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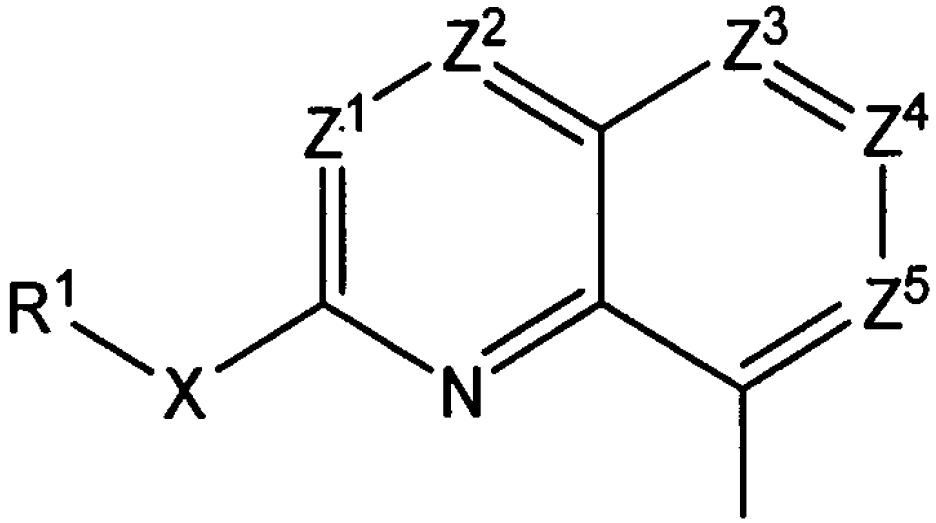
(54) Title: 8-SUBSTITUTED QUINOLINES AND RELATED ANALOGS AS SIRTUIN MODULATORS



(I)

(57) Abrégé/Abstract:
Provided herein are 8-substituted quinolines and related analogues as sirtuin-modulating compounds of Structural Formula (I) and methods of use thereof. The sirtuin-modulating compounds may be used for increasing the lifespan of a cell, and treating and/or preventing a wide variety of diseases and disorders including, for example, diseases or disorders related to aging or stress, diabetes, obesity, neurodegenerative diseases, cardiovascular disease, blood clotting disorders, inflammation, cancer, and/or flushing as well as diseases or disorders that would benefit from increased mitochondrial activity. Also provided are compositions comprising a sirtuin-modulating compound in combination with another therapeutic agent.

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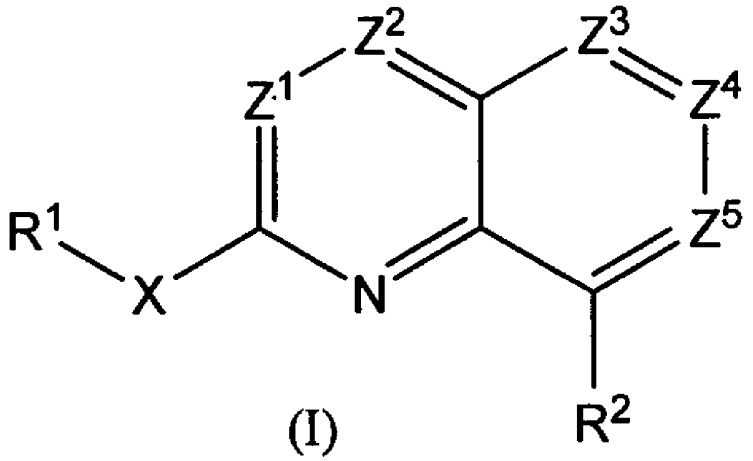
(54) Title: 8-SUBSTITUTED QUINOLINES AND RELATED ANALOGS AS SIRTUIN MODULATORS



(I)

(57) Abstract: Provided herein are 8-substituted quinolines and related analogues as sirtuin-modulating compounds of Structural Formula (I) and methods of use thereof. The sirtuin-modulating compounds may be used for increasing the lifespan of a cell, and treating and/or preventing a wide variety of diseases and disorders including, for example, diseases or disorders related to aging or stress, diabetes, obesity, neurodegenerative diseases, cardiovascular disease, blood clotting disorders, inflammation, cancer, and/or flushing as well as diseases or disorders that would benefit from increased mitochondrial activity. Also provided are compositions comprising a sirtuin-modulating compound in combination with another therapeutic agent.

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8-SUBSTITUTED QUINOLINES AND RELATED ANALOGS AS SIRTUIN MODULATORS

REFERENCE TO RELATED APPLICATION

This application claims the benefit of U.S. Provisional Application No. 61/156,749, filed March 2, 2009 , the disclosure of which is incorporated herein by reference thereto.

BACKGROUND

The Silent Information Regulator (SIR) family of genes represents a highly conserved group of genes present in the genomes of organisms ranging from archaebacteria to a eukaryotes. The encoded SIR proteins are involved in diverse processes from regulation of gene silencing to DNA repair. The proteins encoded by members of the SIR gene family show high sequence conservation in a 250 amino acid core domain. A well-characterized gene in this family is S. cerevisiae SIR2, which is involved in silencing HM loci that contain information specifying yeast mating type, telomere position effects and cell aging. The yeast Sir2 protein belongs to a family of histone deacetylases. The Sir2 homolog, CobB, in Salmonella typhimurium, functions as an NAD (nicotinamide adenine dinucleotide)-dependent ADP-ribosyl transferase.

The Sir2 protein is a class III deacetylase which uses NAD as a cosubstrate. Unlike other deacetylases, many of which are involved in gene silencing, Sir2 is insensitive to class I and II histone deacetylase inhibitors like trichostatin A (TSA).

Deacetylation of acetyl-lysine by Sir2 is tightly coupled to NAD hydrolysis, producing nicotinamide and a novel acetyl-ADP ribose compound. The NAD-dependent deacetylase activity of Sir2 is essential for its functions which can connect its biological role with cellular metabolism in yeast. Mammalian Sir2 homologs have NAD-dependent histone deacetylase activity.

Biochemical studies have shown that Sir2 can readily deacetylate the amino-terminal tails of histones H3 and H4, resulting in the formation of 1-O-acetyl-ADP-ribose and nicotinamide. Strains with additional copies of SIR2 display increased rDNA silencing and a 30% longer life span. It has recently been shown that

additional copies of the C. elegans SIR2 homolog, sir-2.1, and the D. melanogaster dSir2 gene greatly extend life span in those organisms. This implies that the SIR2-dependent regulatory pathway for aging arose early in evolution and has been well conserved. Today, Sir2 genes are believed to have evolved to enhance an organism's health and stress resistance to increase its chance of surviving adversity.

In humans, there are seven Sir2-like genes (SIRT1-SIRT7) that share the conserved catalytic domain of Sir2. SIRT1 is a nuclear protein with the highest degree of sequence similarity to Sir2. SIRT1 regulates multiple cellular targets by deacetylation including the tumor suppressor p53, the cellular signaling factor NF-kB, and the FOXO transcription factor.

SIRT3 is a homolog of SIRT1 that is conserved in prokaryotes and eukaryotes. The SIRT3 protein is targeted to the mitochondrial cristae by a unique domain located at the N-terminus. SIRT3 has NAD+-dependent protein deacetylase activity and is ubiquitously expressed, particularly in metabolically active tissues. Upon transfer to the mitochondria, SIRT3 is believed to be cleaved into a smaller, active form by a mitochondrial matrix processing peptidase (MPP).

Caloric restriction has been known for over 70 years to improve the health and extend the lifespan of mammals. Yeast life span, like that of metazoans, is also extended by interventions that resemble caloric restriction, such as low glucose. The discovery that both yeast and flies lacking the SIR2 gene do not live longer when calorically restricted provides evidence that SIR2 genes mediate the beneficial health effects of a restricted calorie diet. Moreover, mutations that reduce the activity of the yeast glucose-responsive cAMP (adenosine 3',5'-monophosphate)-dependent (PKA) pathway extend life span in wild type cells but not in mutant sir2 strains, demonstrating that SIR2 is likely to be a key downstream component of the caloric restriction pathway.

SUMMARY

Provided herein are novel sirtuin-modulating compounds and methods of use thereof.

In one aspect, the invention provides sirtuin-modulating compounds of Structural Formulas (I) to (VIII) as are described in detail below.

In another aspect, the invention provides methods for using sirtuin-modulating compounds, or compositions comprising sirtuin-modulating compounds. In certain embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for a variety of therapeutic applications including, for example, increasing the lifespan of a cell, and treating and/or preventing a wide variety of diseases and disorders including, for example, diseases or disorders related to aging or stress, diabetes, obesity, neurodegenerative diseases, chemotherapeutic induced neuropathy, neuropathy associated with an ischemic event, ocular diseases and/or disorders, cardiovascular disease, blood clotting disorders, inflammation, and/or flushing, etc. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used for treating a disease or disorder in a subject that would benefit from increased mitochondrial activity, for enhancing muscle performance, for increasing muscle ATP levels, or for treating or preventing muscle tissue damage associated with hypoxia or ischemia. In other embodiments, sirtuin-modulating compounds that decrease the level and/or activity of a sirtuin protein may be used for a variety of therapeutic applications including, for example, increasing cellular sensitivity to stress, increasing apoptosis, treatment of cancer, stimulation of appetite, and/or stimulation of weight gain, etc. As described further below, the methods comprise administering to a subject in need thereof a pharmaceutically effective amount of a sirtuin-modulating compound.

In certain aspects, the sirtuin-modulating compounds may be administered alone or in combination with other compounds, including other sirtuin-modulating compounds, or other therapeutic agents.

DETAILED DESCRIPTION

1. Definitions

As used herein, the following terms and phrases shall have the meanings set forth below. Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood to one of ordinary skill in the art.

The term “agent” is used herein to denote a chemical compound, a mixture of chemical compounds, a biological macromolecule (such as a nucleic acid, an antibody, a protein or portion thereof, e.g., a peptide), or an extract made from biological materials such as bacteria, plants, fungi, or animal (particularly

mammalian) cells or tissues. The activity of such agents may render it suitable as a "therapeutic agent" which is a biologically, physiologically, or pharmacologically active substance (or substances) that acts locally or systemically in a subject.

The term "bioavailable" when referring to a compound is art-recognized and refers to a form of a compound that allows for it, or a portion of the amount of compound administered, to be absorbed by, incorporated into, or otherwise physiologically available to a subject or patient to whom it is administered.

"Biologically active portion of a sirtuin" refers to a portion of a sirtuin protein having a biological activity, such as the ability to deacetylate. Biologically active portions of a sirtuin may comprise the core domain of sirtuins. Biologically active portions of SIRT1 having GenBank Accession No. NP_036370 that encompass the NAD+ binding domain and the substrate binding domain, for example, may include without limitation, amino acids 62-293 of GenBank Accession No. NP_036370, which are encoded by nucleotides 237 to 932 of GenBank Accession No. NM_012238. Therefore, this region is sometimes referred to as the core domain. Other biologically active portions of SIRT1, also sometimes referred to as core domains, include about amino acids 261 to 447 of GenBank Accession No. NP_036370, which are encoded by nucleotides 834 to 1394 of GenBank Accession No. NM_012238; about amino acids 242 to 493 of GenBank Accession No. NP_036370, which are encoded by nucleotides 777 to 1532 of GenBank Accession No. NM_012238; or about amino acids 254 to 495 of GenBank Accession No. NP_036370, which are encoded by nucleotides 813 to 1538 of GenBank Accession No. NM_012238.

The term "companion animals" refers to cats and dogs. As used herein, the term "dog(s)" denotes any member of the species Canis familiaris, of which there are a large number of different breeds. The term "cat(s)" refers to a feline animal including domestic cats and other members of the family Felidae, genus Felis.

"Diabetes" refers to high blood sugar or ketoacidosis, as well as chronic, general metabolic abnormalities arising from a prolonged high blood sugar status or a decrease in glucose tolerance. "Diabetes" encompasses both the type I and type II (Non Insulin Dependent Diabetes Mellitus or NIDDM) forms of the disease. The risk factors for diabetes include the following factors: waistline of more than 40

inches for men or 35 inches for women, blood pressure of 130/85 mmHg or higher, triglycerides above 150 mg/dl, fasting blood glucose greater than 100 mg/dl or high-density lipoprotein of less than 40 mg/dl in men or 50 mg/dl in women.

The term "ED_{50}" refers to the art-recognized measure of effective dose In certain embodiments, ED_{50} means the dose of a drug which produces 50% of its maximum response or effect, or alternatively, the dose which produces a pre-determined response in 50% of test subjects or preparations. The term "LD_{50}" refers to the art-recognized measure of lethal dose. In certain embodiments, LD_{50} means the dose of a drug which is lethal in 50% of test subjects. The term "therapeutic index" is an art-recognized term which refers to the therapeutic index of a drug, defined as LD_{50}/ED_{50}.

The term "hyperinsulinemia" refers to a state in an individual in which the level of insulin in the blood is higher than normal.

The term "insulin resistance" refers to a state in which a normal amount of insulin produces a subnormal biologic response relative to the biological response in a subject that does not have insulin resistance.

An "insulin resistance disorder," as discussed herein, refers to any disease or condition that is caused by or contributed to by insulin resistance. Examples include: diabetes, obesity, metabolic syndrome, insulin-resistance syndromes, syndrome X, insulin resistance, high blood pressure, hypertension, high blood cholesterol, dyslipidemia, hyperlipidemia, atherosclerotic disease including stroke, coronary artery disease or myocardial infarction, hyperglycemia, hyperinsulinemia and/or hyperproinsulinemia, impaired glucose tolerance, delayed insulin release, diabetic complications, including coronary heart disease, angina pectoris, congestive heart failure, stroke, cognitive functions in dementia, retinopathy, peripheral neuropathy, nephropathy, glomerulonephritis, glomerulosclerosis, nephrotic syndrome, hypertensive nephrosclerosis some types of cancer (such as endometrial, breast, prostate, and colon), complications of pregnancy, poor female reproductive health (such as menstrual irregularities, infertility, irregular ovulation, polycystic ovarian syndrome (PCOS)), lipodystrophy, cholesterol related disorders, such as gallstones,

cholecystitis and cholelithiasis, gout, obstructive sleep apnea and respiratory problems, osteoarthritis, and bone loss, e.g. osteoporosis in particular.

The term "livestock animals" refers to domesticated quadrupeds, which includes those being raised for meat and various byproducts, e.g., a bovine animal including cattle and other members of the genus Bos, a porcine animal including domestic swine and other members of the genus Sus, an ovine animal including sheep and other members of the genus Ovis, domestic goats and other members of the genus Capra; domesticated quadrupeds being raised for specialized tasks such as use as a beast of burden, e.g., an equine animal including domestic horses and other members of the family Equidae, genus Equus.

The term "mammal" is known in the art, and exemplary mammals include humans, primates, livestock animals (including bovines, porcines, etc.), companion animals (e.g., canines, felines, etc.) and rodents (e.g., mice and rats).

"Obese" individuals or individuals suffering from obesity are generally individuals having a body mass index (BMI) of at least 25 or greater. Obesity may or may not be associated with insulin resistance.

The terms "parenteral administration" and "administered parenterally" are art-recognized and refer to modes of administration other than enteral and topical administration, usually by injection, and includes, without limitation, intravenous, intramuscular, intraarterial, intrathecal, intracapsular, intraorbital, intracardiac, intradermal, intraperitoneal, transtracheal, subcutaneous, subcuticular, intra-articular, subcapsular, subarachnoid, intraspinal, and intrasternal injection and infusion.

A "patient", "subject", "individual" or "host" refers to either a human or a non-human animal.

The term "pharmaceutically acceptable carrier" is art-recognized and refers to a pharmaceutically-acceptable material, composition or vehicle, such as a liquid or solid filler, diluent, excipient, solvent or encapsulating material, involved in carrying or transporting any subject composition or component thereof. Each carrier must be "acceptable" in the sense of being compatible with the subject composition and its components and not injurious to the patient. Some examples of materials

which may serve as pharmaceutically acceptable carriers include: (1) sugars, such as lactose, glucose and sucrose; (2) starches, such as corn starch and potato starch; (3) cellulose, and its derivatives, such as sodium carboxymethyl cellulose, ethyl cellulose and cellulose acetate; (4) powdered tragacanth; (5) malt; (6) gelatin; (7) talc; (8) excipients, such as cocoa butter and suppository waxes; (9) oils, such as peanut oil, cottonseed oil, safflower oil, sesame oil, olive oil, corn oil and soybean oil; (10) glycols, such as propylene glycol; (11) polyols, such as glycerin, sorbitol, mannitol and polyethylene glycol; (12) esters, such as ethyl oleate and ethyl laurate; (13) agar; (14) buffering agents, such as magnesium hydroxide and aluminum hydroxide; (15) alginic acid; (16) pyrogen-free water; (17) isotonic saline; (18) Ringer's solution; (19) ethyl alcohol; (20) phosphate buffer solutions; and (21) other non-toxic compatible substances employed in pharmaceutical formulations.

The term "preventing" is art-recognized, and when used in relation to a condition, such as a local recurrence (e.g., pain), a disease such as cancer, a syndrome complex such as heart failure or any other medical condition, is well understood in the art, and includes administration of a composition which reduces the frequency of, or delays the onset of, symptoms of a medical condition in a subject relative to a subject which does not receive the composition. Thus, prevention of cancer includes, for example, reducing the number of detectable cancerous growths in a population of patients receiving a prophylactic treatment relative to an untreated control population, and/or delaying the appearance of detectable cancerous growths in a treated population versus an untreated control population, e.g., by a statistically and/or clinically significant amount. Prevention of an infection includes, for example, reducing the number of diagnoses of the infection in a treated population versus an untreated control population, and/or delaying the onset of symptoms of the infection in a treated population versus an untreated control population. Prevention of pain includes, for example, reducing the magnitude of, or alternatively delaying, pain sensations experienced by subjects in a treated population versus an untreated control population.

The term "prophylactic" or "therapeutic" treatment is art-recognized and refers to administration of a drug to a host. If it is administered prior to clinical manifestation of the unwanted condition (e.g., disease or other unwanted state of the

host animal) then the treatment is prophylactic, i.e., it protects the host against developing the unwanted condition, whereas if administered after manifestation of the unwanted condition, the treatment is therapeutic (i.e., it is intended to diminish, ameliorate or maintain the existing unwanted condition or side effects therefrom).

The term "pyrogen-free", with reference to a composition, refers to a composition that does not contain a pyrogen in an amount that would lead to an adverse effect (e.g., irritation, fever, inflammation, diarrhea, respiratory distress, endotoxic shock, etc.) in a subject to which the composition has been administered. For example, the term is meant to encompass compositions that are free of, or substantially free of, an endotoxin such as, for example, a lipopolysaccharide (LPS).

"Replicative lifespan" of a cell refers to the number of daughter cells produced by an individual "mother cell." "Chronological aging" or "chronological lifespan," on the other hand, refers to the length of time a population of non-dividing cells remains viable when deprived of nutrients. "Increasing the lifespan of a cell" or "extending the lifespan of a cell," as applied to cells or organisms, refers to increasing the number of daughter cells produced by one cell; increasing the ability of cells or organisms to cope with stresses and combat damage, e.g., to DNA, proteins; and/or increasing the ability of cells or organisms to survive and exist in a living state for longer under a particular condition, e.g., stress (for example, heatshock, osmotic stress, high energy radiation, chemically-induced stress, DNA damage, inadequate salt level, inadequate nitrogen level, or inadequate nutrient level). Lifespan can be increased by at least about 10%, 20%, 30%, 40%, 50%, 60% or between 20% and 70%, 30% and 60%, 40% and 60% or more using methods described herein.

"Sirtuin-activating compound" refers to a compound that increases the level of a sirtuin protein and/or increases at least one activity of a sirtuin protein. In an exemplary embodiment, a sirtuin-activating compound may increase at least one biological activity of a sirtuin protein by at least about 10%, 25%, 50%, 75%, 100%, or more. Exemplary biological activities of sirtuin proteins include deacetylation, e.g., of histones and p53; extending lifespan; increasing genomic

stability; silencing transcription; and controlling the segregation of oxidized proteins between mother and daughter cells.

"Sirtuin protein" refers to a member of the sirtuin deacetylase protein family, or preferably to the sir2 family, which include yeast Sir2 (GenBank Accession No. P53685), *C. elegans* Sir-2.1 (GenBank Accession No. NP_501912), and human SIRT1 (GenBank Accession No. NM_012238 and NP_036370 (or AF083106)) and SIRT2 (GenBank Accession No. NM_012237, NM_030593, NP_036369, NP_085096, and AF083107) proteins. Other family members include the four additional yeast Sir2-like genes termed "*HST* genes" (homologues of Sir two) HST1, HST2, HST3 and HST4, and the five other human homologues hSIRT3, hSIRT4, hSIRT5, hSIRT6 and hSIRT7 (Brachmann et al. (1995) Genes Dev. 9:2888 and Frye et al. (1999) BBRC 260:273). Preferred sirtuins are those that share more similarities with SIRT1, i.e., hSIRT1, and/or Sir2 than with SIRT2, such as those members having at least part of the N-terminal sequence present in SIRT1 and absent in SIRT2 such as SIRT3 has.

"SIRT1 protein" refers to a member of the sir2 family of sirtuin deacetylases. In one embodiment, a SIRT1 protein includes yeast Sir2 (GenBank Accession No. P53685), C. elegans Sir-2.1 (GenBank Accession No. NP_501912), human SIRT1 (GenBank Accession No. NM_012238 or NP_036370 (or AF083106)), and equivalents and fragments thereof. In another embodiment, a SIRT1 protein includes a polypeptide comprising a sequence consisting of, or consisting essentially of, the amino acid sequence set forth in GenBank Accession Nos. NP_036370, NP_501912, NP_085096, NP_036369, or P53685. SIRT1 proteins include polypeptides comprising all or a portion of the amino acid sequence set forth in GenBank Accession Nos. NP_036370, NP_501912, NP_085096, NP_036369, or P53685; the amino acid sequence set forth in GenBank Accession Nos. NP_036370, NP_501912, NP_085096, NP_036369, or P53685 with 1 to about 2, 3, 5, 7, 10, 15, 20, 30, 50, 75 or more conservative amino acid substitutions; an amino acid sequence that is at least 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% identical to GenBank Accession Nos. NP_036370, NP_501912, NP_085096, NP_036369, or P53685, and functional fragments thereof. Polypeptides of the invention also include homologs

(e.g., orthologs and paralogs), variants, or fragments, of GenBank Accession Nos. NP_036370, NP_501912, NP_085096, NP_036369, or P53685.

As used herein "SIRT2 protein", "SIRT3 protein", "SIRT4 protein", SIRT 5 protein", "SIRT6 protein", and "SIRT7 protein" refer to other mammalian, e.g. human, sirtuin deacetylase proteins that are homologous to SIRT1 protein, particularly in the approximately 275 amino acid conserved catalytic domain. For example, "SIRT3 protein" refers to a member of the sirtuin deacetylase protein family that is homologous to SIRT1 protein. In one embodiment, a SIRT3 protein includes human SIRT3 (GenBank Accession No. AAH01042, NP_036371, or NP_001017524) and mouse SIRT3 (GenBank Accession No. NP_071878) proteins, and equivalents and fragments thereof. In another embodiment, a SIRT3 protein includes a polypeptide comprising a sequence consisting of, or consisting essentially of, the amino acid sequence set forth in GenBank Accession Nos. AAH01042, NP_036371, NP_001017524, or NP_071878. SIRT3 proteins include polypeptides comprising all or a portion of the amino acid sequence set forth in GenBank Accession AAH01042, NP_036371, NP_001017524, or NP_071878; the amino acid sequence set forth in GenBank Accession Nos. AAH01042, NP_036371, NP_001017524, or NP_071878 with 1 to about 2, 3, 5, 7, 10, 15, 20, 30, 50, 75 or more conservative amino acid substitutions; an amino acid sequence that is at least 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% identical to GenBank Accession Nos. AAH01042, NP_036371, NP_001017524, or NP_071878, and functional fragments thereof. Polypeptides of the invention also include homologs (e.g., orthologs and paralogs), variants, or fragments, of GenBank Accession Nos. AAH01042, NP_036371, NP_001017524, or NP_071878. In one embodiment, a SIRT3 protein includes a fragment of SIRT3 protein that is produced by cleavage with a mitochondrial matrix processing peptidase (MPP) and/or a mitochondrial intermediate peptidase (MIP).

The terms "systemic administration," "administered systemically," "peripheral administration" and "administered peripherally" are art-recognized and refer to the administration of a subject composition, therapeutic or other material other than directly into the central nervous system, such that it enters the patient's system and, thus, is subject to metabolism and other like processes.

The term "tautomer" as used herein is art-regcognized and refers to the formal migration of a hydrogen atom, i.e., proton, accompanied by a swap of a single bond and adjacent double bond. When used herein to describe a compound or genus of compounds, tautomer includes any portion of a compound or the entire compound such as a single substituent of a compound, multiple substiutents of a compound or, for example, the entire compound. For example, the tautomer of a compound that includes a hydroxyl-substituted pyridine ring (A) may also include within the scope of the invention the tautomerized form of the substituted ring (B):

A B

The term “therapeutic agent” is art-recognized and refers to any chemical moiety that is a biologically, physiologically, or pharmacologically active substance that acts locally or systemically in a subject. The term also means any substance intended for use in the diagnosis, cure, mitigation, treatment or prevention of disease or in the enhancement of desirable physical or mental development and/or conditions in an animal or human.

The term “therapeutic effect” is art-recognized and refers to a local or systemic effect in animals, particularly mammals, and more particularly humans caused by a pharmacologically active substance. The phrase “therapeutically-effective amount” means that amount of such a substance that produces some desired local or systemic effect at a reasonable benefit/risk ratio applicable to any treatment. The therapeutically effective amount of such substance will vary depending upon the subject and disease condition being treated, the weight and age of the subject, the severity of the disease condition, the manner of administration and the like, which can readily be determined by one of ordinary skill in the art. For example, certain compositions described herein may be administered in a sufficient amount to produce a desired effect at a reasonable benefit/risk ratio applicable to such treatment.

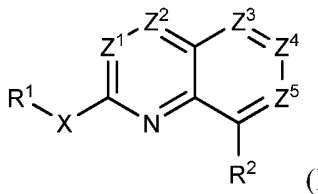
“Treating” a condition or disease refers to curing as well as ameliorating at least one symptom of the condition or disease.

The term "vision impairment" refers to diminished vision, which is often only partially reversible or irreversible upon treatment (e.g., surgery). Particularly severe vision impairment is termed "blindness" or "vision loss", which refers to a complete loss of vision, vision worse than 20/200 that cannot be improved with corrective lenses, or a visual field of less than 20 degrees diameter (10 degrees radius).

