



(51) International Patent Classification:

A61K 31/549 (2006.01) A61P 11/02 (2006.01)

A61K 45/06 (2006.01) A61P 27/02 (2006.01)

(21) International Application Number:

PCT/US2024/020422

(22) International Filing Date:

18 March 2024 (18.03.2024)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

63/452,724 17 March 2023 (17.03.2023) US

(71) Applicants: **EMORY UNIVERSITY** [US/US]; Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **CHILDREN'S HEALTH-CARE OF ATLANTA, INC.** [US/US]; 1575 Northeast Expressway, Atlanta, Georgia 30329 (US).

(72) Inventors: **SORSCHER, Eric**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **MANFREDI, Candela**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **JOSHI, Disha**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **RAB, Andras**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **PONNALURI, Sadhana Srinagasai**; c/o Emory University Office of Technol-

ogy Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **HONG, Jeong**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **HIGGIN, Michelle**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US). **DAVIES, Huw M. L.**; c/o Emory University Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (US).

(74) Agent: **MASON, James C.** et al.; Emory University, Office of Technology Transfer, 1599 Clifton Road NE, 4th Floor, Atlanta, Georgia 30322 (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, CV, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IQ, IR, IS, IT, JM, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, MG, MK, MN, MU, 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, CV, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SC, 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,

(54) Title: PHOSPHODIESTERASE 4 INHIBITORS FOR MANAGING RESPIRATORY DISEASES OR CONDITIONS

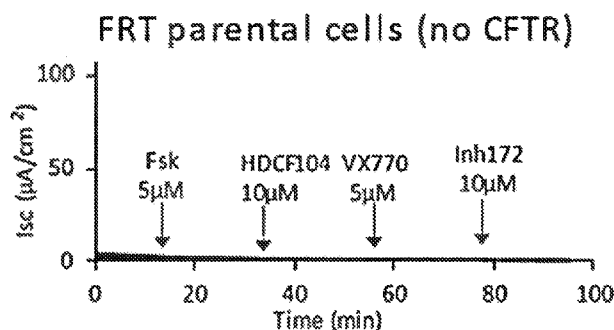


FIG. 1

(57) Abstract: Disclosed herein are methods of treating or preventing respiratory diseases or conditions such as rhinosinusitis, chronic obstructive pulmonary disease, or bronchiectasis in a subject with or without cystic fibrosis (CF). In certain embodiments, this disclosure relates pharmaceutical compositions having compounds disclosed herein and uses thereof. In certain embodiments, the compound is a phosphodiesterase (PDE) 4 inhibitor. In certain embodiments, the compound is 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCE-104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivatives, or salts thereof.



DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT,
LU, LV, MC, ME, 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))*
- *with sequence listing part of description (Rule 5.2(a))*

PHOSPHODIESTERASE 4 INHIBITORS FOR MANAGING RESPIRATORY DISEASES OR CONDITIONS

CROSS-REFERENCE TO RELATED APPLICATIONS

5 This application claims the benefit of U.S. Provisional Application No. 63/452,724 filed March 17, 2023. The entirety of this application is hereby incorporated by reference for all purposes.

INCORPORATION-BY-REFERENCE OF MATERIAL SUBMITTED AS AN XML FILE 10 VIA THE OFFICE ELECTRONIC FILING SYSTEM

 The Sequence Listing associated with this application is provided in XML format and is hereby incorporated by reference into the specification. The name of the XML file containing the Sequence Listing is 23061PCT.xml. The XML file is 4 KB, was created on February 28, 2024 and is being submitted electronically via the USPTO patent electronic filing system.

BACKGROUND

 Rhinosinusitis, also referred to as sinusitis, is a condition where paranasal sinuses become inflamed and often swollen. Viral or bacterial infections typically cause rhinosinusitis; however, rhinosinusitis can also be a result of growths in the sinuses or other conditions (e.g., allergies).
20 Symptoms typically include obstructed nasal passages, pain, tenderness, runny nose, drainage down the back of the throat (post-nasal drip), and/or swelling around eyes, cheeks, nose, forehead. Acute sinusitis is often short-lasting sinusitis; however, chronic sinusitis, also referred to as longer-lasting sinusitis, often results in bacterial infections necessitating antibiotic therapies.

 Cystic fibrosis (CF) is a disease associated with mutations in the cystic fibrosis gene which
25 primarily affects the airways, lungs, gastrointestinal track, and pancreas, but other tissues are also affected. CF patients develop chronic rhinosinusitis (CRS). Cystic fibrosis associated rhinosinusitis is extremely challenging to manage because of the underlying cause is genetic, and the patients are often refractory to standard therapies and surgical treatments. Thus, there is a need to identify improved treatment methods.

30 Noel et al. report PDE5 inhibitors as potential tools in the treatment of cystic fibrosis. *Frontiers in Pharmacology*, 2012, 3:167, 13 pages.

Janosova et al. report phosphodiesterase 4 inhibitors in allergic rhinitis/rhinosinusitis. Frontiers in Pharmacology, 2020, 11:1135, 6 pages.

Turner et al. report the dual phosphodiesterase 3/4 inhibitor RPL554 stimulates rare class III and IV CFTR mutants. Am J Physiol Lung Cell Mol Physiol, 2020, 318: L908–L920.

5 See also US Pat. Pub. No. 2023/0293498.

References cited herein are not an admission of prior art.

SUMMARY

10 In certain embodiments, this disclosure relates to methods of treating or preventing respiratory diseases or conditions such as rhinosinusitis, chronic obstructive pulmonary disease, or bronchiectasis in a subject with or without cystic fibrosis (CF) comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor and/or other compound disclosed herein. In certain embodiments, the phosphodiesterase (PDE) inhibitor is 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof.

15 In certain embodiments, this disclosure relates to methods of treating or preventing Sjogren's anosmia, xerostomia, xerophthalmia, a lacrimal disorder, to reduce salivation, or to reduce tearing in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor and/or other compound disclosed herein. In certain 20 embodiments, the phosphodiesterase (PDE) inhibitor is selected from 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof to a subject in need thereof.

25 In certain embodiments, rhinosinusitis is chronic. In certain embodiments, administration is through the mouth and/or nose by inhalation or a topical application. In certain embodiments, the subject is diagnosed with cystic fibrosis. In certain embodiments, the subject is not diagnosed with cystic fibrosis.

30 In certain embodiments, the subject is diagnosed with pancreatic sufficiency. In certain embodiments, the subject is diagnosed with moderately elevated sweat chloride and the moderately elevated sweat chloride at a concentration between 30 to 60 millimoles per liter.

In certain embodiments, a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and no mutation was identified in the sample, thereby providing a subject diagnosed without a known cystic fibrosis transmembrane conductance regulator mutation.

5 In certain embodiments, a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and a pathogenic mutation was identified on a single allele in the sample, thereby providing a subject diagnosed with a heterozygous (carrier) cystic fibrosis transmembrane conductance regulator mutation.

10 In certain embodiments, the heterozygous cystic fibrosis transmembrane conductance regulator mutation is a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene. In certain embodiments, the heterozygous cystic fibrosis transmembrane conductance regulator mutation is N1303K, W1282X, G551D, and/or G542X.

15 In certain embodiments, a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and a pathogenic mutation was identified on both alleles in the sample, thereby providing a subject diagnosed with homozygous cystic fibrosis transmembrane conductance regulator mutations.

In certain embodiments, the homozygous cystic fibrosis transmembrane conductance regulator mutation is a F508del mutation in both copies of the cystic fibrosis transmembrane conductance regulator (CFTR) gene. In certain embodiments, the cystic fibrosis transmembrane conductance regulator mutation is N1303K, W1282X, G551D, or G542X in one or both copies of the CFTR gene.

20 In certain embodiments, methods further comprising administering to the subject a phosphodiesterase (PDE) inhibitor selected from 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof in combination with a cystic fibrosis drug.

25 In certain embodiments, the cystic fibrosis drug is CFTR activator or modulator. In certain embodiments, the cystic fibrosis drug is elexacaftor, ivacaftor, tezacaftor, or combination thereof. In certain embodiments, the cystic fibrosis drug is a combination of ivacaftor and tezacaftor or is a combination of ivacaftor and lumacaftor. In certain embodiments, the cystic fibrosis drug is a combination of elexacaftor, ivacaftor, and tezacaftor.

In certain embodiments, 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof is administered in combination with another active agent such as a bronchodilator, corticosteroid, antimuscarinic, antibiotic, and/or combinations thereof.

In certain embodiments, this disclosure relates to compounds and pharmaceutical compositions comprising compounds disclosed herein.

BRIEF DESCRIPTION OF THE SEVERAL VIEWS OF THE DRAWINGS

Figures 1A and 1B show data on acute potentiation of wild type CFTR with HDCF104. Using channel analysis in the Fischer rat thyroid (FRT) model shows activation of WT CFTR by HDCF104, a compound from the HDCF series. Acute drug additions included amiloride (epithelial sodium channel blocker), forskolin (cAMP-mediation dependent CFTR activator), VX-770 (CFTR gating potentiator), and inhibitor-172 (CFTR blocker).

Figure 1A shows data using parental FRT cells (no CFTR expression).

Figure 1B shows data from representative tracing demonstrating robust activation of wild type CFTR and strong enhancement of ivacaftor (VX-770) when HDCF104 is applied in combination.

Figure 2A shows data on the G551D rescue by an HDCF analog compound on FRT cell background. Activation of G551D CFTR in the FRT model by a drug in the HDCF series (uHTS_159 (10 μ M)) is shown, including substantial potentiator activity and strong enhancement of Isc (short circuit current) beyond VX-770 (irrespective of the order in which compounds are added).

Figure 2B shows data indicating uHTS159 activates elxacaftor/tezacaftor/ivacaftor (TCT) rescued F508del CFTR in dose-dependent manner - uHTS159 was administered between 0.1 μ M and 100 μ M.

Figure 3 provides evidence that HDCF104 acutely activates F508del CFTR corrected by VX-661 in combination with VX-445. Following pharmaco-correction by VX-661 (5 μ M) and VX-445 (5 μ M), HDCF104 activates CFTR dependent short circuit current in homozygous F508del human bronchial epithelial monolayers. Panels show representative tracings. Acute treatments are amiloride (100 μ M), VX-770 (5 μ M), HDCF104 (20 μ M), and inh-172 (10 μ M).

Figure 4A shows data on the rescue of N1303K CFTR by HDCF104 in primary nasal airway epithelium. Drug additions include amiloride (100 μ M), forskolin (5 μ M), HDCF104 (10 μ M), curcumin (40 μ M) to augment certain CFTR variants, inh-172 (10 μ M), VX-770 (5 μ M). UTP (activates non-CFTR dependent chloride ion transport; 100 μ M).

5 Figure 4B shows data indicating HDCF104 activates short circuit current in N1303K/N1303K homozygous airway epithelial monolayers.

Figures 5A-C show data indicating HDCF104 is blocked by pretreatment with PDE4 inhibitory compounds. Compounds added acutely include amiloride (100 μ M), forskolin (5 μ M), inh-172 (10 μ M), and UTP (100 μ M). To assure saturating drug concentrations, IBMX (broad spectrum PDE inhibitor) was added at 200 μ M, whereas rolipram (PDE4 specific inhibitor) and
10 cilostamide (PDE3 inhibitor) were tested at 10 μ M and 1 μ M, respectively. The studies show particularly robust wild type CFTR activation in non-CF primary nasal cells.

Figure 5A shows data indicating loss of HDCF104 effect on CFTR following pretreatment with rolipram or IBMX. HDCF104 was added at none saturating concentration (10 μ M).

15 Figure 5B shows data indicating pretreatment with HDCF104 attenuated the CFTR activation by IBMX. HDCF104 was added at a saturating concentration (100 μ M).

Figure 5C shows data indicating pretreatment with HDCF104 attenuated CFTR activation by rolipram. HDCF104 was added at a saturating concentration (100 μ M). Cilostamide, a specific PDE3 inhibitor, did not activate CFTR at the tested concentrations and did not alter HDCF104-
20 induced CFTR activation.

Figure 6A illustrates a general synthesis route for the compounds of this disclosure to evaluate the effects of C3 aryl substituents on co-potential of G551D CFTR.

Figure 6B shows data using compounds with C3 aryl substituents tested for co-potential of G551D CFTR. C3-appended dichloro-, dimethoxy-, or trimethoxyarene led to a series of
25 compounds that were efficacious in combination with VX-770.

Figure 6C shows data on C6 alkyl moiety in relation to highly active C3 arene motifs.

Figure 6D shows derivatives with improved solubility (cLogP) and maintained potency together in co-potential of G551D CFTR.

DETAILED DISCUSSION

Before the present disclosure is described in greater detail, it is to be understood that this disclosure is not limited to particular embodiments described, and as such may, of course, vary. "Embodiments" refer to an example, and it is contemplated that the embodiments are not necessarily limited to the example. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to be limiting, since the scope of the present disclosure will be limited only by the appended claims or as amended during prosecution.

Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. Although any methods and materials similar or equivalent to those described herein can also be used in the practice or testing of the present disclosure, the preferred methods and materials are now described.

All publications and patents cited in this specification are herein incorporated by reference as if each individual publication or patent were specifically and individually indicated to be incorporated by reference and are incorporated herein by reference to disclose and describe the methods and/or materials in connection with which the publications are cited.

As will be apparent to those of skill in the art upon reading this disclosure, each of the individual embodiments described and illustrated herein has discrete components and features which may be readily separated from or combined with the features of any of the other several embodiments without departing from the scope or spirit of the present disclosure. Any recited method can be carried out in the order of events recited or in any other order that is logically possible.

Embodiments of the present disclosure will employ, unless otherwise indicated, techniques of medicine, organic chemistry, biochemistry, molecular biology, pharmacology, and the like, which are within the skill of the art. Such techniques are explained fully in the literature.

It must be noted that, as used in the specification and the appended claims, the singular forms "a," "an," and "the" include plural referents unless the context clearly dictates otherwise. In this specification and in the claims that follow, reference will be made to a number of terms that shall be defined to have the following meanings unless a contrary intention is apparent. Also, the term "or" is used in its inclusive sense (and not in its exclusive sense) so that when used, for

example, to connect a list of elements, the term "or" means one, some, or all of the elements in the list.

As used herein, the term "about" is synonymous with the term "approximately." Illustratively, the use of the term "about" indicates that a value includes values slightly outside the cited values. Variation may be due to conditions such as experimental error, manufacturing tolerances, variations in equilibrium conditions, and the like. In some embodiments, the term "about" includes the cited value plus or minus 10%. In all cases, where the term "about" has been used to describe a value, it should be appreciated that this disclosure also supports the exact value.

The terms "comprising," "including," "having," and the like are synonymous and are used inclusively, in an open-ended fashion, and do not exclude additional elements, features, acts, operations, and so forth.

"Consisting essentially of" or "consists of" or the like, when applied to methods and compositions encompassed by the present disclosure refers to compositions and methods like those disclosed herein that exclude certain prior art elements to provide an inventive feature of a claim, but which may contain additional composition components or method steps, etc., that do not materially affect the basic and novel characteristic(s) of the compositions or methods, compared to those of the corresponding compositions or methods disclosed herein.

"Subject" refers to any animal, preferably a human patient, livestock, rodent, monkey, or domestic pet.

"Rhinosinusitis" refers to a condition where the mucus membranes within the sinuses become infected and inflamed.

The terms "pancreatic insufficient" (PI) and "pancreatic sufficient" (PS), and the like, relate to whether a person has enough pancreatic function to grow and maintain health without supplemental pancreatic enzyme therapy (PERT) due to a failure to properly digest food because of a lack of digestive enzymes made by the pancreas.

As used herein, the terms "treat" and "treating" are not limited to the case where the subject (e.g., human patient) is cured and the disease is eradicated. Rather, embodiments of the present disclosure also contemplate treatment that merely reduces symptoms, and/or delays disease progression.

As used herein, the term "in combination with," when referring to two or more compounds, agents, or additional active pharmaceutical ingredients, means the administration of two or more

compounds, agents, or active pharmaceutical ingredients to the patient prior to, concurrent with, or subsequent to each other such that they are contained/circulating in the patient at the same time, e.g., considering half-lives.

