated alkane (F6H8)	Co-solvent (ethanol)	Measured saturating concentraion by HPLC/UV
99.75%	0.25%	0.031
99.50%	0.50%	0.089
99.25%	0.75%	0.108
Semifluorinated alkane (F6H8)	Co-solvent (ethanol)	Observed stable concentration after 10X serial dilution of the saturating concentration
99.25%	0.75%	0.0108
99.25%	0.75%	0.00108
99.25%	0.75%	0.000108

Compositions with isopropanol as co-solvent

Topical ophthalmological composition		Atropine concentraion (w/w %)
Semifluorinated alkane (F6H8)	Co-solvent (isopropanol)	Measured saturating concentraion by HPLC/UV
99.75%	0.25%	0.046
99.50%	0.50%	0.055
99.25%	0.75%	0.082
98.00%	2.00%	0.153
Semifluorinated alkane (F6H8)	Co-solvent (isopropanol)	Observed stable concentration after 10X serial dilution of the saturating concentration
99.25%	0.75%	0.0082
99.25%	0.75%	0.00082
99.25%	0.75%	0.000082

[0050] Example 2: The atropine F6H8 formulation is stable over time

[0051] Methods

[0052] Atropine is dissolved in F6H8 as described in Example 1. The level of atropine is measure by the HPLC method at 25°C at 1, 3, 6, 9 and 12 months. The atropine in the formulation is defined as stable if the level is maintained between 90-110% of the original level.

[0053] Results

[0054] During the study period, atropine is stable as shown in Table 3 below.

Table 3: Atropine stability in F6H8 formulation

Time point (month)	1	3	6	9	12
Remaining 90-110% of time zero (yes/no)	yes	yes	yes	yes	yes

[0055] Example 3: The atropine F6H8 formulation is tolerable in a rabbit study

[0056] Methods

[0057] The atropine F6H8 formulation is evaluated in rabbits for ocular tolerability. The study design and assessments are shown in Tables 4 and 5.

Table 4: Experiment Design

Group	Number of Animals & Sex	Right Eye	Left Eye	Dose Frequency
3	3F	Vehicle	0.01% atropine	Four Times per day, 4 hrs apart

Table 5: Study Assessments

Parameters	**Descriptions**
Viability	Twice daily
Clinical Observation	Once during the predose and once daily during the dosing phase after the last daily dose.

Body weight	Once during the predose, and on Day 1, Day 7 and Day 14
Food consumption	Once daily during predose and dosing phase
Ocular Discomfort observation	Twice (on different days) during the predose phase, daily during the dosing phase after the last daily dose. Both eyes will be grossly examined and graded using a modified Hackett-McDonald grading scale by technical staff.
Ocular Irritation Observation (Modified Hackette McDonald)	Twice (on different days) during the predose phase, daily during the dosing phase after the third daily dose. Both eyes will be grossly examined and graded using a modified Hackett-McDonald grading scale by technical staff.
Cornea Examination	Once predose phase and once after the last daily dose on Day 1 and Day 14. Both eyes will be examined for corneal opacity and % of corneal opacity using slit lamp and will be taken photos.

[0058] Results and conclusions

[0059] The atropine formulation is well tolerated in rabbits with no significant irritation and discomfort issues.

[0060] Example 4: Stability of atropine in F6H8

[0061] 0.0125% atropine was prepared by dissolving appropriate amount of atropine free base in 100% F6H8. The stability of atropine formulation over time was assessed at 25°C. At 1 month, 2 month and 3 month time point, atropine was extracted with acetonitrile twice and quantitated by HPLC as described in Example 1. Table 6 below showed that atropine levels remained stable at least for 3 months with minimum change from the target concentration. This example, disclosed for the first time, demonstrated that the atropine formulation in the invention was stable for at room temperature.