2. Sirtuin Modulators

In one aspect, the invention provides novel sirtuin-modulating compounds for treating and/or preventing a wide variety of diseases and disorders including, for example, diseases or disorders related to aging or stress, diabetes, obesity, neurodegenerative diseases, ocular diseases and disorders, cardiovascular disease, blood clotting disorders, inflammation, cancer, and/or flushing, etc. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used for treating a disease or disorder in a subject that would benefit from increased mitochondrial activity, for enhancing muscle performance, for increasing muscle ATP levels, or for treating or preventing muscle tissue damage associated with hypoxia or ischemia. Other compounds disclosed herein may be suitable for use in a pharmaceutical composition and/or one or more methods disclosed herein.

In certain embodiments, sirtuin-modulating compound of the invention are represented by structural formula (I):



(I),

or a salt thereof, wherein:

each of Z^1, Z^2, Z^3, Z^4, and Z^5 are independently selected from N and CR, wherein:

each of Z^3, Z^4 and Z^5 is independently CR, or

at least one of Z^1 or Z^2 is N and no more than two of Z^1-Z^5 are simultaneously N, or

two of Z^3, Z^4 and Z^5 are N and each other of Z^1-Z^5 is independently CR;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl), and a second heterocycle, and when R^2 is phenyl, R^2 is

also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1;

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl.

In certain embodiments, each of Z^3, Z^4 and Z^5 is CR.

In certain embodiments, X is -C(O)-NR^6-†. In certain embodiments, where each of Z^3, Z^4 and Z^5 is CR, X is -C(O)-NR^6-†. In particular embodiments, X is -C(O)-NH-†.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy;

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, wherein the saturated heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated

heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl), and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -C(=O)-NH-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-C(=O)NR^4-†, -NR^4-C(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^{4}R^5-†, -CR^{4}R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†, -C(=O)-NH-CR^{4}R^5-†, -CR^{4}R^5-NH-C(O)-†, -NH-C(=S)-CR^{4}R^5-†, -CR^{4}R^5-C(=S)-NH-†, -NH-S(O)-CR^{4}R^5-†, -CR^{4}R^5-S(O)-NH-†, -NH-S(O)$_2$-CR^{4}R^5-†, -CR^{4}R^5-S(O)$_2$-NH-†, -NH-C(=O)-O-CR^{4}R^5-†, -CR^{4}R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^{4}R^5-†, -NH-C(=O)-CR^{4}R^5-†, and -CR^{4}R^5-NH-C(=O)-O-†; and

each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

In certain embdodiments, each of Z^3, Z^4 and Z^5 is CR in the compound represented by Structural Formula (I). In certain such embodiments, each of Z^1-Z^5 is CR.

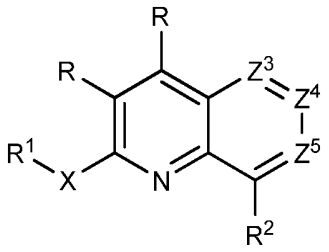
In certain embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl. In particular embodiments, R^1 is selected from optionally

substituted thiazole, pyridine, pyrazine, and phenyl. Suitable values of R^2 include phenyl, pyridyl and benzomorpholine, such as when R^1 has the values indicated previously.

In certain embodiments, X is selected from $-NR^6$-C(=O)-†, $-NR^6$-C(=O)-CR^4R^5-NR^6-†, $-NR^6$-C(=O)-CR^4R^5-†, $-NR^6$-S(=O)$_2$-†, $-NR^6$-S(=O)$_2$-CR^4R^5-†, $-NR^6$-C(=O)-NR^6-†, -C(=O)-NR^6-†, -C(=O)-NR^6-$(CR^4R^5)_{1-3}$-†, $-NR^6$-C(=O)-CR^4R^5-O-†, $-NR^6$-C(=O)-O-†,-CR^4R^5-NR^6-†, $-NR^6$-C(=NR^6)-NR^6-†, $-NR^6$-C(=NR^6)-† and -C(=NR^6)-NR^6-†. In certain embodiments, X is -C(O)-NR^6-†.

In certain embodiments, X is -C(O)-NH-†. In particular embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl and X is –C(=O)-NH-†. In certain embodiments, Z^3, Z^4 and Z^5 is CR and X is -C(O)-NH-†. In particular embodiments, each of Z^1-Z^5 is CR, R^1 is selected from thiazole, pyridine, pyrazine and phenyl, R^2 is selected from phenyl, pyridyl, and benzomorpholine and X is -C(O)-NH-†,

In certain embodiments, sirtuin-modulating compounds of the invention are represented by structural formula (II):



(II),

or a salt thereof, wherein:

each of Z^3, Z^4, and Z^5 are independently selected from N and CR, wherein only one of Z^3, Z^4, and Z^5 is N, wherein:

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N,

C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when X is not -C(=O)-NH-†, R^1 is also optionally substituted with =O and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted

C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, and

X is selected from -NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-†, and when Z^3 or Z^5 is N, X is also selected from: -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†,

wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl.

In certain embodiments, Z^3 or Z^5 is N. In certain embodiments, Z^3 or Z^5 is N and X is selected from: -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when X is not -C(=O)-NH-†, R^1 is also optionally substituted with =O and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy;

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, wherein the saturated heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4

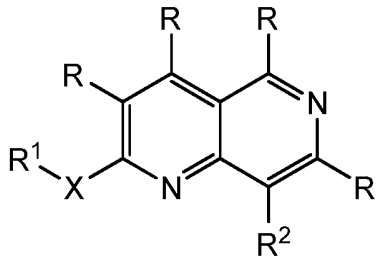
alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -C(=O)-NH-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-C(=O)NR^4-†, -NR^4-C(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^{4}R^5-†, -CR^{4}R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†, -CR^{4}R^5-NH-C(O)-†, -NH-C(=S)-CR^{4}R^5-†, -CR^{4}R^5-C(=S)-NH-†, -NH-S(O)-CR^{4}R^5-†, -CR^{4}R^5-S(O)-NH-†, -NH-S(O)$_2$-CR^{4}R^5-†, -CR^{4}R^5-S(O)$_2$-NH-†, -NH-C(=O)-O-CR^{4}R^5-†, -CR^{4}R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^{4}R^5-†, -NH-C(=O)-CR^{4}R^5-†, and -CR^{4}R^5-NH-C(=O)-O-†; and

each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

In certain embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl. In certain embodiments X is -C(O)-NH-†. In particular embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl and X is –C(=O)-NH-†.

In certain embodiments, sirtuin-modulating compounds of the invention are represented by Structural Formula (III):



(III),

or a salt thereof, wherein:

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O- saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when X is not -C(=O)-NH-†, R^1 is also optionally substituted with =O and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -SO_2-(C_1-C_4 alkyl)-, -O-phenyl, phenyl, and a second heterocycle, and when R^2 is phenyl, R^2 is

also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl and –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NH-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NR^6-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl;

and each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when X is not -C(=O)-NH-†, R^1 is also optionally substituted with =O and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, wherein the saturated heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl; and

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one to two substitutents independently

selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -SO_2-(C_1-C_4 alkyl)-, -O-phenyl, phenyl, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

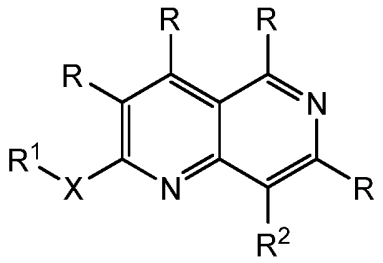
X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -C(=O)-NH-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-C(=O)NR^4-†, -NR^4-C(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^4R^5-†, -CR^4R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†,-CR^4R^5-NH-C(O)-†, -NH-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NH-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NH-S(O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NH-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^4R^5-†, -NH-C(=O)-CR^4R^5-†, and -CR^4R^5-NH-C(=O)-O-†, wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

In certain embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl. In certain embodiments X is -C(O)-NH-†. In particular embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl and X is –C(=O)-NH-†.

In certain embodiments, compounds of the invention are a subset of the compounds of Structural Formula (III) represented by Structural Formula (IV):



(IV),

or a salt thereof, wherein:

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O- saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), and -N($CH_2CH_2OCH_3$)$_2$ and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl and –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl),and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with
-O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, and

X is selected from –NR^6-C(=S)-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†,

-NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† and when R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, X is additionally selected from -C(=S)-NR^6-† and -NR^6-C(=O)-NR^6-†, and when R^1 is optionally substituted cycloalkyl or saturated heterocyclyl X is additionally selected from –NR^6-C(=O)-†, wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy;

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, wherein the saturated

heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

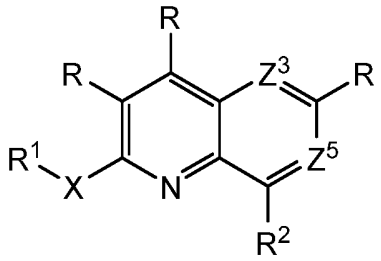
R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl),and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

X is selected from -NH-C(=S)-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^4R^5-†, -CR^4R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†, -CR^4R^5-NH-C(O)-†, -NH-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NH-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NH-S(O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NH-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^4R^5-†, -NH-C(=O)-CR^4R^5-†, and -CR^4R^5-NH-C(=O)-O-†, and when R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, X is additionally selected from -C(=S)-NH-†, -NH-C(=O)NR^4-†, and -NR^4-C(=O)NH-†, and when R^1 is optionally substituted cycloalkyl or saturated heterocyclyl X is additionally selected from -NH-C(=O)-†; and

each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

In certain embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl. In certain embodiments X is -C(O)-NH-†. In particular embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl and X is –C(=O)-NH-†.

In certain embodiments, sirtuin-modulating compounds of the invention are represented by structural formula (V):



(V),

or a salt thereof, wherein:

one of Z^3 or Z^5 is N and the other is CR;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2,

-NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), and -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, , -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-†, wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro,

$-NH_2$, $-NH(C_1-C_4$ alkyl), $-N(C_1-C_4$ alkyl$)_2$, $-NH(CH_2CH_2OCH_3)$, or $-N(CH_2CH_2OCH_3)_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), $S(=O)_2$, and O, wherein the saturated heterocycle is optionally substituted at a carbon atom with -OH, $-C_1-C_4$ alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, $-NH_2$, $-NH(C_1-C_4$ alkyl), $-N(C_1-C_4$ alkyl$)_2$, $-NH(CH_2CH_2OCH_3)$, or $-N(CH_2CH_2OCH_3)_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

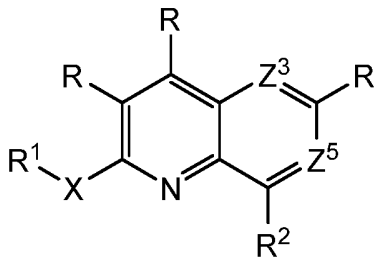
R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, $-O-R^3$, $-S-R^3$, $-(C_1-C_4$ alkyl$)-N(R^3)(R^3)$, $-N(R^3)(R^3)$, $-O-(C_1-C_4$ alkyl$)-N(R^3)(R^3)$, $-(C_1-C_4$ alkyl$)-O-(C_1-C_4$ alkyl$)-N(R^3)(R^3)$, $-C(O)-N(R^3)(R^3)$, $-(C_1-C_4$ alkyl$)-C(O)-N(R^3)(R^3)$, -O-phenyl, phenyl, $-SO_2-(C_1-C_4$ alkyl$)-$, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, $-O-(C_1-C_2)$ fluoro-substituted alkyl, $-O-(C_1-C_4)$ alkyl, $-S-(C_1-C_4)$ alkyl, $-S-(C_1-C_2)$ fluoro-substituted alkyl, $-NH-(C_1-C_4)$ alkyl, and $-N-(C_1-C_4)_2$ alkyl;

X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -C(=O)-NH-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, $-S(=O)_2-NH-$†, $-NH-S(=O)_2-$†, $-NH-S(O)_2-NR^4-$†, $-NR^4-S(O)_2-NH-$†, -NH-C(=O)O-†, -OC(=O)NH-†, $-NH-C(=O)NR^4-$†, $-NR^4-C(=O)NH-$†, $-NH-NR^4-$†, $-NR^4-NH-$†, -O-NH-†, -NH-O-†, $-NH-CR^4R^5-$†, $-CR^4R^5-NH-$†, $-NH-C(=NR^4)-$†, $-C(=NR^4)-NH-$†,$-CR^4R^5-NH-C(O)-$†, $-NH-C(=S)-CR^4R^5-$†, $-CR^4R^5-C(=S)-NH-$†, $-NH-S(O)-CR^4R^5-$†, $-CR^4R^5-S(O)-NH-$†, $-NH-S(O)_2-CR^4R^5-$†, $-CR^4R^5-S(O)_2-NH-$†, $-NH-C(=O)-O-CR^4R^5-$†, $-CR^4R^5-O-C(=O)-NH-$†, $-NH-C(=O)-NR^4-CR^4R^5-$†, $-NH-C(=O)-CR^4R^5-$†, and $-CR^4R^5-NH-C(=O)-O-$†, wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

In certain embodiments, compounds of the invention are a subset of the compounds of Structural Formula (V) represented by Structural Formula (VI):



(VI),

or a salt thereof, wherein:

one of Z^3 or Z^5 is N and the other is CR;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2,

-NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro-or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro- substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), $S(=O)_2$, and O; and

X is selected from $-NR^6$-C(=O)-†, $-NR^6$-C(=S)-†, -NH-S(=O)-†, -S(=O)-NH-†, $-S(=O)_2-NR^6$-†, $-NR^6-S(=O)_2$-†, $-NR^6-S(O)_2-NR^6$-†, $-NR^6$-C(=O)-O-†, -O-C(=O)-NR^6-†, $-NR^6-NR^6$-†, -O-NH-†, -NH-O-†, $-NR^6-CR^4R^5$-†, $-CR^4R^5-NR^6$-†, $-NR^6$-C(=NR^6)-†, -C(=NR^6)-NR^6-†, $-NR^6$-C(=NR^6)-NR^6-†, -C(=O)-NR^6-$(CR^4R^5)_{1-3}$-†, $-CR^4R^5-NR^6$-C(O)-†, $-NR^6$-C(=S)-CR^4R^5-†, $-CR^4R^5$-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, $-CR^4R^5$-S(O)-NH-†, $-NR^6-S(O)_2-CR^4R^5$-†, $-CR^4R^5-S(O)_2-NR^6$-†, -NH-C(=O)-O-CR^4R^5-†, $-CR^4R^5$-O-C(=O)-NH-†, -NH-C(=O)-$NR^6-CR^4R^5$-†, $-NR^6$-C(=O)-CR^4R^5-†, $-CR^4R^5$-NH-C(=O)-O-†, $-NR^6$-C(=O)-$CR^4R^5-NR^6$-†, and $-NR^6$-C(=O)-CR^4R^5-O-†, and when R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, X is additionally selected from -C(=O)-NR^6-†, -C(=S)-NR^6-† and $-NR^6$-C(=O)-NR^6-†, wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated

heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy;

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$.; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, wherein the saturated heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

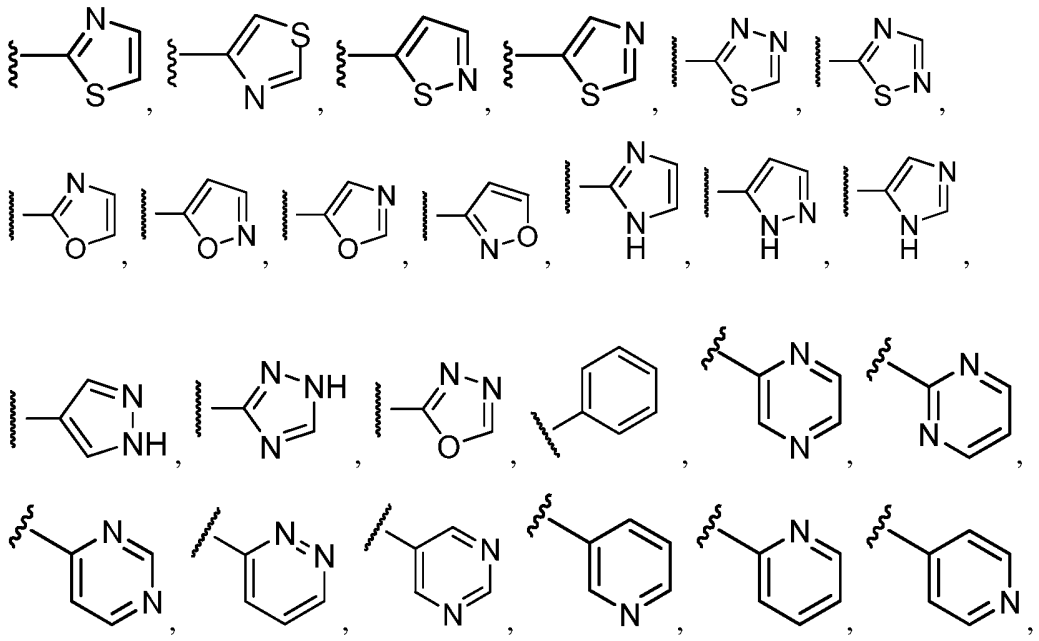
X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^4R^5-†, -CR^4R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†, -C(=O)-NH-CR^4R^5-†, -CR^4R^5-NH-C(O)-†, -NH-C(=S)-CR^4R^5-†,

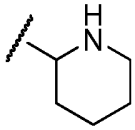
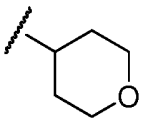
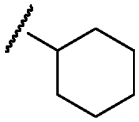
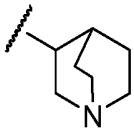
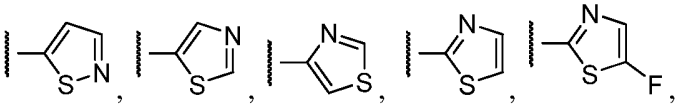
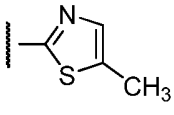
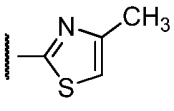
-CR^4R^5-C(=S)-NH-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NH-$S(O)_2$-CR^4R^5-†, -CR^4R^5-$S(O)_2$-NH-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^4R^5-†, -NH-C(=O)-CR^4R^5-†, and -CR^4R^5-NH-C(=O)-O-†, and when R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, X is additionally selected from -C(=O)-NH-†, -C(=S)-NH-†, -NH-C(=O)NR^4-†, and -NR^4-C(=O)NH-†; and

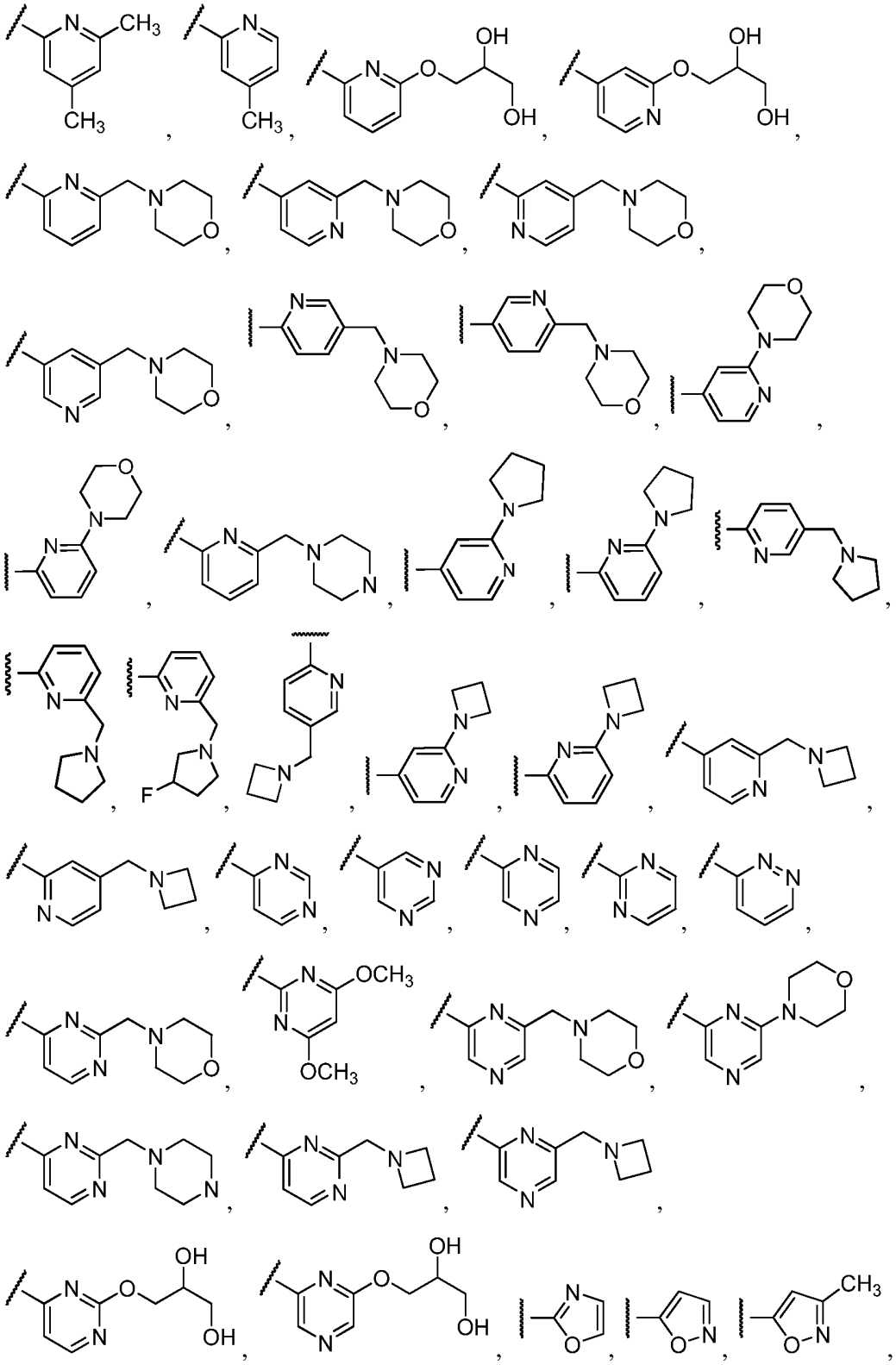
each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

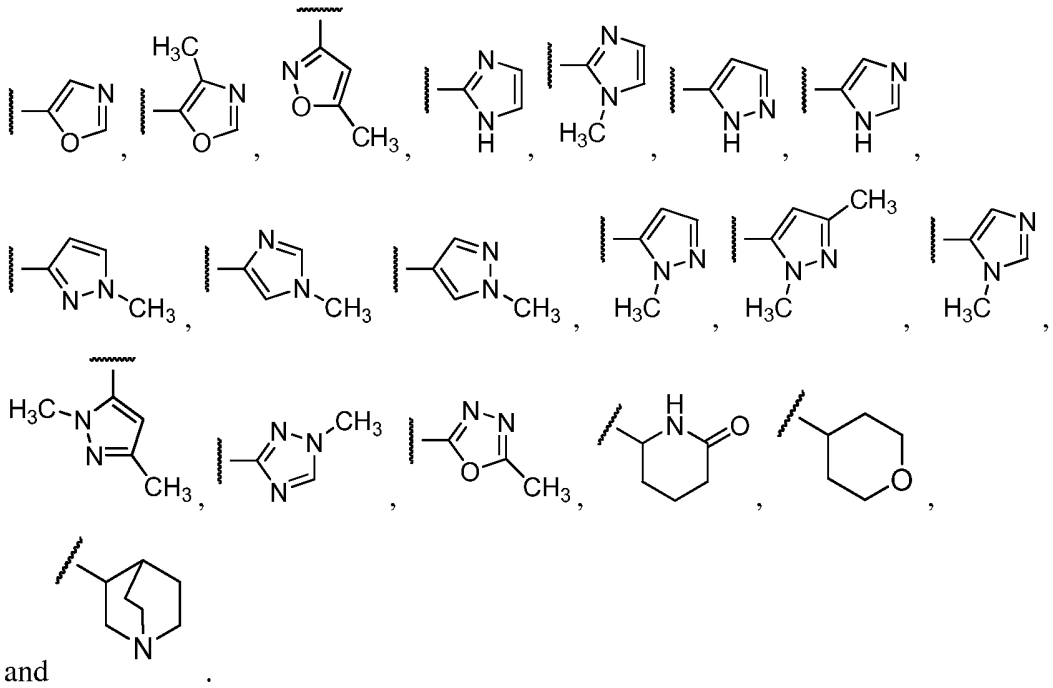
In certain embodiments for any of the above-described structural formulas (as appropriate), X is selected from -NR^6-C(=O)-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, -NR^6-C(=O)-CR^4R^5-†, -NR^6-$S(=O)_2$-†, -NR^6-$S(=O)_2$-CR^4R^5-†, -NR^6-C(=O)-NR^6-†, -C(=O)-NR^6-†, -NR^6-C(=O)-CR^4R^5-O-†, -NR^6-C(=O)-O-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-† and -C(=NR^6)-NR^6-†. In certain embodiments, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, and X is -C(=O)-NR^6-†.