A "cystic fibrosis drug" or "modulator" refers to a CFTR activator, corrector, potentiator, stabilizers, amplifiers, read-through agents, or combinations thereof. A "CFTR corrector" is a compound that acts by increasing the delivery to and amount of functional CFTR protein at the cell surface, resulting in enhanced ion transport. Examples include elexacaftor, lumacaftor, tezacaftor. A "CFTR potentiator" is a compound that acts in the presence of endogenous or pharmacological CFTR activators to increase the channel gating activity of cell-surface localized CFTR, resulting in enhanced ion transport. An example is ivacaftor. As used herein, a "CFTR activator" is a compound that acts to stimulate CFTR-mediated ion transport, and includes agents that increase cAMP levels, such as beta-adrenergic agonists, adenylate cyclase activators, and phosphodiesterase inhibitors. Other examples of CFTR activators include lubiprostone, unoprostone, or cobiprostone, forskolin, beta-2-agonists (such as albuterol and/or isoproterenol), genistein, pyrrolo[2,3-b]pyrazines derivatives (such as RP-107), 4-chlorobenzo[f]isoquinoline (CBIQ), 2-thioxo-4-amino-thiazoles (such as A01 and A02), 5-((Z)-2-(2-(allyloxy)phenyl)-1-cyanovinyl)-3-amino-1H-pyrazole-4-carbonitrile (Cact-A1).

CFTR function can be characterized at the cellular (in vitro) level using cell-based assays, such as an FRT-based assay (Van Goor, et al., (2009) PNAS 106(44): 18825-30; and Van Goor, et al. (2011) PNAS 108(46):18843-46) to measure the amount of chloride transport through the mutated or wild type CFTR channels. Residual function mutations result in a reduction of CFTR-dependent ion transport. For example, residual function mutations may result in more or less than about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80% or about 90% reduction of CFTR activity in the FRT-based assay.

Any compound disclosed herein may be in a pharmaceutically acceptable salt thereof. As used herein, a "pharmaceutically acceptable salt" refers to any salt or salt of an ester of a compound of this disclosure that, upon administration to a recipient, is capable of providing, either directly or indirectly, a compound of this disclosure. Pharmaceutically acceptable salts are well known in the art. For example, pharmaceutically acceptable salts may include the clinically approved salts: acetate, aluminum, benzenesulfonate, benzathine, benzoate, bicarbonate, bitartrate, bromide, calcium, calcium edetate, camsylate, carbonate, chloride, choline, citrate, diethanolamine,

dihydrochloride, edetate, edisylate, esylate, ethylenediamine, fumarate, gluceptate, gluconate, glutamate, hydrobromide, hydrochloride, hydroxynaphthoate, iodide, isethionate, lactate, lactobionate, lithium, magnesium, malate, maleate, mandelate, meglumine, mesylate, nitrate, pamoate, pantothenate, phosphate/diphosphate, polygalacturonate, potassium, salicylate, sodium, stearate, subacetate, succinate, sulfate, tannate, tartrate, and zinc.

As used herein, the term "derivative" refers to a structurally similar compound that retains sufficient functional attributes of the identified analogue. The derivative may be structurally similar because it is lacking one or more atoms, substituted, a salt, a prodrug, in different hydration/oxidation states, or because one or more atoms within the molecule are switched, such as, but not limited to, adding a hydroxyl group, replacing an oxygen atom with a sulfur atom, or replacing an amino group with a hydroxyl group, oxidizing a hydroxyl group to a carbonyl group, reducing a carbonyl group to a hydroxyl group, and reducing a carbon-to-carbon double bond to an alkyl group or oxidizing a carbon-to-carbon single bond to a double bond. A derivative includes a compound wherein one or more hydrogens are deuterated. A derivative optionally has one or more, the same or different, substitutions. Derivatives may be prepared by any variety of synthetic methods or appropriate adaptations presented in synthetic or organic chemistry textbooks, such as those provide in March's Advanced Organic Chemistry: Reactions, Mechanisms, and Structure, Wiley, 6th Edition (2007) Michael B. Smith or Domino Reactions in Organic Synthesis, Wiley (2006) Lutz F. Tietze hereby incorporated by reference.

The term "prodrug" refers to an agent that is converted into a biologically active form in vivo. Prodrugs are often useful because, in some situations, they may be easier to administer than the parent compound. They may, for instance, be bioavailable by oral administration whereas the parent compound is not. The prodrug may also have improved solubility in pharmaceutical compositions over the parent drug. A prodrug may be converted into the parent drug by various mechanisms, including enzymatic processes and metabolic hydrolysis. Typical prodrugs are pharmaceutically acceptable esters. Prodrugs include compounds wherein a hydroxy, amino or mercapto group is bonded to any group that, when the prodrug of the active compound is administered to a subject, cleaves to form a free hydroxy, free amino or free mercapto group, respectively. Examples of prodrugs include, but are not limited to, acetate, formate, and benzoate derivatives of an alcohol or acetamide, formamide and benzamide derivatives of an amine functional group in the active compound and the like.

As used herein, "alkyl" means a noncyclic, straight chain or branched chain, unsaturated or saturated hydrocarbon such as those containing from 1 to 25 carbon atoms. For example, a "C₈-C₁₈" refers to an alkyl containing 8 to 18 carbon atoms. Likewise, a "C₆-C₂₂" refers to an alkyl containing 6 to 22 carbon atoms. Representative saturated straight chain alkyls include methyl, ethyl, n-propyl, n-butyl, n-pentyl, n-hexyl, n-septyl, n-octyl, n-nonyl, and the like; while saturated branched alkyls include isopropyl, sec-butyl, isobutyl, tert-butyl, isopentyl, and the like. Unsaturated alkyls contain at least one double or triple bond between adjacent carbon atoms (referred to as an "alkenyl" or "alkynyl", respectively). Representative straight chain and branched alkenyls include ethylenyl, propylenyl, 1-butenyl, 2-butenyl, isobutylenyl, 1-pentenyl, 2-pentenyl, 3-methyl-1-butenyl, 2-methyl-2-butenyl, 2,3-dimethyl-2-butenyl, and the like; while representative straight chain and branched alkynyls include acetylenyl, propynyl, 1-butyne, 2-butyne, 1-pentyne, 2-pentyne, 3-methyl-1-butyne, and the like.

As used herein, "heteroalkyl" refers to alkyl groups where one or more carbon atoms are replaced with a heteroatom, such as, O, N, or S. Similar to alkyl groups, heteroalkyl groups can be straight or branched, saturated or unsaturated. Optionally, the nitrogen and/or sulfur heteroatom(s) can be oxidized, and the nitrogen heteroatom(s) can be quaternized. Suitable heteroalkyl groups may contain 1 to 25 carbon atoms and 1 to 4 heteroatoms.

Non-aromatic, mono or polycyclic alkyls are referred to herein as "carbocycles" or "carbocyclyl" groups. Representative saturated carbocycles include cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, and the like; while representative unsaturated carbocycles include cyclopentenyl, cyclohexenyl, and the like. In polycyclic carbocyclyl groups, the rings can be attached together in a pendant manner (*i.e.*, two rings are connected by a single bond), in a spiro manner (*i.e.*, two rings are connected through a defining single common atom), in a fused manner (*i.e.*, two rings share two adjacent atoms; in other words, two rings share one covalent bond), in a bridged manner (*i.e.*, two rings share three or more atoms, separating the two bridgehead atoms by a bridge containing at least one atom), or a combination thereof. The number of "members" of a carbocyclyl group refers to the total number of carbon atoms in the ring(s) of the carbocyclyl group.

"Heterocarbocycles" or "heterocarbocyclyl" groups are carbocycles which contain from 1 to 4 heteroatoms independently selected from nitrogen, oxygen, and sulphur, wherein the nitrogen and/or sulphur heteroatom(s) may be optionally oxidized, and the nitrogen heteroatom(s) may be

optionally quaternized. Heterocarbocycles may be saturated or unsaturated (but not aromatic), monocyclic or polycyclic. Exemplary heterocarbocycles include morpholinyl, pyrrolidinonyl, pyrrolidinyl, piperidinyl, hydantoinyl, valerolactamyl, oxiranyl, oxetanyl, tetrahydrofuranyl, tetrahydropyranyl, tetrahydropyridinyl, tetrahydroprimidinyl, tetrahydrothiophenyl, tetrahydrothiopyranyl, and the like. The number of "members" of a heterocarbocyclyl group refers to the total number of carbon atoms and heteroatoms in the ring(s) of the heterocarbocyclyl group.

The term "aryl" refers to aromatic homocyclic (*i.e.*, hydrocarbon) mono-, bi- or tricyclic ring-containing groups, preferably having 6 to 12 members, such as phenyl, naphthyl and biphenyl. Optionally, the aryl group is phenyl. In polycyclic aryl groups, the rings can be attached together in a pendant manner or can be fused. The number of "members" of an alkyl group refers to the total number of carbon atoms in the ring(s) of the alkyl group.

As used herein, "heteroaryl" or "heteroaromatic" refers an aromatic heterocarbocycle having 1 to 4 heteroatoms selected from nitrogen, oxygen and sulfur, and containing at least 1 carbon atom, including both mono- and polycyclic ring systems. Polycyclic ring systems may, but are not required to, contain one or more non-aromatic rings, as long as one of the rings is aromatic. Representative heteroaryls are furyl, benzofuranyl, thiophenyl, benzothiophenyl, pyrrolyl, indolyl, isoindolyl, azaindolyl, pyridyl, quinoliny, isoquinoliny, oxazolyl, isooxazolyl, benzoxazolyl, pyrazolyl, imidazolyl, benzimidazolyl, thiazolyl, benzothiazolyl, isothiazolyl, pyridazinyl, pyrimidinyl, pyrazinyl, triazinyl, cinnoliny, phthalazinyl, and quinazolinyl. It is contemplated that the use of the term "heteroaryl" includes *N*-alkylated derivatives such as a 1-methylimidazol-5-yl substituent. The number of "members" of a heteroaryl group refers to the total number of carbon atoms and heteroatoms in the ring(s) of the heteroaryl group.

As used herein, "heterocycle" or "heterocyclyl" refers to mono- and polycyclic ring systems having 1 to 4 heteroatoms selected from nitrogen, oxygen and sulfur, and containing at least 1 carbon atom. The mono- and polycyclic ring systems may be aromatic, non-aromatic or mixtures of aromatic and non-aromatic rings. Heterocycle includes heterocarbocycles, heteroaryls, and the like. In polycyclic heterocyclyl groups, the rings can be attached together in a pendant manner (*i.e.*, two rings are connected by a single bond), in a spiro manner (*i.e.*, two rings are connected through a defining single common atom), in a fused manner (*i.e.*, two rings share two adjacent atoms; in other words, two rings share one covalent bond), in a bridged manner (*i.e.*, two rings share three or more atoms, separating the two bridgehead atoms by a bridge containing at

least one atom), or a combination thereof. The number of “members” of a heterocyclyl group refers to the total number of carbon atoms and heteroatoms in the ring(s) of the heterocyclyl group.

“Alkoxy” or “alkyloxy” refers to an alkyl group as defined above with the indicated number of carbon atoms attached through an oxygen bridge. Examples of alkoxy include, but are not limited to, methoxy, ethoxy, n-propoxy, i-propoxy, n-butoxy, s-butoxy, t-butoxy, n-pentoxo, and s-pentoxo. Preferred alkoxy groups are methoxy, ethoxy, n-propoxy, i-propoxy, n-butoxy, s-butoxy, t-butoxy.

“Alkoxyalkyl” refers an alkoxy group as defined above with the indicated number of carbon atoms attached through an alkyl bridge (*i.e.*, $-\text{CH}_2-\text{O}-\text{CH}_2\text{CH}_3$).

“Alkylamino” refers an alkyl group as defined above with the indicated number of carbon atoms attached through an amino bridge. An example of an alkylamino is methylamino (*i.e.*, $-\text{NH}-\text{CH}_3$).

“Alkylthio” refers to an alkyl group as defined above with the indicated number of carbon atoms attached through a sulfur bridge. An example of an alkylthio is methylthio (*i.e.*, $-\text{S}-\text{CH}_3$).

“Alkanoyl” refers to an alkyl as defined above with the indicated number of carbon atoms attached through a carbonyl bridge (*i.e.*, $-(\text{C}=\text{O})\text{alkyl}$).

The terms “cycloalkyl” and “cycloalkenyl” refer to mono-, bi-, or tri homocyclic ring groups of 3 to 15 carbon atoms which are, respectively, fully saturated and partially unsaturated (but not aromatic).

The terms “halogen” or “Hal” refer to fluorine, chlorine, bromine, and iodine.

The term “substituted” refers to a molecule wherein at least one hydrogen atom is replaced with a substituent. The molecule may be multiply substituted. In the case of an oxo substituent (“=O”), two hydrogen atoms are replaced. Example substituents within this context may include halogen, hydroxy, alkyl, alkoxy, nitro, cyano, oxo, carbocyclyl, carbocycloalkyl, heterocarbocyclyl, heterocarbocycloalkyl, aryl, arylalkyl, heteroaryl, heteroarylalkyl, $-\text{NR}^a\text{R}^b$, $-\text{NR}^a\text{C}(=\text{O})\text{R}^b$, $-\text{NR}^a\text{C}(=\text{O})\text{NR}^a\text{R}^b$, $-\text{NR}^a\text{C}(=\text{O})\text{OR}^b$, $-\text{NR}^a\text{SO}_2\text{R}^b$, $-\text{C}(=\text{O})\text{R}^a$, $-\text{C}(=\text{O})\text{OR}^a$, $-\text{C}(=\text{O})\text{NR}^a\text{R}^b$, $-\text{OC}(=\text{O})\text{NR}^a\text{R}^b$, $-\text{OR}^a$, $-\text{SR}^a$, $-\text{SOR}^a$, $-\text{S}(=\text{O})_2\text{R}^a$, $-\text{OS}(=\text{O})_2\text{R}^a$, and $-\text{S}(=\text{O})_2\text{OR}^a$. R_a and R_b in this context may be the same or different, and independently hydrogen, halogen hydroxyl, alkyl, alkoxy, alkyl, amino, alkylamino, dialkylamino, carbocyclyl, carbocycloalkyl, heterocarbocyclyl, heterocarbocycloalkyl, aryl, arylalkyl, heteroaryl, or heteroarylalkyl.

Treatment or prevention diseases or conditions using phosphodiesterase (PDE) inhibitors

Rhinosinusitis is commonly found in individuals with cystic fibrosis (CF). However, non-cystic fibrosis rhinosinusitis treatments are contemplated. These individuals are not considered to have a diagnosis of cystic fibrosis (CF) because they do not meet diagnostic criteria for CF. Some patients with rhinosinusitis may have slightly elevated sweat chloride – but do not carry a diagnosis of CF or have known, disease-causing mutations in CFTR. They are generally believed to have a cause of rhinosinusitis due to factors unrelated to mutations in CFTR.

In certain embodiments, this disclosure relates to methods of treating airway diseases such as rhinosinusitis, chronic obstructive pulmonary disease, and bronchiectasis in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor.

In certain embodiments, this disclosure relates to methods of treating or preventing Sjogren's anosmia, xerostomia, xerophthalmia, a lacrimal disorder, to reduce salivation, or to reduce tearing in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor.

In certain embodiments, this disclosure relates to methods of treating or preventing Sjogren's anosmia, xerostomia, xerophthalmia, or lacrimal disorder in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor.

In certain embodiments, the phosphodiesterase (PDE) inhibitor is a PDE4 inhibitor.

In certain embodiments, the phosphodiesterase (PDE) inhibitor is administered in combination with a CFTR activator or modulator.

In certain embodiments, the phosphodiesterase (PDE) inhibitor is administered in a topical or aerosolized form in the nasal passages.

In certain embodiments, the phosphodiesterase (PDE) inhibitor is administered by pulmonary administration.

In certain embodiments, administration is by inhalation of an aerosol of a phosphodiesterase (PDE) inhibitor and a CFTR activator or modulator in the pulmonary airway. In certain embodiments, administration is by inhalation of a phosphodiesterase (PDE) inhibitor and a CFTR activator or modulator through the mouth and/or nose.

In certain embodiments, administration is by inhalation of an aerosol of a phosphodiesterase (PDE) inhibitor and an oral CFTR activator or modulator to provide mucus clearance in the pulmonary airway. In certain embodiments, administration is by inhalation of a

phosphodiesterase (PDE) inhibitor and a CFTR activator or modulator through the mouth and/or nose while a CFTR activator or modulator is given orally.

In certain embodiments, administration is by a metered-dose inhaler. In certain embodiments, administration is by a single or multiple dose dry powder inhaler. In certain
5 embodiments, administration is by a nebulizer or exhaled delivery system or bidirectional dosing mechanism.

In certain embodiments, administration is by instillation or administration through an implanted controlled release device.

In certain embodiments, this disclosure relates to pharmaceutical compositions, containers,
10 and kits comprising a phosphodiesterase (PDE) inhibitor and optionally an oral CFTR activator or modulator for use in pulmonary administration and optionally comprising another active agent. In certain embodiments, the container is a pressurized or unpressurized container. In certain embodiments, the container is a manual pump spray, inhaler, meter-dosed inhaler, dry powder inhaler, nebulizer, vibrating mesh nebulizer, jet nebulizer, or ultrasonic wave nebulizer.