Table 6: Stability of 0.0125% atropine free base in 100% F6H8

Sample Name	Time point	Target Conc.(ug/ml)	Actual Conc.(ug/ml)
S20210525-01	1 Month	125	120.34
	2 Month	125	123.31
	3 Month	125	133.00

[0062] In a separate experiment, 0.01% atropine was dissolved in F6H8 and 0.25% phenylethyl alcohol. The stability of the atropine formulation over time was assessed at 25°C and 40°C. At selected time points, atropine was extracted with acetonitrile twice and quantitated by HPLC as describe in Example 1. Table 7 below showed that atropine levels remained stable at Days 32 and 84 without significant change from the baseline at Day 0. The results were similar at both room temperature and accelerated temperature. The stability at accelerated temperature indicated that the formulation can be potentially stored at room temperature for months or years without significant loss of atropine. This example, disclosed for the first time, demonstrated that the atropine formulation in the invention was stable for prolonged storage at room temperature.

Table 7

Stability of atropine in F6H8 and 0.25% phenylethyl alcohol				
Time (days)	**25 °C**		**40 °C**	
	Concentration (µg/mL)	**% of time 0**	**Concentration (µg/mL)**	**% of time 0**
0	99.2		99.2	
32	94.0	94.8%	94.5	95.3%
84	110.8	111.7%	114.3	115.2%

[0063] Example 5: *In vivo* ocular tolerability in rabbits

[0064] Study Design:

[0065] Three (3) female Dutch belted rabbits were given 40 µL of Control Article (0.01% atropine sulfate monohydrate in normal saline) to the right eyes and 40 µL of 0.012% atropine free base in 0.25% phenylethyl alcohol (PEA) in F6H8 to the left eyes, 1 drop/eye, twice per day, 12 hrs apart for 14 consecutive days. Ocular discomfort observation and ocular

irritation observation were performed for all animals at predose (twice, on different days) and daily during the dosing phase after the last daily dose. Cornea examination were performed for all animals at predose (once) phase and once after the last daily dose on Day 1 and Day 14. The first dosing day were designated as Day 1.

[0066] The ocular irritation scores on Day 14 were shown Table 8 below. Other time points had similar or better results.

Table 8

Day 14													
Animal ID	Subject	Cornea				Iris		Conjunctiva					
		Opacity intensity		Opacity area				Congestion		Swelling		Discharge	
		OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS
C0866	1501	0	0	0	0	0	0	0	0	1	1	0	0
C0863	1502	0	0	0	0	0	0	0	0	1	1	0	0
C0867	1503	0	0	0	0	0	0	0	0	1	1	0	0

[0067] McDonald-Shadduck scoring (categories with positive scores) was shown in Table 9 below.

Table 9

Day 1																			
Animal ID	Subject	Conjunctiva						Aqueous Flare		Iris		Cornea						Fluorescein	
		Congestion				Discharge						Cloudiness		Cloudiness Area		Pannus			
		OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS
C0866	1501	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0
C0863	1502	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
C0867	1503	0	0	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Day 14																			
Animal ID	Subject	Conjunctiva						Aqueous Flare		Iris		Cornea						Fluorescein	
		Congestion				Discharge						Cloudiness		Cloudiness Area		Pannus			
		OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS
C0866	1501	0	0	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0
C0863	1502	0	0	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	1
C0867	1503	0	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	1	0

[0068] Conclusion: The atropine formulation was well tolerated in all animals. No significant ocular irritation or ophthalmic findings were observed in any animals. There were no test article-related effects on body weights and food consumption during the studies in both species. There were no other test article-related ophthalmologic findings during the

scheduled examinations for all animals. This Example demonstrated the safety of the claimed novel formulation of atropine for ocular use.

[0069] Example 6: *In vivo* ocular tolerability in dogs

[0070] Study design

[0071] Three (3) male Beagle dogs were given 40 µL of Control Article (0.01% atropine sulfate monohydrate in normal saline) to the right eyes and 40 µL of 0.012% atropine free base in 0.25% phenylethyl alcohol (PEA) in F6H8 to the left eyes, 1 drop/eye, twice per day, 12 hrs apart for 14 consecutive days. Ocular discomfort observation and ocular irritation observation were performed for all animals at predose (twice, on different days) and daily during the dosing phase after the last daily dose. Cornea examination were performed for all animals at predose (once) phase and once after the last daily dose on Day 1 and Day 14. The first dosing day was designated as Day 1.