In certain embodiments for any of the above-described structural formulas, R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl. In certain embodiments, R^1 for any of structural formulas (I) to (VI) is selected from:

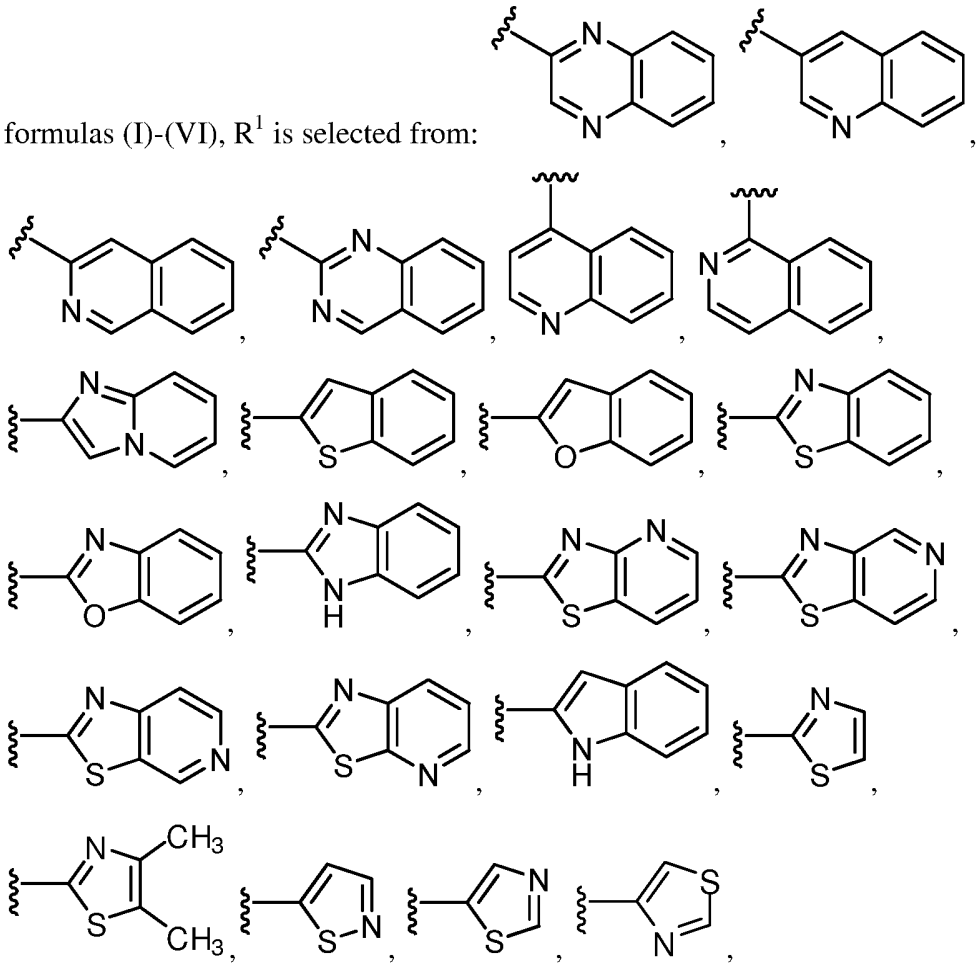


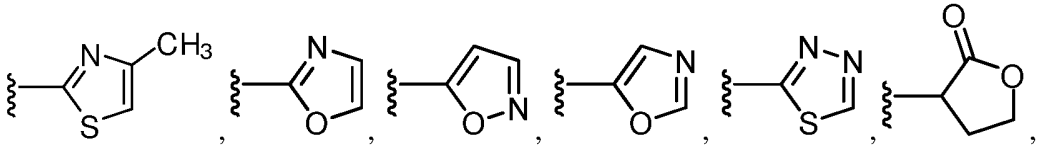
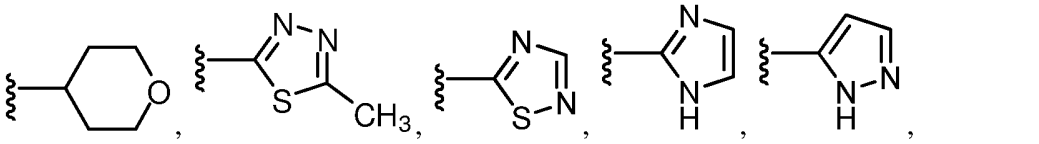
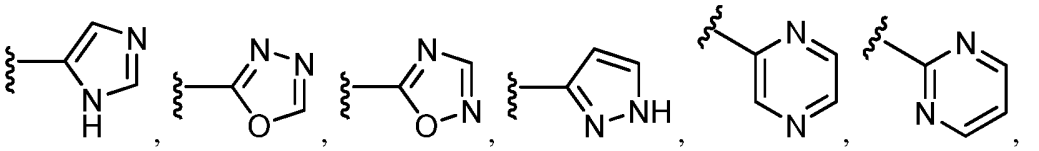
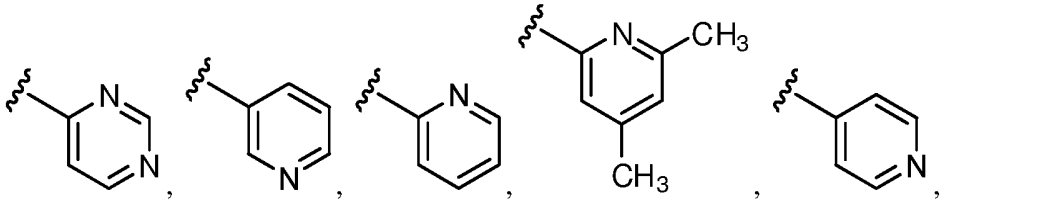
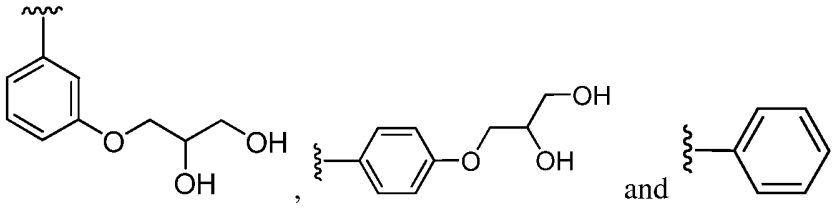
 ,  ,  , and  , wherein R^1 is optionally substituted with one or two substituents independently selected from halo, C_1-C_4 alkyl, -(C_1-C_4 alkyl)-N(R^3)(R^3), =O, -N(R^3)(R^3), and -O-R^3. In certain embodiments, R^1 is selected from:  ,  ,  ,

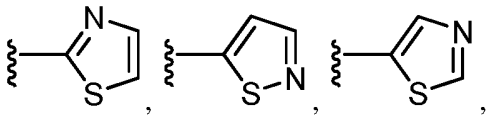
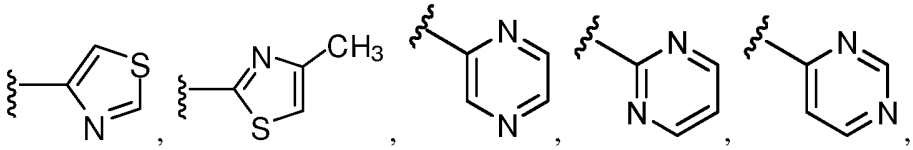
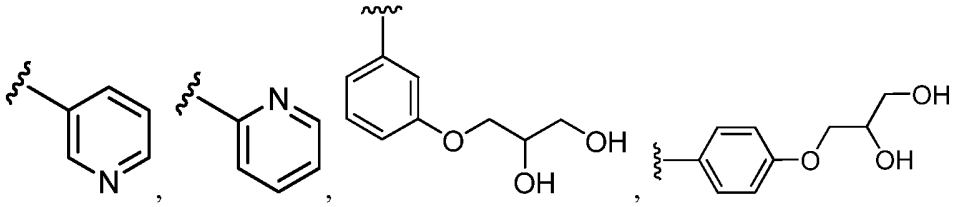




In certain embodiments, for a compound selected from any one of structural formulas (I)-(VI), R^1 is selected from:








and

. In particular embodiments, R^1 is selected from:



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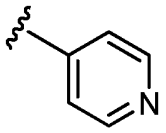
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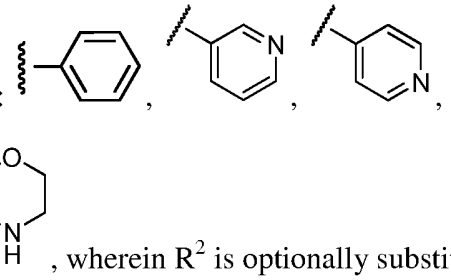
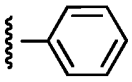
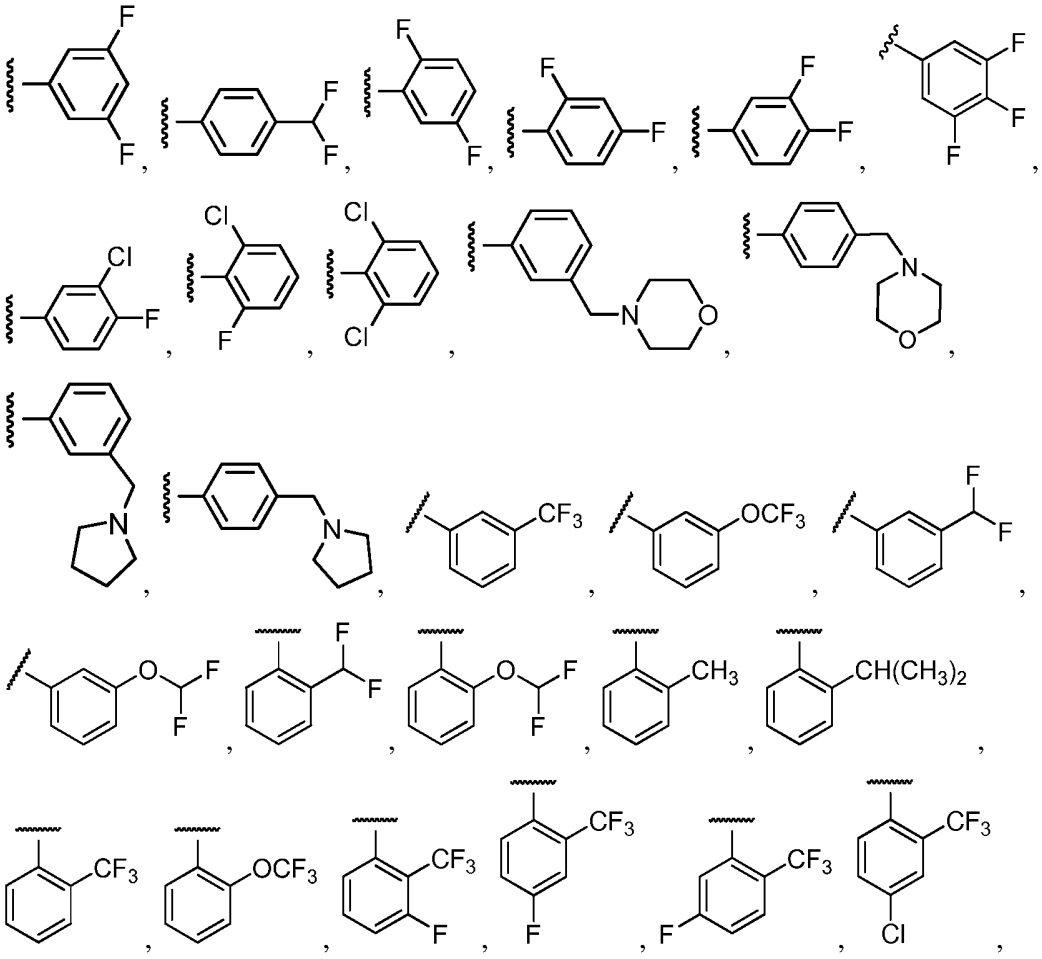
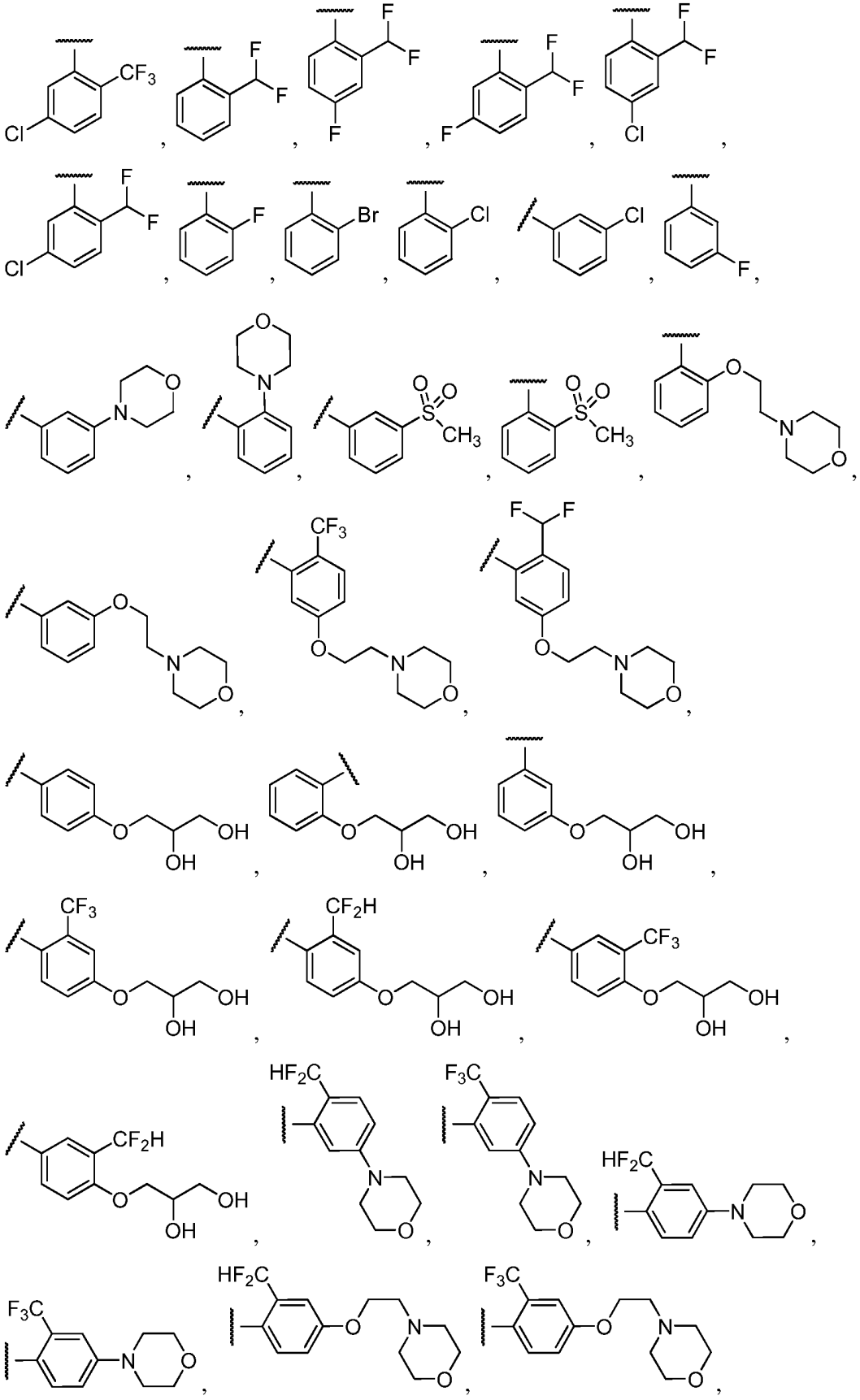
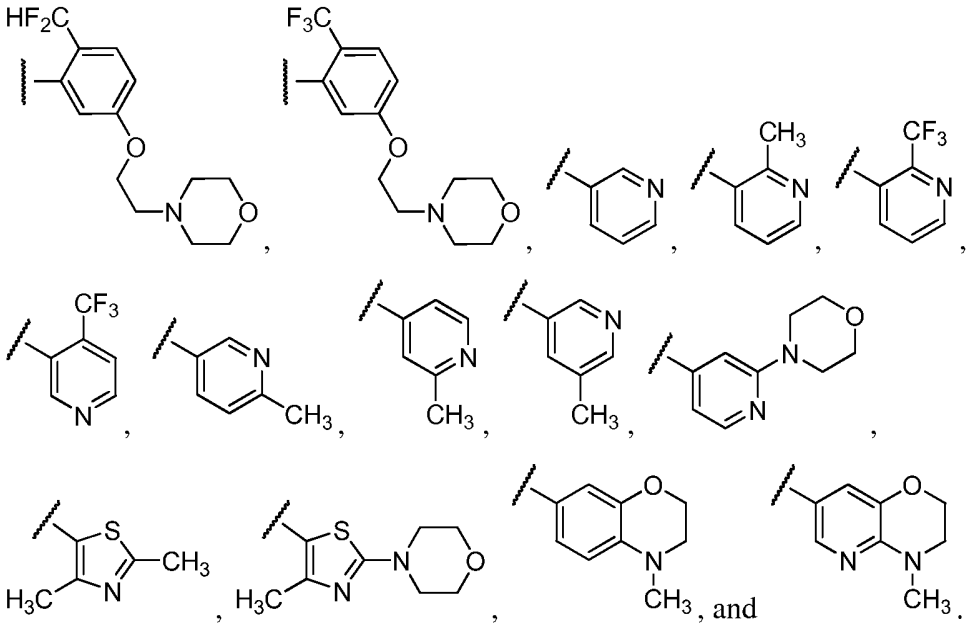
In certain embodiments, R^2 of any of structural formulas (I) to (VI) is selected from optionally substituted aryl and optionally substituted heteroaryl. In

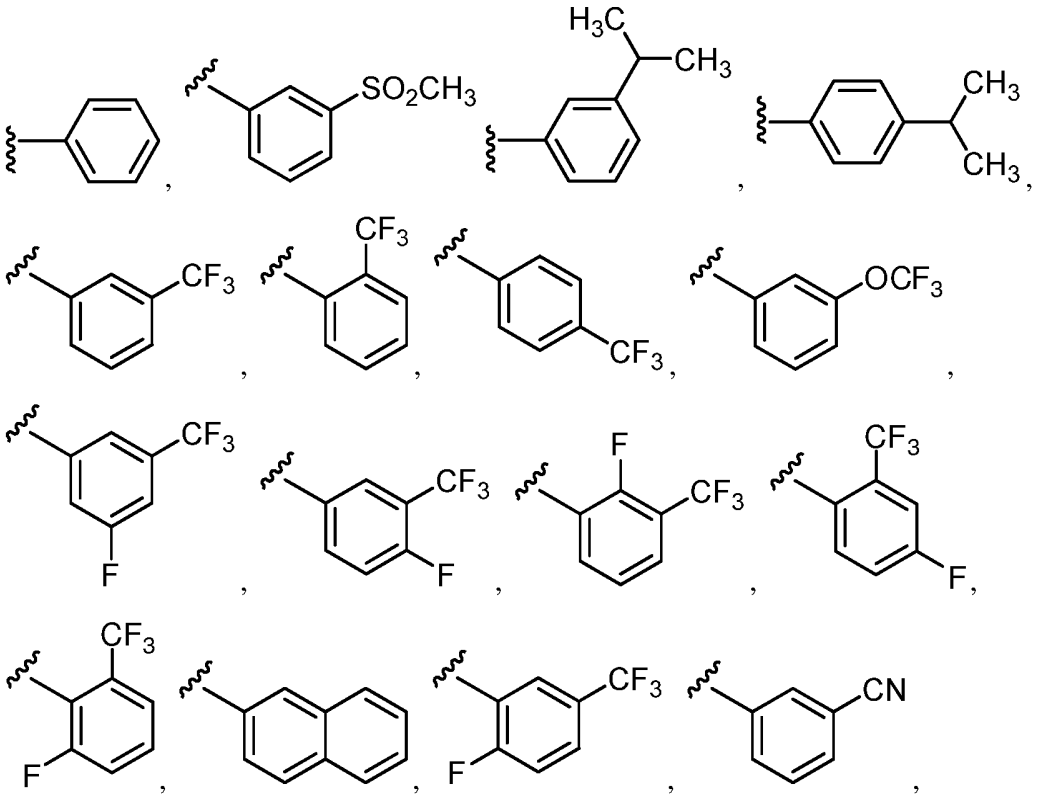
certain embodiments, R^2 is selected from:  , wherein R^2 is optionally substituted with one or more groups independently selected from halo, C_1-C_4 alkyl, -(C_1-C_4 alkyl)-N(R^3)(R^3), C_1-C_2 fluoro-substituted alkyl, -O-R^3, -SO_2-R^3, -N(R^3)(R^3), and -O-(C_1-C_4 alkyl)-N(R^3)(R^3). In certain embodiments, R^2 is meta-substituted relative to the attachment of R^2 to the rest of the compound, and wherein R^2 is optionally further substituted. In certain embodiments, R^2 is selected from:  ,

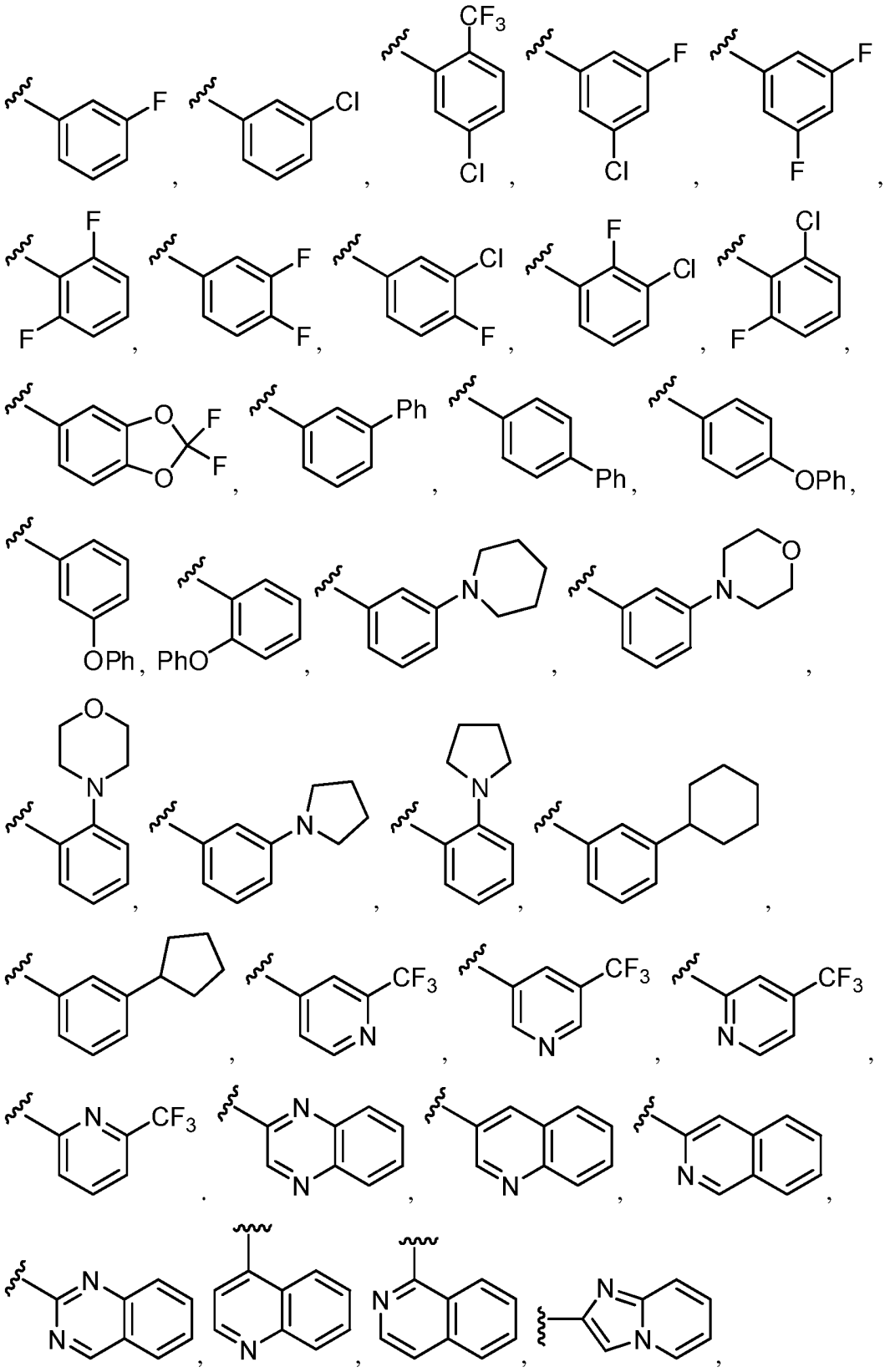


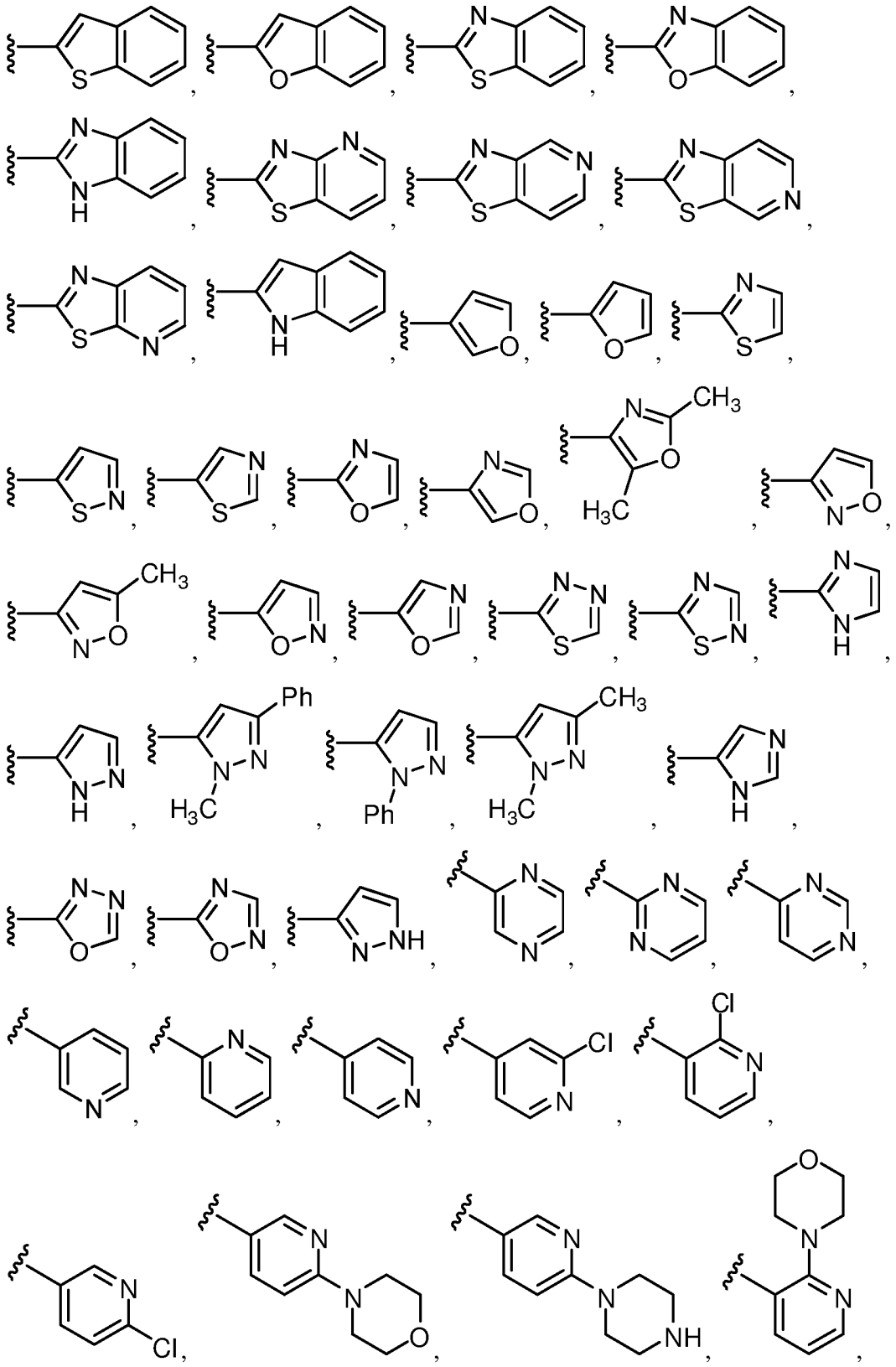


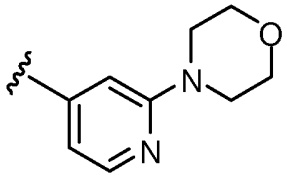
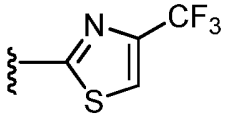
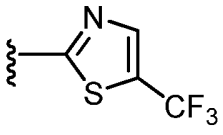


In certain embodiments, for a compound selected from any one of structural formulas (I)-(VI), R^2 is selected from optionally substituted aryl and optionally substituted heteroaryl. In certain such embodiments, R^2 is selected from:








, 
, 
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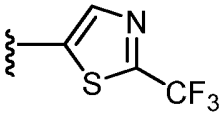
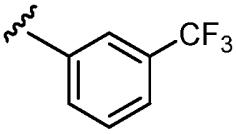

.