15 In certain embodiments, rhinosinusitis is chronic.

In certain embodiments, administration is through the mouth and/or nose by inhalation to provide a topical application in the lungs.

In certain embodiments, the subject is diagnosed with cystic fibrosis.

In certain embodiments, the subject is not diagnosed with cystic fibrosis.

20 In certain embodiments, the subject is diagnosed with pancreatic sufficiency.

In certain embodiments, the subject is diagnosed with moderately elevated sweat chloride and the moderately elevated sweat chloride at a concentration between 30 to 60 millimoles per liter.

In certain embodiments, a sample of the subject was tested for presence of a known cystic
25 fibrosis transmembrane conductance regulator mutation and no mutation was identified in the sample, thereby providing a subject diagnosed without a known cystic fibrosis transmembrane conductance regulator mutation.

In certain embodiments, a sample of the subject was tested for presence of a known cystic
30 fibrosis transmembrane conductance regulator mutation and a mutation was identified on a single allele in the sample, thereby providing a subject diagnosed with a carrier status.

In certain embodiments, a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and different pathologic mutations were identified on the two alleles in the sample, thereby providing a subject diagnosed with heterozygous cystic fibrosis transmembrane conductance regulator mutation.

5 In certain embodiments, the heterozygous cystic fibrosis transmembrane conductance regulator mutation is a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

In certain embodiments, the heterozygous cystic fibrosis transmembrane conductance regulator mutation is N1303K, W1282X, G551D, or G542X.

10 In certain embodiments, a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and the same mutation was identified on a both alleles in the sample, thereby providing a subject diagnosed with homozygous cystic fibrosis transmembrane conductance regulator mutations.

In certain embodiments, the homozygous cystic fibrosis transmembrane conductance regulator mutation is a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

15 In certain embodiments, the homozygous cystic fibrosis transmembrane conductance regulator mutation is N1303K, W1282X, G551D, and/or G542X.

In certain embodiments, methods further comprising administering the phosphodiesterase (PDE) inhibitor in combination with a cystic fibrosis drug. In certain embodiments, the cystic fibrosis drug is an CFTR activator or modulator.

In certain embodiments, the cystic fibrosis drug is elexacaftor, ivacaftor, tezacaftor, or a combination thereof.

25 In certain embodiments, the cystic fibrosis drug is a combination of ivacaftor and lumacaftor.

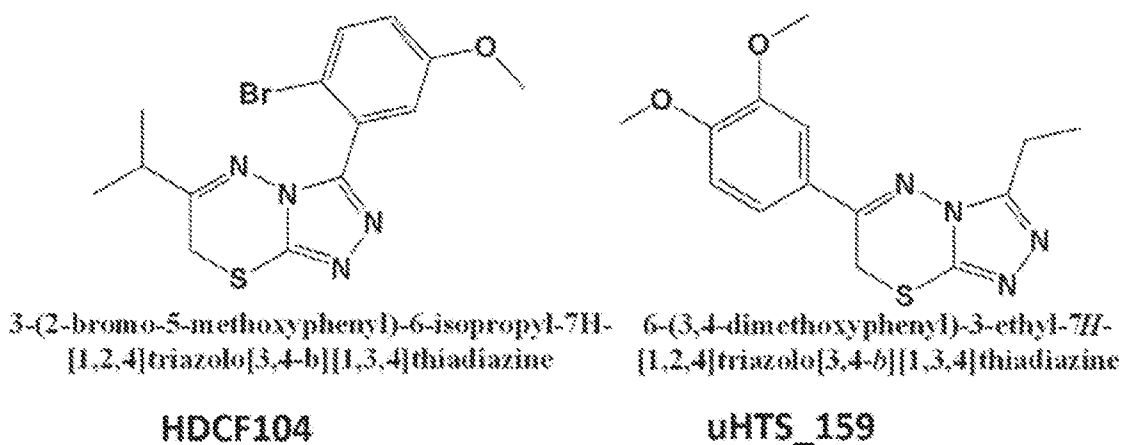
In certain embodiments, the cystic fibrosis drug is a combination of ivacaftor and tezacaftor.

In certain embodiments, the cystic fibrosis drug is a combination of elexacaftor, ivacaftor, and tezacaftor.

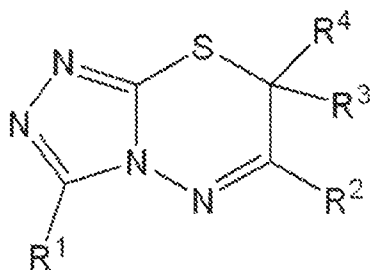
30 In certain embodiments, 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104) and/or 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-

[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159) or salt thereof is administered in combination with another active agent such as a bronchodilator, corticosteroid, antimuscarinic, antibiotic, or combinations thereof.

In certain embodiments, the phosphodiesterase (PDE) inhibitor is 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159) or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104) or salt thereof to a subject diagnosed with rhinosinusitis, chronic obstructive pulmonary disease, bronchiectasis, and/or CF.



In certain embodiments, the phosphodiesterase (PDE) inhibitor is a compound of formula I,



Formula I,

derivative or salt thereof, wherein

R^1 is alkyl, aryl or heterocyclyl, wherein R^1 is optionally substituted with one or more, the same or different, R^{10} ;

R² is hydrogen, alkyl, halogenated alkyl, halogen, hydroxy, alkoxy, amino, alkylamino, (alkyl)₂amino, cyano, formyl, alkanoyl, benzoyl, carboxy, carbamoyl, carbocyclyl, aryl, or heterocyclyl, wherein R¹ is optionally substituted with one or more, the same or different, R¹⁰;

R³ and R⁴ are each, individually and independently, hydrogen, alkyl, halogen, hydroxy, alkoxy, amino, alkylamino, (alkyl)₂amino, cyano, formyl, alkanoyl, benzoyl, carboxy, carbamoyl, carbocyclyl, aryl, or heterocyclyl, wherein R³ and R⁴ are optionally substituted with one or more, the same or different, R¹⁰;

R¹⁰ is alkyl, halogen, nitro, cyano, hydroxy, amino, mercapto, formyl, carboxy, carbamoyl, alkoxy, alkylthio, alkylamino, (alkyl)₂amino, alkylsulfinyl, alkylsulfonyl, arylsulfonyl, alkanoyl, benzoyl, carbocyclyl, aryl, or heterocyclyl, wherein R¹⁰ is optionally substituted with one or more, the same or different, R¹¹; and

R¹¹ is halogen, nitro, cyano, hydroxy, trifluoromethoxy, trifluoromethyl, amino, formyl, carboxy, carbamoyl, mercapto, sulfamoyl, methyl, ethyl, propyl, tert-butyl, methoxy, ethoxy, acetyl, acetoxyl, methylamino, ethylamino, dimethylamino, diethylamino, N-methyl-N-ethylamino, acetylamino, N-methylcarbamoyl, N-ethylcarbamoyl, N,N-dimethylcarbamoyl, N,N-diethylcarbamoyl, N-methyl-N-ethylcarbamoyl, methylthio, ethylthio, methylsulfinyl, ethylsulfinyl, mesyl, ethylsulfonyl, methoxycarbonyl, ethoxycarbonyl, N-methylsulfamoyl, N-ethylsulfamoyl, N,N-dimethylsulfamoyl, N,N-diethylsulfamoyl, N-methyl-N-ethylsulfamoyl, carbocyclyl, aryl, and heterocyclyl.

In certain embodiments, R¹ is phenyl optionally substituted with one or more, the same or different, R¹⁰.

In certain embodiments, R² is alkyl optionally substituted with one or more, the same or different, R¹⁰.

In certain embodiments, R² is a branched alkyl of six or less carbons.

In certain embodiments, R² is carbocyclyl optionally substituted with one or more, the same or different, R¹⁰.

In certain embodiments, R² is a cyclopropyl or methylcyclopropyl.

In certain embodiments, R³ and R⁴ are hydrogen.

In certain embodiments, R¹ is phenyl optionally substituted with one or more, the same or different, R¹⁰, and R³ and R⁴ are hydrogen.

In certain embodiments, R² is alkyl optionally substituted with one or more, the same or different, R¹⁰, and R³ and R⁴ are hydrogen.

In certain embodiments, R² is a branched alkyl of six or less carbons, and R³ and R⁴ are hydrogen.

5 In certain embodiments, R² is carbocyclyl optionally substituted with one or more, the same or different, R¹⁰, and R³ and R⁴ are hydrogen.

In certain embodiments, R² is a cyclopropyl or methylcyclopropyl, and R³ and R⁴ are hydrogen.

10 In certain embodiments, R¹ is phenyl optionally substituted with one or more, the same or different, R¹⁰, R² is alkyl optionally substituted with one or more, the same or different, R¹⁰, and R³ and R⁴ are hydrogen.

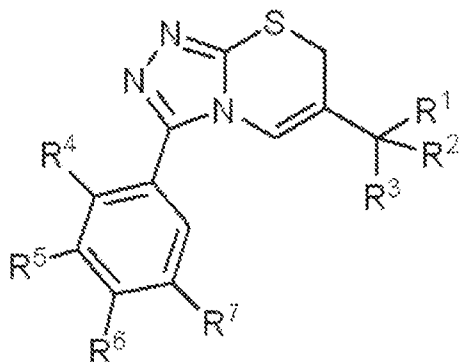
In certain embodiments, R¹ is phenyl optionally substituted with one or more, the same or different, R¹⁰, R² is a branched alkyl of six or less carbons, and R³ and R⁴ are hydrogen.

15 In certain embodiments, R¹ is phenyl optionally substituted with one or more, the same or different, R¹⁰, R² is carbocyclyl optionally substituted with one or more, the same or different, R¹⁰, and R³ and R⁴ are hydrogen.

In certain embodiments, R¹ is phenyl optionally substituted with one or more, the same or different, R¹⁰, R² is a cyclopropyl or methylcyclopropyl, and R³ and R⁴ are hydrogen.

20 In certain embodiments, the compound of formula I is 3-(2,3-dichlorophenyl)-6-(tert-pentyl)-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF 83), 6-(tert-butyl)-3-(2,4-dichlorophenyl)-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF 89), 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104) or 3-(2-bromo-5-chlorophenyl)-6-(tert-butyl)-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF 95).

25 In certain embodiments, the phosphodiesterase (PDE) inhibitor is a compound of Formula IA,



Formula IA

derivative or salt thereof, wherein

R^1 is hydrogen, deuterium, halogen, or alkyl;

5 R^2 is hydrogen, deuterium, halogen, or alkyl;

or R^1 and R^2 and the attached atom form a carbocyclic ring;

or R^1 and R^2 and R^3 and the attached atom form adamantanyl;

R^3 is hydrogen, deuterium, halogen, or alkyl;

R^4 is hydrogen, deuterium, halogen or alkoxy;

10 R^5 is hydrogen, deuterium, halogen or alkoxy;

R^6 is hydrogen, deuterium, halogen or alkoxy; and

R^7 is hydrogen, deuterium, halogen or alkoxy.

In certain embodiments, the compound is:

3-(2-bromo-5-chlorophenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine

15 (HDCF94/9c),

3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine

(HDCF104/10a),

3-(2-fluoro-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine

(10b),

20 3-(2,5-dichlorophenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (10e) or salts thereof.

In certain embodiments, the subject is diagnosed with moderately elevated sweat chloride and pancreatic sufficiency. In certain embodiments, a sample of the subject is tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation no mutation is identified in the sample. In certain embodiments, a sample of the subject is tested for presence of a known

25

cystic fibrosis transmembrane conductance regulator mutation and a mutation is identified in the sample. In certain embodiments, moderately elevated sweat chloride at a concentration between about 30 to 60 millimoles per liter (mEq/L). In certain embodiments, the subject has a normal sweat chloride at a concentration of less than or about 30 mEq/L.

5 In certain embodiments, the subject is diagnosed to have or not have a W1282X and/or N1303K mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

In certain embodiments, the subject is diagnosed to have or not have a N1303K and/ or G542X mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

10 In certain embodiments, the subject is diagnosed to have or not have a G551D mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

In certain embodiments, the subject is diagnosed to have or not have a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene and/or having or not having any of the following mutations 3141del9, E822K, G1069R, L967S, R117L, S912L, 546insCTA, F191V, G1244E, L997F, R117P, S945L, A46D, F311del, G1249R, L1077P, R170H, 15 S977F, A120T, F311L, G1349D, L1324P, R258G, S1159F, A234D, F508C, H139R, L1335P, R334L, S1159P, A349V, F508C, H199Y, L1480P, R334Q, S1251N, A455E, H939R, M152V, R347H, S1255P, A554E, F575Y, H1054D, M265R, R347L, T338I, A1006E, F1016S, H1085P, M952I, R347P, T1036N, A1067T, F1052V, H1085R, M952T, R352Q, T1053I, D110E, F1074L, H1375P, M1101K, R352W, V201M, D110H, F1099L, I148T, P5L, R553Q, V232D, D192G, 20 G27R, I175V, P67L, R668C, V456A, D443Y, G85E, I336K, P205S, R751L, V456F, D443Y, R668C, G126D, I502T, P574H, R792G, V562I, D579G, G178E, I601F, Q98R, R933G, V754M, D614G, G178R, I618T, Q237E, R1066H, V1153E, D836Y, G194R, I807M, Q237H, R1070Q, V1240G, D924N, G194V, I980K, Q359R, R1070W, V1293G, D979V, G314E, I1027T, Q1291R, R1162L, W361R, D1152H, G463V, I1139V, R31L, R1283M, W1098C, D1270N, G480C, I1269N, 25 R74Q, R1283S, W1282R, E56K, G551D, I1366N, R74W, S13F, Y109N, E60K, G551S, K1060T, D1270N, S341P, Y161D, E92K, G576A, L15P, S364P, Y161S, E116K, R668C, L165S, D1270N, S492F, Y563N, E193K, G622D, L206W, R75Q, S549N, Y1014C, E403D, G628R, L320V, R117C, S549R, Y1032C, E474K, G970D, L346P, R117G, S589N, E588V, G1061R, L453S, R117H, and S737F or other CFTR mutations known to cause clinical CF.

30 In certain embodiments, a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and a mutation or no mutation or only one

mutation was identified in the sample, thereby providing a subject diagnosed without two known cystic fibrosis transmembrane conductance regulator mutations and carrying a diagnosis of airway disease or condition such as rhinosinusitis associated with cystic fibrosis or diagnosed with an airway disease or condition such as rhinosinusitis not associated with cystic fibrosis, i.e., non-cystic fibrosis rhinosinusitis, chronic obstructive pulmonary disease, or bronchiectasis.

In certain embodiments, the subject for treatment is diagnosed with one copy of a CFTR mutation (i.e. heterozygote) optionally having moderately elevated sweat chloride at a concentration between about 30 to 60 millimoles per liter (mEq/L).

In certain embodiments, the phosphodiesterase (PDE) inhibitor is 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), or 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof and is administered as a pharmaceutical composition comprising the drug(s), or a pharmaceutically acceptable salt thereof optionally in combination with a cystic fibrosis drug a CFTR corrector, a CFTR potentiator, a CFTR activator or modulator, or combinations thereof.

In certain embodiments, the cystic fibrosis drug is lumacaftor, elexacaftor, ivacaftor, tezacaftor, or other CFTR activator or modulator or combinations, derivative, or salt thereof. In certain embodiments, the cystic fibrosis drug is a combination of elexacaftor, ivacaftor, and tezacaftor. In certain embodiments, the cystic fibrosis drug is a combination of ivacaftor and lumacaftor. In certain embodiments, the cystic fibrosis drug is a combination of ivacaftor and tezacaftor.

In certain embodiments, the cystic fibrosis drug is lumacaftor, elexacaftor, ivacaftor, tezacaftor, cavosonstat, olacaftor, posenacaftor, galicaftor, navocaftor, deutivacaftor, nesolicaftor, bamocaftor, ataluren, icenticaftor (QBW-251), N-(3-carbamoyl-5,5,7,7-tetramethyl-4,7-dihydro-5H-thieno[2,3-c]pyran-2-yl)-1H-pyrazole-5-carboxamide (GLPG-1837), (S)-3-amino-N-(2-hydroxypropyl)-5-((4-(trifluoromethoxy)phenyl)sulfonyl)picolinamide (GLPG-2451), N-(3-carbamoyl-5,5,7,7-tetramethyl-4,7-dihydro-5H-thieno[2,3-c]pyran-2-yl)-1H-pyrazole-3-carboxamide (ABBV 974), or combinations, or salts thereof.

Bronchiectasis is a complex, chronic respiratory condition, characterized by frequent cough and shortness of breath due to a range of conditions that include inherited mucociliary defects, inhalational airway injury, immunodeficiency states and prior respiratory infections. Bronchiectasis is characterized as a thickening and dilation of the walls of the bronchi from

inflammation, infection, or other etiologies which result in the inability to clear mucus from the airway. Affected individuals are then more susceptible to repeated lung infections.