[0072] The ocular irritation scores on Day 14 were shown in Table 10 below. Other time points had similar or better results.

Table 10

Day 14													
Animal ID	Subject	Cornea				Iris		Conjunctiva					
		Opacity intensity		Opacity area				Congestion		Swelling		Discharge	
		OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS
8525538	1001	0	0	0	0	0	0	1	1	0	0	0	0
8370950	1002	0	0	0	0	0	0	0	0	0	0	0	0
8473172	1003	0	0	0	0	0	0	1	1	0	0	0	0

[0073] McDonald-Shadduck scoring (categories with positive scores) was shown in Table 11 below.

Table 11

Day 1																			
Animal ID	Subject	Conjunctiva						Aqueous Flare		Iris		Cornea						Fluorescein	
		Congestion				Discharge						Cloudiness		Cloudiness Area		Pannus			
		OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS
8525538	1001	0	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0	0	1
8370950	1002	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1
8473172	1003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Day 14																			
Animal ID	Subject	Conjunctiva						Aqueous Flare		Iris		Cornea						Fluorescein	
		Congestion				Discharge						Cloudiness		Cloudiness Area		Pannus			
		OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS	OD	OS
8525538	1001	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8370950	1002	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8473172	1003	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

[0074] Conclusion: The atropine formulation was well tolerated in all animals. No significant ocular irritation or ophthalmic findings were observed in any animals. There were no test article-related effects on body weights and food consumption during the studies in both species. There were no other test article-related ophthalmologic findings during the scheduled examinations for all animals. This Example demonstrated the safety of the claimed novel formulation of atropine for ocular use.

[0075] Example 7: *In vivo* pharmacological potency in a rabbit model

[0076] The pharmacological potency of the atropine formulation in F6H8 and 0.25% phenylethyl alcohol was tested in a rabbit model. The potency was measured as pupil dilation in normal naïve rabbits. Three concentrations of the F6H8 formulation of atropine (0.012%, 0.01%, 0.08%) were compared to that of an aqueous formulation of 0.01% atropine which was known to have good pupil dilation effects. One drop of each formulation was dosed into the eye and pupil size was measured during the following 8 hours.

[0077] Study design

[0078] Fifteen (15) female Dutch belted rabbits were assigned to five groups, which included 3 animals/group. Three (3) female Dutch belted rabbits were randomly assigned to each group by Provantis or Excel based on body weight. The dosing of animals was performed in 2 phases, Phase 1 and Phase 2.

[0079] In phase 1, each animal was given 40 µL of testing article (see Table 12 below) to both eyes. First day of dosing was designated as Day 1. The pupil size of both

eyes of all animals were measured at baseline (30 minutes before dosing), 0.5h, 1h, 2h, 3h, 4h, 6h, 8h after dosing on day 1. The pupil size measurement data were analyzed for efficacy to determine which doses of atropine free base in Vehicle was equivalent to the dose of the control group of 0.01% atropine sulfate monohydrate in normal saline. Animals were allowed 2 days for wash-out period.

[0080] In phase 2, each animal was given 40 µL of testing article (see Table 13 below) to both eyes for 14 days. First day of dosing in Phase 2 was designated as Day 4. The pupil size of both eyes of all animals were measured at baseline (30 minutes before dosing), 0.5h, 1h, 2h, 3h, 4h, 6h, 8h after dosing on Day 4 and Day 17.