In certain emboditments, R^2 is meta-substituted relative to the attachment of R^2 to the rest of the compound, and R^2 is optionally further substituted. In certain such embodiments, R^2 is selected from: 
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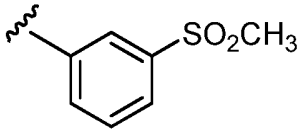
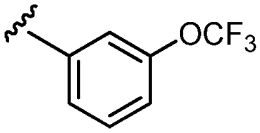
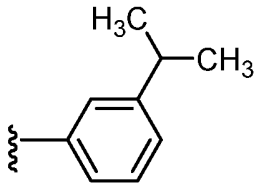

, 
, 
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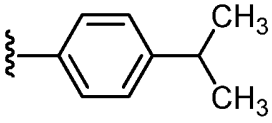
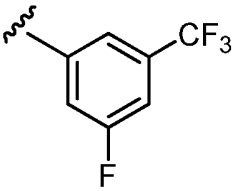
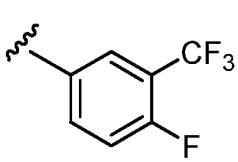
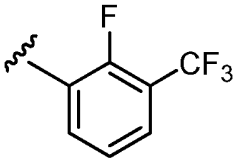

, 
, 
, 
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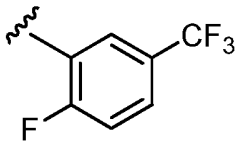
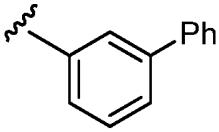
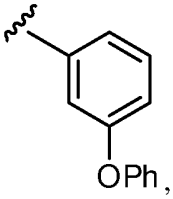
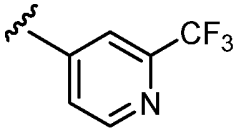

, 
, 
, 
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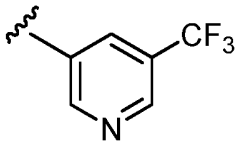
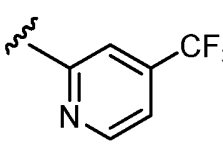
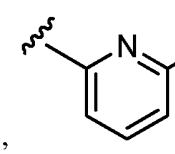
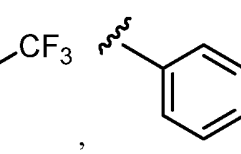

, 
, 
, 
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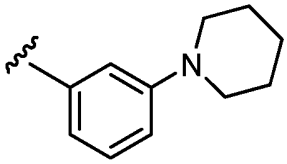
, 
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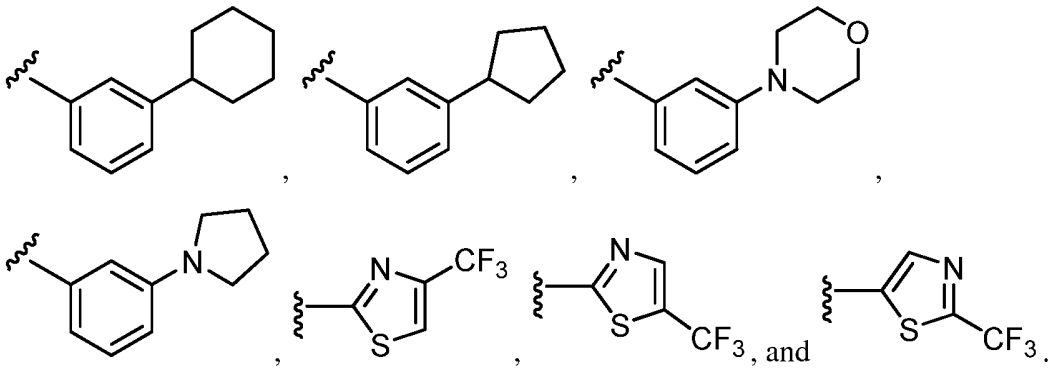


The embodiments described below apply to compounds of any of Structural Formulas (I)-(VI).

In certain aspects, the compound of any of structures (I)-(VI) is a free base.

Compounds of the invention, including novel compounds of the invention, can also be used in the methods described herein.

Sirtuin-modulating compounds of the invention advantageously modulate the level and/or activity of a sirtuin protein, particularly the deacetylase activity of the sirtuin protein.

Separately or in addition to the above properties, certain sirtuin-modulating compounds of the invention do not substantially have one or more of the following activities: inhibition of PI3-kinase, inhibition of aldoreductase, inhibition of tyrosine kinase, transactivation of EGFR tyrosine kinase, coronary dilation, or spasmolytic activity, at concentrations of the compound that are effective for modulating the deacetylation activity of a sirtuin protein (e.g., such as a SIRT1 and/or a SIRT3 protein).

An alkyl group is a straight chained or branched non-aromatic hydrocarbon which is completely saturated. Typically, a straight chained or branched alkyl group has from 1 to about 20 carbon atoms, preferably from 1 to about 10. Examples of straight chained and branched alkyl groups include methyl, ethyl, n-propyl, iso-propyl, n-butyl, sec-butyl, tert-butyl, pentyl, hexyl, pentyl and octyl. A C_1-C_4 straight chained or branched alkyl group is also referred to as a "lower alkyl" group.

The terms alkenyl and alkynyl refer to unsaturated aliphatic groups analogous in length and possible substitution to the alkyl groups described above, but that contain at least one double or triple bond respectively.

The terms alkoxyl or alkoxy as used herein refers to an alkyl group having an oxygen radical attached thereto. Representative alkoxyl groups include methoxy, ethoxy, propyloxy, tert-butoxy and the like.

A cycloalkyl group is a cyclic hydrocarbon which is completely saturated. Typically, a cycloalkyl group has from 3 to about 10 carbon atoms, more typically 3 to 8 carbon atoms.

The terms “heterocycle”, and “heterocyclic”, as used herein, refers to a saturated or unsaturated ring comprising one or more heteroatoms selected from, for example, N, O, and S atoms. Heterocycles include 4-7 membered monocyclic and 8-12 membered bicyclic rings. Each ring of a bicyclic heterocycle may be selected from saturated, unsaturated and aromatic rings. In an exemplary embodiment, an aromatic ring, e.g., pyridyl, may be fused to a saturated or unsaturated ring, e.g., cyclohexane, cyclopentane, or cyclohexene.The terms “heterocyclyl” and “heterocyclic” also include polycyclic ring systems having two or more cyclic rings in which two or more carbons or heteroatoms are common to two adjoining rings wherein at least one of the rings is heterocyclic, e.g., the other cyclic rings can be cycloalkyls, cycloalkenyls, cycloalkynyls, aryls, heteroaryls, and/or heterocycloalkyls. Heterocyclyl groups include, for example, piperidine, piperazine, pyrrolidine, morpholine, lactones, and lactams.

The term “heteroaryl” includes substituted or unsubstituted aromatic single ring structures, preferably 5- to 7-membered rings, more preferably 5- to 6-membered rings, whose ring structures include at least one heteroatom, preferably one to four heteroatoms, more preferably one or two heteroatoms. The terms “heteroaryl” and “hetaryl” also include polycyclic ring systems having two or more cyclic rings in which two or more carbons or heteroatoms are common to two adjoining rings wherein at least one of the rings is heteroaromatic, e.g., the other cyclic rings can be cycloalkyls, cycloalkenyls, cycloalkynyls, aryls, heteroaryls, and/or heterocyclyls. Heteroaryl groups include, for example, pyrrole, furan, thiophene, imidazole, oxazole, thiazole, pyrazole, pyridine, pyrazine, pyridazine, and pyrimidine.

Monocyclic rings include 5-7 membered aryl or heteroaryl, 3-7 membered cycloalkyl, and 5-7 membered non-aromatic heterocyclyl. Monocyclic rings are

optionally substituted with one or more substituents such as halo, cyano, lower alkoxy, lower alkyl, hydroxyl, amino, lower alkylamino and lower dialkylamino. Exemplary monocyclic groups include substituted or unsubstituted heterocycles or carbocycles such as thiazolyl, oxazolyl, oxazinyl, thiazinyl, dithianyl, dioxanyl, isoxazolyl, isothiazolyl, triazolyl, furanyl, tetrahydrofuranyl, dihydrofuranyl, pyranyl, tetrazolyl, pyrazolyl, pyrazinyl, pyridazinyl, imidazolyl, pyridinyl, pyrrolyl, dihydropyrrolyl, pyrrolidinyl, piperidinyl, piperazinyl, pyrimidinyl, morpholinyl, tetrahydrothiophenyl, thiophenyl, cyclohexyl, cyclopentyl, cyclopropyl, cyclobutyl, cycloheptanyl, azetidinyl, oxetanyl, thiiranyl, oxiranyl, aziridinyl, and thiomorpholinyl.

Aromatic (aryl) groups include carbocyclic aromatic groups such as phenyl, naphthyl, and anthracyl, and heteroaryl groups such as imidazolyl, thienyl, furyl, pyridyl, pyrimidyl, pyranyl, pyrazolyl, pyrrolyl, pyrazinyl, thiazolyl, oxazolyl, and tetrazolyl.

Aromatic groups also include fused polycyclic aromatic ring systems in which a carbocyclic aromatic ring or heteroaryl ring is fused to one or more other heteroaryl rings. Examples include benzothienyl, benzofuryl, indolyl, quinolinyl, benzothiazole, benzoxazole, benzimidazole, quinolinyl, isoquinolinyl and isoindolyl.

Azabicyclo refers to a bicyclic molecule that contains a nitrogen atom in the ring skeleton. The two rings of the bicycle may be fused, at two mutually bonded atoms, e.g., indole, across a sequence of atoms, e.g., azabicyclo[2.2.1]heptane, or at a single atom, e.g., spirocycle.

Bridged azabicyclo refers to a bicyclic molecule that contains a nitrogen atom and two fused rings wherein the fusion occurs across a sequence of atoms, i.e., bridgehead atoms. Bridged bicyclo compounds comprise at least one bridge of one or more atoms connecting two bridgehead atoms.

Suitable substituents on a heterocyclyl or heterocyclylmethyl group include -OH, halogen (-Br, -Cl, -I and -F), $-OR^a$, $-O-COR^a$, $-COR^a$, $-C(O)R^a$, -CN, $-NO^2$, -COOH, $-COOR^a$, $-OCO_2R^a$, $-C(O)NR^aR^b$, $-OC(O)NR^aR^b$, $-SO_3H$, $-NH_2$, $-NHR^a$, $-N(R^aR^b)$, $-COOR^a$, -CHO, $-CONH_2$, $-CONHR^a$, $-CON(R^aR^b)$, $-NHCOR^a$, $-NRCOR^a$, $-NHCONH_2$, $-NHCONR^aH$, $-NHCON(R^aR^b)$, $-NR^cCONH_2$, $-NR^cCONR^aH$, $-NR^cCON(R^aR^b)$, $-C(=NH)-NH_2$, $-C(=NH)-NHR^a$, $-C(=NH)-N(R^aR^b)$, $-C(=NR^c)-$

NH_2, -C(=NR^c)-NHR^a, -C(=NR^c)-N(R^aR^b), -NH-C(=NH)-NH_2, -NH-C(=NH)-NHR^a, -NH-C(=NH)-N(R^aR^b), -NH-C(=NR^c)-NH_2, -NH-C(=NR^c)-NHR^a, -NH-C(=NR^c)-N(R^aR^b), -NR^dH-C(=NH)-NH_2, -NR^d-C(=NH)-NHR^a, -NR^d-C(=NH)-N(R^aR^b), -NR^d-C(=NR^c)-NH_2, -NR^d-C(=NR^c)-NHR^a, -NR^d-C(=NR^c)-N(R^aR^b), -$NHNH_2$, -$NHNHR^a$, -NHR^aR^b, -SO_2NH_2, -SO_2NHR_a, -$SO_2NR^aR^b$, -CH=CHR^a, -CH=CR^aR^b, -CR^c=CR^aR^b, CR^c=CHR^a, -CR^c=CR^aR^b, -CCR^a, -SH, -SO_kR^a (k is 0, 1 or 2), -$S(O)_kOR^a$ (k is 0, 1 or 2) and -NH-C(=NH)-NH_2. R^a-R^d are each independently an optionally substituted group selected from an aliphatic, benzyl, or aromatic group, preferably an alkyl, benzylic or aryl group. Optional substituents on R^a-R^d are selected from NH_2, NH(C_{1-4}aliphatic), N(C_{1-4}aliphatic)$_2$, halogen, C_{1-4}aliphatic, OH, O(C_{1-4}aliphatic), NO_2, CN, CO_2H, CO_2(C_{1-4}aliphatic), O(haloC_{1-4} aliphatic), or haloC_{1-4}aliphatic, wherein each of the foregoing C_{1-4}aliphatic groups of is unsubstituted. In addition, -NR^aR^b, taken together, can also form a substituted or unsubstituted non-aromatic heterocyclic group. A substituted aliphatic or substituted aryl group can have more than one substituent.

Halo-substituted includes from one halo substituent up to per-halo substitution. Exemplary halo substituted C_1-C_2 alkyl includes -CIH_2, CF_2H, -CCl_3, -CH_2CH_2Br, -CH_2CHCl_2, -$CHBrCH_2Br$, and -CF_2CHCl_2. Per-halo-substituted C_1-C_2 alkyl, for example, includes –CCl_3 and –CCl_2CF_3.

Fluoro-substituted includes from one fluoro substituent up to per-fluoro-substitution. Exemplary fluoro-substituted C_1-C_2 alkyl includes -CFH_2, CF_2H, -CF_3, -CH_2CH_2F, -CH_2CHF_2, -$CHFCH_3$, and -CF_2CHF_2. Per-fluoro-substituted C_1-C_2 alkyl, for example, includes -CF_3 and –CF_2CF_3.

Combinations of substituents and variables envisioned by this invention are only those that result in the formation of stable compounds. As used herein, the term “stable” refers to compounds that possess stability sufficient to allow manufacture and that maintain the integrity of the compound for a sufficient period of time to be useful for the purposes detailed herein.

The compounds disclosed herein also include partially and fully deuterated variants. In certain embodiments, deuterated variants may be used for kinetic studies. One of ordinary skill in the art can select the sites at which such deuterium atoms are present.

Also included in the present invention are salts, particularly pharmaceutically acceptable salts, of the sirtuin-modulating compounds described herein. The compounds of the present invention that possess a sufficiently acidic, a sufficiently basic, or both functional groups, can react with any of a number of inorganic bases, and inorganic and organic acids, to form a salt. Alternatively, compounds that are inherently charged, such as those with a quaternary nitrogen, can form a salt with an appropriate counterion (e.g., a halide such as bromide, chloride, or fluoride, particularly bromide).

Acids commonly employed to form acid addition salts are inorganic acids such as hydrochloric acid, hydrobromic acid, hydroiodic acid, sulfuric acid, phosphoric acid, and the like, and organic acids such as p-toluenesulfonic acid, methanesulfonic acid, oxalic acid, p-bromophenyl-sulfonic acid, carbonic acid, succinic acid, citric acid, benzoic acid, acetic acid, and the like. Examples of such salts include the sulfate, pyrosulfate, bisulfate, sulfite, bisulfite, phosphate, monohydrogenphosphate, dihydrogenphosphate, metaphosphate, pyrophosphate, chloride, bromide, iodide, acetate, propionate, decanoate, caprylate, acrylate, formate, isobutyrate, caproate, heptanoate, propiolate, oxalate, malonate, succinate, suberate, sebacate, fumarate, maleate, butyne-1,4-dioate, hexyne-1,6-dioate, benzoate, chlorobenzoate, methylbenzoate, dinitrobenzoate, hydroxybenzoate, methoxybenzoate, phthalate, sulfonate, xylenesulfonate, phenylacetate, phenylpropionate, phenylbutyrate, citrate, lactate, gamma-hydroxybutyrate, glycolate, tartrate, methanesulfonate, propanesulfonate, naphthalene-1-sulfonate, naphthalene-2-sulfonate, mandelate, and the like.

Base addition salts include those derived from inorganic bases, such as ammonium or alkali or alkaline earth metal hydroxides, carbonates, bicarbonates, and the like. Such bases useful in preparing the salts of this invention thus include sodium hydroxide, potassium hydroxide, ammonium hydroxide, potassium carbonate, and the like.

According to another embodiment, the present invention provides methods of producing the above-defined sirtuin-modulating compounds. The compounds may be synthesized using conventional techniques. Advantageously, these compounds are conveniently synthesized from readily available starting materials.

Synthetic chemistry transformations and methodologies useful in synthesizing the sirtuin-modulating compounds described herein are known in the art and include, for example, those described in R. Larock, *Comprehensive Organic Transformations* (1989); T. W. Greene and P. G. M. Wuts, *Protective Groups in Organic Synthesis, 2d. Ed.* (1991); L. Fieser and M. Fieser, *Fieser and Fieser's Reagents for Organic Synthesis* (1994); and L. Paquette, ed., *Encyclopedia of Reagents for Organic Synthesis* (1995).

In an exemplary embodiment, a sirtuin-modulating compound may traverse the cytoplasmic membrane of a cell. For example, a compound may have a cell-permeability of at least about 20%, 50%, 75%, 80%, 90% or 95%.

Sirtuin-modulating compounds described herein may also have one or more of the following characteristics: the compound may be essentially non-toxic to a cell or subject; the sirtuin-modulating compound may be an organic molecule or a small molecule of 2000 amu or less, 1000 amu or less; a compound may have a half-life under normal atmospheric conditions of at least about 30 days, 60 days, 120 days, 6 months or 1 year; the compound may have a half-life in solution of at least about 30 days, 60 days, 120 days, 6 months or 1 year; a sirtuin-modulating compound may be more stable in solution than resveratrol by at least a factor of about 50%, 2 fold, 5 fold, 10 fold, 30 fold, 50 fold or 100 fold; a sirtuin-modulating compound may promote deacetylation of the DNA repair factor Ku70; a sirtuin-modulating compound may promote deacetylation of RelA/p65; a compound may increase general turnover rates and enhance the sensitivity of cells to TNF-induced apoptosis.

In certain embodiments, a sirtuin-modulating compound does not have any substantial ability to inhibit a histone deacetylase (HDACs) class I, a HDAC class II, or HDACs I and II, at concentrations (e.g., in vivo) effective for modulating the deacetylase activity of the sirtuin. For instance, in preferred embodiments the sirtuin-modulating compound is a sirtuin-activating compound and is chosen to have an EC_{50} for activating sirtuin deacetylase activity that is at least 5 fold less than the EC_{50} for inhibition of an HDAC I and/or HDAC II, and even more preferably at least 10 fold, 100 fold or even 1000 fold less. Methods for assaying HDAC I and/or HDAC II activity are well known in the art and kits to perform

such assays may be purchased commercially. See e.g., BioVision, Inc. (Mountain View, CA; world wide web at biovision.com) and Thomas Scientific (Swedesboro, NJ; world wide web at tomassci.com).

In certain embodiments, a sirtuin-modulating compound does not have any substantial ability to modulate sirtuin homologs. In one embodiment, an activator of a human sirtuin protein may not have any substantial ability to activate a sirtuin protein from lower eukaryotes, particularly yeast or human pathogens, at concentrations (e.g., in vivo) effective for activating the deacetylase activity of human sirtuin. For example, a sirtuin-activating compound may be chosen to have an EC_{50} for activating a human sirtuin, such as SIRT1 and/or SIRT3, deacetylase activity that is at least 5 fold less than the EC_{50} for activating a yeast sirtuin, such as Sir2 (such as Candida, S. cerevisiae, etc.), and even more preferably at least 10 fold, 100 fold or even 1000 fold less. In another embodiment, an inhibitor of a sirtuin protein from lower eukaryotes, particularly yeast or human pathogens, does not have any substantial ability to inhibit a sirtuin protein from humans at concentrations (e.g., in vivo) effective for inhibiting the deacetylase activity of a sirtuin protein from a lower eukaryote. For example, a sirtuin-inhibiting compound may be chosen to have an IC_{50} for inhibiting a human sirtuin, such as SIRT1 and/or SIRT3, deacetylase activity that is at least 5 fold less than the IC_{50} for inhibiting a yeast sirtuin, such as Sir2 (such as Candida, S. cerevisiae, etc.), and even more preferably at least 10 fold, 100 fold or even 1000 fold less.

In certain embodiments, a sirtuin-modulating compound may have the ability to modulate one or more sirtuin protein homologs, such as, for example, one or more of human SIRT1, SIRT2, SIRT3, SIRT4, SIRT5, SIRT6, or SIRT7. In one embodiment, a sirtuin-modulating compound has the ability to modulate both a SIRT1 and a SIRT3 protein.

In other embodiments, a SIRT1 modulator does not have any substantial ability to modulate other sirtuin protein homologs, such as, for example, one or more of human SIRT2, SIRT3, SIRT4, SIRT5, SIRT6, or SIRT7, at concentrations (e.g., in vivo) effective for modulating the deacetylase activity of human SIRT1. For example, a sirtuin-modulating compound may be chosen to have an ED_{50} for modulating human SIRT1 deacetylase activity that is at least 5 fold less than the

ED_{50} for modulating one or more of human SIRT2, SIRT3, SIRT4, SIRT5, SIRT6, or SIRT7, and even more preferably at least 10 fold, 100 fold or even 1000 fold less. In one embodiment, a SIRT1 modulator does not have any substantial ability to modulate a SIRT3 protein.

In other embodiments, a SIRT3 modulator does not have any substantial ability to modulate other sirtuin protein homologs, such as, for example, one or more of human SIRT1, SIRT2, SIRT4, SIRT5, SIRT6, or SIRT7, at concentrations (e.g., in vivo) effective for modulating the deacetylase activity of human SIRT3. For example, a sirtuin-modulating compound may be chosen to have an ED_{50} for modulating human SIRT3 deacetylase activity that is at least 5 fold less than the ED_{50} for modulating one or more of human SIRT1, SIRT2, SIRT4, SIRT5, SIRT6, or SIRT7, and even more preferably at least 10 fold, 100 fold or even 1000 fold less. In one embodiment, a SIRT3 modulator does not have any substantial ability to modulate a SIRT1 protein.

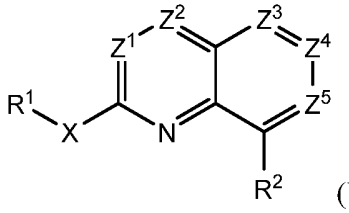
In certain embodiments, a sirtuin-modulating compound may have a binding affinity for a sirtuin protein of about 10^{-9}M, 10^{-10}M, 10^{-11}M, 10^{-12}M or less. A sirtuin-modulating compound may reduce (activator) or increase (inhibitor) the apparent Km of a sirtuin protein for its substrate or NAD+ (or other cofactor) by a factor of at least about 2, 3, 4, 5, 10, 20, 30, 50 or 100. In certain embodiments, Km values are determined using the mass spectrometry assay described herein. Preferred activating compounds reduce the Km of a sirtuin for its substrate or cofactor to a greater extent than caused by resveratrol at a similar concentration or reduce the Km of a sirtuin for its substrate or cofactor similar to that caused by resveratrol at a lower concentration. A sirtuin-modulating compound may increase the Vmax of a sirtuin protein by a factor of at least about 2, 3, 4, 5, 10, 20, 30, 50 or 100. A sirtuin-modulating compound may have an ED50 for modulating the deacetylase activity of a SIRT1 and/or SIRT3 protein of less than about 1 nM, less than about 10 nM, less than about 100 nM, less than about 1 μM, less than about 10 μM, less than about 100 μM, or from about 1-10 nM, from about 10-100 nM, from about 0.1-1 μM, from about 1-10 μM or from about 10-100 μM. A sirtuin-modulating compound may modulate the deacetylase activity of a SIRT1 and/or SIRT3 protein by a factor of at least about 5, 10, 20, 30, 50, or 100, as measured in

a cellular assay or in a cell based assay. A sirtuin-activating compound may cause at least about 10%, 30%, 50%, 80%, 2 fold, 5 fold, 10 fold, 50 fold or 100 fold greater induction of the deacetylase activity of a sirtuin protein relative to the same concentration of resveratrol. A sirtuin-modulating compound may have an ED50 for modulating SIRT5 that is at least about 10 fold, 20 fold, 30 fold, 50 fold greater than that for modulating SIRT1 and/or SIRT3.

3. Exemplary Uses

In certain aspects, the invention provides methods for modulating the level and/or activity of a sirtuin protein and methods of use thereof.

In certain embodiments, the invention provides methods for increasing sirtuin-1 activity in a cell comprising the step of contacting the cell with a compound represented by Structural Formula (VII):



(VII),

or a salt thereof, wherein:

each of Z^1-Z^5 is independently selected from N and CR, wherein no more than two of Z^1-Z^5 are simultaneously N;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O- saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle),

-O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen, and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NH-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle),

fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy;

each R^3 is independently selected from hydrogen, and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, wherein the saturated heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

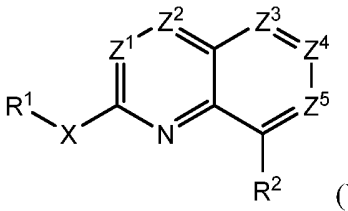
X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -C(=O)-NH-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-C(=O)NR^4-†, -NR^4-C(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^4R^5-†, -CR^4R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†,

-C(=O)-NH-CR^4R^5-†, -CR^4R^5-NH-C(O)-†, -NH-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NH-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NH-$S(O)_2$-CR^4R^5-†, -CR^4R^5-$S(O)_2$-NH-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^4R^5-†, -NH-C(=O)-CR^4R^5-†, and -CR^4R^5-NH-C(=O)-O-†; and

each R^4 and R^5 is independently selected from hydrogen, C_1-C_4 alkyl, -CF_3 and (C_1-C_3 alkyl)-CF_3.