Bronchiectasis is commonly found in individuals with cystic fibrosis. However, non-cystic fibrosis bronchiectasis (NCFBE) is prevalent worldwide. These individuals are not considered to have a diagnosis of cystic fibrosis (CF) because they do not meet diagnostic criteria for CF. Some patients with NCFBE may have slightly elevated sweat chloride – but do not carry a diagnosis of CF or have known, disease-causing mutations in CFTR. They are generally believed to have a cause of bronchiectasis due to factors unrelated to mutations in CFTR.

In certain embodiments, this disclosure relates to the use of a pharmaceutical composition comprising 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), or 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159) and/or derivatives, or combinations or salts as reported herein, in the preparation of a medicament for the treatment of rhinosinusitis, chronic obstructive pulmonary disease, bronchiectasis, CF and/or other condition reported herein.

In certain embodiments, this disclosure relates to the use of a pharmaceutical composition in the treatment of rhinosinusitis, chronic obstructive pulmonary disease, bronchiectasis, CF, or other condition reported herein comprising a drug or combination of drugs reported herein. In certain embodiments, this disclosure relates to the manufacture of a medicament for use in the treatment of rhinosinusitis, chronic obstructive pulmonary disease, bronchiectasis, CF and/or other condition reported herein comprising a drug or combination of drugs reported herein.

In certain embodiments, this disclosure contemplates a non-personalized use of drugs disclosed herein among patients with CF rhinosinusitis, CF chronic obstructive pulmonary disease, and/or CF bronchiectasis in particular those with sweat chloride elevation about 30-60 mEq/L, or above, and without a clinically approved genotype.

In certain embodiments, this disclosure contemplates uses in treating patients with non-CF rhinosinusitis, non-CF chronic obstructive pulmonary disease, non-CF bronchiectasis, and/or other non-CF condition and a mildly elevated sweat chloride (i.e., suggesting diminished CFTR activity) not sufficient to establish a diagnosis of CF (but instead leading to a diagnosis of non-CF rhinosinusitis, non-CF chronic obstructive pulmonary disease, non-CF bronchiectasis, and/or other non-CF condition) by administering 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-

b][1,3,4]thiadiazine (uHTS-159) or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), derivative, or salt thereof.

In certain embodiments, this disclosure contemplates uses in treating patients with non-CF rhinosinusitis, non-CF chronic obstructive pulmonary disease, non-CF bronchiectasis, and/or other non-CF condition, and normal sweat chloride (less than 30 mEq/L, normal CFTR function in the sweat gland) by administering 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), and/or 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative or salt thereof provided activating the endogenous (mutant or non-mutant) CFTR can enhance mucus clearance and/or minimize airway remodeling.

In certain embodiments, this disclosure contemplates uses in treating rhinosinusitis, chronic obstructive pulmonary disease, and/or bronchiectasis in patients without disease-causing CFTR mutations which can nonetheless benefit from a therapy of 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159) derivative or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), derivative, or salt thereof.

In certain embodiments, a sample of the subject is tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and a mutation is found which is not identified as resulting in a defective cystic fibrosis transmembrane conductance regulator or a cause of CF, thereby contributing to a diagnosis of non-cystic fibrosis rhinosinusitis or bronchiectasis. In certain embodiments, the mutation is a conservative substitution or non-conservative substitution inside or outside of a functional domain. In certain embodiments, the mutation is a conservative substitution or non-conservative substitution outside a functional domain or a conservative substitution inside a functional domain. One type of conservative amino acid substitutions refers to the interchangeability of residues having similar side chains. For example, a group of amino acids having aliphatic side chains is glycine, alanine, valine, leucine, and isoleucine; a group of amino acids having aliphatic-hydroxyl side chains is serine and threonine; a group of amino acids having amide-containing side chains is asparagine and glutamine; a group of amino acids having aromatic side chains is phenylalanine, tyrosine, and tryptophan; a group of amino acids having basic side chains is lysine, arginine, and histidine; and a group of amino acids having sulfur-containing side chains is cysteine and methionine. Preferred

conservative amino acids substitution groups are valine-leucine-isoleucine, phenylalanine-tyrosine, lysine-arginine, alanine-valine, and asparagine-glutamine.

The human cystic fibrosis transmembrane conductance regulator has NCBI Reference Sequence number NP_000483.3 starting a position 1: MQRSPLEKAS VVSKLFFSWT

5 RPILRKGYRQ RLELSDIYQI PSVDSADNLS EKLEREWDRE 61 LASKKNPKLI
NALRRCCFFWR FMFYGIFLYL GEVTKAVQPL LLGRIIASYD PDNKEERSIA 121
IYLGIGLCLL FIVRTLHHP AIFGLHHIGM QMRIAMFSLI YKKTLLKSSR VLDKISIGQL
181 VSLLSNNLNK FDEGLALAHF VWIAPLQVAL LMGLIWELLQ ASAFCLGLFL
IVLALFQAGL 241 GRMMMKEYRDQ RAGKISERLV ITSEMIENIQ SVKAYCWEEA

10 MEKMIENLRQ TELKLTRKAA 301 YVRYFNSSAF FFSGFFVVFL SVLPYALIKG
IILRKIFTTI SFCIVLRMAV TRQFPWAVQT 361 WYDSLGAINK IQDFLQKQEY
KTLEYNLTTT EVVMENVTAF WEEGFGELFE KAKQNNNNRK 421 TSNGDDSLFF
SNFSLGTPV LKDINFKIER GQLLAVAGST GAGKTSLLMV IMGELEPSEG 481
KIKHSGRISF CSQFSWIMPG TIKENIIFGV SYDEYRYRSV IKACQLEEDI SKFAEKDNIV

15 541 LGEGGITLSG GQRARISLAR AVYKDADLYL LDSPFGYLDV LTEKEIFESC
VCKLMANKTR 601 ILVTSKMEHL KKADKILILH EGSSYFYGTF SELQNLQPDF
SSKLMGCDSF DQFSAERRNS 661 ILTETLHRFS LEGDAPVSWT ETKKQSFQKT
GEFGEKRKNS ILNPINSIRK FSIVQKTPLQ 721 MNGIEEDSDE PLERRLSLVP
DSEQGEAILP RISVISTGPT LQARRRQSVL NLMTHSVNQG 781 QNIHRKTTAS

20 TRKVS LAPQA NLTELDIYSR RLSQETGLEI SEEINEEDLK ECFDDMESI 841
PAVTTWNTYL RYITVHKSLI FVLIWCLVIF LAEVAASLVV LWLLGNTPLQ
DKGNSTHSRN 901 NSYAVIITST SSYYVFYIYV GVADTLLAMG FFRGLPLVHT
LITVSKILHH KMLHSV LQAP 961 MSTLNTLKAG GILNRFSKDI AILDDLPLT
IFDFIQLLLI VIGAI VAV LQPYIFVATV 1021 PVIVAFIMLR AYFLQTSQQL

25 KQLESEGRSP IFTHLVTSK GLWTLRAFGR QPYFETLFHK 1081 ALNLHTANWF
LYLSTLRWFQ MRIEMIFVIF FIAVTFISIL TTGEGEGRVG IILTLAMNIM 1141
STLQWAVNSS IDVD SLMRSV SRVFKFIDMP TEGKPTKSTK PYKNGQLSKV
MIENSHVKK 1201 DDIWPSGGQM TVKDLTAKYT EGGNAILENI SFSISPGQRV
GLLGRTGSGK STLLSAFLRL 1261 LNTEGEIQID GVSWDSITLQ QWRKAFGVIP

30 QKVFIFSGTF RKNLDPYEQW SDQEIWKVAD 1321 EVGLRSVIEQ FPGKLDFVLV
DGGCVLSHGH KQLMCLARSV LSKAKILLLD EPSAHLDPVT 1381 YQIIRRTLKQ

AFADCTVILC EHRIEAMLEC QQFLVIEENK VRQYDSIQKL LNERSLFRQA 1441
ISPSDRVKLF PHRNSSKCKS KPQIAALKEE TEEEVQDTRL (SEQ ID NO: 1).

Functional domains may include the membrane spanning domain 1 having amino acids of the transmembrane regions: 78 to 98, 123 to 146, 196 to 216, 223 to 243, 299 to 319, 340 to 358; nucleotide binding domain 1 having amino acids 389 to 670; a regulatory domain having amino acids 639 to 849; a membrane spanning domain 2 having amino acid of the transmembrane regions: 866 to 879, 919 to 939, 991 to 1011, 1014 to 1034, 1096 to 1116, 1131 to 1151; and a nucleotide binding domain 2 having amino acids 1208 to 1480 (PDZ-binding region amino acids 1478 to 1480).

In certain embodiments, the subject is diagnosed with a mutation outside or inside a functional domain with sweat chloride elevation 30-60 mEq/L, and without a clinically approved genotype. In certain embodiments, the subject is diagnosed with a heterologous mutation outside or inside a functional domain. In certain embodiments, the subject is diagnosed with homologous mutations inside or outside a functional domain.

In certain embodiments, this disclosure contemplates treating patients with a diagnosis of rhinosinusitis, chronic obstructive pulmonary disease, bronchiectasis and/or other condition reported herein that have elevated sweat chloride, e.g., greater than 60 mEq/L, without a diagnosis of a clinically approved cystic fibrosis genotype, unable to identify a known cystic fibrosis transmembrane conductance regulator mutation, pancreatic insufficiency, or combinations thereof that respond to treatment with drugs 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), derivative, or salt thereof optionally in combination with other CF drug(s), e.g., lumacaftor, elexacaftor, ivacaftor, tezacaftor, or combinations thereof.

In certain embodiments, 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), derivative or salt thereof is administered in combination with another active agent such as a bronchodilator, corticosteroid, antimuscarinic, antibiotic, or combinations thereof. In certain embodiments, the active agent is administered as a pill, an injection, a tablet, or syrup or for using an inhaler or nebulizer,

compressed gas or liquid to be released into a mist, optionally using an aqueous saline solution, or inhaled through a mouthpiece or face mask.

In certain embodiments, the bronchodilator is a beta-2 agonist, such as salbutamol, salmeterol, formoterol and vilanterol or an anticholinergic, such as ipratropium, tiotropium, acclidinium, or glycopyrronium, or an antimuscarinic such as atropine or scopolamine. In certain
5 embodiments, the drug is administered in combination with a bronchodilator albuterol, formoterol, or levalbuterol or salts thereof.

In certain embodiments, a compound disclosed herein is administered in combination with a mucolytic agent such as bromhexine or salts thereof.

10 In certain embodiments, a compound disclosed herein is administered in combination with an anti-inflammatory agent such as a corticosteroid, fluticasone, or salts thereof. Suitable anti-inflammatory compounds include both steroidal and non-steroidal structures. Suitable non-limiting examples of steroidal anti-inflammatory compounds are corticosteroids such as hydrocortisone, cortisol, triamcinolone, alpha-methyl dexamethasone, dexamethasone-phosphate,
15 beclomethasone dipropionates, clobetasol valerate, desonide, desoximetasone, desoxycorticosterone acetate, dexamethasone, diflorasone diacetate, diflucortolone valerate, fludrenolone, flucorolone acetonide, fludrocortisone, flumethasone pivalate, fluocinolone acetonide, fluocinonide, fluocortin butyl ester, fluocortolone, fluprednidene acetate, flurandrenolone, halcinonide, hydrocortisone acetate, hydrocortisone butyrate,
20 methylprednisolone, triamcinolone acetonide, cortisone, cortodoxone, fludrocortisone, fludrocortisone, fluocinolone, medrysone, betamethasone and the balance of its esters, chloroprednisone, dichlorisone, flunisolide, fluperolone, fluprednisolone, hydrocortisone valerate, hydrocortamate, prednisone, paramethasone, prednisolone, prednisone, beclomethasone dipropionate, and triamcinolone. Mixtures of the above steroidal anti-inflammatory compounds
25 may also be used.

In certain embodiments, a compound disclosed herein is administered in combination with one or more antibiotics. Suitable antibiotics include, without limitation, such as 2,4-diaminopyrimidines, including baquiloprim, brodimoprim, iclaprim, ormetoprim, pyrimethamine, tetroxoprim, trimethoprim; aminocoumarins, including novobiocin; aminocyclitols, including
30 spectinomycin; aminoglycosides, including amikacin, apramycin, arbekacin, bekanamycin, butirosin, dibekacin, dihydrostreptomycin, etimicin, fortimicins, astromicin, framycetin,

gentamicin, hygromycin B, isepamicin, kanamycin, micromonicin, neomycin, netilmicin, paromomycin, plazomicin, ribostamycin, sisomicin, streptomycin, tobramycin; aminomethylcyclines, including omadacycline; amphenicols, including azidamfenicol, chloramphenicol, florfenicol, thiamphenicol; ansamycins, including rifabutin, rifampicin, rifapentine, rifaximin; antitubercular agents, including cycloserine, delamanid, ethambutol, ethionamide, morinamide, p-aminosalicylic acid (PAS), protionamide, pyrazinamide, terizidone, thioacetazone; bacteriocins, including nisin; b-lactam carbapenems, including loracarbef, biapenem, doripenem, ertapenem, faropenem, imipenem, meropenem, panipenem, razupenem, sulopenem, tebipenem, tomopenem; b-lactam cephalosporins, including cefacetrile, cefaclor, cefadroxil, cefalexin, cefalonium, cefaloridine, cefamandole, cefapirin, cefatrizine, cefazaflur, cefazedone, cefazolin, cefcapene, cefdinir, cefditoren, cefepime, cefetamet, cefixime, cefmenoxime, cefodizime, cefonicid, cefoperazone, ceforanide, cefoselis, cefotaxime, cefotiam, cefovecin, cefozopran, cefpimizole, cefpiramide, cefpirome, cefpodoxime, cefprozil, cefquinome, cefradine, cefroxadine, cefsulodin, ceftaroline, ceftazidime, cefteram, ceftazole, ceftibuten, ceftiofur, ceftizoxime, ceftobiprole, ceftolozane, ceftriaxone, cefuroxime, cefuzonam; b-lactam cephamycin, including cefbuperazone, cefmetazole, cefotetan, cefoxitin; b-lactam monobactams, including aztreonam, carumonam, tigemonam; b-lactam oxacephems, including flomoxef, latamoxef, moxalactam; b-lactam penicillins, including amdinocillin, amoxicillin, ampicillin, apalcillin, azidocillin, azlocillin, bacampicillin, carbenicillin, ciclacillin, clemizole penicillin, cloxacillin, cyclacillin, dicloxacillin, epicillin, fenbenicillin, floxacillin, hetacillin, mecillinam, metampicillin, methicillin sodium, mezlocillin, nafcillin, oxacillin, penamecillin, penethamate hydroiodide, penicillin G, penicillin G benzathine, penicillin G procaine, penicillin N, penicillin O, penicillin V, phenethicillin potassium, piperacillin, pivampicillin, pivmecillinam, propicillin, quinacillin, sultamicillin, talampicillin, temocillin, ticarcillin; b-lactam in combination with b-lactamase inhibitors, including avibactam, clavulanic acid, relebactam, sulbactam, tazobactam, vaborbactam, zidebactam; bicyclomycins, including bicozamycin; bis-benzimidazoles including ridinilazole; cyclic esters, including fosfomicin; fluoroquinolones, including avarofloxacin, balofloxacin, besifloxacin, cinoxacin, ciprofloxacin, clinafloxacin, danofloxacin, delafloxacin, difloxacin, enoxacin, enrofloxacin, finafloxacin, fleroxacin, flumequine, garenoxacin, gatifloxacin, gemifloxacin, grepafloxacin, levofloxacin, lomefloxacin, marbofloxacin, miloxacin, moxifloxacin, nadifloxacin, norfloxacin, ofloxacin, orbifloxacin, pazufloxacin, pefloxacin,

pradofloxacin, prulifloxacin, rosoxacin, rufloxacin, sarafloxacin, sitafloxacin, sparfloxacin, temafloxacin, tosufloxacin, trovafloxacin, zabofloxacin; fusidane-type, including helvolic acid, fusidic acid and cephalosporin; glycolipodepsipeptides, including ramoplanin; glycopeptides, including avoparcin, dalbavancin, norvancomycin, oritavancin, teicoplanin, telavancin, 5 vancomycin; glycyclcyclines, including tigecycline; hybrids, oxazolidinone-quinolone; lincosamides, including clindamycin, lincomycin, pirlimycin; lipopeptides, including daptomycin, surotomycin; macrolides, including azithromycin, cethromycin, clarithromycin, dirithromycin, erythromycin, fidaxomicin, flurithromycin, gamithromycin, josamycin, kitasamycin, leucomycin, miocamycin, oleandomycin, primycin, rokitamycin, rosaramicin, roxithromycin, solithromycin, 10 spiramycin, telithromycin, tildipirosin, tilmicosin, troleandomycin, tulathromycin, tylosin, tylvalosin; nitrofurans, including furaltadone, furazidin, furazolidone, nifuroxazide, nifurtinol, nifurzide, nitrofur, nitrofurantoin, nitrofurazone; nitroimidazoles, including dimetridazole, metronidazole, ornidazole, secnidazole, tinidazole; oligosaccharides, including avilamycin, everninomicin; polymyxins, including polymyxin E (colistin), polymyxin B; polypeptides, 15 including amphomycin, bacitracin, capreomycin, enduracidin, enramycin, gramicidin(s), ristocetin, tuberactinomycin, tyrocidine, tyrothricin, viomycin; pseudomonic acids including mupirocin; quinoxalines including carbadox, olaquinox; sulfonamides including acetyl sulfamethoxypyrazine, chloramine-B, chloramine-T, dichloramine T, mafenide, sulfacetamide, sulfadiazine, sulfadimidine, sulfamethazine, sulfamethizole, sulfapyridine, sulfathiazole, 20 sulfisomidine, sulfisoxazole; sulfones, including dapsone sodium, sulfanilic acid; tetracycline including chlortetracycline, clomocycline, demeclocycline, doxycycline, lymecycline, meclocycline, methacycline, minocycline, oxytetracycline, rolitetracycline, tetracycline; triazaacenaphthylene including gepotidacin, or any combination thereof.