Table 12: The study design of phase 1

Group/ Code Color	Animals[a]	Treatment	Animal Number	Dosage & Frequency
		Both eyes	Female	Once a day, 1 drop/eye, on Day 1 followed by 2 days wash-out period
1/White	3	0.01% atropine sulfate monohydrate in normal saline	1501-1503	
2/Green	3	0.012% atropine free base in Vehicle	2501-2503	
3/Yellow	3	0.008% atropine free base in Vehicle	3501-3503	
4/Red	3	0.005% atropine free base in Vehicle	4501-4503	
5/Cyan	3	Vehicle	5501-5503	

Note: [a] Replacement animals, if any, will be numbered per Testing Facility SOP and will be included in the study report.
Vehicle: 0.25% phenylethyl alcohol in 1-(perfluorohexyl)octane

Table 13: The study design of phase 2

Group/ Code Color	Animals[a]	Treatment	Animal Number	Dosage & Frequency
		Both eyes	Female	Once a day, 1 drop/eye, on Day 4 to Day 17. Pupil size will be measured on Day 4 and Day 17 only.
1/White	3	0.01% atropine sulfate monohydrate in normal saline	1501-1503	
2/Green	3	Dose to be determined after Day 1[b]	2501-2503	
3/Yellow	3	Dose to be determined after Day 1[b]	3501-3503	
4/Red	3	Dose to be determined after Day 1[b]	4501-4503	
5/Cyan	3	Vehicle	5501-5503	

Note: [a] Replacement animals, if any, will be numbered per Testing Facility SOP and will be included in the study report.

[b]Equivalent dose is determined from Phase 1 efficacy data. The optimized concentrations of atropine free base in Vehicle that gives equivalent efficacy as 0.01% atropine sulfate monohydrate in normal saline.

Vehicle: 0.25% phenylethyl alcohol in 1-(perfluorohexyl)octane

[0081] Results

[0082] As shown in Figure 2, the F6H8 formulation of atropine increased pupil size with similar potency to that of the water formulation. The 0.01% F6H8 formulation was slightly more effective than the water formulation. This observation indicated that the novel F6H8 of atropine was as effective as a proven atropine formulation and can be used for the treatment of diseases with water-based formulations. Figure 2 showed pupil dilation effect in rabbit: CBT-009 = atropine F6H8 formulation; Comparator = atropine water formulation.

[0083] It will be apparent to those skilled in the art that various modifications and variations can be made in the present invention without departing from the spirit or scope of the invention. Thus, it is intended that the present invention cover the modifications and variations of this invention provided they come within the scope of the appended claims and their equivalents.

References

Berton B, Chennell P, Yessaad M, Bouattour Y, Jouannet M, Wasiak M, Sautou V. Stability of Ophthalmic Atropine Solutions for Child Myopia Control. Pharmaceutics. 2020 Aug 17;12(8):E781.

Cooper J, Tkatchenko AV. A Review of Current Concepts of the Etiology and Treatment of Myopia. Eye Contact Lens. 2018 Jul;44(4):231-247.

Li FF, Yam JC. Low-Concentration Atropine Eye Drops for Myopia Progression. Asia Pac J Ophthalmol (Phila). 2019 Sep-Oct;8(5):360-365.

Saito J, Imaizumi H, Yamatani A. Physical, chemical, and microbiological stability study of diluted atropine eye drops. J Pharm Health Care Sci. 2019 Dec 5;5:25.

Wu PC, Chuang MN, Choi J, Chen H, Wu G, Ohno-Matsui K, Jonas JB, Cheung CMG. Update in myopia and treatment strategy of atropine use in myopia control. Eye (Lond). 2019 Jan;33(1):3-13.

Yam JC, Li FF, Zhang X, Tang SM, Yip BHK, Kam KW, Ko ST, Young AL, Tham CC, Chen LJ, Pang CP. Two-Year Clinical Trial of the Low-Concentration Atropine for Myopia Progression (LAMP) Study: Phase 2 Report. Ophthalmology. 2020 Jul;127(7):910-919.

WHAT IS CLAIMED IS:

1. A topical ophthalmological composition comprising:

a therapeutically effective amount of a muscarinic receptor antagonist as an active pharmaceutical ingredient; and

a semifluorinated alkane, as a liquid vehicle,

wherein the topical ophthalmological composition treats an ocular disease.