Other indications described below may also be treated by a compound of Structural Formula (VII).

In certain embodiments, the invention provides methods for treating a subject suffering from or susceptible to insulin resistance, a metabolic syndrome, diabetes, or complications thereof, or for increasing insulin sensitivity in a subject, comprising administering to the subject in need thereof a compound represented by Structural Formula (VIII):



(VIII),

or a salt thereof, wherein:

each of Z^1-Z^5 is independently selected from N and CR, wherein no more than two of Z^1-Z^5 are simultaneously N;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O- saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is

also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom

with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen, and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NH-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

Other indications described below may also be treated by a compound of Structural Formula (VIII).

In certain embodiments, each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl;

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl,

fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with O-(saturated heterocycle), fluoro-substituted -O-(saturated heterocycle), C_1-C_4 alkyl-substituted O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy;

each R^3 is independently selected from hydrogen, and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, the saturated heterocycle is optionally substituted at a carbon atom with -OH, -C_1-C_4 alkyl, fluoro, fluoro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and the saturated heterocycle is optionally substituted at a nitrogen atom with C_1-C_4 alkyl or fluoro-substituted C_1-C_4 alkyl;

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one to two substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, C_1-C_2 fluoro-substituted alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with O-(saturated heterocycle), 3,4-methylenedioxy, fluoro-substituted 3,4-methylenedioxy, 3,4-ethylenedioxy, or fluoro-substituted 3,4-ethylenedioxy, wherein any phenyl, saturated heterocycle, or second heterocycle substituent of R^2 is optionally substituted with halo, -C≡N, C_1-C_4 alkyl, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

X is selected from -NH-C(=O)-†, -NH-C(=S)-†, -C(=O)-NH-†, -C(=S)-NH-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NH-†, -NH-S(=O)$_2$-†, -NH-S(O)$_2$-NR^4-†, -NR^4-S(O)$_2$-NH-†, -NH-C(=O)O-†, -OC(=O)NH-†, -NH-C(=O)NR^4-†, -NR^4-C(=O)NH-†, -NH-NR^4-†, -NR^4-NH-†, -O-NH-†, -NH-O-†, -NH-CR^{4}R^5-†, -CR^{4}R^5-NH-†, -NH-C(=NR^4)-†, -C(=NR^4)-NH-†, -C(=O)-NH-CR^{4}R^5-†, -CR^{4}R^5-NH-C(O)-†, -NH-C(=S)-CR^{4}R^5-†, -CR^{4}R^5-C(=S)-NH-†, -NH-S(O)-CR^{4}R^5-†, -CR^{4}R^5-S(O)-NH-†, -NH-S(O)$_2$-CR^{4}R^5-†, -CR^{4}R^5-S(O)$_2$-NH-†, -NH-C(=O)-O-CR^{4}R^5-†, -CR^{4}R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^4-CR^{4}R^5-†, -NH-C(=O)-CR^{4}R^5-†, and -CR^{4}R^5-NH-C(=O)-O-†; and

each R^4 and R^5 is independently selected from hydrogen, C$_1$-C$_4$ alkyl, -CF$_3$ and (C$_1$-C$_3$ alkyl)-CF$_3$.

Other indications described below may also be treated by a compound of Structural Formula (VIII).

In certain embodiments, the invention provides methods for using sirtuin-modulating compounds wherein the sirtuin-modulating compounds activate a sirtuin protein, e.g., increase the level and/or activity of a sirtuin protein. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be useful for a variety of therapeutic applications including, for example, increasing the lifespan of a cell, and treating and/or preventing a wide variety of diseases and disorders including, for example, diseases or disorders related to aging or stress, diabetes, obesity, neurodegenerative diseases, cardiovascular disease, blood clotting disorders, inflammation, cancer, and/or flushing, etc. The methods comprise administering to a subject in need thereof a pharmaceutically effective amount of a sirtuin-modulating compound, e.g., a sirtuin-activating compound.

Without wishing to be bound by theory, it is believed that activators of the instant invention may interact with a sirtuin at the same location within the sirtuin protein (e.g., active site or site affecting the Km or Vmax of the active site). It is believed that this is the reason why certain classes of sirtuin activators and inhibitors can have substantial structural similarity.

In certain embodiments, the sirtuin-modulating compounds described herein may be taken alone or in combination with other compounds. In one embodiment, a

mixture of two or more sirtuin-modulating compounds may be administered to a subject in need thereof. In another embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered with one or more of the following compounds: resveratrol, butein, fisetin, piceatannol, or quercetin. In an exemplary embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered in combination with nicotinic acid. In another embodiment, a sirtuin-modulating compound that decreases the level and/or activity of a sirtuin protein may be administered with one or more of the following compounds: nicotinamide (NAM), suramin; NF023 (a G-protein antagonist); NF279 (a purinergic receptor antagonist); Trolox (6-hydroxy-2,5,7,8,tetramethylchroman-2-carboxylic acid); (-)-epigallocatechin (hydroxy on sites 3,5,7,3',4', 5'); (-)-epigallocatechin gallate (Hydroxy sites 5,7,3',4',5' and gallate ester on 3); cyanidin chloride (3,5,7,3',4'-pentahydroxyflavylium chloride); delphinidin chloride (3,5,7,3',4',5'-hexahydroxyflavylium chloride); myricetin (cannabiscetin; 3,5,7,3',4',5'-hexahydroxyflavone); 3,7,3',4',5'-pentahydroxyflavone; gossypetin (3,5,7,8,3',4'-hexahydroxyflavone), sirtinol; and splitomicin. In yet another embodiment, one or more sirtuin-modulating compounds may be administered with one or more therapeutic agents for the treatment or prevention of various diseases, including, for example, cancer, diabetes, neurodegenerative diseases, cardiovascular disease, blood clotting, inflammation, flushing, obesity, aging, stress, etc. In various embodiments, combination therapies comprising a sirtuin-modulating compound may refer to (1) pharmaceutical compositions that comprise one or more sirtuin-modulating compounds in combination with one or more therapeutic agents (e.g., one or more therapeutic agents described herein); and (2) co-administration of one or more sirtuin-modulating compounds with one or more therapeutic agents wherein the sirtuin-modulating compound and therapeutic agent have not been formulated in the same compositions (but may be present within the same kit or package, such as a blister pack or other multi-chamber package; connected, separately sealed containers (e.g., foil pouches) that can be separated by the user; or a kit where the sirtuin modulating compound(s) and other therapeutic agent(s) are in separate vessels). When using separate formulations, the sirtuin-modulating compound may be

administered at the same, intermittent, staggered, prior to, subsequent to, or combinations thereof, with the administration of another therapeutic agent.

In certain embodiments, methods for reducing, preventing or treating diseases or disorders using a sirtuin-modulating compound may also comprise increasing the protein level of a sirtuin, such as human SIRT1, SIRT2 and/or SIRT3, or homologs thereof. Increasing protein levels can be achieved by introducing into a cell one or more copies of a nucleic acid that encodes a sirtuin. For example, the level of a sirtuin can be increased in a mammalian cell by introducing into the mammalian cell a nucleic acid encoding the sirtuin, e.g., increasing the level of SIRT1 by introducing a nucleic acid encoding the amino acid sequence set forth in GenBank Accession No. NP_036370 and/or increasing the level of SIRT3 by introducing a nucleic acid encoding the amino acid sequence set forth in GenBank Accession No. AAH01042.

A nucleic acid that is introduced into a cell to increase the protein level of a sirtuin may encode a protein that is at least about 80%, 85%, 90%, 95%, 98%, or 99% identical to the sequence of a sirtuin, e.g., SIRT1 and/or SIRT3 protein. For example, the nucleic acid encoding the protein may be at least about 80%, 85%, 90%, 95%, 98%, or 99% identical to a nucleic acid encoding a SIRT1 (e.g. GenBank Accession No. NM_012238) and/or SIRT3 (e.g., GenBank Accession No. BC001042) protein. The nucleic acid may also be a nucleic acid that hybridizes, preferably under stringent hybridization conditions, to a nucleic acid encoding a wild-type sirtuin, e.g., SIRT1 and/or SIRT3 protein. Stringent hybridization conditions may include hybridization and a wash in 0.2 x SSC at 65 °C. When using a nucleic acid that encodes a protein that is different from a wild-type sirtuin protein, such as a protein that is a fragment of a wild-type sirtuin, the protein is preferably biologically active, e.g., is capable of deacetylation. It is only necessary to express in a cell a portion of the sirtuin that is biologically active. For example, a protein that differs from wild-type SIRT1 having GenBank Accession No. NP_036370, preferably contains the core structure thereof. The core structure sometimes refers to amino acids 62-293 of GenBank Accession No. NP_036370, which are encoded by nucleotides 237 to 932 of GenBank Accession No. NM_012238, which encompasses the NAD binding as well as the substrate binding domains. The core

domain of SIRT1 may also refer to about amino acids 261 to 447 of GenBank Accession No. NP_036370, which are encoded by nucleotides 834 to 1394 of GenBank Accession No. NM_012238; to about amino acids 242 to 493 of GenBank Accession No. NP_036370, which are encoded by nucleotides 777 to 1532 of GenBank Accession No. NM_012238; or to about amino acids 254 to 495 of GenBank Accession No. NP_036370, which are encoded by nucleotides 813 to 1538 of GenBank Accession No. NM_012238. Whether a protein retains a biological function, e.g., deacetylation capabilities, can be determined according to methods known in the art.

In certain embodiments, methods for reducing, preventing or treating diseases or disorders using a sirtuin-modulating compound may also comprise decreasing the protein level of a sirtuin, such as human SIRT1, SIRT2 and/or SIRT3, or homologs thereof. Decreasing a sirtuin protein level can be achieved according to methods known in the art. For example, an siRNA, an antisense nucleic acid, or a ribozyme targeted to the sirtuin can be expressed in the cell. A dominant negative sirtuin mutant, e.g., a mutant that is not capable of deacetylating, may also be used. For example, mutant H363Y of SIRT1, described, e.g., in Luo et al. (2001) Cell 107:137 can be used. Alternatively, agents that inhibit transcription can be used.

Methods for modulating sirtuin protein levels also include methods for modulating the transcription of genes encoding sirtuins, methods for stabilizing/destabilizing the corresponding mRNAs, and other methods known in the art.

Aging/Stress

In one embodiment, the invention provides a method extending the lifespan of a cell, extending the proliferative capacity of a cell, slowing aging of a cell, promoting the survival of a cell, delaying cellular senescence in a cell, mimicking the effects of calorie restriction, increasing the resistance of a cell to stress, or preventing apoptosis of a cell, by contacting the cell with a sirtuin-modulating compound of the invention that increases the level and/or activity of a sirtuin protein. In an exemplary embodiment, the methods comprise contacting the cell with a sirtuin-activating compound.

The methods described herein may be used to increase the amount of time that cells, particularly primary cells (i.e., cells obtained from an organism, e.g., a human), may be kept alive in a cell culture. Embryonic stem (ES) cells and pluripotent cells, and cells differentiated therefrom, may also be treated with a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein to keep the cells, or progeny thereof, in culture for longer periods of time. Such cells can also be used for transplantation into a subject, e.g., after *ex vivo* modification.

In one embodiment, cells that are intended to be preserved for long periods of time may be treated with a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. The cells may be in suspension (e.g., blood cells, serum, biological growth media, etc.) or in tissues or organs. For example, blood collected from an individual for purposes of transfusion may be treated with a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein to preserve the blood cells for longer periods of time. Additionally, blood to be used for forensic purposes may also be preserved using a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. Other cells that may be treated to extend their lifespan or protect against apoptosis include cells for consumption, e.g., cells from non-human mammals (such as meat) or plant cells (such as vegetables).

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be applied during developmental and growth phases in mammals, plants, insects or microorganisms, in order to, e.g., alter, retard or accelerate the developmental and/or growth process.

In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used to treat cells useful for transplantation or cell therapy, including, for example, solid tissue grafts, organ transplants, cell suspensions, stem cells, bone marrow cells, etc. The cells or tissue may be an autograft, an allograft, a syngraft or a xenograft. The cells or tissue may be treated with the sirtuin-modulating compound prior to administration/implantation, concurrently with administration/implantation, and/or post administration/implantation into a subject. The cells or tissue may be treated

prior to removal of the cells from the donor individual, *ex vivo* after removal of the cells or tissue from the donor individual, or post implantation into the recipient. For example, the donor or recipient individual may be treated systemically with a sirtuin-modulating compound or may have a subset of cells/tissue treated locally with a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. In certain embodiments, the cells or tissue (or donor/recipient individuals) may additionally be treated with another therapeutic agent useful for prolonging graft survival, such as, for example, an immunosuppressive agent, a cytokine, an angiogenic factor, etc.

In yet other embodiments, cells may be treated with a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein *in vivo*, e.g., to increase their lifespan or prevent apoptosis. For example, skin can be protected from aging (e.g., developing wrinkles, loss of elasticity, etc.) by treating skin or epithelial cells with a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. In an exemplary embodiment, skin is contacted with a pharmaceutical or cosmetic composition comprising a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. Exemplary skin afflictions or skin conditions that may be treated in accordance with the methods described herein include disorders or diseases associated with or caused by inflammation, sun damage or natural aging. For example, the compositions find utility in the prevention or treatment of contact dermatitis (including irritant contact dermatitis and allergic contact dermatitis), atopic dermatitis (also known as allergic eczema), actinic keratosis, keratinization disorders (including eczema), epidermolysis bullosa diseases (including pemphigus), exfoliative dermatitis, seborrheic dermatitis, erythemas (including erythema multiforme and erythema nodosum), damage caused by the sun or other light sources, discoid lupus erythematosus, dermatomyositis, psoriasis, skin cancer and the effects of natural aging. In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for the treatment of wounds and/or burns to promote healing, including, for example, first-, second- or third-degree burns and/or thermal, chemical or electrical burns. The formulations may be administered topically, to the skin or mucosal tissue.

Topical formulations comprising one or more sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used as preventive, e.g., chemopreventive, compositions. When used in a chemopreventive method, susceptible skin is treated prior to any visible condition in a particular individual.

Sirtuin-modulating compounds may be delivered locally or systemically to a subject. In one embodiment, a sirtuin-modulating compound is delivered locally to a tissue or organ of a subject by injection, topical formulation, etc.

In another embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be used for treating or preventing a disease or condition induced or exacerbated by cellular senescence in a subject; methods for decreasing the rate of senescence of a subject, e.g., after onset of senescence; methods for extending the lifespan of a subject; methods for treating or preventing a disease or condition relating to lifespan; methods for treating or preventing a disease or condition relating to the proliferative capacity of cells; and methods for treating or preventing a disease or condition resulting from cell damage or death. In certain embodiments, the method does not act by decreasing the rate of occurrence of diseases that shorten the lifespan of a subject. In certain embodiments, a method does not act by reducing the lethality caused by a disease, such as cancer.

In yet another embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered to a subject in order to generally increase the lifespan of its cells and to protect its cells against stress and/or against apoptosis. It is believed that treating a subject with a compound described herein is similar to subjecting the subject to hormesis, i.e., mild stress that is beneficial to organisms and may extend their lifespan.

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered to a subject to prevent aging and aging-related consequences or diseases, such as stroke, heart disease, heart failure, arthritis, high blood pressure, and Alzheimer's disease. Other conditions that can be treated include ocular disorders, e.g., associated with the aging of the eye, such as cataracts, glaucoma, and macular degeneration. Sirtuin-modulating compounds that

increase the level and/or activity of a sirtuin protein can also be administered to subjects for treatment of diseases, e.g., chronic diseases, associated with cell death, in order to protect the cells from cell death. Exemplary diseases include those associated with neural cell death, neuronal dysfunction, or muscular cell death or dysfunction, such as Parkinson's disease, Alzheimer's disease, multiple sclerosis, amyotrophic, amniotropic lateral sclerosis, and muscular dystrophy; AIDS; fulminant hepatitis; diseases linked to degeneration of the brain, such as Creutzfeld-Jakob disease, retinitis pigmentosa and cerebellar degeneration; myelodysplasia such as aplastic anemia; ischemic diseases such as myocardial infarction and stroke; hepatic diseases such as alcoholic hepatitis, hepatitis B and hepatitis C; joint-diseases such as osteoarthritis; atherosclerosis; alopecia; damage to the skin due to UV light; lichen planus; atrophy of the skin; cataract; and graft rejections. Cell death can also be caused by surgery, drug therapy, chemical exposure or radiation exposure.

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can also be administered to a subject suffering from an acute disease, e.g., damage to an organ or tissue, e.g., a subject suffering from stroke or myocardial infarction or a subject suffering from a spinal cord injury. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used to repair an alcoholic's liver.

Cardiovascular Disease

In another embodiment, the invention provides a method for treating and/or preventing a cardiovascular disease by administering to a subject in need thereof a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein.

Cardiovascular diseases that can be treated or prevented using the sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein include cardiomyopathy or myocarditis; such as idiopathic cardiomyopathy, metabolic cardiomyopathy, alcoholic cardiomyopathy, drug-induced cardiomyopathy, ischemic cardiomyopathy, and hypertensive cardiomyopathy. Also treatable or preventable using compounds and methods described herein are atheromatous disorders of the major blood vessels (macrovascular disease) such as

the aorta, the coronary arteries, the carotid arteries, the cerebrovascular arteries, the renal arteries, the iliac arteries, the femoral arteries, and the popliteal arteries. Other vascular diseases that can be treated or prevented include those related to platelet aggregation, the retinal arterioles, the glomerular arterioles, the vasa nervorum, cardiac arterioles, and associated capillary beds of the eye, the kidney, the heart, and the central and peripheral nervous systems. The sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used for increasing HDL levels in plasma of an individual.

Yet other disorders that may be treated with sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein include restenosis, e.g., following coronary intervention, and disorders relating to an abnormal level of high density and low density cholesterol.

In one embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered as part of a combination therapeutic with another cardiovascular agent. In one embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered as part of a combination therapeutic with an anti-arrhythmia agent. In another embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered as part of a combination therapeutic with another cardiovascular agent.

Cell Death/Cancer

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered to subjects who have recently received or are likely to receive a dose of radiation or toxin. In one embodiment, the dose of radiation or toxin is received as part of a work-related or medical procedure, e.g., administered as a prophylactic measure. In another embodiment, the radiation or toxin exposure is received unintentionally. In such a case, the compound is preferably administered as soon as possible after the exposure to inhibit apoptosis and the subsequent development of acute radiation syndrome.

Sirtuin-modulating compounds may also be used for treating and/or preventing cancer. In certain embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for treating and/or

preventing cancer. Calorie restriction has been linked to a reduction in the incidence of age-related disorders including cancer. Accordingly, an increase in the level and/or activity of a sirtuin protein may be useful for treating and/or preventing the incidence of age-related disorders, such as, for example, cancer. Exemplary cancers that may be treated using a sirtuin-modulating compound are those of the brain and kidney; hormone-dependent cancers including breast, prostate, testicular, and ovarian cancers; lymphomas, and leukemias. In cancers associated with solid tumors, a modulating compound may be administered directly into the tumor. Cancer of blood cells, e.g., leukemia, can be treated by administering a modulating compound into the blood stream or into the bone marrow. Benign cell growth, e.g., warts, can also be treated. Other diseases that can be treated include autoimmune diseases, e.g., systemic lupus erythematosus, scleroderma, and arthritis, in which autoimmune cells should be removed. Viral infections such as herpes, HIV, adenovirus, and HTLV-1 associated malignant and benign disorders can also be treated by administration of sirtuin-modulating compound. Alternatively, cells can be obtained from a subject, treated ex vivo to remove certain undesirable cells, e.g., cancer cells, and administered back to the same or a different subject.

Chemotherapeutic agents may be co-administered with modulating compounds described herein as having anti-cancer activity, e.g., compounds that induce apoptosis, compounds that reduce lifespan or compounds that render cells sensitive to stress. Chemotherapeutic agents may be used by themselves with a sirtuin-modulating compound described herein as inducing cell death or reducing lifespan or increasing sensitivity to stress and/or in combination with other chemotherapeutics agents.

In addition to conventional chemotherapeutics, the sirtuin-modulating compounds described herein may also be used with antisense RNA, RNAi or other polynucleotides to inhibit the expression of the cellular components that contribute to unwanted cellular proliferation.

Combination therapies comprising sirtuin-modulating compounds and a conventional chemotherapeutic agent may be advantageous over combination therapies known in the art because the combination allows the conventional chemotherapeutic agent to exert greater effect at lower dosage. In a preferred

embodiment, the effective dose (ED_{50}) for a chemotherapeutic agent, or combination of conventional chemotherapeutic agents, when used in combination with a sirtuin-modulating compound is at least 2 fold less than the ED_{50} for the chemotherapeutic agent alone, and even more preferably at 5 fold, 10 fold or even 25 fold less. Conversely, the therapeutic index (TI) for such chemotherapeutic agent or combination of such chemotherapeutic agent when used in combination with a sirtuin-modulating compound described herein can be at least 2 fold greater than the TI for conventional chemotherapeutic regimen alone, and even more preferably at 5 fold, 10 fold or even 25 fold greater.

Neuronal Diseases/Disorders

In certain aspects, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be used to treat patients suffering from neurodegenerative diseases, and traumatic or mechanical injury to the central nervous system (CNS), spinal cord or peripheral nervous system (PNS). Neurodegenerative disease typically involves reductions in the mass and volume of the human brain, which may be due to the atrophy and/or death of brain cells, which are far more profound than those in a healthy person that are attributable to aging. Neurodegenerative diseases can evolve gradually, after a long period of normal brain function, due to progressive degeneration (e.g., nerve cell dysfunction and death) of specific brain regions. Alternatively, neurodegenerative diseases can have a quick onset, such as those associated with trauma or toxins. The actual onset of brain degeneration may precede clinical expression by many years. Examples of neurodegenerative diseases include, but are not limited to, Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's disease (HD), amyotrophic lateral sclerosis (ALS; Lou Gehrig's disease), diffuse Lewy body disease, chorea-acanthocytosis, primary lateral sclerosis, ocular diseases (ocular neuritis), chemotherapy-induced neuropathies (e.g., from vincristine, paclitaxel, bortezomib), diabetes-induced neuropathies and Friedreich's ataxia. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be used to treat these disorders and others as described below.

AD is a CNS disorder that results in memory loss, unusual behavior, personality changes, and a decline in thinking abilities. These losses are related to

the death of specific types of brain cells and the breakdown of connections and their supporting network (e.g. glial cells) between them. The earliest symptoms include loss of recent memory, faulty judgment, and changes in personality. PD is a CNS disorder that results in uncontrolled body movements, rigidity, tremor, and dyskinesia, and is associated with the death of brain cells in an area of the brain that produces dopamine. ALS (motor neuron disease) is a CNS disorder that attacks the motor neurons, components of the CNS that connect the brain to the skeletal muscles.

HD is another neurodegenerative disease that causes uncontrolled movements, loss of intellectual faculties, and emotional disturbance. Tay-Sachs disease and Sandhoff disease are glycolipid storage diseases where GM2 ganglioside and related glycolipidssubstrates glycolipids substrates for β-hexosaminidase accumulate in the nervous system and trigger acute neurodegeneration.

It is well-known that apoptosis plays a role in AIDS pathogenesis in the immune system. However, HIV-1 also induces neurological disease, which can be treated with sirtuin-modulating compounds of the invention.

Neuronal loss is also a salient feature of prion diseases, such as Creutzfeldt-Jakob disease in human, BSE in cattle (mad cow disease), Scrapie Disease in sheep and goats, and feline spongiform encephalopathy (FSE) in cats. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be useful for treating or preventing neuronal loss due to these prior diseases.

In another embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be used to treat or prevent any disease or disorder involving axonopathy. Distal axonopathy is a type of peripheral neuropathy that results from some metabolic or toxic derangement of peripheral nervous system (PNS) neurons. It is the most common response of nerves to metabolic or toxic disturbances, and as such may be caused by metabolic diseases such as diabetes, renal failure, deficiency syndromes such as malnutrition and alcoholism, or the effects of toxins or drugs. Those with distal axonopathies usually present with symmetrical glove-stocking sensori-motor disturbances. Deep tendon reflexes and autonomic nervous system (ANS) functions are also lost or diminished in affected areas.

Diabetic neuropathies are neuropathic disorders that are associated with diabetes mellitus. Relatively common conditions which may be associated with diabetic neuropathy include third nerve palsy; mononeuropathy; mononeuritis multiplex; diabetic amyotrophy; a painful polyneuropathy; autonomic neuropathy; and thoracoabdominal neuropathy.

Peripheral neuropathy is the medical term for damage to nerves of the peripheral nervous system, which may be caused either by diseases of the nerve or from the side-effects of systemic illness. Major causes of peripheral neuropathy include seizures, nutritional deficiencies, and HIV, though diabetes is the most likely cause.

In an exemplary embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be used to treat or prevent multiple sclerosis (MS), including relapsing MS and monosymptomatic MS, and other demyelinating conditions, such as, for example, chromic inflammatory demyelinating polyneuropathy (CIDP), or symptoms associated therewith.

In yet another embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be used to treat trauma to the nerves, including, trauma due to disease, injury (including surgical intervention), or environmental trauma (e.g., neurotoxins, alcoholism, etc.).

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be useful to prevent, treat, and alleviate symptoms of various PNS disorders. The term “peripheral neuropathy” encompasses a wide range of disorders in which the nerves outside of the brain and spinal cord—peripheral nerves—have been damaged. Peripheral neuropathy may also be referred to as peripheral neuritis, or if many nerves are involved, the terms polyneuropathy or polyneuritis may be used.

PNS diseases treatable with sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein include: diabetes, leprosy, Charcot-Marie-Tooth disease, Guillain-Barré syndrome and Brachial Plexus Neuropathies (diseases of the cervical and first thoracic roots, nerve trunks, cords, and peripheral nerve components of the brachial plexus.

In another embodiment, a sirtuin activating compound may be used to treat or prevent a polyglutamine disease. Exemplary polyglutamine diseases include Spinobulbar muscular atrophy (Kennedy disease), Huntington's Disease (HD), Dentatorubral-pallidoluysian atrophy (Haw River syndrome), Spinocerebellar ataxia type 1, Spinocerebellar ataxia type 2, Spinocerebellar ataxia type 3 (Machado-Joseph disease), Spinocerebellar ataxia type 6, Spinocerebellar ataxia type 7, and Spinocerebellar ataxia type 17.

In certain embodiments, the invention provides a method to treat a central nervous system cell to prevent damage in response to a decrease in blood flow to the cell. Typically the severity of damage that may be prevented will depend in large part on the degree of reduction in blood flow to the cell and the duration of the reduction. In one embodiment, apoptotic or necrotic cell death may be prevented. In still a further embodiment, ischemic-mediated damage, such as cytoxic edema or central nervous system tissue anoxemia, may be prevented. In each embodiment, the central nervous system cell may be a spinal cell or a brain cell.