In certain embodiments, 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b] 25 [1,3,4]thiadiazine (uHTS-159) derivative or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), derivative, or salt thereof is administered in combination with an antibiotic agent such as macrolides, azithromycin, antipseudomonal, fluoroquinolones, ciprofloxacin, levofloxacin, ceftazidime, piperacillin and tazobactam, imipenem, aminoglycosides, aztreonam, tobramycin, colistin, colistimethate sodium, 30 or salt thereof.

In certain embodiments, 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salt thereof and/or 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), derivative, or salt thereof is administered in combination with a reversible inhibitor of dipeptidyl peptidase I (DPP1) such as brensocatic.

Pharmaceutical compositions

In certain embodiment, this disclosure contemplates pharmaceutical compositions comprising compounds disclosed herein and pharmaceutically acceptable excipient. In certain embodiments, this disclosure contemplates the production of a medicament comprising compounds disclosed herein and uses for methods disclosed herein.

Pharmaceutical compositions typically comprise an effective amount of compounds and a suitable pharmaceutical acceptable carrier. The preparations can be prepared in a manner known per se, which usually involves mixing the compounds according to the disclosure with the one or more pharmaceutically acceptable carriers, and, if desired, in combination with other pharmaceutical active compounds, when necessary under aseptic conditions. Reference is made to U.S. Pat. No. 6,372,778, U.S. Pat. No. 6,369,086, U.S. Pat. No. 6,369,087 and U.S. Pat. No. 6,372,733 and the further references mentioned above, as well as to the standard handbooks, such as the latest edition of Remington's Pharmaceutical Sciences.

In certain embodiments, pharmaceutical composition is in the form of a neutral pH buffered solution optionally comprising isotonic saline or a pharmaceutically acceptable mono or polysaccharide. In certain embodiments, pharmaceutical composition is contained in a device in the form of a powder, liquid, or gas, e.g., contained in pressurized container (sterilized). In certain embodiments, the device is configured to dispense an aerosol spray.

In certain embodiments, the pharmaceutically acceptable excipient is selected from lactose, sucrose, mannitol, triethyl citrate, dextrose, cellulose, methyl cellulose, ethyl cellulose, hydroxyl propyl cellulose, hydroxypropyl methylcellulose, carboxymethylcellulose, croscarmellose sodium, polyvinyl N-pyrrolidone, crospovidone, ethyl cellulose, povidone, methyl and ethyl acrylate copolymer, polyethylene glycol, fatty acid esters of sorbitol, lauryl sulfate, gelatin, glycerin, glyceryl monooleate, silicon dioxide, titanium dioxide, talc, corn starch, carnauba wax, stearic acid, sorbic acid, magnesium stearate, calcium stearate, castor oil, mineral oil, calcium

phosphate, starch, carboxymethyl ether of starch, iron oxide, triacetin, acacia gum, esters, or salts thereof.

In certain embodiments, the disclosure relates to pharmaceutical compositions comprising compounds disclosed herein and a pharmaceutically acceptable excipient. In certain embodiments, the composition is a powder or liquid, in the form of a gel, particles, pill, tablet, or in a capsule (gel capsule) or the composition is an aqueous buffer, e.g., a pH between 6 and 8. In certain embodiments, the pharmaceutically acceptable excipient is selected from a filler, glidant, binder, disintegrant, lubricant, and saccharide.

Compositions suitable for parenteral injection may comprise physiologically acceptable sterile aqueous or nonaqueous solutions, dispersions, suspensions or emulsions, and sterile powders for reconstitution into sterile injectable solutions or dispersions. Examples of suitable aqueous and nonaqueous carriers, diluents solvents or vehicles include water, ethanol, polyols (propylene glycol, polyethylene glycol, glycerol, and the like), suitable mixtures thereof, vegetable (such as olive oil, sesame oil) and injectable organic esters such as ethyl oleate.

Prevention of the action of microorganisms may be controlled by addition of any of various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, sorbic acid, and the like. It may also be desirable to include isotonic agents, for example sugars, sodium chloride, and the like. Prolonged absorption of the injectable pharmaceutical form can be brought about by the use of agents delaying absorption, for example, aluminum monostearate and gelatin.

Solid dosage forms for oral administration include capsules, tablets, pills, powders and granules. In such solid dosage forms, the compounds may be admixed with at least one inert customary excipient (or carrier) such as sodium citrate or dicalcium phosphate or: (a) fillers or extenders, as for example, starches, lactose, sucrose, glucose, mannitol and silicic acid, (b) binders, as for example, carboxymethylcellulose, alginates, gelatin, polyvinylpyrrolidone, sucrose, and acacia, (c) humectants, as for example, glycerol (d) disintegrating agents, as for example, agar-agar, calcium carbonate, potato or tapioca starch, alginic acid, certain complex silicates, and sodium carbonate, (e) solution retarders, as for example paraffin, (f) absorption accelerators, as for example, quaternary ammonium compounds, (g) wetting agents, as for example cetyl alcohol, and glycerol monostearate, (h) adsorbents, as for example, kaolin and bentonite, and (i) lubricants, as for example, talc, calcium stearate, magnesium stearate, solid polyethylene glycols, sodium lauryl

sulfate, or mixtures thereof. In the case of capsules, tablets, and pills, the dosage forms may also comprise buffering agents.

Liquid dosage forms for oral administration include pharmaceutically acceptable emulsions, solutions, suspensions, syrups, and elixirs. In addition to the compounds, the liquid dosage forms may contain inert diluents commonly used in the art, such as water or other solvents, solubilizing agents and emulsifiers, for example, ethyl alcohol, isopropyl alcohol, ethyl carbonate, ethyl acetate, benzyl alcohol, benzyl benzoate, propylene glycol, 1,3-butylene glycol, dimethylformamide, oils, in particular, cottonseed oil, groundnut oil, corn germ oil, olive oil, castor oil and sesame oil, glycerol, tetrahydrofurfuryl alcohol, polyethylene glycols and fatty acid esters of sorbitan or mixtures of these substances, and the like.

Optimizing the strong effects on CFTR function in combination with VX-770

Experiments were performed to evaluate the effects of compounds disclosed herein on CFTR function in combination with VX-770. An initial probing of the series produced by the high throughput screen revealed that a small alkyl group at C6 of the triazolothiadiazine core and an arene at C3 produced compounds that co-potentiated mutant CFTR (G551D), i.e., more than with VX-770 alone. Experiments indicated that a disubstituted arene with an ortho-substitution was strongly correlated with efficacy. Thus, the size of the C6 alkyl group was varied. Interestingly, a slightly larger tert-butyl substituent produced compounds (8d and 9d) (see Figure 6C) that were quite potent and an improvement compared to VX-770 alone.

In order to balance cLogP and efficacy in compounds (10a, 10b, and 10e) (see Figure 6D) were produced with cLogP of less than 5 and potency reaching and surpassing 200% CFTR function compared to VX-770 as a single agent. The ability of these compounds to augment VX-770 stimulation of a relatively common CFTR variant (G551D) by close to 400% is surprising and indicates a distinct mechanism of action with potential usefulness against CFTR-related illnesses for patients.

Clinically approved CFTR potentiators and correctors are available for the majority of the CF population. Use of alternative CFTR modulators as reported herein are contemplated for patients that encode rare or refractory CFTR variants, and for patients approved for modulator treatment who respond poorly, or experience toxicity. Mutant CFTRs were treated with saturating concentrations of a CFTR potentiator, VX-770. These compounds were evaluated using a well-

validated Fischer rat thyroid model that has been extensively utilized for cystic fibrosis drug discovery. It is contemplated that compounds reported herein can be used for managing CF, non-CF, and CFTR-related diseases and conditions such as CF or non-CF chronic pancreatitis, CF or non-CF rhinosinusitis, CF or non-CF bronchiectasis, CF or non-CF Sjogren's anemia, CF or non-CF xerostomia, CF or non-CF xerophthalmia and CF or non-CF chronic obstructive pulmonary disease.

Phosphodiesterase 4 Inhibitors for Treatment of Chronic Sinusitis

Chronic rhinosinusitis is a significant cause of morbidity in patients with cystic fibrosis (PwCF) and believed to seed lower airways, precluding pathogen eradication, and compromising pulmonary graft integrity post lung transplant. Activating mutant CFTR by modulator therapy improves CF CRS, although sinus opacification, polymicrobial infection, and inflammation persist despite drug treatment. Recent clinical results furnish rationale for testing other (including topically administered vs systemically) CFTR activating compounds as part of treatment for CF CRS. Moreover, the ability of CFTR modulators to strongly augment mucociliary clearance in nasal airways and improve CF CRS suggests activation of wild type CFTR could provide a means to address non-CF CRS. This may particularly be true among frequent individuals who are silent carriers for CFTR mutations and at increased risk of non-CF CRS. Some patients with non-CF CRS are silent carrier for the common F508del CFTR mutation. Experiments indicate such patients may benefit from a phosphodiesterase 4 (PDE) inhibitor treatment that may be optimized for mutant and wild type CFTR stimulation in sinonasal epithelium. Certain compounds would also be expected to blunt inflammation in diseased nasal airways and sinuses. This provides a treatment strategy for PwCF – over 90% of whom experience CRS – including those without access to a modular treatment but encoding PDE4i-sensitive CFTR mutations. The strategy also points to a new approach for robust therapy of chronic rhinosinusitis in the general (non-CF) population.

An effective modulator treatment (MT) for CF is clinically approved for many mutations in the cystic fibrosis transmembrane conductance regulator (CFTR) which are established as disease-causing variants. Triple combination therapy (TCT; comprised of elexacaftor (VX-445), tezacaftor (VX-661), and ivacaftor (VX-770) confers improvement in lung function, chronic rhinosinusitis (CRS), and markers of CFTR activity. A subset of patients fails TCT (e.g., about 5% encoding the F508del variant may not benefit) or exhibit limiting toxicity (for example, hepatic

impairment). Effective therapies are not available for patients with certain CFTR mutations, such as refractory defects of folding and maturational processing (e.g., N1303K) or premature truncation codons (PTCs, including W1282X). There remains a need to identify improvements for individuals with these variants.

5 There are upper and lower respiratory tract manifestations of CF. Patients with cystic fibrosis experience tissue damage throughout the respiratory tract, including nasal passages, sinuses, sub-mucosal glands, and pulmonary parenchyma. Involvement of the upper respiratory tract is common (radiographic evidence of sinusitis), with impaired mucociliary clearance, mucus obstruction, chronic infection (by *P. aeruginosa*, *H. influenza*, *Staphylococcus aureus*,
10 *Burkholderia cepacia* and other pathogens; many of which are also present in CF lungs), glandular hypertrophy, scarring, remodeling, and pronounced immune-mediated damage. Chronic rhinosinusitis in CF is a cause of significant morbidity, with sequelae that include persistent and/or severe facial pain, headache, congestion, increased cough, rhinorrhea, anosmia, halitosis, and disruption of sleep patterns. Diagnosed CRS-associated conditions such as nasal polyposis, sinus
15 dysgenesis, inflammation, and scarring sometimes necessitate surgical intervention to relieve obstructed ostia, facilitate sinus drainage, excise polyps, etc.

 Pulmonary tissues typically exhibit similar clinical findings to those in the sinonasal tract in CF. For example, both nasal and lower airways exhibit similar mucociliary defects, bacterial pathogens, bioelectric abnormalities, and immunologic hyperactivity in vivo. Epithelial
20 monolayers from both nasal and lower airways are used as tools to understand upper respiratory tract responses and modulator activity in lungs. Chronically infected nasal sinuses may serve as a bacterial reservoir that infects the lungs, impairs pulmonary graft viability during systemic immune suppression, and contributes to failure of attempts to eradicate lower airway colonization.

 Clinically approved MT treatments are, in some instances, inadequate. Normalization of
25 CFTR function and activation of mucociliary clearance can substantially improve chronic rhinosinusitis in CF. It is contemplated that activating wild type CFTR among individuals without CF but exhibiting partial CFTR deficiency (e.g., attributable to silent carrier status of the F508del mutation, which confers increased risk of non-CF CRS) will benefit from robust CFTR activation by augmenting CFTR activity. Chronic infection, inflammation, cigarette smoke, environmental
30 toxin exposures, and other sinonasal insults cause diminished CFTR activity. Thus, it is contemplated that clinical improvement can be achieved by measures that activate wild type CFTR

in the setting of these insults. There is a need to identify improved treatments for both CF and non-CF sinusitis.

Reported herein are PDE4 inhibitors developed specifically for activating CFTR in sinonasal epithelium which are well suited as therapies for chronic rhinosinusitis. Although it is not intended that certain embodiments of this disclosure be limited by any particular mechanism, certain compounds described here appear to function via PDE4 inhibition and are contemplated to confer both mucus clearance and immune-attenuating properties in human airways.

A CFTR activator suitable for respiratory mucosal administration and treatment of chronic rhinosinusitis in cystic fibrosis and other clinical settings

Agents were screened from a library of bioactive compounds for rescue of N1303K or W1282X CFTR, variants with measurable functions. The screen was performed using stably transduced Fisher rat thyroid (FRT) cells, a model validated in successful CFTR pharmaceutical screening programs. Following confirmation and dose-dependence analysis, as well as in silico profiling to exclude candidate drugs with inherent chemical liabilities, several compound series were identified. Candidates were tested in complementary cell systems. The most promising agents were advanced at early stage to validation in primary airway epithelial monolayers grown on permeable supports.

A compound active in two distinct HRP-based screens (W1282X, N1303K) was also shown to acutely stimulate short circuit current (Isc) utilizing wildtype CFTR in FRT cells. Because strong and immediate enhancement of CFTR stimulation was observed for this compound series, studies of acute CFTR potentiation were pursued in detail. HDCF104 augmented wild type CFTR 150% above what could be achieved with a clinically approved potentiator (ivacaftor) alone (Figure 1B). Other structural analogs led to enhanced wild type CFTR potentiation and augmented ivacaftor alone. Importantly, drugs in the series also acutely stimulate disease-causing CFTR variants. For example, Figures 2A and 2B show activity of compound uHTS159, an HDCF104 analog, against G551D CFTR. The drug is roughly equivalent to VX-770, and when saturating concentrations of the potentiator are applied. The HDCF series agent significantly enhances Isc compared to using VX-770 alone.

Experiments were performed to determine HDCF-based stimulation of F508del CFTR following triple combination therapy (TCT). The FRT model is predictive of in vivo clinical

response – particularly when a test compound achieves greater than 10 % wild type levels of activity during mutant CFTR stimulation. Primary CF airway epithelial cells are also utilized as important indicators of patient benefit. HDCF compounds were examined using airway epithelium from individuals with or without cystic fibrosis. Figure 3 provides an example showing significant (10-20 $\mu\text{A}/\text{cm}^2$) CFTR-dependent anion current mediated by HDCF104 following pharmacocorrection of F508del CFTR. Importantly, HDCF104 was found to elevate the I_{sc} beyond what can be achieved using standard concentrations of VX-770 by approximately 23-65%, depending on order of addition.

Experiments were performed on the N1303K CFTR mutation. Figures 4A-4B show acute activation of N1303K CFTR by HDCF104 in primary airway epithelial monolayers. Nasal airway epithelial cells from an individual with genotype N1303K/N1303K exhibit 4-5 $\mu\text{A}/\text{cm}^2$ stimulation following corrector treatment (versus negligible activation by VX-770).