2. The topical ophthalmological composition of claim 1, wherein the muscarinic receptor antagonist is selected from the group consisting of atropine, pirenzepine, aclidinium bromide, benztropine, cyclopentolate, diphenhydramine, doxylamine, dimenhydrinate, dicyclomine, darifenacin, flavoxate, hydroxyzine, ipratropium, mebeverine, oxybutynin, procyclidine, scopolamine, solifenacin, tropicamide, tiotropium, trihexyphenidyl, and tolterodine.

3. The topical ophthalmological composition of claim 2, wherein the muscarinic receptor antagonist is atropine.

4. The topical ophthalmological composition of claim 3, wherein the atropine is in a free base form or a salt form.

5. The topical ophthalmological composition of claim 4, wherein the concentration of the atropine in a free base form is from about 0.0001% to about 1.0% (w/w), preferably, from about 0.001% to about 0.1% (w/w), more preferably, about 0.01% to about 0.1% (w/w).

6. The topical ophthalmological composition of claim 1, wherein the semifluorinated alkane is a compound of formula RFRH or of formula RFRHRF, wherein RF is a perfluorinated hydrocarbon with 15 or less carbon atoms, and wherein RH is a non-fluorinated hydrocarbon with 15 or less carbon atoms.

7. The topical ophthalmological composition of claim 6, wherein the semifluorinated alkane is selected from the group consisting of F4H5, F4H6, F6H4, F6H6, F6H8 and F6H10.

8. The topical ophthalmological composition of claim 7, wherein the semifluorinated alkane is F6H8 (perfluorohexyloctane).

9. The topical ophthalmological composition of claim 1, further comprising an organic cosolvent selected from the group consisting of phenylethyl alcohol, ethanol, isopropanol, glycerol, propylene glycol, and polyethylene glycol.

10. The topical ophthalmological composition of claim 9, wherein the organic cosolvent is ethanol or phenylethyl alcohol.

11. The topical ophthalmological composition of claim 10, wherein the concentration of ethanol is about 1% (w/w) or less; or the concentration of phenylethyl alcohol is about 1% (w/w) or less.

12. The topical ophthalmological composition of claim 1, wherein the topical ophthalmological composition is a non-aqueous solution, a suspension, or an emulsion.

13. The topical ophthalmological composition of claim 12, wherein the atropine in the topical ophthalmological composition is chemically stable for at least 0.5 year, for at least 1 year, or for at least 2 years.

14. The topical ophthalmological composition of claim 1, wherein the topical ophthalmological composition is adapted for topically administering as eye drops to an eye of a patient.

15. The topical ophthalmological composition of claim 14, wherein the topical ophthalmological composition causes minimal irritation in the eye.

16. The topical ophthalmological composition of claims 1, wherein the ocular disease is myopia.

17. The topical ophthalmological composition of claim 16, wherein the topical ophthalmological the composition slows a myopia progression.