Another aspect encompasses administrating a sirtuin activating compound to a subject to treat a central nervous system ischemic condition. A number of central nervous system ischemic conditions may be treated by the sirtuin activating compounds described herein. In one embodiment, the ischemic condition is a stroke that results in any type of ischemic central nervous system damage, such as apoptotic or necrotic cell death, cytoxic edema or central nervous system tissue anoxia. The stroke may impact any area of the brain or be caused by any etiology commonly known to result in the occurrence of a stroke. In one alternative of this embodiment, the stroke is a brain stem stroke. In another alternative of this embodiment, the stroke is a cerebellar stroke. In still another embodiment, the stroke is an embolic stroke. In yet another alternative, the stroke may be a hemorrhagic stroke. In a further embodiment, the stroke is a thrombotic stroke.

In yet another aspect, a sirtuin activating compound may be administered to reduce infarct size of the ischemic core following a central nervous system ischemic condition. Moreover, a sirtuin activating compound may also be beneficially administered to reduce the size of the ischemic penumbra or transitional zone following a central nervous system ischemic condition.

In one embodiment, a combination drug regimen may include drugs or compounds for the treatment or prevention of neurodegenerative disorders or secondary conditions associated with these conditions. Thus, a combination drug regimen may include one or more sirtuin activators and one or more anti-neurodegeneration agents.

Blood Coagulation Disorders

In other aspects, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be used to treat or prevent blood coagulation disorders (or hemostatic disorders). As used interchangeably herein, the terms "hemostasis", "blood coagulation," and "blood clotting" refer to the control of bleeding, including the physiological properties of vasoconstriction and coagulation. Blood coagulation assists in maintaining the integrity of mammalian circulation after injury, inflammation, disease, congenital defect, dysfunction or other disruption. Further, the formation of blood clots does not only limit bleeding in case of an injury (hemostasis), but may lead to serious organ damage and death in the context of atherosclerotic diseases by occlusion of an important artery or vein. Thrombosis is thus blood clot formation at the wrong time and place.

Accordingly, the present invention provides anticoagulation and antithrombotic treatments aiming at inhibiting the formation of blood clots in order to prevent or treat blood coagulation disorders, such as myocardial infarction, stroke, loss of a limb by peripheral artery disease or pulmonary embolism.

As used interchangeably herein, "modulating or modulation of hemostasis" and "regulating or regulation of hemostasis" includes the induction (e.g., stimulation or increase) of hemostasis, as well as the inhibition (e.g., reduction or decrease) of hemostasis.

In one aspect, the invention provides a method for reducing or inhibiting hemostasis in a subject by administering a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. The compositions and methods disclosed herein are useful for the treatment or prevention of thrombotic disorders. As used herein, the term "thrombotic disorder" includes any disorder or condition characterized by excessive or unwanted coagulation or hemostatic activity, or a hypercoagulable state. Thrombotic disorders include diseases or disorders involving

platelet adhesion and thrombus formation, and may manifest as an increased propensity to form thromboses, e.g., an increased number of thromboses, thrombosis at an early age, a familial tendency towards thrombosis, and thrombosis at unusual sites.

In another embodiment, a combination drug regimen may include drugs or compounds for the treatment or prevention of blood coagulation disorders or secondary conditions associated with these conditions. Thus, a combination drug regimen may include one or more sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein and one or more anti-coagulation or anti-thrombosis agents.

Weight Control

In another aspect, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for treating or preventing weight gain or obesity in a subject. For example, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used, for example, to treat or prevent hereditary obesity, dietary obesity, hormone related obesity, obesity related to the administration of medication, to reduce the weight of a subject, or to reduce or prevent weight gain in a subject. A subject in need of such a treatment may be a subject who is obese, likely to become obese, overweight, or likely to become overweight. Subjects who are likely to become obese or overweight can be identified, for example, based on family history, genetics, diet, activity level, medication intake, or various combinations thereof.

In yet other embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered to subjects suffering from a variety of other diseases and conditions that may be treated or prevented by promoting weight loss in the subject. Such diseases include, for example, high blood pressure, hypertension, high blood cholesterol, dyslipidemia, type 2 diabetes, insulin resistance, glucose intolerance, hyperinsulinemia, coronary heart disease, angina pectoris, congestive heart failure, stroke, gallstones, cholecystitis and cholelithiasis, gout, osteoarthritis, obstructive sleep apnea and respiratory problems, some types of cancer (such as endometrial, breast, prostate, and colon), complications of pregnancy, poor female reproductive health (such as menstrual irregularities,

infertility, irregular ovulation), bladder control problems (such as stress incontinence); uric acid nephrolithiasis; psychological disorders (such as depression, eating disorders, distorted body image, and low self esteem). Finally, patients with AIDS can develop lipodystrophy or insulin resistance in response to combination therapies for AIDS.

In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for inhibiting adipogenesis or fat cell differentiation, whether in vitro or in vivo. Such methods may be used for treating or preventing obesity.

In other embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for reducing appetite and/or increasing satiety, thereby causing weight loss or avoidance of weight gain. A subject in need of such a treatment may be a subject who is overweight, obese or a subject likely to become overweight or obese. The method may comprise administering daily or, every other day, or once a week, a dose, e.g., in the form of a pill, to a subject. The dose may be an "appetite reducing dose."

In an exemplary embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered as a combination therapy for treating or preventing weight gain or obesity. For example, one or more sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered in combination with one or more anti-obesity agents.

In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered to reduce drug-induced weight gain. For example, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be administered as a combination therapy with medications that may stimulate appetite or cause weight gain, in particular, weight gain due to factors other than water retention.

Metabolic Disorders/Diabetes

In another aspect, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for treating or preventing a metabolic

disorder, such as insulin-resistance, a pre-diabetic state, type II diabetes, and/or complications thereof. Administration of a sirtuin-modulating compounds that increases the level and/or activity of a sirtuin protein may increase insulin sensitivity and/or decrease insulin levels in a subject. A subject in need of such a treatment may be a subject who has insulin resistance or other precursor symptom of type II diabetes, who has type II diabetes, or who is likely to develop any of these conditions. For example, the subject may be a subject having insulin resistance, e.g., having high circulating levels of insulin and/or associated conditions, such as hyperlipidemia, dyslipogenesis, hypercholesterolemia, impaired glucose tolerance, high blood glucose sugar level, other manifestations of syndrome X, hypertension, atherosclerosis and lipodystrophy.

In an exemplary embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered as a combination therapy for treating or preventing a metabolic disorder. For example, one or more sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered in combination with one or more anti-diabetic agents.

Inflammatory Diseases

In other aspects, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be used to treat or prevent a disease or disorder associated with inflammation. Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered prior to the onset of, at, or after the initiation of inflammation. When used prophylactically, the compounds are preferably provided in advance of any inflammatory response or symptom. Administration of the compounds may prevent or attenuate inflammatory responses or symptoms.

In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used to treat or prevent allergies and respiratory conditions, including asthma, bronchitis, pulmonary fibrosis, allergic rhinitis, oxygen toxicity, emphysema, chronic bronchitis, acute respiratory distress syndrome, and any chronic obstructive pulmonary disease (COPD). The compounds may be used to treat chronic hepatitis infection, including hepatitis B and hepatitis C.

Additionally, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used to treat autoimmune diseases, and/or inflammation associated with autoimmune diseases such as arthritis, including rheumatoid arthritis, psoriatic arthritis, and ankylosing spondylitis, as well as organ-tissue autoimmune diseases (e.g., Raynaud's syndrome), ulcerative colitis, Crohn's disease, oral mucositis, scleroderma, myasthenia gravis, transplant rejection, endotoxin shock, sepsis, psoriasis, eczema, dermatitis, multiple sclerosis, autoimmune thyroiditis, uveitis, systemic lupus erythematosis, Addison's disease, autoimmune polyglandular disease (also known as autoimmune polyglandular syndrome), and Grave's disease.

In certain embodiments, one or more sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be taken alone or in combination with other compounds useful for treating or preventing inflammation.

Flushing

In another aspect, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for reducing the incidence or severity of flushing and/or hot flashes which are symptoms of a disorder. For instance, the subject method includes the use of sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein, alone or in combination with other agents, for reducing incidence or severity of flushing and/or hot flashes in cancer patients. In other embodiments, the method provides for the use of sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein to reduce the incidence or severity of flushing and/or hot flashes in menopausal and post-menopausal woman.

In another aspect, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used as a therapy for reducing the incidence or severity of flushing and/or hot flashes which are side-effects of another drug therapy, e.g., drug-induced flushing. In certain embodiments, a method for treating and/or preventing drug-induced flushing comprises administering to a patient in need thereof a formulation comprising at least one flushing inducing compound and at least one sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein. In other embodiments, a method for treating drug

induced flushing comprises separately administering one or more compounds that induce flushing and one or more sirtuin-modulating compounds, e.g., wherein the sirtuin-modulating compound and flushing inducing agent have not been formulated in the same compositions. When using separate formulations, the sirtuin-modulating compound may be administered (1) at the same as administration of the flushing inducing agent, (2) intermittently with the flushing inducing agent, (3) staggered relative to administration of the flushing inducing agent, (4) prior to administration of the flushing inducing agent, (5) subsequent to administration of the flushing inducing agent, and (6) various combination thereof. Exemplary flushing inducing agents include, for example, niacin, raloxifene, antidepressants, anti-psychotics, chemotherapeutics, calcium channel blockers, and antibiotics.

In one embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used to reduce flushing side effects of a vasodilator or an antilipemic agent (including anticholesteremic agents and lipotropic agents). In an exemplary embodiment, a sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein may be used to reduce flushing associated with the administration of niacin.

In another embodiment, the invention provides a method for treating and/or preventing hyperlipidemia with reduced flushing side effects. In another representative embodiment, the method involves the use of sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein to reduce flushing side effects of raloxifene. In another representative embodiment, the method involves the use of sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein to reduce flushing side effects of antidepressants or anti-psychotic agent. For instance, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be used in conjunction (administered separately or together) with a serotonin reuptake inhibitor, or a 5HT2 receptor antagonist.

In certain embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used as part of a treatment with a serotonin reuptake inhibitor (SRI) to reduce flushing. In still another representative embodiment, sirtuin-modulating compounds that increase the level and/or activity of

a sirtuin protein may be used to reduce flushing side effects of chemotherapeutic agents, such as cyclophosphamide and tamoxifen.

In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used to reduce flushing side effects of calcium channel blockers, such as amlodipine.

In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used to reduce flushing side effects of antibiotics. For example, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be used in combination with levofloxacin.

Ocular Disorders

One aspect of the present invention is a method for inhibiting, reducing or otherwise treating vision impairment by administering to a patient a therapeutic dosage of sirtuin modulator selected from a compound disclosed herein, or a pharmaceutically acceptable salt, prodrug or a metabolic derivative thereof.

In certain aspects of the invention, the vision impairment is caused by damage to the optic nerve or central nervous system. In particular embodiments, optic nerve damage is caused by high intraocular pressure, such as that created by glaucoma. In other particular embodiments, optic nerve damage is caused by swelling of the nerve, which is often associated with an infection or an immune (e.g., autoimmune) response such as in optic neuritis.

In certain aspects of the invention, the vision impairment is caused by retinal damage. In particular embodiments, retinal damage is caused by disturbances in blood flow to the eye (e.g., arteriosclerosis, vasculitis). In particular embodiments, retinal damage is caused by disruption of the macula (e.g., exudative or non-exudative macular degeneration).

Exemplary retinal diseases include Exudative Age Related Macular Degeneration, Nonexudative Age Related Macular Degeneration, Retinal Electronic Prosthesis and RPE Transplantation Age Related Macular Degeneration, Acute Multifocal Placoid Pigment Epitheliopathy, Acute Retinal Necrosis, Best Disease, Branch Retinal Artery Occlusion, Branch Retinal Vein Occlusion, Cancer Associated and Related Autoimmune Retinopathies, Central Retinal Artery

Occlusion, Central Retinal Vein Occlusion, Central Serous Chorioretinopathy, Eales Disease, Epimacular Membrane, Lattice Degeneration, Macroaneurysm, Diabetic Macular Edema, Irvine-Gass Macular Edema, Macular Hole, Subretinal Neovascular Membranes, Diffuse Unilateral Subacute Neuroretinitis, Nonpseudophakic Cystoid Macular Edema, Presumed Ocular Histoplasmosis Syndrome, Exudative Retinal Detachment, Postoperative Retinal Detachment, Proliferative Retinal Detachment, Rhegmatogenous Retinal Detachment, Tractional Retinal Detachment, Retinitis Pigmentosa, CMV Retinitis, Retinoblastoma, Retinopathy of Prematurity, Birdshot Retinopathy, Background Diabetic Retinopathy, Proliferative Diabetic Retinopathy, Hemoglobinopathies Retinopathy, Purtscher Retinopathy, Valsalva Retinopathy, Juvenile Retinoschisis, Senile Retinoschisis, Terson Syndrome and White Dot Syndromes.

Other exemplary diseases include ocular bacterial infections (e.g. conjunctivitis, keratitis, tuberculosis, syphilis, gonorrhea), viral infections (e.g. Ocular Herpes Simplex Virus, Varicella Zoster Virus, Cytomegalovirus retinitis, Human Immunodeficiency Virus (HIV)) as well as progressive outer retinal necrosis secondary to HIV or other HIV-associated and other immunodeficiency-associated ocular diseases. In addition, ocular diseases include fungal infections (e.g. Candida choroiditis, histoplasmosis), protozoal infections (e.g. toxoplasmosis) and others such as ocular toxocariasis and sarcoidosis.

One aspect of the invention is a method for inhibiting, reducing or treating vision impairment in a subject undergoing treatment with a chemotherapeutic drug (e.g., a neurotoxic drug, a drug that raises intraocular pressure such as a steroid), by administering to the subject in need of such treatment a therapeutic dosage of a sirtuin modulator disclosed herein.

Another aspect of the invention is a method for inhibiting, reducing or treating vision impairment in a subject undergoing surgery, including ocular or other surgeries performed in the prone position such as spinal cord surgery, by administering to the subject in need of such treatment a therapeutic dosage of a sirtuin modulator disclosed herein. Ocular surgeries include cataract, iridotomy and lens replacements.

Another aspect of the invention is the treatment, including inhibition and prophylactic treatment, of age related ocular diseases include cataracts, dry eye, age-related macular degeneration (AMD), retinal damage and the like, by administering to the subject in need of such treatment a therapeutic dosage of a sirtuin modulator disclosed herein.

Another aspect of the invention is the prevention or treatment of damage to the eye caused by stress, chemical insult or radiation, by administering to the subject in need of such treatment a therapeutic dosage of a sirtuin modulator disclosed herein. Radiation or electromagnetic damage to the eye can include that caused by CRT's or exposure to sunlight or UV.

In one embodiment, a combination drug regimen may include drugs or compounds for the treatment or prevention of ocular disorders or secondary conditions associated with these conditions. Thus, a combination drug regimen may include one or more sirtuin activators and one or more therapeutic agents for the treatment of an ocular disorder.

In one embodiment, a sirtuin modulator can be administered in conjunction with a therapy for reducing intraocular pressure. In another embodiment, a sirtuin modulator can be administered in conjunction with a therapy for treating and/or preventing glaucoma. In yet another embodiment, a sirtuin modulator can be administered in conjunction with a therapy for treating and/or preventing optic neuritis. In one embodiment, a sirtuin modulator can be administered in conjunction with a therapy for treating and/or preventing CMV Retinopathy. In another embodiment, a sirtuin modulator can be administered in conjunction with a therapy for treating and/or preventing multiple sclerosis.

Mitochondrial-Associated Diseases and Disorders

In certain embodiments, the invention provides methods for treating diseases or disorders that would benefit from increased mitochondrial activity. The methods involve administering to a subject in need thereof a therapeutically effective amount of a sirtuin activating compound. Increased mitochondrial activity refers to increasing activity of the mitochondria while maintaining the overall numbers of mitochondria (e.g., mitochondrial mass), increasing the numbers of mitochondria thereby increasing mitochondrial activity (e.g., by stimulating mitochondrial

biogenesis), or combinations thereof. In certain embodiments, diseases and disorders that would benefit from increased mitochondrial activity include diseases or disorders associated with mitochondrial dysfunction.

In certain embodiments, methods for treating diseases or disorders that would benefit from increased mitochondrial activity may comprise identifying a subject suffering from a mitochondrial dysfunction. Methods for diagnosing a mitochondrial dysfunction may involve molecular genetic, pathologic and/or biochemical analyses. Diseases and disorders associated with mitochondrial dysfunction include diseases and disorders in which deficits in mitochondrial respiratory chain activity contribute to the development of pathophysiology of such diseases or disorders in a mammal. Diseases or disorders that would benefit from increased mitochondrial activity generally include for example, diseases in which free radical mediated oxidative injury leads to tissue degeneration, diseases in which cells inappropriately undergo apoptosis, and diseases in which cells fail to undergo apoptosis.

In certain embodiments, the invention provides methods for treating a disease or disorder that would benefit from increased mitochondrial activity that involves administering to a subject in need thereof one or more sirtuin activating compounds in combination with another therapeutic agent such as, for example, an agent useful for treating mitochondrial dysfunction or an agent useful for reducing a symptom associated with a disease or disorder involving mitochondrial dysfunction.

In exemplary embodiments, the invention provides methods for treating diseases or disorders that would benefit from increased mitochondrial activity by administering to a subject a therapeutically effective amount of a sirtuin activating compound. Exemplary diseases or disorders include, for example, neuromuscular disorders (e.g., Friedreich's Ataxia, muscular dystrophy, multiple sclerosis, etc.), disorders of neuronal instability (e.g., seizure disorders, migraine, etc.), developmental delay, neurodegenerative disorders (e.g., Alzheimer's Disease, Parkinson's Disease, amyotrophic lateral sclerosis, etc.), ischemia, renal tubular acidosis, age-related neurodegeneration and cognitive decline, chemotherapy fatigue, age-related or chemotherapy-induced menopause or irregularities of menstrual cycling or ovulation, mitochondrial myopathies, mitochondrial damage

(e.g., calcium accumulation, excitotoxicity, nitric oxide exposure, hypoxia, etc.), and mitochondrial deregulation.

Muscular dystrophy refers to a family of diseases involving deterioration of neuromuscular structure and function, often resulting in atrophy of skeletal muscle and myocardial dysfunction, such as Duchenne muscular dystrophy. In certain embodiments, sirtuin activating compounds may be used for reducing the rate of decline in muscular functional capacities and for improving muscular functional status in patients with muscular dystrophy.

In certain embodiments, sirtuin modulating compounds may be useful for treatment mitochondrial myopathies. Mitochondrial myopathies range from mild, slowly progressive weakness of the extraocular muscles to severe, fatal infantile myopathies and multisystem encephalomyopathies. Some syndromes have been defined, with some overlap between them. Established syndromes affecting muscle include progressive external ophthalmoplegia, the Kearns-Sayre syndrome (with ophthalmoplegia, pigmentary retinopathy, cardiac conduction defects, cerebellar ataxia, and sensorineural deafness), the MELAS syndrome (mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes), the MERFF syndrome (myoclonic epilepsy and ragged red fibers), limb-girdle distribution weakness, and infantile myopathy (benign or severe and fatal).

In certain embodiments, sirtuin activating compounds may be useful for treating patients suffering from toxic damage to mitochondria, such as, toxic damage due to calcium accumulation, excitotoxicity, nitric oxide exposure, drug induced toxic damage, or hypoxia.

In certain embodiments, sirtuin activating compounds may be useful for treating diseases or disorders associated with mitochondrial deregulation.

Muscle Performance

In other embodiments, the invention provides methods for enhancing muscle performance by administering a therapeutically effective amount of a sirtuin activating compound. For example, sirtuin activating compounds may be useful for improving physical endurance (e.g., ability to perform a physical task such as exercise, physical labor, sports activities, etc.), inhibiting or retarding physical

fatigues, enhancing blood oxygen levels, enhancing energy in healthy individuals, enhance working capacity and endurance, reducing muscle fatigue, reducing stress, enhancing cardiac and cardiovascular function, improving sexual ability, increasing muscle ATP levels, and/or reducing lactic acid in blood. In certain embodiments, the methods involve administering an amount of a sirtuin activating compound that increase mitochondrial activity, increase mitochondrial biogenesis, and/or increase mitochondrial mass.

Sports performance refers to the ability of the athlete's muscles to perform when participating in sports activities. Enhanced sports performance, strength, speed and endurance are measured by an increase in muscular contraction strength, increase in amplitude of muscle contraction, shortening of muscle reaction time between stimulation and contraction. Athlete refers to an individual who participates in sports at any level and who seeks to achieve an improved level of strength, speed and endurance in their performance, such as, for example, body builders, bicyclists, long distance runners, short distance runners, etc. Enhanced sports performance in manifested by the ability to overcome muscle fatigue, ability to maintain activity for longer periods of time, and have a more effective workout.

In the arena of athlete muscle performance, it is desirable to create conditions that permit competition or training at higher levels of resistance for a prolonged period of time.

It is contemplated that the methods of the present invention will also be effective in the treatment of muscle related pathological conditions, including acute sarcopenia, for example, muscle atrophy and/or cachexia associated with burns, bed rest, limb immobilization, or major thoracic, abdominal, and/or orthopedic surgery.

In certain embodiments, the invention provides novel dietary compositions comprising sirtuin modulators, a method for their preparation, and a method of using the compositions for improvement of sports performance. Accordingly, provided are therapeutic compositions, foods and beverages that have actions of improving physical endurance and/or inhibiting physical fatigues for those people involved in broadly-defined exercises including sports requiring endurance and labors requiring repeated muscle exertions. Such dietary compositions may additional comprise electrolytes, caffeine, vitamins, carbohydrates, etc.

Other Uses

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for treating or preventing viral infections (such as infections by influenza, herpes or papilloma virus) or as antifungal agents. In certain embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered as part of a combination drug therapy with another therapeutic agent for the treatment of viral diseases. In another embodiment, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be administered as part of a combination drug therapy with another anti-fungal agent.

Subjects that may be treated as described herein include eukaryotes, such as mammals, e.g., humans, ovines, bovines, equines, porcines, canines, felines, non-human primate, mice, and rats. Cells that may be treated include eukaryotic cells, e.g., from a subject described above, or plant cells, yeast cells and prokaryotic cells, e.g., bacterial cells. For example, modulating compounds may be administered to farm animals to improve their ability to withstand farming conditions longer.

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used to increase lifespan, stress resistance, and resistance to apoptosis in plants. In one embodiment, a compound is applied to plants, e.g., on a periodic basis, or to fungi. In another embodiment, plants are genetically modified to produce a compound. In another embodiment, plants and fruits are treated with a compound prior to picking and shipping to increase resistance to damage during shipping. Plant seeds may also be contacted with compounds described herein, e.g., to preserve them.

In other embodiments, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may be used for modulating lifespan in yeast cells. Situations in which it may be desirable to extend the lifespan of yeast cells include any process in which yeast is used, e.g., the making of beer, yogurt, and bakery items, e.g., bread. Use of yeast having an extended lifespan can result in using less yeast or in having the yeast be active for longer periods of time. Yeast or other mammalian cells used for recombinantly producing proteins may also be treated as described herein.

Sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used to increase lifespan, stress resistance and resistance to apoptosis in insects. In this embodiment, compounds would be applied to useful insects, e.g., bees and other insects that are involved in pollination of plants. In a specific embodiment, a compound would be applied to bees involved in the production of honey. Generally, the methods described herein may be applied to any organism, e.g., eukaryote, which may have commercial importance. For example, they can be applied to fish (aquaculture) and birds (e.g., chicken and fowl).

Higher doses of sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein may also be used as a pesticide by interfering with the regulation of silenced genes and the regulation of apoptosis during development. In this embodiment, a compound may be applied to plants using a method known in the art that ensures the compound is bio-available to insect larvae, and not to plants.

At least in view of the link between reproduction and longevity, sirtuin-modulating compounds that increase the level and/or activity of a sirtuin protein can be applied to affect the reproduction of organisms such as insects, animals and microorganisms.

4. Assays

Yet other methods contemplated herein include screening methods for identifying compounds or agents that modulate sirtuins. An agent may be a nucleic acid, such as an aptamer. Assays may be conducted in a cell based or cell free format. For example, an assay may comprise incubating (or contacting) a sirtuin with a test agent under conditions in which a sirtuin can be modulated by an agent known to modulate the sirtuin, and monitoring or determining the level of modulation of the sirtuin in the presence of the test agent relative to the absence of the test agent. The level of modulation of a sirtuin can be determined by determining its ability to deacetylate a substrate. Exemplary substrates are acetylated peptides which can be obtained from BIOMOL (Plymouth Meeting, PA). Preferred substrates include peptides of p53, such as those comprising an acetylated K382. A particularly preferred substrate is the Fluor de Lys-SIRT1 (BIOMOL), i.e., the acetylated peptide

Arg-His-Lys-Lys. Other substrates are peptides from human histones H3 and H4 or an acetylated amino acid. Substrates may be fluorogenic. The sirtuin may be SIRT1, Sir2, SIRT3, or a portion thereof. For example, recombinant SIRT1 can be obtained from BIOMOL. The reaction may be conducted for about 30 minutes and stopped, e.g., with nicotinamide. The HDAC fluorescent activity assay/drug discovery kit (AK-500, BIOMOL Research Laboratories) may be used to determine the level of acetylation. Similar assays are described in Bitterman et al. (2002) J. Biol. Chem. 277:45099. The level of modulation of the sirtuin in an assay may be compared to the level of modulation of the sirtuin in the presence of one or more (separately or simultaneously) compounds described herein, which may serve as positive or negative controls. Sirtuins for use in the assays may be full length sirtuin proteins or portions thereof. Since it has been shown herein that activating compounds appear to interact with the N-terminus of SIRT1, proteins for use in the assays include N-terminal portions of sirtuins, e.g., about amino acids 1-176 or 1-255 of SIRT1; about amino acids 1-174 or 1-252 of Sir2.

In one embodiment, a screening assay comprises (i) contacting a sirtuin with a test agent and an acetylated substrate under conditions appropriate for the sirtuin to deacetylate the substrate in the absence of the test agent ; and (ii) determining the level of acetylation of the substrate, wherein a lower level of acetylation of the substrate in the presence of the test agent relative to the absence of the test agent indicates that the test agent stimulates deacetylation by the sirtuin, whereas a higher level of acetylation of the substrate in the presence of the test agent relative to the absence of the test agent indicates that the test agent inhibits deacetylation by the sirtuin.