Because drugs in the HDCF group were identified by unbiased, compound library screening, mechanism of action was not initially known. Since many CFTR variants (as well as wild type CFTR) are activated by the series – including CFTR mutations with severe gating defects such as G551D – it was contemplated that HDCF104 was directly binding to CFTR protein. For example, an increase in surface HRP-CFTR activity observed in the initial drug screen might be taken to suggest enhanced steady-state protein levels at the cell surface, but could also support other mechanisms of short circuit current enhancement (complementary to available modulators) such as increased CFTR plasma membrane insertion or decreased retrieval/ endocytosis (i.e., due to altered vesicle trafficking in primary airway cells). Because plasma membrane recycling of CFTR can be strongly influenced by compartmental cAMP, co-administration of PDE4 inhibitors such as IBMX (broad spectrum PDE blocker) and rolipram (PDE4-specific inhibitor) were tested. These experiments indicated overlapping activity with HDCF104, whereas PDE3-specific blockers such as cilostamide had a negligible effect (Figure 5A-5C).

Direct measurements of PDE inhibition by HDCF compounds were performed. The HDCF compounds were evaluated by a conventional cell-free enzymatic assay using purified PDE protein in which luciferase serves as an ATP “sink” over the linear range of luminescence. Because ATP is also a substrate for PKA, inhibition of PDE4 (with a resultant rise in cAMP and PKA activity) elicits a decrease in ATP available to luciferase and decreased cellular luminescence. IC_{50} determinations for HDCF104 were made with purified PDE4B or PDE3A, respectively. PDE

enzymes were purchased from Abcam and used in PDE-Glo™ Phosphodiesterase Assay (Promega). IC₅₀ of HDCF104 = 2.9 μM for PDE4B and greater than 100 μM for PDE3A. IC₅₀ of uHTS_159 (in the HDCF series) was 14.0 μM for PDE3A and low nM range for PDE4B.

The inhibitory profile of HDCF104 showed IC₅₀ of 2.9 μM for PDE4B – a concentration that agrees well with the half-maximal activation levels of HDCF104 needed to stimulate mutant or wild type CFTR in primary airway cells. Drug concentrations in this range can be utilized for mucosal administration to the human respiratory tract (unlike oral agents, where greater potency may be required to avoid systemic toxicity). For example, levels of approved antibiotics or other compounds administered by aerosol to CF lungs may be 100-200 μM in CF airway secretions in vivo.

HDCF104 was shown to 1) substantially augment the level of activation achieved with clinically approved modulators against clinically important CFTR variants (e.g., F508del, G551D, N1303K; as well as WT CFTR), and 2) activate short circuit current in epithelial monolayers encoding variants for which no CFTR modulators are presently available (e.g., N1303K). Therapy for CF CRS often includes oral antibiotics, sinus lavage and installation of mucosal steroids, as well as topical antibiotics. Antihistamines and decongestants are also utilized. Thus, these agents are contemplated to be used in combination with HDCF104 and other compounds disclosed herein.

Compounds in the HDCF series have a number of favorable attributes relevant to CF sinus disease including use as topical (mucosal) drugs for CFTR modulation, activation of mutant CFTR (including “corrected” F508del CFTR in model systems of nasal airway epithelium to levels above what can be achieved with ivacaftor (VX-770) alone), and potential for amelioration of immune-mediated lung injury. Experiments indicate clinical development is desirable for nasal airway/sinus administration. Contemplated administration of compounds disclosed herein include positive pressure irrigation or exhaled delivery system (EDS) technology, e.g., an inhaler with a nosepiece is specifically shaped to seal tightly in the nostril, acting as a stent in the nasal valve, and allowing for the transfer of pressure from the mouth into the nose, wherein a flexible mouthpiece enables a patients to utilize the device while maintaining a seal in the nostril providing a means of activating CFTR and mucociliary clearance, as well as blunting inflammation, in both CF and non-CF chronic rhinosinusitis.

Contemplated methods of treatment with compounds disclosed herein include use for refractory CFTR mutations and CF chronic rhinosinusitis. A threshold that predicts clinically

relevant rescue of CFTR variants in vitro is often taken as 30-40% wild type activity in bronchial airway monolayers. Ten to fifteen (10-15) $\mu\text{A}/\text{cm}^2$ is frequently observed in wild type nasal epithelial monolayers from many patient samples, although larger CFTR currents can be obtained from certain patients. Thus, in certain embodiment, selecting a patient for treatment includes those
5 diagnosed with less than 15 or 20 $\mu\text{A}/\text{cm}^2$ current after assaying nasal epithelia monolayers.

With regard to levels of CFTR activation for rare variants without approved modulators such as N1303K, combination drug treatment (e.g., as with patients encoding F508del) is contemplated to achieve clinically relevant levels of rescue. CFTR activators of N1303K CFTR and HDCF104 in combination are contemplated. Among gating mutations such as G551D,
10 experiments reported herein indicate HDCF104 complements ivacaftor. For certain PwCF, a drug such as HDCF104 may provide considerable benefit particularly for patients that fail or are ineligible for other therapies.

Although it is not intended that embodiments of this disclosure be limited by any particular mechanism, compounds disclosed herein believed to be PDE4 inhibitors (PDE4is). These PDE4is
15 are contemplated for use in human airway CFTR therapeutic applications. PDE4is are activators of wild type CFTR. Enzymatically and pharmacologically distinct members of the PDE family (including PDE subtypes 1-8) are expressed in mammalian cells or tissues and hydrolyze/inactivate cyclic nucleotides. Blockade of certain PDEs elicits compartmental elevation of cyclic AMP, which (in a manner dependent on PDE category, tissue, cell type, subcellular
20 location, etc.) can strongly activate protein kinase A and stimulate CFTR through phosphorylation of the regulatory or other protein domains. Experiments indicate that non-CF obstructive airway diseases such as CRS, bronchiectasis, and COPD – which share a number of key pathogenic features with CF lung disease – are more likely to occur among carriers of cystic fibrosis. These same diseases are also associated with diminished wild type CFTR activity. In patients with CF
25 lung disease and/or CRS, activation of CFTR and resulting enhancement of mucociliary clearance with compound drugs such as ivacaftor provide clinical benefit. Such findings provide rationale for mucosal activation of CFTR in the setting of COPD, bronchiectasis, or CRS. In this context, an aerosolized PDE3/PDE4 inhibitor ensifentrine (RPL445) was developed and has completed clinical testing and shown efficacy in patients with COPD. In certain embodiments, this disclosure
30 contemplates topical use of compounds disclosed herein for CFTR activation as a means to address obstructive airway diseases. Mucosal administration of CFTR activators can be used in the

treatment of respiratory conditions such as CF, bronchiectasis, COPD, and CRS. In certain embodiments, compounds disclosed herein may be administered in combination with ensifentrine (RPL445).

5 In certain embodiments, this disclosure contemplates topical use of compounds disclosed herein for PDE inhibition as an effective means to diminish immune hyper-responsiveness in human lower airways. The PDE inhibitors are contemplated to blunt inflammation for treating asthma, mediate bronchodilation and suppress inflammatory response associated with COPD, diminish immune activity of eosinophils in human bronchial specimens from subjects with bronchitis, repress inflammation in human airway epithelial cells, decrease inflammatory effects
10 attributable to alveolar macrophages, enhance lung clearance of pathogens by recruitment of macrophages, diminish an array of inflammatory modifiers in the respiratory tract, and mediate additional effects. The PDE4 inhibitor, roflumilast was developed for treatment of COPD. Compounds disclosed herein are contemplated for use as aerosolized PDE blockers for COPD. In certain embodiments, this disclosure contemplates use of compounds disclosed herein in
15 combination with roflumilast.

In certain embodiments, this disclosure contemplates use of compounds disclosed herein in combination with tanimilast, e.g., to treat chronic rhinosinusitis (CRS), chronic bronchitis, emphysema or other lung disease or condition reported herein. In certain embodiments, this disclosure contemplates use of compounds disclosed herein in lower airways of COPD or
20 bronchiectasis patients. In certain embodiments, this disclosure contemplates use of compounds disclosed herein in therapy of upper airway disease (i.e., CRS) e.g., as a topical PDE inhibitor. In certain embodiments, this disclosure contemplates use of compounds disclosed herein delivered by mucosal administration (intranasal or oral) in combination with a CFTR activator or modulator therapy to improved treatment of CF CRS, to diminish bacterial seeding of CF lungs, promote
25 Pseudomonas eradication (i.e., by limiting the upper airway “reservoir”), and/or to use in lower airways of PwCF. Patients suffer from non-CF CRS which has been linked to both mutant CFTR carrier status and acquired defects in CFTR activity. Compounds disclosed herein are contemplated for use in blunting respiratory inflammation in humans in either CF or non-CF CRS patients. Palliation of CF CRS using a topical PDE4 blocker are contemplated to reduce morbidity. In
30 certain embodiments, this disclosure contemplates use of compounds disclosed herein as clinical benefit in CRS by promoting mucus clearance and attenuating airway inflammation.

CLAIMS

1. A method of treating or preventing rhinosinusitis, chronic obstructive pulmonary disease, or bronchiectasis in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor.
2. The method of claim 1, wherein the phosphodiesterase (PDE) inhibitor is a phosphodiesterase (PDE) 4 inhibitor.
3. The method of claim 1, wherein the phosphodiesterase (PDE) inhibitor is selected from 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salts thereof to the subject in need thereof.
4. The method of claim 1, wherein rhinosinusitis is chronic.
5. The method of claim 1, wherein administration is through the mouth and/or nose by inhalation or topical application.
6. The method of claim 1, wherein the subject is diagnosed with cystic fibrosis.
7. The method of claim 1, wherein the subject is not diagnosed with cystic fibrosis.
8. The method of claim 1, wherein the subject is diagnosed with pancreatic sufficiency.
9. The method of claim 1, wherein the subject is diagnosed with moderately elevated sweat chloride and the moderately elevated sweat chloride at a concentration between 30 to 60 millimoles per liter.
10. The method of claim 1, wherein a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and no mutation was identified in

the sample, thereby providing a subject diagnosed without a known cystic fibrosis transmembrane conductance regulator mutation.

11. The method of claim 1, wherein a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and a mutation was identified on a single allele in the sample, thereby providing a subject diagnosed with a heterozygous cystic fibrosis transmembrane conductance regulator mutation.

12. The method of claim 11 wherein the heterozygous cystic fibrosis transmembrane conductance regulator mutation is a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

13. The method of claim 12 wherein the heterozygous cystic fibrosis transmembrane conductance regulator mutation is N1303K, W1282X, G551D, or G542X.

14. The method of claim 1, wherein a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and the mutation was identified on both alleles in the sample, thereby providing a subject diagnosed with homozygous cystic fibrosis transmembrane conductance regulator mutations.

15. The method of claim 14 wherein the cystic fibrosis transmembrane conductance regulator mutation involves a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

16. The method of claim 14 wherein the cystic fibrosis transmembrane conductance regulator mutation involves an N1303K, W1282X, G551D, or G542X mutation.

17. The method of claim 1 wherein the phosphodiesterase (PDE) inhibitor is administered in combination with a cystic fibrosis drug.

18. The method of claim 17, wherein the cystic fibrosis drug is CFTR activator or modulator.

19. The method of claim 17, wherein the cystic fibrosis drug is elexacaftor, ivacaftor, tezacaftor, or combination thereof.
20. The method of claim 17, wherein the cystic fibrosis drug is a combination of ivacaftor and tezacaftor or is a combination of ivacaftor and lumacaftor.
21. The method of claim 17, wherein the cystic fibrosis drug is a combination of elexacaftor, ivacaftor, and tezacaftor.
22. The method of claim 1 wherein the phosphodiesterase (PDE) inhibitor is administered in combination with another active agent such as a bronchodilator, corticosteroid, antimuscarinic, antibiotic, or combinations thereof.
21. A method of treating or preventing Sjogren's anosmia, xerostomia, xerophthalmia, or lacrimal disorder in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor.
22. The method of claim 21, wherein the phosphodiesterase (PDE) inhibitor is a phosphodiesterase (PDE) 4 inhibitor.
23. The method of claim 21, wherein the phosphodiesterase (PDE) inhibitor is selected from 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF104), 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159), derivative, or salts thereof to the subject in need thereof.
24. The method of claim 21, wherein xerostomia, xerophthalmia, or lacrimal disorder is chronic.
25. The method of claim 21, wherein administration is through the mouth and/or nose by inhalation or topical application.

26. The method of claim 21, wherein the subject is diagnosed with cystic fibrosis.
27. The method of claim 21, wherein the subject is not diagnosed with cystic fibrosis.
28. The method of claim 21, wherein the subject is diagnosed with pancreatic sufficiency.
29. The method of claim 21, wherein the subject is diagnosed with moderately elevated sweat chloride and the moderately elevated sweat chloride at a concentration between 30 to 60 millimoles per liter.
30. The method of claim 21, wherein a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and no mutation was identified in the sample, thereby providing a subject diagnosed without a known cystic fibrosis transmembrane conductance regulator mutation.
31. The method of claim 21, wherein a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and a mutation was identified on a single allele in the sample, thereby providing a subject diagnosed with a heterozygous cystic fibrosis transmembrane conductance regulator mutation.
32. The method of claim 31 wherein the heterozygous cystic fibrosis transmembrane conductance regulator mutation is a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.
33. The method of claim 32 wherein the heterozygous cystic fibrosis transmembrane conductance regulator mutation is N1303K, W1282X, G551D, or G542X.
34. The method of claim 31, wherein a sample of the subject was tested for presence of a known cystic fibrosis transmembrane conductance regulator mutation and the mutation was

identified on both alleles in the sample, thereby providing a subject diagnosed with homozygous cystic fibrosis transmembrane conductance regulator mutations.

35. The method of claim 34 wherein the cystic fibrosis transmembrane conductance regulator mutation involves a F508del mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene.

36. The method of claim 34 wherein the cystic fibrosis transmembrane conductance regulator mutation involves an N1303K, W1282X, G551D, or G542X mutation.

37. The method of claim 21 wherein the phosphodiesterase (PDE) inhibitor is administered in combination with a cystic fibrosis drug.

38. The method of claim 37, wherein the cystic fibrosis drug is CFTR activator or modulator.

39. The method of claim 37, wherein the cystic fibrosis drug is elexacaftor, ivacaftor, tezacaftor, or combination thereof.

40. The method of claim 37, wherein the cystic fibrosis drug is a combination of ivacaftor and tezacaftor or is a combination of ivacaftor and lumacaftor.

41. The method of claim 37, wherein the cystic fibrosis drug is a combination of elexacaftor, ivacaftor, and tezacaftor.

42. The method of claim 21 wherein the phosphodiesterase (PDE) inhibitor is administered in combination with another active agent such as a bronchodilator, corticosteroid, antimuscarinic, antibiotic, or combinations thereof.