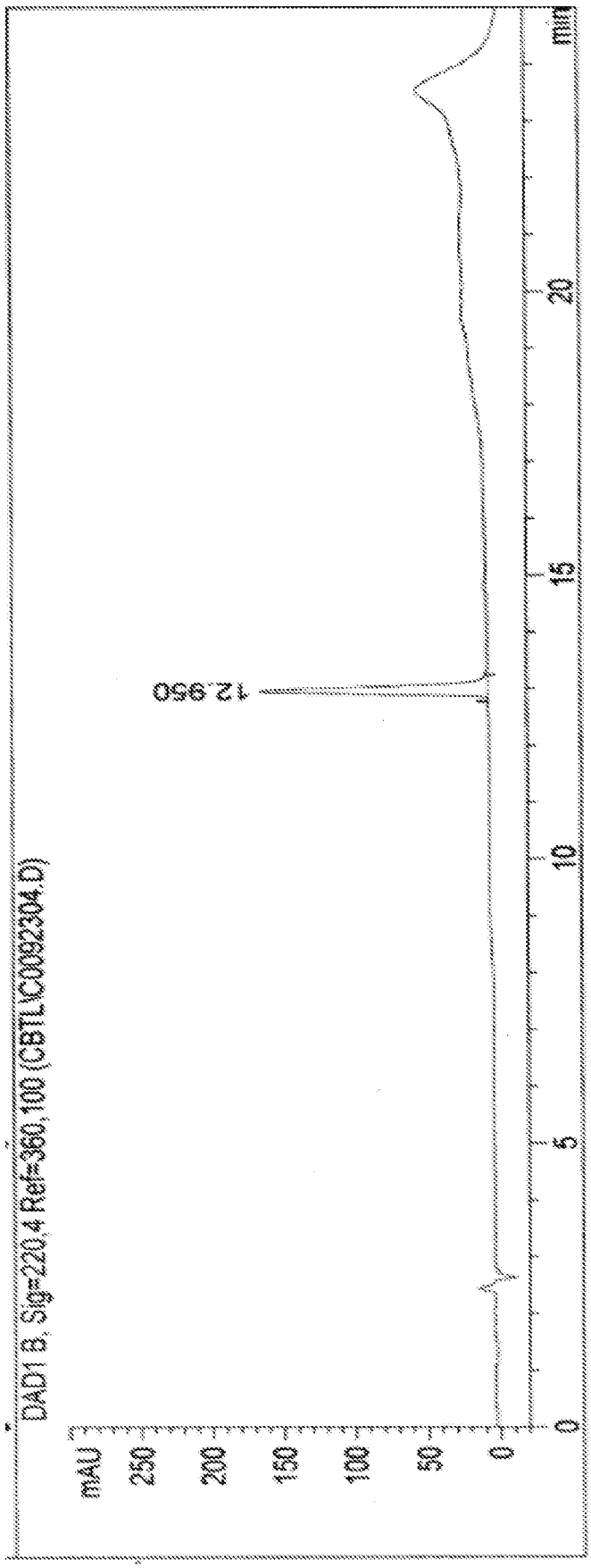

DAD1 B, Sig=220,4 Ref=360,100 (CBTLIC009230 4.D)
mAU
250
200
150
100
50
0
12.950
0
5
10
15
20
min