Methods for identifying an agent that modulates, e.g., stimulates, sirtuins *in vivo* may comprise (i) contacting a cell with a test agent and a substrate that is capable of entering a cell in the presence of an inhibitor of class I and class II HDACs under conditions appropriate for the sirtuin to deacetylate the substrate in the absence of the test agent ; and (ii) determining the level of acetylation of the substrate, wherein a lower level of acetylation of the substrate in the presence of the test agent relative to the absence of the test agent indicates that the test agent stimulates deacetylation by the sirtuin, whereas a higher level of acetylation of the

substrate in the presence of the test agent relative to the absence of the test agent indicates that the test agent inhibits deacetylation by the sirtuin. A preferred substrate is an acetylated peptide, which is also preferably fluorogenic, as further described herein. The method may further comprise lysing the cells to determine the level of acetylation of the substrate. Substrates may be added to cells at a concentration ranging from about 1μM to about 10mM, preferably from about 10μM to 1mM, even more preferably from about 100μM to 1mM, such as about 200μM. A preferred substrate is an acetylated lysine, e.g., ε-acetyl lysine (Fluor de Lys, FdL) or Fluor de Lys-SIRT1. A preferred inhibitor of class I and class II HDACs is trichostatin A (TSA), which may be used at concentrations ranging from about 0.01 to 100μM, preferably from about 0.1 to 10μM, such as 1μM. Incubation of cells with the test compound and the substrate may be conducted for about 10 minutes to 5 hours, preferably for about 1-3 hours. Since TSA inhibits all class I and class II HDACs, and that certain substrates, e.g., Fluor de Lys, is a poor substrate for SIRT2 and even less a substrate for SIRT3-7, such an assay may be used to identify modulators of SIRT1 *in vivo*.

5. Pharmaceutical Compositions

The sirtuin-modulating compounds described herein may be formulated in a conventional manner using one or more physiologically or pharmaceutically acceptable carriers or excipients. For example, sirtuin-modulating compounds and their pharmaceutically acceptable salts and solvates may be formulated for administration by, for example, injection (e.g. SubQ, IM, IP), inhalation or insufflation (either through the mouth or the nose) or oral, buccal, sublingual, transdermal, nasal, parenteral or rectal administration. In one embodiment, a sirtuin-modulating compound may be administered locally, at the site where the target cells are present, i.e., in a specific tissue, organ, or fluid (e.g., blood, cerebrospinal fluid, etc.).

Sirtuin-modulating compounds can be formulated for a variety of modes of administration, including systemic and topical or localized administration. Techniques and formulations generally may be found in Remington's Pharmaceutical Sciences, Meade Publishing Co., Easton, PA. For parenteral administration, injection is preferred, including intramuscular, intravenous,

intraperitoneal, and subcutaneous. For injection, the compounds can be formulated in liquid solutions, preferably in physiologically compatible buffers such as Hank's solution or Ringer's solution. In addition, the compounds may be formulated in solid form and redissolved or suspended immediately prior to use. Lyophilized forms are also included.

For oral administration, the pharmaceutical compositions may take the form of, for example, tablets, lozenges, or capsules prepared by conventional means with pharmaceutically acceptable excipients such as binding agents (e.g., pregelatinized maize starch, polyvinylpyrrolidone or hydroxypropyl methylcellulose); fillers (e.g., lactose, microcrystalline cellulose or calcium hydrogen phosphate); lubricants (e.g., magnesium stearate, talc or silica); disintegrants (e.g., potato starch or sodium starch glycolate); or wetting agents (e.g., sodium lauryl sulphate). The tablets may be coated by methods well known in the art. Liquid preparations for oral administration may take the form of, for example, solutions, syrups or suspensions, or they may be presented as a dry product for constitution with water or other suitable vehicle before use. Such liquid preparations may be prepared by conventional means with pharmaceutically acceptable additives such as suspending agents (e.g., sorbitol syrup, cellulose derivatives or hydrogenated edible fats); emulsifying agents (e.g., lecithin or acacia); non-aqueous vehicles (e.g., almond oil, oily esters, ethyl alcohol or fractionated vegetable oils); and preservatives (e.g., methyl or propyl-p-hydroxybenzoates or sorbic acid). The preparations may also contain buffer salts, flavoring, coloring and sweetening agents as appropriate. Preparations for oral administration may be suitably formulated to give controlled release of the active compound.

For administration by inhalation (e.g., pulmonary delivery), sirtuin-modulating compounds may be conveniently delivered in the form of an aerosol spray presentation from pressurized packs or a nebuliser, with the use of a suitable propellant, e.g., dichlorodifluoromethane, trichlorofluoromethane, dichlorotetrafluoroethane, carbon dioxide or other suitable gas. In the case of a pressurized aerosol the dosage unit may be determined by providing a valve to deliver a metered amount. Capsules and cartridges of e.g., gelatin, for use in an

inhaler or insufflator may be formulated containing a powder mix of the compound and a suitable powder base such as lactose or starch.

Sirtuin-modulating compounds may be formulated for parenteral administration by injection, e.g., by bolus injection or continuous infusion. Formulations for injection may be presented in unit dosage form, e.g., in ampoules or in multi-dose containers, with an added preservative. The compositions may take such forms as suspensions, solutions or emulsions in oily or aqueous vehicles, and may contain formulatory agents such as suspending, stabilizing and/or dispersing agents. Alternatively, the active ingredient may be in powder form for constitution with a suitable vehicle, e.g., sterile pyrogen-free water, before use.

Sirtuin-modulating compounds may also be formulated in rectal compositions such as suppositories or retention enemas, e.g., containing conventional suppository bases such as cocoa butter or other glycerides.

In addition to the formulations described previously, sirtuin-modulating compounds may also be formulated as a depot preparation. Such long acting formulations may be administered by implantation (for example subcutaneously or intramuscularly) or by intramuscular injection. Thus, for example, sirtuin-modulating compounds may be formulated with suitable polymeric or hydrophobic materials (for example as an emulsion in an acceptable oil) or ion exchange resins, or as sparingly soluble derivatives, for example, as a sparingly soluble salt. Controlled release formula also includes patches.

In certain embodiments, the compounds described herein can be formulated for delivery to the central nervous system (CNS) (reviewed in Begley, Pharmacology & Therapeutics 104: 29-45 (2004)). Conventional approaches for drug delivery to the CNS include: neurosurgical strategies (e.g., intracerebral injection or intracerebroventricular infusion); molecular manipulation of the agent (e.g., production of a chimeric fusion protein that comprises a transport peptide that has an affinity for an endothelial cell surface molecule in combination with an agent that is itself incapable of crossing the BBB) in an attempt to exploit one of the endogenous transport pathways of the BBB; pharmacological strategies designed to increase the lipid solubility of an agent (e.g., conjugation of water-soluble agents to lipid or cholesterol carriers); and the transitory disruption of the integrity of the BBB

by hyperosmotic disruption (resulting from the infusion of a mannitol solution into the carotid artery or the use of a biologically active agent such as an angiotensin peptide).

Liposomes are a further drug delivery system which is easily injectable. Accordingly, in the method of invention the active compounds can also be administered in the form of a liposome delivery system. Liposomes are well-known by a person skilled in the art. Liposomes can be formed from a variety of phospholipids, such as cholesterol, stearylamine of phosphatidylcholines. Liposomes being usable for the method of invention encompass all types of liposomes including, but not limited to, small unilamellar vesicles, large unilamellar vesicles and multilamellar vesicles.

Another way to produce a formulation, particularly a solution, of a sirtuin modulator such as resveratrol or a derivative thereof, is through the use of cyclodextrin. By cyclodextrin is meant α-, β-, or γ-cyclodextrin. Cyclodextrins are described in detail in Pitha et al., U.S. Pat. No. 4,727,064, which is incorporated herein by reference. Cyclodextrins are cyclic oligomers of glucose; these compounds form inclusion complexes with any drug whose molecule can fit into the lipophile-seeking cavities of the cyclodextrin molecule.

Rapidly disintegrating or dissolving dosage forms are useful for the rapid absorption, particularly buccal and sublingual absorption, of pharmaceutically active agents. Fast melt dosage forms are beneficial to patients, such as aged and pediatric patients, who have difficulty in swallowing typical solid dosage forms, such as caplets and tablets. Additionally, fast melt dosage forms circumvent drawbacks associated with, for example, chewable dosage forms, wherein the length of time an active agent remains in a patient's mouth plays an important role in determining the amount of taste masking and the extent to which a patient may object to throat grittiness of the active agent.

Pharmaceutical compositions (including cosmetic preparations) may comprise from about 0.00001 to 100% such as from 0.001 to 10% or from 0.1% to 5% by weight of one or more sirtuin-modulating compounds described herein. In other embodiments, the pharmaceutical composition comprises: (i) 0.05 to 1000 mg

of the compounds of the invention, or a pharmaceutically acceptable salt thereof, and (ii) 0.1 to 2 grams of one or more pharmaceutically acceptable excipients.

In one embodiment, a sirtuin-modulating compound described herein, is incorporated into a topical formulation containing a topical carrier that is generally suited to topical drug administration and comprising any such material known in the art. The topical carrier may be selected so as to provide the composition in the desired form, e.g., as an ointment, lotion, cream, microemulsion, gel, oil, solution, or the like, and may be comprised of a material of either naturally occurring or synthetic origin. It is preferable that the selected carrier not adversely affect the active agent or other components of the topical formulation. Examples of suitable topical carriers for use herein include water, alcohols and other nontoxic organic solvents, glycerin, mineral oil, silicone, petroleum jelly, lanolin, fatty acids, vegetable oils, parabens, waxes, and the like.

Formulations may be colorless, odorless ointments, lotions, creams, microemulsions and gels.

Sirtuin-modulating compounds may be incorporated into ointments, which generally are semisolid preparations which are typically based on petrolatum or other petroleum derivatives. The specific ointment base to be used, as will be appreciated by those skilled in the art, is one that will provide for optimum drug delivery, and, preferably, will provide for other desired characteristics as well, e.g., emolliency or the like. As with other carriers or vehicles, an ointment base should be inert, stable, nonirritating and nonsensitizing.

Sirtuin-modulating compounds may be incorporated into lotions, which generally are preparations to be applied to the skin surface without friction, and are typically liquid or semiliquid preparations in which solid particles, including the active agent, are present in a water or alcohol base. Lotions are usually suspensions of solids, and may comprise a liquid oily emulsion of the oil-in-water type.

Sirtuin-modulating compounds may be incorporated into creams, which generally are viscous liquid or semisolid emulsions, either oil-in-water or water-in-oil. Cream bases are water-washable, and contain an oil phase, an emulsifier and an aqueous phase. The oil phase is generally comprised of petrolatum and a fatty alcohol such as cetyl or stearyl alcohol; the aqueous phase usually, although not

necessarily, exceeds the oil phase in volume, and generally contains a humectant. The emulsifier in a cream formulation, as explained in Remington 's, *supra*, is generally a nonionic, anionic, cationic or amphoteric surfactant.

Sirtuin-modulating compounds may be incorporated into microemulsions, which generally are thermodynamically stable, isotropically clear dispersions of two immiscible liquids, such as oil and water, stabilized by an interfacial film of surfactant molecules (Encyclopedia of Pharmaceutical Technology (New York: Marcel Dekker, 1992), volume 9).

Sirtuin-modulating compounds may be incorporated into gel formulations, which generally are semisolid systems consisting of either suspensions made up of small inorganic particles (two-phase systems) or large organic molecules distributed substantially uniformly throughout a carrier liquid (single phase gels). Although gels commonly employ aqueous carrier liquid, alcohols and oils can be used as the carrier liquid as well.

Other active agents may also be included in formulations, e.g., other anti-inflammatory agents, analgesics, antimicrobial agents, antifungal agents, antibiotics, vitamins, antioxidants, and sunblock agents commonly found in sunscreen formulations including, but not limited to, anthranilates, benzophenones (particularly benzophenone-3), camphor derivatives, cinnamates (e.g., octyl methoxycinnamate), dibenzoyl methanes (e.g., butyl methoxydibenzoyl methane), p-aminobenzoic acid (PABA) and derivatives thereof, and salicylates (e.g., octyl salicylate).

In certain topical formulations, the active agent is present in an amount in the range of approximately 0.25 wt. % to 75 wt. % of the formulation, preferably in the range of approximately 0.25 wt. % to 30 wt. % of the formulation, more preferably in the range of approximately 0.5 wt. % to 15 wt. % of the formulation, and most preferably in the range of approximately 1.0 wt. % to 10 wt. % of the formulation.

Conditions of the eye can be treated or prevented by, e.g., systemic, topical, intraocular injection of a sirtuin-modulating compound, or by insertion of a sustained release device that releases a sirtuin-modulating compound. A sirtuin-modulating compound that increases the level and/or activity of a sirtuin protein

may be delivered in a pharmaceutically acceptable ophthalmic vehicle, such that the compound is maintained in contact with the ocular surface for a sufficient time period to allow the compound to penetrate the corneal and internal regions of the eye, as for example the anterior chamber, posterior chamber, vitreous body, aqueous humor, vitreous humor, cornea, iris/ciliary, lens, choroid/retina and sclera. The pharmaceutically-acceptable ophthalmic vehicle may, for example, be an ointment, vegetable oil or an encapsulating material. Alternatively, the compounds of the invention may be injected directly into the vitreous and aqueous humour. In a further alternative, the compounds may be administered systemically, such as by intravenous infusion or injection, for treatment of the eye.

Sirtuin-modulating compounds described herein may be stored in oxygen free environment. For example, resveratrol or analog thereof can be prepared in an airtight capsule for oral administration, such as Capsugel from Pfizer, Inc.

Cells, e.g., treated *ex vivo* with a sirtuin-modulating compound, can be administered according to methods for administering a graft to a subject, which may be accompanied, e.g., by administration of an immunosuppressant drug, e.g., cyclosporin A. For general principles in medicinal formulation, the reader is referred to Cell Therapy: Stem Cell Transplantation, Gene Therapy, and Cellular Immunotherapy, by G. Morstyn & W. Sheridan eds, Cambridge University Press, 1996; and Hematopoietic Stem Cell Therapy, E. D. Ball, J. Lister & P. Law, Churchill Livingstone, 2000.

Toxicity and therapeutic efficacy of sirtuin-modulating compounds can be determined by standard pharmaceutical procedures in cell cultures or experimental animals. The LD_{50} is the dose lethal to 50% of the population. The ED_{50} is the dose therapeutically effective in 50% of the population. The dose ratio between toxic and therapeutic effects (LD_{50}/ED_{50}) is the therapeutic index. Sirtuin-modulating compounds that exhibit large therapeutic indexes are preferred. While sirtuin-modulating compounds that exhibit toxic side effects may be used, care should be taken to design a delivery system that targets such compounds to the site of affected tissue in order to minimize potential damage to uninfected cells and, thereby, reduce side effects.

The data obtained from the cell culture assays and animal studies can be used in formulating a range of dosage for use in humans. The dosage of such compounds may lie within a range of circulating concentrations that include the ED_{50} with little or no toxicity. The dosage may vary within this range depending upon the dosage form employed and the route of administration utilized. For any compound, the therapeutically effective dose can be estimated initially from cell culture assays. A dose may be formulated in animal models to achieve a circulating plasma concentration range that includes the IC_{50} (i.e., the concentration of the test compound that achieves a half-maximal inhibition of symptoms) as determined in cell culture. Such information can be used to more accurately determine useful doses in humans. Levels in plasma may be measured, for example, by high performance liquid chromatography.

6. Kits

Also provided herein are kits, e.g., kits for therapeutic purposes or kits for modulating the lifespan of cells or modulating apoptosis. A kit may comprise one or more sirtuin-modulating compounds, e.g., in premeasured doses. A kit may optionally comprise devices for contacting cells with the compounds and instructions for use. Devices include syringes, stents and other devices for introducing a sirtuin-modulating compound into a subject (e.g., the blood vessel of a subject) or applying it to the skin of a subject.

In yet another embodiment, the invention provides a composition of matter comprising a sirtuin modulator of this invention and another therapeutic agent (the same ones used in combination therapies and combination compositions) in separate dosage forms, but associated with one another. The term “associated with one another” as used herein means that the separate dosage forms are packaged together or otherwise attached to one another such that it is readily apparent that the separate dosage forms are intended to be sold and administered as part of the same regimen. The agent and the sirtuin modulator are preferably packaged together in a blister pack or other multi-chamber package, or as connected, separately sealed containers (such as foil pouches or the like) that can be separated by the user (e.g., by tearing on score lines between the two containers).

In still another embodiment, the invention provides a kit comprising in separate vessels, a) a sirtuin modulator of this invention; and b) another therapeutic agent such as those described elsewhere in the specification.

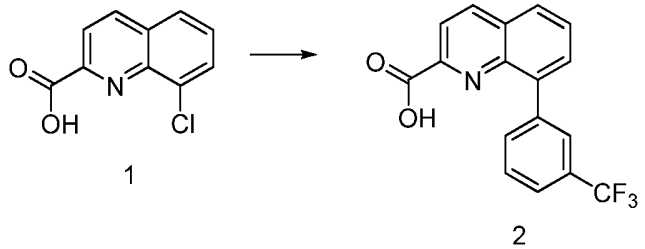
The practice of the present methods will employ, unless otherwise indicated, conventional techniques of cell biology, cell culture, molecular biology, transgenic biology, microbiology, recombinant DNA, and immunology, which are within the skill of the art. Such techniques are explained fully in the literature. See, for example, Molecular Cloning A Laboratory Manual, 2nd Ed., ed. by Sambrook, Fritsch and Maniatis (Cold Spring Harbor Laboratory Press: 1989); DNA Cloning, Volumes I and II (D. N. Glover ed., 1985); Oligonucleotide Synthesis (M. J. Gait ed., 1984); Mullis et al. U.S. Patent No: 4,683,195; Nucleic Acid Hybridization (B. D. Hames & S. J. Higgins eds. 1984); Transcription And Translation (B. D. Hames & S. J. Higgins eds. 1984); Culture Of Animal Cells (R. I. Freshney, Alan R. Liss, Inc., 1987); Immobilized Cells And Enzymes (IRL Press, 1986); B. Perbal, A Practical Guide To Molecular Cloning (1984); the treatise, Methods In Enzymology (Academic Press, Inc., N.Y.); Gene Transfer Vectors For Mammalian Cells (J. H. Miller and M. P. Calos eds., 1987, Cold Spring Harbor Laboratory); Methods In Enzymology, Vols. 154 and 155 (Wu et al. eds.), Immunochemical Methods In Cell And Molecular Biology (Mayer and Walker, eds., Academic Press, London, 1987); Handbook Of Experimental Immunology, Volumes I-IV (D. M. Weir and C. C. Blackwell, eds., 1986); Manipulating the Mouse Embryo, (Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1986).

EXEMPLIFICATION

The invention now being generally described, it will be more readily understood by reference to the following examples which are included merely for purposes of illustration of certain aspects and embodiments of the present invention, and are not intended to limit the invention in any way.

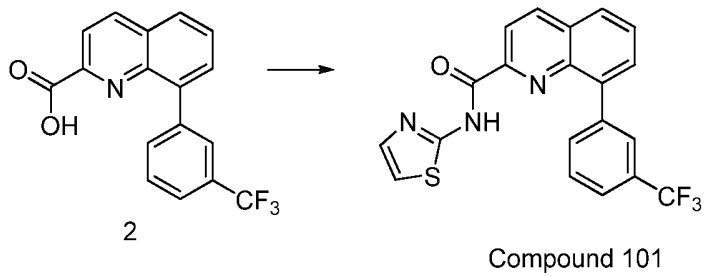
Example 1. Preparation of *N*-(thiazol-2-yl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide (Compound 101):

Step 1) Synthesis of 8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxylic acid (2):



8-Chloroquinoline-2-carboxylic acid (**1**; 100 mg, 0.483 mmol) was taken up in 4 mL of *N,N*-dimethylformamide along with 3-trifluoromethylphenyl boronic acid (92 mg, 0.483 mmol), [1,1'-bis(diphenylphosphino)ferrocene]dichloropalladium (II), 1:1 complex with CH_2Cl_2 (39 mg, 10 mol%) and Cs_2CO_3 (472 mg, 1.4 mmol). The reaction mixture was stirred at 160 °C in a microwave reactor for 50 min. It was then diluted with enough 6 N HCl to bring the pH = 4. The mixture was diluted further with EtOAc. The organic layer was separated and the aqueous layer was further extracted with EtOAc. The combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure. Purification by preparative HPLC using aqueous CH_3CN that had been buffered with 0.1% TFA afforded 20 mg (13%) of 8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxylic acid **2**. MS (ESI) calcd for $C_{17}H_{10}F_3NO_2$: 317.07; found: 318 [M+H].

Step 2) Synthesis of N-(thiazol-2-yl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide (Compound 101):



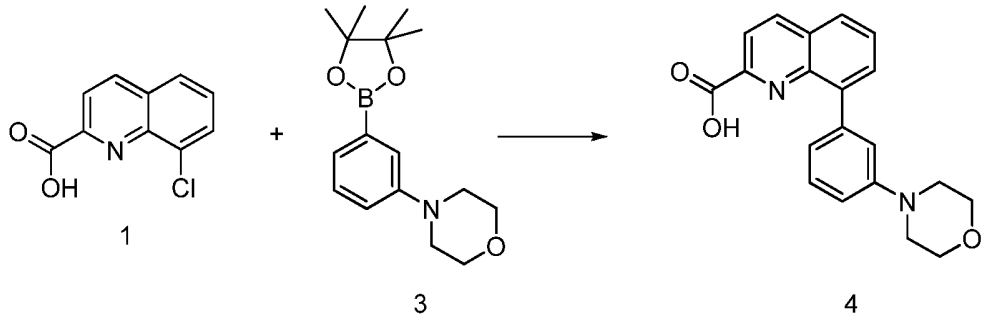
8-(3-(Trifluoromethyl)phenyl)quinoline-2-carboxylic acid (**2**; 16 mg, 0.0505 mmol) was taken up in 1 mL of *N,N*-dimethylformamide along with 2-aminothiazole (5 mg, 0.0505 mmol), *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (38 mg, 0.1 mmol) and DIEA (18 μL, 0.1 mmol). The reaction mixture was stirred at room temperature for 3 h. It was then diluted with EtOAc (10 mL) and washed with water (2 x 5 mL). The organic layer was dried (Na_2SO_4) and concentrated under reduced pressure. Purification by preparative HPLC using aqueous CH_3CN that had been buffered with 0.1% TFA afforded 6 mg (30%) of N-

(thiazol-2-yl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide. MS (ESI) calcd for $C_{20}H_{12}F_3N_3OS$: 399.07; found: 400 [M+H].

This general procedure is used to produce any of the amide derivatives of the invention, including those shown in Table 1, by using the appropriate amine component in the place of 2-aminothiazole.

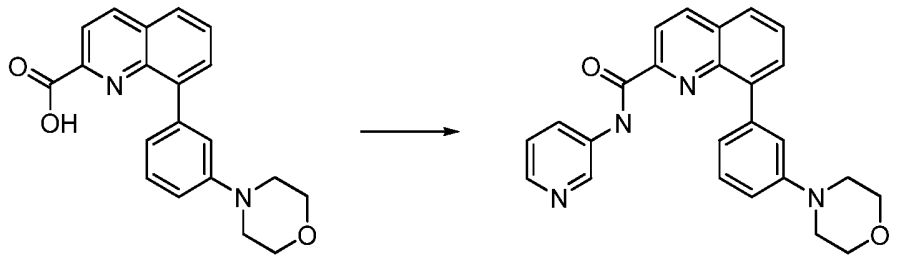
Example 2. Preparation of 8-(3-morpholinophenyl)-*N*-(pyridin-3-yl)quinoline-2-carboxamide (Compound 204):

Step 1) Synthesis of 8-(3-morpholinophenyl)quinoline-2-carboxylic acid (4):



8-Chloroquinoline-2-carboxylic acid (**1**; 100 mg, 0.48 mmol), 4-(3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)morpholine (**3**; 204 mg, 0.72 mmol), dicyclohexyl(2',6'-dimethoxybiphenyl-2-yl)phosphine (16mg, 0.04 mmol) and K_3PO_4 (307 mg, 1.44 mmol) were suspended in dioxane (2 mL) and water (0.2 mL). Tris(dibenzylideneacetone)dipalladium(0) (18mg, 0.02 mmol) was added, and nitrogen was bubbled through the solution for 5 min. The tube was then sealed and the reaction was heated in the microwave with stirring for 1.5 h at 120ºC. Water was added (40 mL) and enough 5N HCl was added to bring the pH to 4. The mixture was extracted with ethyl acetate (3 x 25mL) and the organics were washed with brine, dried with sodium sulfate, filtered and concentrated. Purification by silica gel chromatography (0-10% gradient methanol in dichloromethane) afforded 39.3 mg of 8-(3-morpholinophenyl)quinoline-2-carboxylic acid **4** (36%). MS (ESI) calcd for $C_{20}H_{18}N_2O_3$: 334.13; found: 335 [M+H].

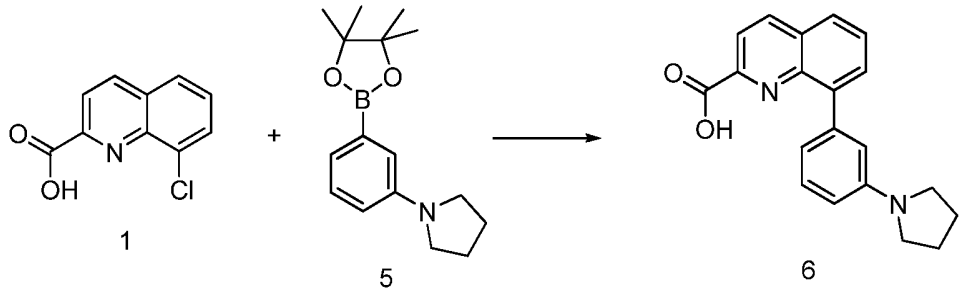
Step 2) Synthesis of 8-(3-morpholinophenyl)-N-(pyridin-3-yl)quinoline-2-carboxamide (Compound 204):



8-(3-morpholinophenyl)quinoline-2-carboxylic acid (**4**; 55 mg, 0.16 mmol) and *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (62 mg, 0.16 mmol) were dissolved in *N,N*-dimethylformamide (1.3 mL). Diisopropylethlyamine (0.11 mL, 0.65 mmol) was added, followed by 3-aminopyridine (15 mg, 0.16 mmol) in DMF (1.3 mL). The reaction was warmed to 50°C for 5 h. Saturated aqueous sodium bicarbonate solution was added (7 mL) and the reaction stirred 10 min. Water was added (10mL) and the mixture was extracted with dichloromethane (3 x 25 mL). The organics were washed with brine, dried with sodium sulfate, filtered and concentrated. The residue was purified by preparative HPLC using aqueous acetonitrile that had been buffered with 0.1% TFA, to afford 16.7 mg of 8-(3-morpholinophenyl)-*N*-(pyridin-3-yl)quinoline-2-carboxamide (22%). MS (ESI) calcd for $C_{25}H_{22}N_4O_2$: 410.17; found: 411 [M+H].