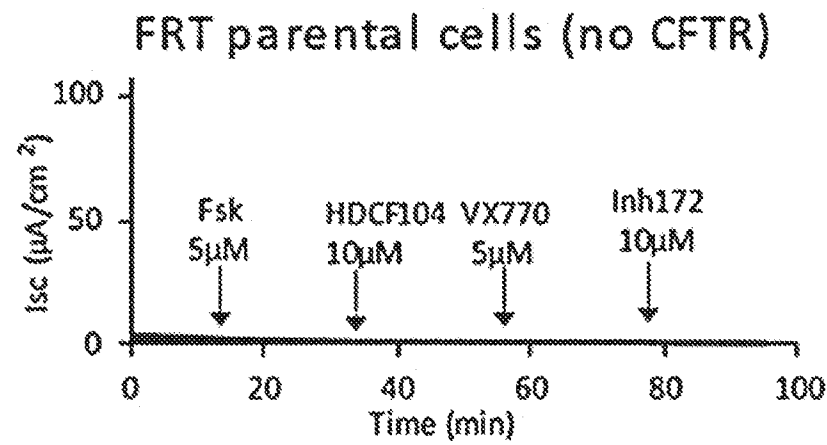


FIG. 1

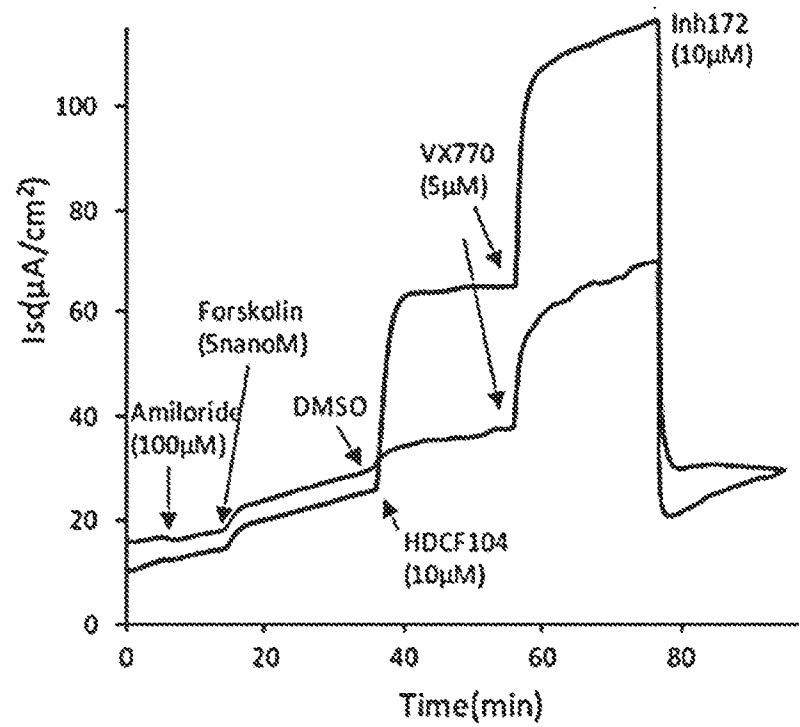


FIG. 1B

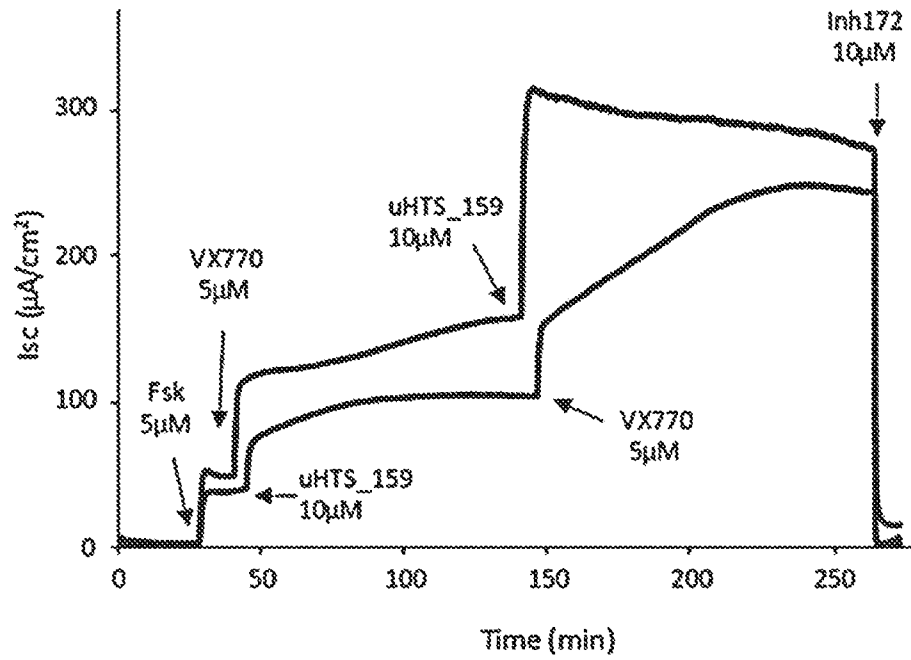


FIG. 2A

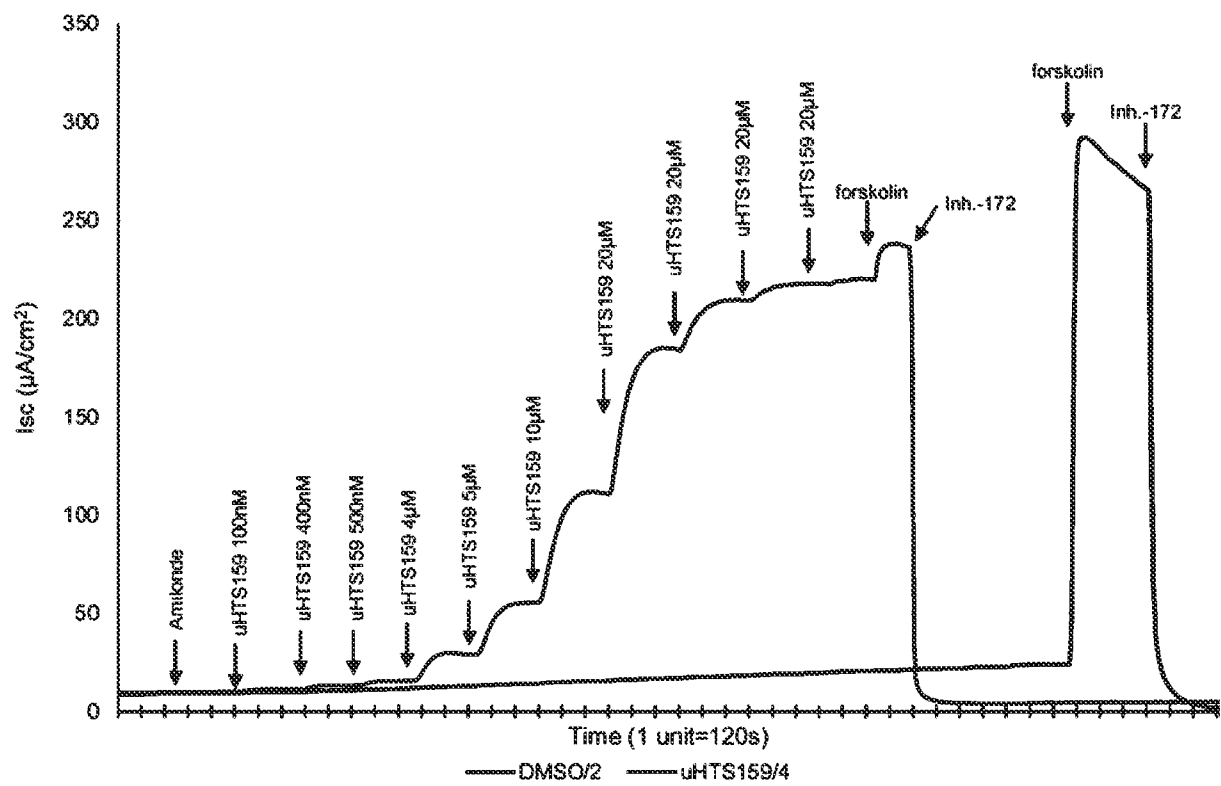


FIG. 2B

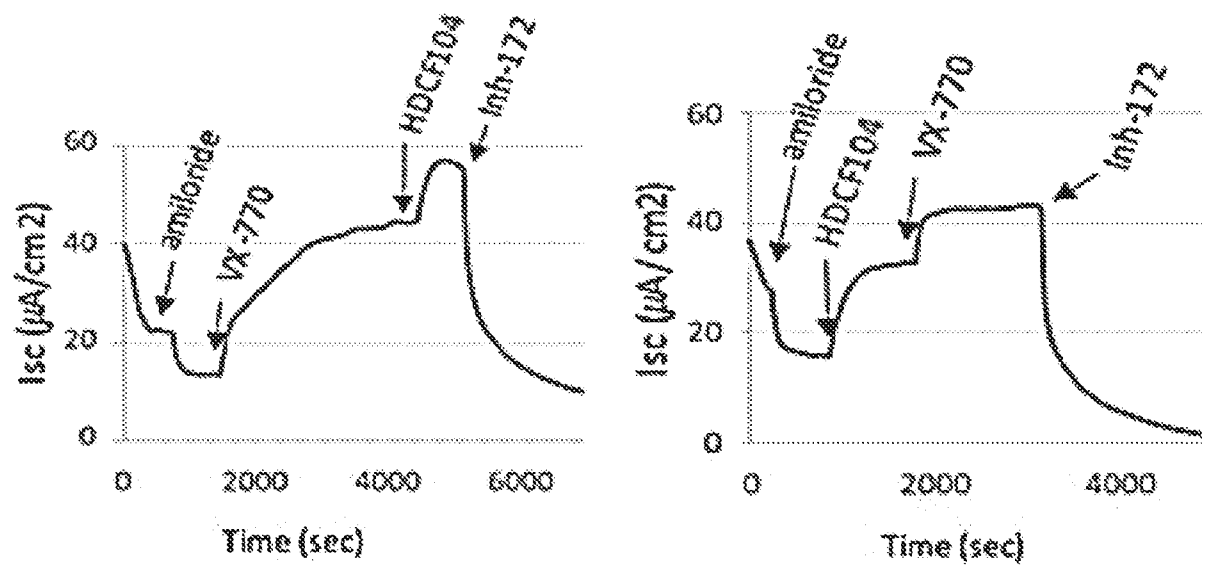


FIG. 3

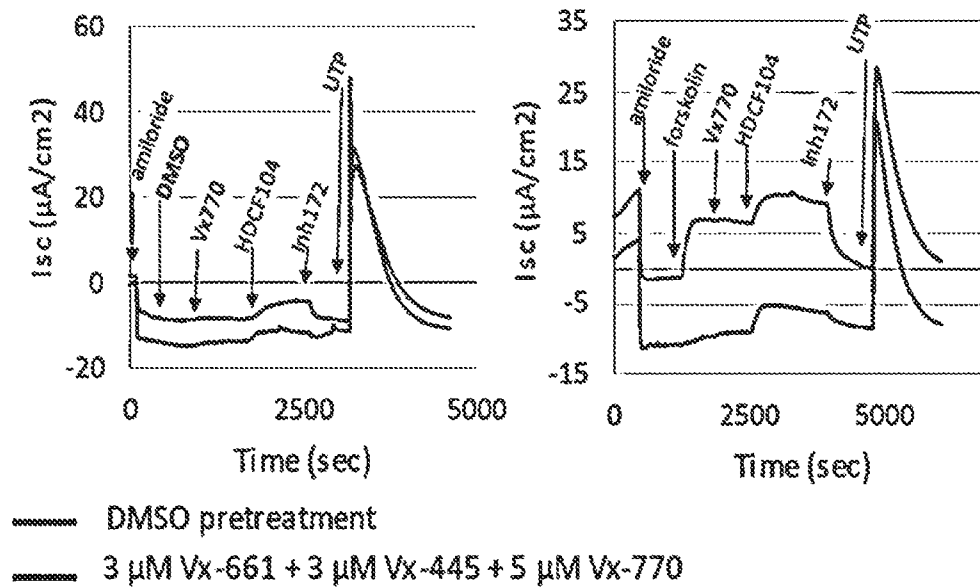


FIG. 4A

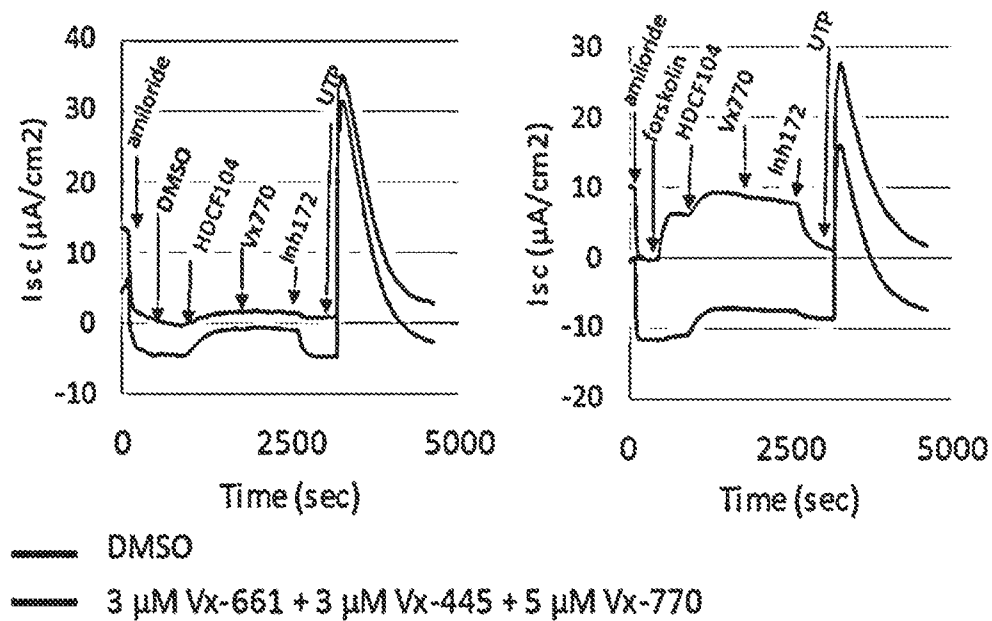


FIG. 4B

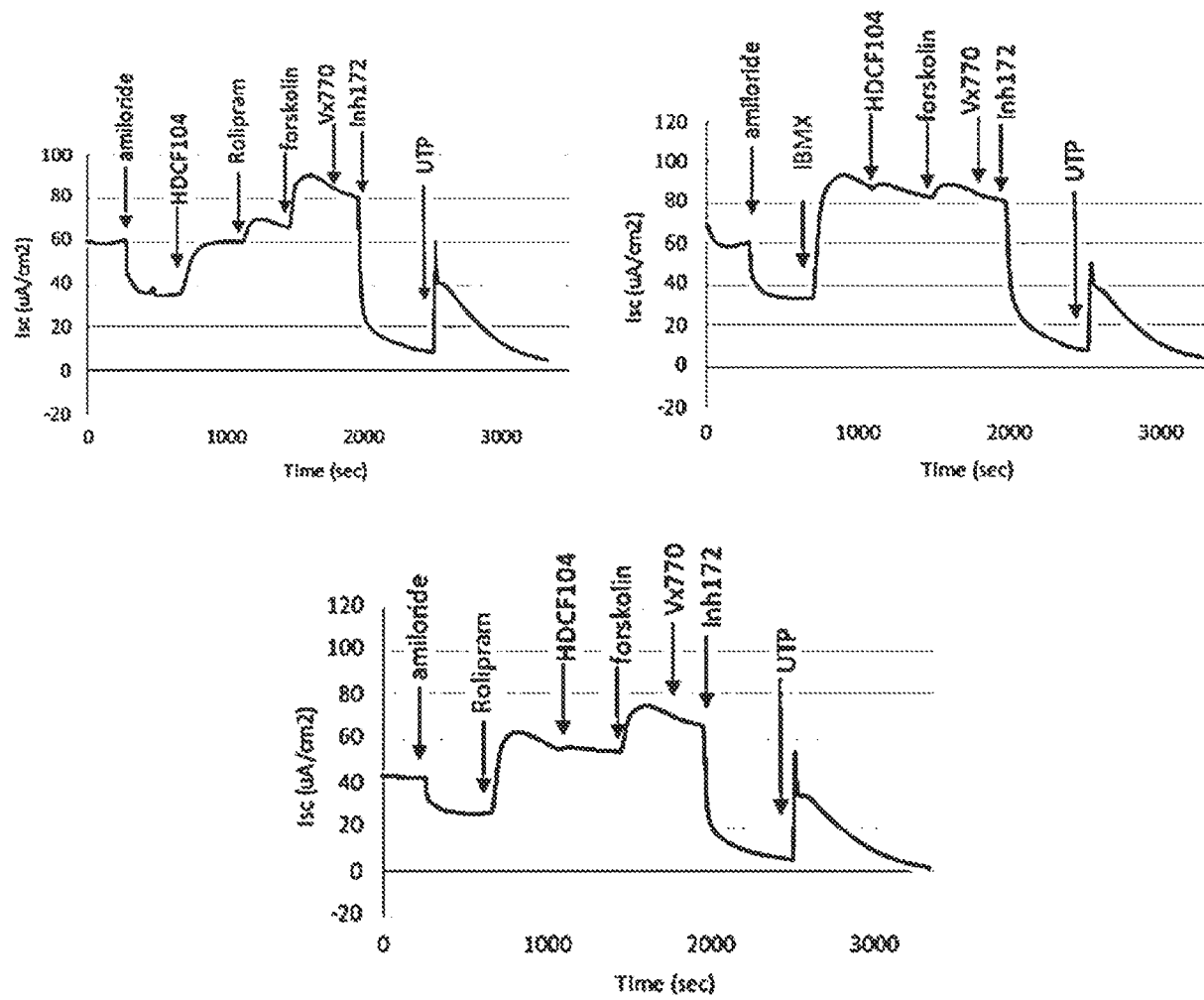


FIG. 5A

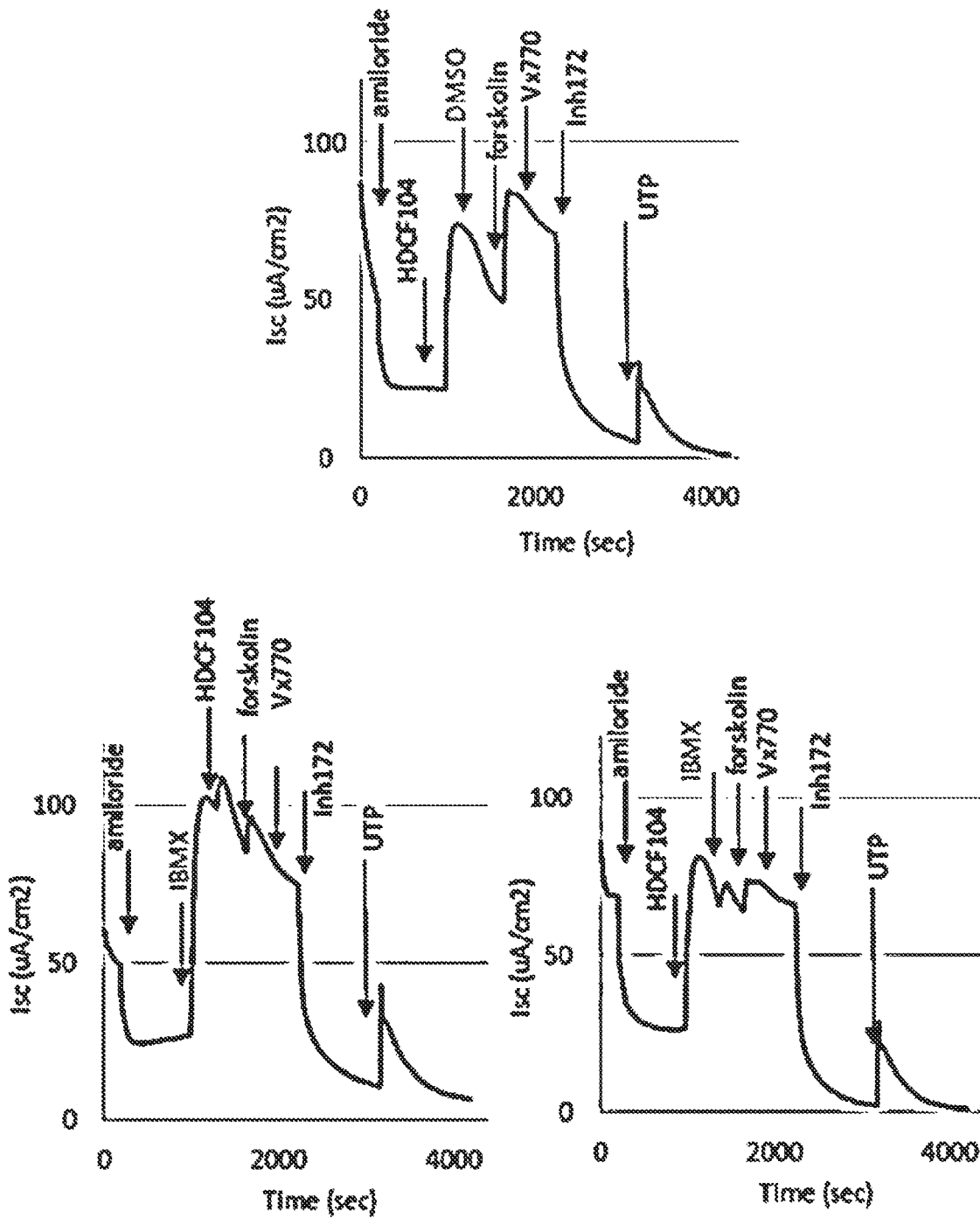


FIG. 5B

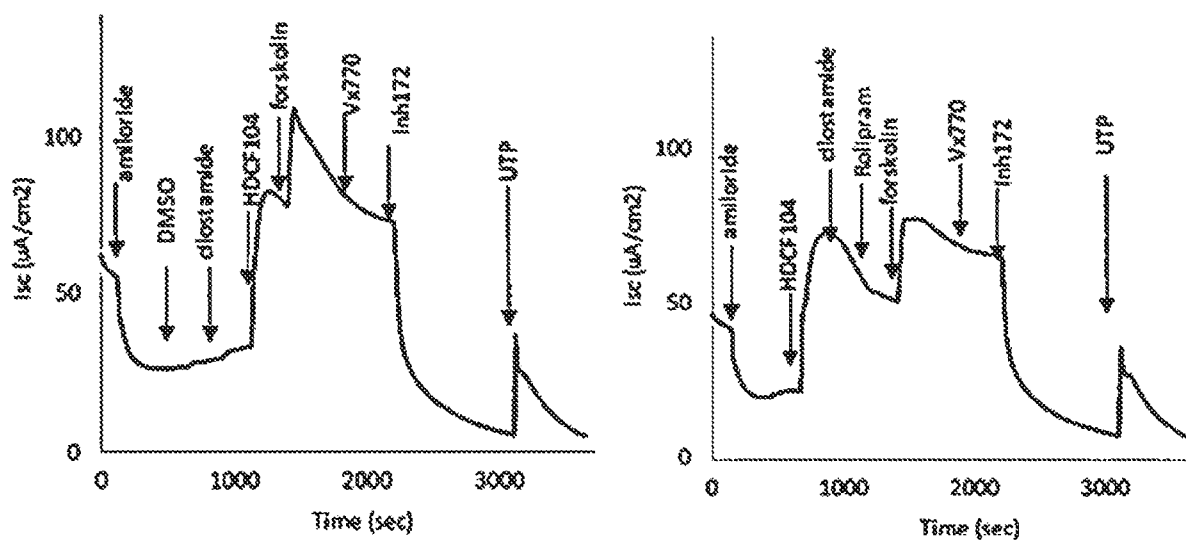


FIG. 5C

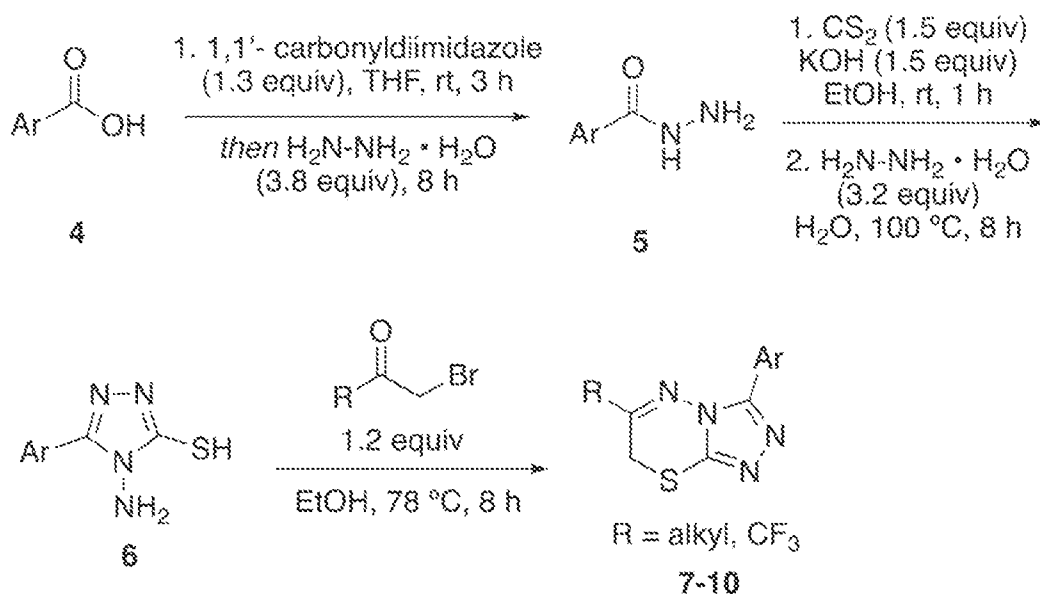
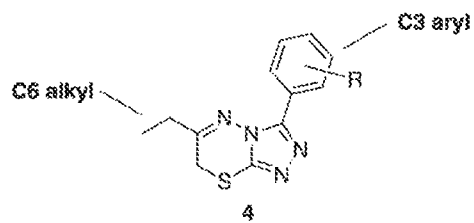


FIG. 6A



C3 Aryl Substituent % Improvement vs. VX-770

	107.1
	118.7
	149.1
	162.4
	186.6
	129.1
	256.9
	118.0

FIG. 6B

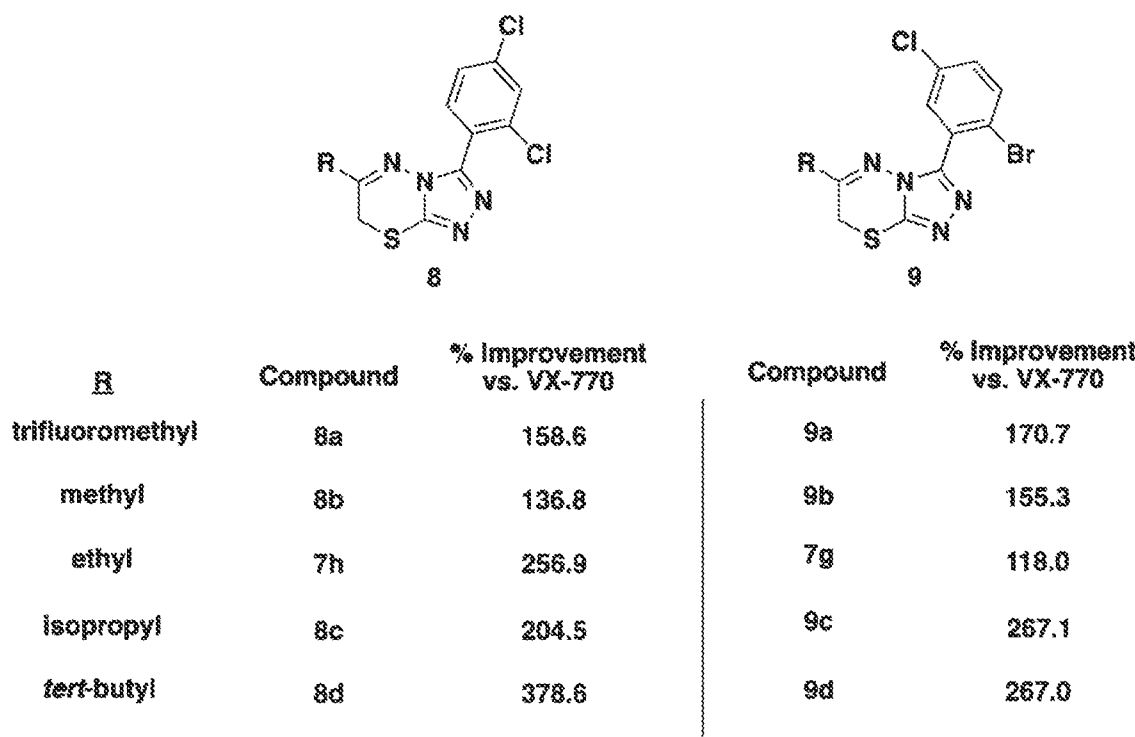
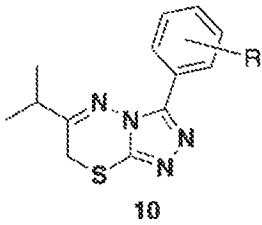


FIG. 6C



Entry	Compound	Aryl Substituent	cLogP	% Improvement vs. VX-770
1	9c		5.26	267.1
2	10a		4.59	231.9
3	10b		3.59	236.7
4	10c		3.37	105.6
5	10d		3.63	166.3
6	10e		3.37	195.6

FIG. 6D

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US2024/020422

A. CLASSIFICATION OF SUBJECT MATTERIPC: **A61K 31/549** (2024.01); **A61K 45/06** (2024.01); **A61P 11/02** (2024.01); **A61P 27/02** (2024.01)CPC: **A61K 31/549**; **A61K 45/06**; **A61P 11/02**; **A61P 27/02**; A61K 2300/00

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	WO 2022/026863 A2 (EMORY UNIVERSITY et al.) 03 February 2022 (03.02.2022) entire document	1-3, 5, 6, 8, 11-20, 21a, 21b, 22a, 22b
Y	entire document	4, 9, 10
Y	US 2017/0319558 A1 (RHIZEN PHARMACEUTICALS SA) 09 November 2017 (09.11.2017) entire document	4
Y	US 2015/0346185 A1 (INSTITUT NATIONAL DE LA SANTE ET DE LA RECHERCHE MEDICALE (INSERM) et al.) 03 December 2015 (03.12.2015) entire document	9, 10



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

28 May 2024 (28.05.2024)

Date of mailing of the international search report

25 June 2024 (25.06.2024)

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US
Commissioner for Patents
P.O. Box 1450, Alexandria, VA 22313-1450

Facsimile No. 571-273-8300

Authorized officer

MATOS
TAINA

Telephone No. 571-272-4300

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

Claims 21 and 22 are objected to under PCT Rule 66.2(a)(v) as lacking clarity under PCT Article 6 because the claims are indefinite for the following reason(s):

Regarding claims 21 and 22, the application contains two claim 21s and 22s, which is improper. For the purposes of the Written Opinion proposed, the first appearances of claim 21 and 22 are analyzed as claim 21b and 22b, respectively, and the second appearances of claim 21 and 22 are analyzed as claim 21b and 22b, respectively, as best interpreted.

This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees need to be paid.

Group I+: claims 1-20, 21a, 21b, 22a, 22b, and 23-42 are drawn to methods of treating or preventing in a subject.

The first invention of Group I+ is restricted to a method of treating or preventing rhinosinusitis in a subject with CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor, wherein the phosphodiesterase (PDE) inhibitor is selected from 3-(2-bromo-5-methoxyphenyl)-6-isopropyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (HDCF 104); and a method of treating or preventing. The first named invention has been selected based on the guidance set forth in section 10.54 of the PCT International Search and Preliminary Examination Guidelines. Specifically, the first named invention was selected based on the first listed element for each of the variables presented in the claims (disease state, PDE inhibitor – claim 1). It is believed that claims 1-6, 8-20, 21a, 21b, 22a, and 22b read on this first named invention and thus these claims will be searched without fee to the extent that they read on the above embodiment.