FIG. 1

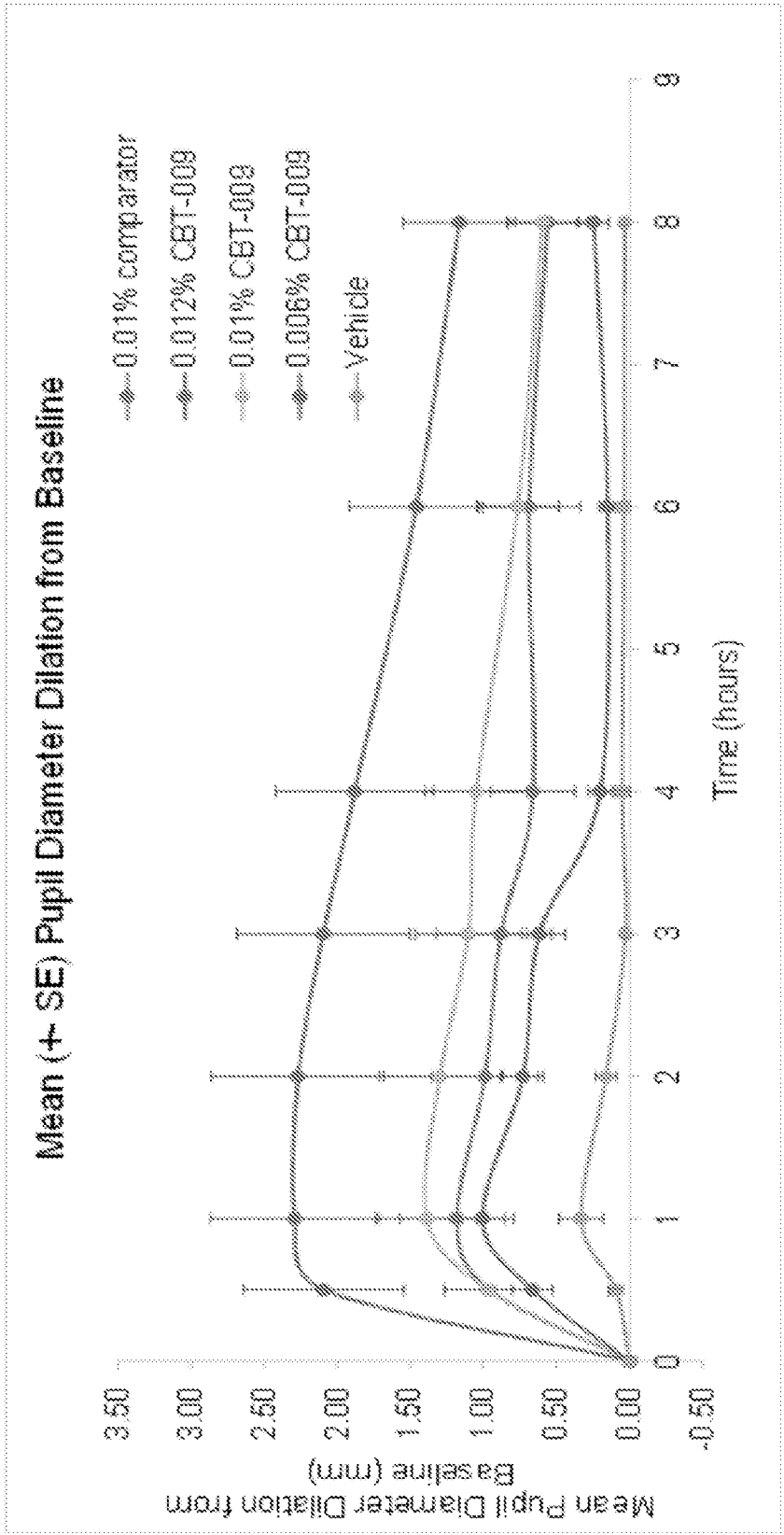

Mean (+- SE) Pupil Diameter Dilation from Baseline
0.01% comparator
0.012% CBT-009
0.01% CBT-009
0.006% CBT-009
Vehicle
Mean Pupil Diameter Dilation from Baseline (mm)
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00
-0.50
0
1
2
3
4
5
6
7
8
9
Time (hours)

FIG. 2

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 21/54221

A. CLASSIFICATION OF SUBJECT MATTER

IPC - A61K 31/02; A61K 9/00 (2021.01)

CPC - A61K 31/02; A61K 47/24; A61K 9/0048

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

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
See Search History document

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

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
See Search History document

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- Y	WO 2020/074697 A1 (NOVALIQ GMBH) 16 April 2020 (16.04.2020) - entire document especially abstract and pg 6, 8, 10, 11	1-17 --- 8, 16-17
X --- Y	US 2020/0268682 A1 (NOVALIQ GMBH) 27 August 2020 (27.08.2020) - entire document especially para [0017], [0026], [0027], [0028], [0664], [0677]	1, 12-13 --- 8
Y	WO 2020/160493 A2 (AERIE PHARMACEUTICALS, INC.) 6 August 2020 (06.08.2020) - entire document especially para [0004] [0048]	16, 17
A	US 2019/0328717 A1 (NOVALIQ GMBH) 31 October 2019 (31.10.2019) - entire document	1-17
A	WO 2020/152046 A1 (NOVALIQ GMBH) 30 July 2020 (30.07.2020) - entire document	1-17
A	US 2019/0343793 A1 (NOVALIQ GMBH) 14 November 2019 (14.11.2019) - entire document	1-17
A	US 2019/0343848 A1 (MEDICON PHARMACEUTICALS, INC.) 14 November 2019 (14.11.2019) - entire document	1-17

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

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"D" document cited by the applicant in the international application
"E" earlier application or patent but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
3 December 2021	JAN 21 2022
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-8300	Authorized officer Kari Rodriquez Telephone No. PCT Helpdesk: 571-272-4300