Applicant is invited to elect additional formula(e) for each additional compound to be searched in a specific combination by paying an additional fee for each set of election. Each additional elected formula(e) requires the selection of a single definition for each compound variable. An exemplary election would be a method of treating or preventing rhinosinusitis in a subject with CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor, wherein the phosphodiesterase (PDE) inhibitor is selected from 6-(3,4-dimethoxyphenyl)-3-ethyl-7H-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazine (uHTS-159); and a method of treating or preventing. Additional formula(e) will be searched upon the payment of additional fees. Applicants must specify the claims that read on any additional elected inventions. Applicants must further indicate, if applicable, the claims which read on the first named invention if different than what was indicated above for this group. Failure to clearly identify how any paid additional invention fees are to be applied to the “+” group(s) will result in only the first claimed invention to be searched/examined.

The inventions listed in Groups I+ do not relate to a single general inventive concept under PCT Rule 13.1, because under PCT Rule 13.2 they lack the same or corresponding special technical features for the following reasons:

The Groups I+ formulae do not share a significant structural element requiring the selection of alternatives for the disease state and PDE inhibitor, and accordingly these groups lack unity a priori.

Additionally, even if Groups I+ were considered to share the technical features of a method of treating or preventing rhinosinusitis, chronic obstructive pulmonary disease, or bronchiectasis in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor; and a method of treating or preventing Sjogren's anosmia, xerostomia, xerophthalmia, or lacrimal disorder in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor, these shared technical features do not represent a contribution over the prior art as disclosed by WO 2022/026863 A2 to Emory University et al.

WO 2022/026863 A2 to Emory University et al. teach a method of treating or preventing rhinosinusitis, chronic obstructive pulmonary disease, or bronchiectasis in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor (Claim 6, method of treating respiratory distress comprising administering to a subject in need thereof an effective amount of a compound of formula I; Claim

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

7, the subject is diagnosed with cystic fibrosis, chronic obstructive pulmonary disease; Pg. 23, Ln. 30, CFTR modulator which is PDE4 inhibitor); and a method of treating or preventing Sjogren's anosmia, xerostomia, xerophthalmia, or lacrimal disorder in a subject with or without CF comprising administering an effective amount of a phosphodiesterase (PDE) inhibitor (Claim 6, method of treating respiratory distress comprising administering to a subject in need thereof an effective amount of a compound of formula I; Claim 7, the subject is diagnosed with cystic fibrosis, chronic obstructive pulmonary disease; Pg. 23, Ln. 30, CFTR modulator which is PDE4 inhibitor; Pg. 22, Lns. 15-16, dry-eye disease, or Sjogren's disease).

The inventions listed in Groups I+ therefore lack unity under Rule 13 because they do not share a same or corresponding special technical feature.

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.: **1-6, 8-20, 21a, 21b, 22a, 22b**

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- ☐ No protest accompanied the payment of additional search fees.