Example 3. Preparation of *N*-(pyridin-3-yl)-8-(3-(pyrrolidin-1-yl)phenyl)quinoline-2-carboxamide (Compound 205):

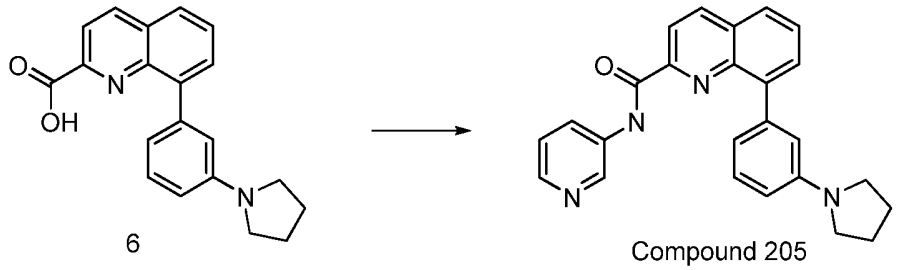
Step 1) Synthesis of 8-(3-(pyrrolidin-1-yl)phenyl)quinoline-2-carboxylic acid (6):



8-Chloroquinoline-2-carboxylic acid (**1**; 100 mg, 0.48 mmol), 1-(3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)pyrrolidine (**5**; 197 mg, 0.72 mmol), dicyclohexyl(2',6'-dimethoxybiphenyl-2-yl)phosphine (16 mg, 0.04 mmol) and K_3PO_4 (307mg, 1.44 mmol) were suspended in dioxane (2 mL) and water (0.2 mL). Tris(dibenzylideneacetone)dipalladium(0) (18 mg, 0.02 mmol) was added, and

nitrogen was bubbled through the solution for 5 min. The tube was then sealed and the reaction was heated in the microwave with stirring for 1.5 h at 120°C. Water was added (40 mL) and enough 5N HCl was added to bring the pH to 4. The mixture was extracted with ethyl acetate (3x25mL) and the organics were washed with brine, dried with sodium sulfate, filtered and concentrated. Purification by silica gel chromatography (0-10% gradient methanol in dichloromethane) afforded 60.2 mg of 8-(3-(pyrrolidin-1-yl)phenyl)quinoline-2-carboxylic acid **6** (39%). MS (ESI) calcd for $C_{20}H_{18}N_2O_2$: 318.14; found: 319 [M+H].

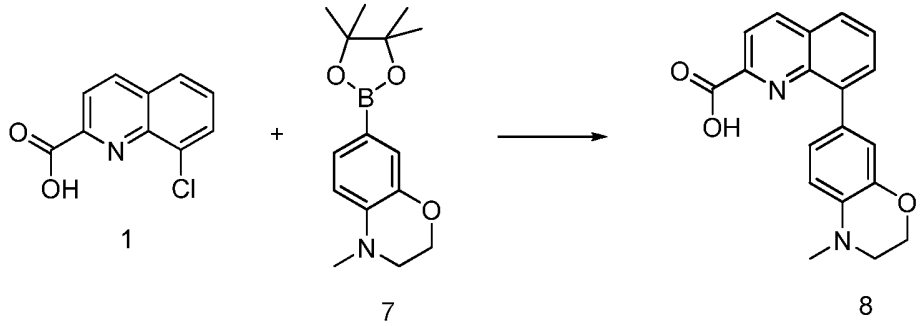
Step 2) Synthesis of N-(pyridin-3-yl)-8-(3-(pyrrolidin-1-yl)phenyl)quinoline-2-carboxamide (Compound 205):



8-(3-(pyrrolidin-1-yl)phenyl)quinoline-2-carboxylic acid (**6**; 34 mg, 0.11 mmol) and *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (81 mg, 0.21 mmol) were dissolved in *N,N*-dimethylformamide (0.7 mL). Diisopropylethlyamine (0.074mL, 0.42 mmol) was added, followed by 3-aminopyridine (10 mg, 0.11 mmol) in DMF (1 mL). The reaction was warmed to 50°C for 2 h. Saturated aqueous sodium bicarbonate solution was added (3mL) and the reaction stirred 10 min. Water was added (5 mL) and the mixture was extracted with dichloromethane (3x15 mL). The organics were washed with brine, dried with sodium sulfate, filtered and concentrated. The residue was purified by preparative HPLC using aqueous acetonitrile that had been buffered with 0.1% TFA, to afford 20.7 mg of *N*-(pyridin-3-yl)-8-(3-(pyrrolidin-1-yl)phenyl)quinoline-2-carboxamide (Compound 205) (45%). MS (ESI) calcd for $C_{25}H_{22}N_4O$: 394.18; found: 395 [M+H].

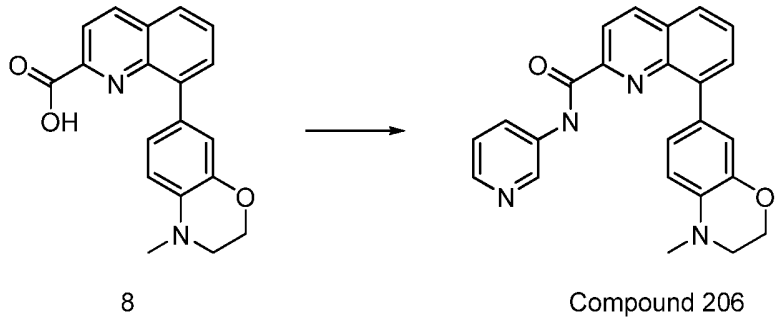
Example 4. Preparation of 8-(4-methyl-3,4-dihydro-2*H*-benzo[*b*][1,4]oxazin-7-yl)-*N*-(pyridin-3-yl)quinoline-2-carboxamide (Compound 206):

Step 1) Synthesis of 8-(4-methyl-3,4-dihydro-2H-benzo[b][1,4]oxazin-7-yl)quinoline-2-carboxylic acid (8):



8-Chloroquinoline-2-carboxylic acid (**1**; 100 mg, 0.48 mmol), 4-methyl-7-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-3,4-dihydro-2*H*-benzo[*b*][1,4]oxazine (**7**; 199 mg, 0.72 mmol), dicyclohexyl(2',6'-dimethoxybiphenyl-2-yl)phosphine (16 mg, 0.04 mmol) and K_3PO_4 (307 mg, 1.44 mmol) were suspended in dioxane (2 mL) and water (0.2 mL). Tris(dibenzylideneacetone)dipalladium(0) (18 mg, 0.02 mmol) was added, and nitrogen was bubbled through the solution for 5 min. The tube was then sealed and the reaction was heated in the microwave with stirring for 2 h at 120ºC. Water was added (40 mL) and enough 5N HCl was added to bring the pH to 4. The mixture was extracted with ethyl acetate (3 x 30 mL) and the organics were washed with brine, dried with sodium sulfate, filtered and concentrated. Purification by silica gel chromatography (0-10% gradient methanol in dichloromethane) afforded 42.4 mg of 8-(4-methyl-3,4-dihydro-2*H*-benzo[*b*][1,4]oxazin-7-yl)quinoline-2-carboxylic acid **8** (27%). MS (ESI) calcd for $C_{19}H_{16}N_2O_3$: 320.12; found: 321 [M+H].

Step 2) Synthesis of 8-(4-methyl-3,4-dihydro-2H-benzo[b][1,4]oxazin-7-yl)-N-(pyridin-3-yl)quinoline-2-carboxamide (Compound 206):

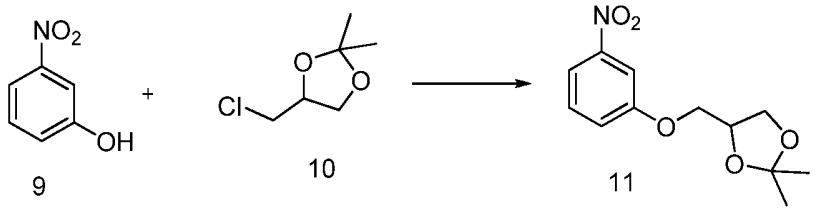


8-(4-methyl-3,4-dihydro-2*H*-benzo[*b*][1,4]oxazin-7-yl)quinoline-2-carboxylic acid (**8**; 42mg, 0.13 mmol) and *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (101 mg, 0.26 mmol) were dissolved in

N,N-dimethylformamide (1.1 mL). Diisopropylethlyamine (0.092 mL, 0.53 mmol) was added, followed by 3-aminopyridine (12 mg, 0.13 mmol) in *N,N*-dimethylformamide (1 mL). The reaction was warmed to 50°C for 2 h. Saturated aqueous sodium bicarbonate solution was added (3mL) and the reaction stirred 10 min. Water was added (5 mL) and the mixture was extracted with dichloromethane (3 x 15 mL). The organics were washed with brine, dried with sodium sulfate, filtered and concentrated. The residue was purified by preparative HPLC using aqueous acetonitrile that had been buffered with 0.1% TFA, to afford 32.1 mg of 8-(4-methyl-3,4-dihydro-2*H*-benzo[*b*][1,4]oxazin-7-yl)-*N*-(pyridin-3-yl)quinoline-2-carboxamide (Compound 206) (56%). MS (ESI) calcd for $C_{24}H_{20}N_4O_2$: 396.16; found: 397 [M+H].

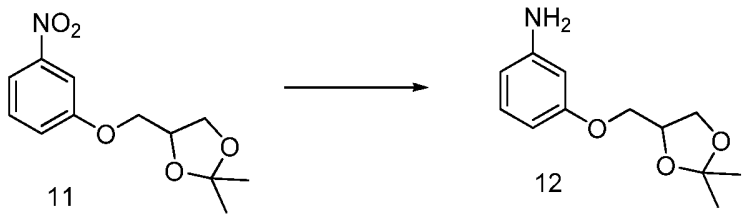
Example 5. Preparation of *N*-(3-(2,3-dihydroxypropoxy)phenyl)-8-(3-(trifluoromethyl)phenyl) quinoline-2-carboxamide (Compound 116):

Step 1) Synthesis of 2,2-dimethyl-4-((3-nitrophenoxy)methyl)-1,3-dioxolane (11):



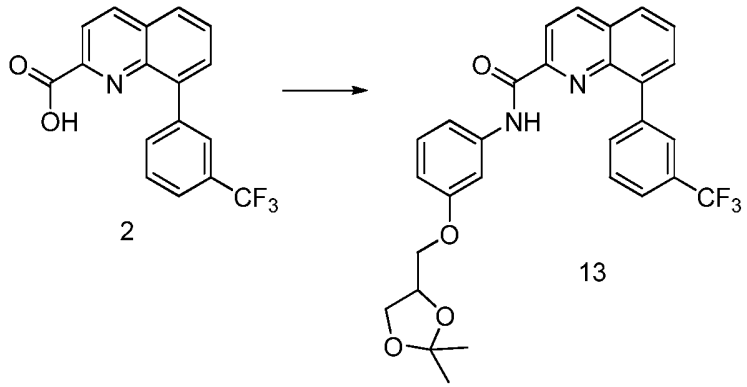
3-Nitrophenol (**9**; 2.0 g, 14.37 mmol) was taken up in 20 mL of anhydrous DMF along with anhydrous potassium carbonate (4.96 g, 35.93 mmol) and 4-(chloromethyl)-2,2-dimethyl-1,3-dioxolane (**10**; 2.55 mL, 18.68 mmol). The resulting reaction mixture was heated in the microwave reactor, with stirring, at 160°C for 4 h. The crude reaction mixture was rinsed with water, filtered and extracted with dichloromethane (3 x 15 mL). The combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure. The resulting residue was purified by chromatography using ethyl acetate: pentanes to obtain 2,2-dimethyl-4-((3-nitrophenoxy)methyl)-1,3-dioxolane as an amber-colored oil **11** (52%). MS (ESI) calcd for $C_{12}H_{15}NO_5$: 253.3; found: 254 [M+H].

Step 2) Synthesis of 3-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)aniline (12):



Under nitrogen, Fe powder (2.38 g, 42.54 mmol) and NH_4Cl (2.38 g, 42.54 mmol) were combined, followed by addition of 2,2-dimethyl-4-((3-nitrophenoxy)methyl)-1,3-dioxolane (**11**; 1.8 g, 7.09 mmol) and a 4:1 mixture of isopropanol:water (30 mL:10 mL). The reaction mixture was stirred under reflux for 18 h. The crude material was filtered through a pad of Celite and the filtrate was concentrated under reduced pressure. The resulting aqueous layer was extracted with dichloromethane (3 x 15 mL). The combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure to afford 3-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy) **12** (1.2 g, 79% yield). The material was used in the next step without any further purification. MS (ESI) calcd for $C_{12}H_{17}NO_3$: 223.3; found: 224[M+H].

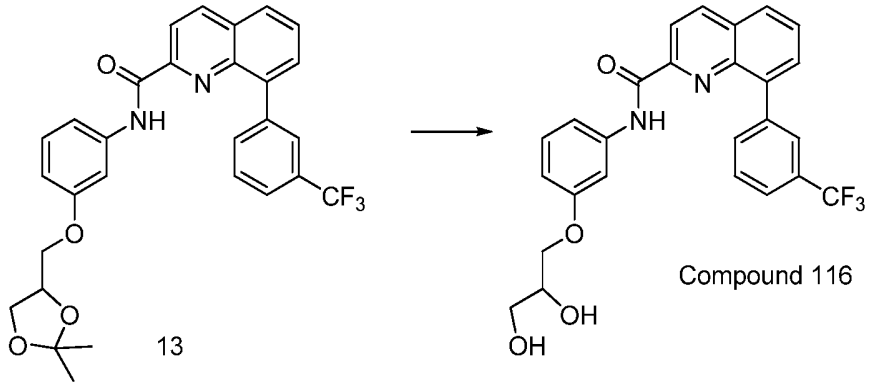
Step 3) Synthesis of N-(3-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)phenyl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide (13):



3-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)aniline (**2**; 31.1 mg, 0.15 mmol) and *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (56.6 mg, 0.15 mmol) were dissolved in *N,N*-dimethylformamide (0.4 mL) under nitrogen atmosphere. Diisopropylethylamine (0.10 mL) was added, followed by 8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxylic acid (47.4 mg, 0.15 mmol) in 2 mL DMF. The reaction was warmed to 50 °C for 4 h. Saturated aqueous sodium bicarbonate solution was added (4 mL), followed by water (10 mL). The mixture was extracted with dichloromethane and the organics were washed with brine, dried

with sodium sulfate, filtered and concentrated. The residue was purified by silica gel column chromatography using a gradient of 0 – 100% ethyl acetate in pentane, to afford *N*-(3-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)phenyl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide **13** (49.9 mg, 64%). MS (ESI) calcd for $C_{29}H_{25}F_3N_2O_4$: 522.18; found: 523 [M+H].

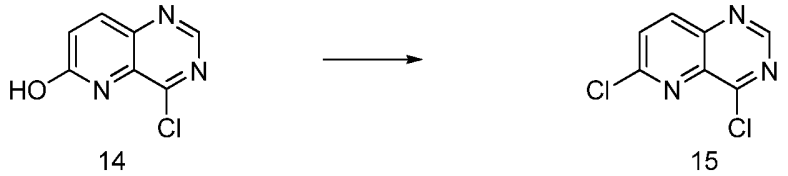
Step 4) Synthesis of N-(3-(2,3-dihydroxypropoxy)phenyl)-8-(3-(trifluoromethyl)phenyl) quinoline-2-carboxamide (Compound 116):



N-(3-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)phenyl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide (**13**; 49.9 mg, 0.096 mmol) was dissolved in THF (2.5 mL). Concentrated hydrochloric acid (0.032 mL, 0.38 mmol) was added and the reaction was allowed to stir at room temperature. After 3 h the mixture was concentrated. purification by preparative HPLC using aqueous acetonitrile that had been buffered with 0.1% TFA afforded *N*-(3-(2,3-dihydroxypropoxy)phenyl)-8-(3-(trifluoromethyl)phenyl) quinoline-2-carboxamide (Compound 116) (29.7 mg, 64%). MS (ESI) calcd for $C_{26}H_{21}F_3N_2O_4$: 482.15; found: 483 [M+H].

Example 6. Preparation of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxylic acid (18), Route 1:

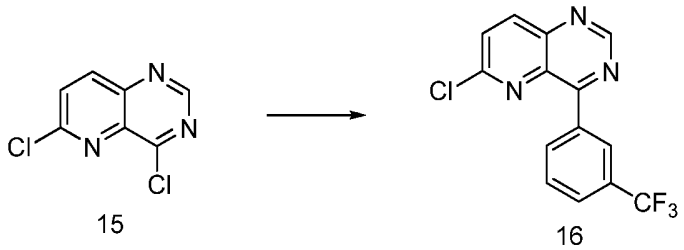
Step 1) Synthesis of 4,6-dichloropyrido[3,2-d]pyrimidine (15):



To a mixture of crude 4-chloropyrido[3,2-d]pyrimidin-6-ol (**14**; 500 mg, 2.76 mmol) and *N*,*N*-Diisopropylethylamine (1.07 g, 8.29 mmol) in 10 ml of toluene was added

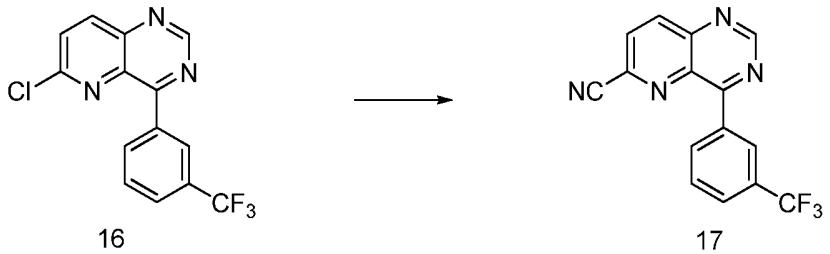
slowly $POCl_3$. (1.3 g, 8.29 mmol). The resulting mixture was refluxed for about 2 hours. After cooling down to room temperature, the solvents were removed under reduced pressure and the residue was dissolved in dichloromethane and washed with cooled diluted sodium bicarbonate solution. The organic layer was dried under anhydrous sodium sulfate, filtered and concentrated in vacuum to yield crude product, which was purified by flash chromatography eluting with ethyl acetate/petroleum=20:1 to give 4,6-dichloropyrido[3,2-d]pyrimidinethe **15** as a white solid (210mg, 37%). MS (ESI) calcd for $C_7H_3Cl_2N_3$: 200; found: 201 [M+H].

Step 2) Synthesis 6-chloro-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine (16):



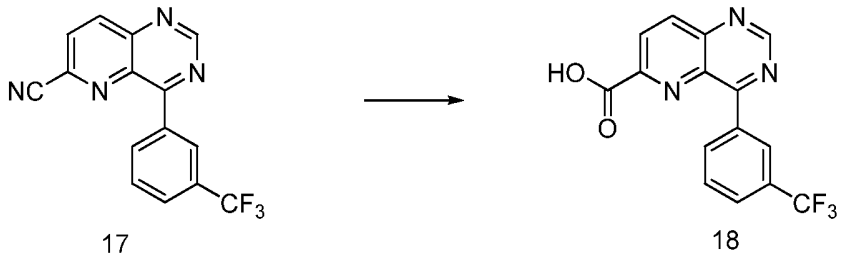
Under a nitrogen atmosphere, a mixture of compound 4,6-dichloropyrido[3,2-d]pyrimidine (**15**; 100 mg, 0.5 mmol), 3-(trifluoromethyl)phenylboronic acid (95 mg, 0.5mmol), $PdCl_2$(dppf) (20 mg, cat., 0.05eq) and cesium carbonate (326 mg, 1.0 mmol) in 5mL of dioxane was stirred at 80°C for 6 hours. Water was added to the reaction mixture and extracted with ethyl acetate (2 times) and the combined organic layers were washed with water and brine, then concentrated in vacuo to give a dark residue, which was purified by column chromatography eluting with ethyl acetate/petroleum =1:3 to afford 6-chloro-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine as a white solid **16** (20 mg, 12.6%). MS (ESI) calcd for $C_{14}H_7ClF_3N_3$: 309.7; found: 311 [M+H].

Step 3) Synthesis of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbonitrile (17):



Under a nitrogen atmosphere, a mixture of compound 6-chloro-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine (**16**; 100 mg, 0.324 mmol), Zn (2.5 mg, 0.039 mmol, 0.12 equiv), $Pd_2(dba)_3$ (29.6 mg, cat., 0.1 equiv), zinc cyanide (23 mg, 0.194 mmol, 0.6eq) and dppf (34 mg, 0.0628 mmol, 0.2 eq) in 5 ml of N,N-Dimethylacetamide was stirred at 120°C for 1 hour. Water was added to the reaction mixture and extracted with ethyl acetate (2 x 1mL) and the combined organic layers were washed with water (2 times) and brine (2 x 1mL), dried and concentrated in vacuum to give a residue, which was purified by preparation TLC to afford 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbonitrile **17** as a yellow solid (20 mg, 12.6%). MS (ESI) calcd for $C_{15}H_7F_3N_4$: 300.2; found: 301 [M+H].

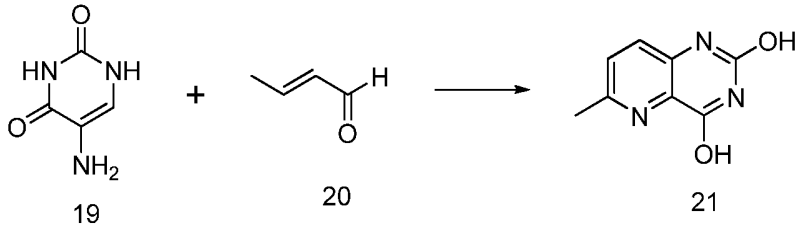
Step 4) Synthesis of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxylic acid (18):



4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbonitrile (**17**; 80 mg, 0.267 mmol) was mixed with 0.5 ml of ethanol, aqueous sodium hydroxide solution (3 mol/L, 0.5 ml) was added to the above mixture at room temperature and the resulting mixture was stirred at reflux for 1.5 hours. After cooling down to r.t, 1 mol/L HCl was added dropwise to the mixture until pH=3~4, the forming precipitate was filtered and dried to afford 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxylic acid **18** as a white solid (56 mg, 66%). MS (ESI) calcd for $C_{15}H_8F_3N_3O_2$: 319.2; found: 320 [M+H].

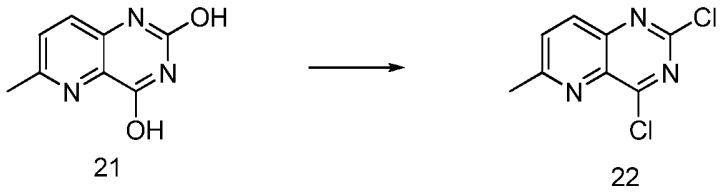
Example 7. Preparation of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxylic acid (18), Route 2:

Step 1) Synthesis of 6-methylpyrido[3,2-d]pyrimidine-2,4-diol (21):



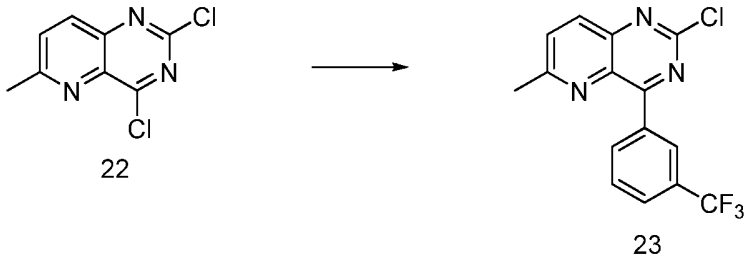
A mixture of 5-aminopyrimidine-2,4(1H,3H)-dione (**19**; 2 g, 4 mmol), 20% HCl (8 mL) and (*E*)-but-2-enal (**20**; 1.6 mL) were refluxed for 1 hour. The solution was evaporated to dryness in vacuo. Water was added to the residue so as to make the mixture just stirrable and then it was triturated with ammonium hydroxide with strong stirring until the pH increased to 10-11. Stirring was continued for another 10 minutes. The precipitate was filtered and was washed with minimal methanol and then chloroform and dried to afford 6-methylpyrido[3,2-d]pyrimidine-2,4-diol **21** (800 mg 28.8%). MS (ESI) calcd for $C_8H_7N_3O_2$: 177.2; found: 178 [M+H].

Step 2) Synthesis of 2,4-dichloro-6-methylpyrido[3,2-d]pyrimidine (22):



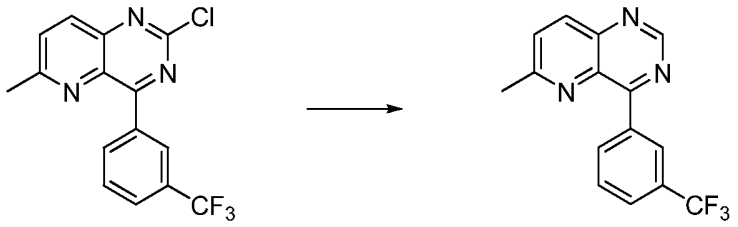
A mixture of 6-methylpyrido[3,2-d]pyrimidine-2,4-diol (**21**; 3 g, 17 mmol), $POCl_3$ (30 mL) and N,N-Diisopropylethylamine (6 mL) was heated to reflux overnight. The $POCl_3$ was removed in vacuo and the residue was dissolved in ethyl acetate. $NaHCO_3$ was added to adjust pH to 8~9. The organic layer was dried and concentrated; the residue was purified by column chromatography to afford 2,4-dichloro-6-methylpyrido[3,2-d]pyrimidine **22** (1.7 g, 46%). MS (ESI) calcd for $C_8H_5Cl_2N_3$: 214.1; found: 215 [M+H].

Step 3) Synthesis of 2-chloro-6-methyl-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine (23):



To a nitrogen degassed solution of 2,4-dichloro-6-methylpyrido[3,2-d]pyrimidine (**22**; 1.7 g, 7.9 mmol) in toluene (10 mL) were successively added 3-trifluoromethylphenylboronic acid (1.46 g, 7.9 mmol), potassium carbonate (1.65 g, 11.8 mmol, 1.5equiv) and $Pd(PPh_3)_4$ (459 mg, 0.4 mmol, 0.05 equiv). The reaction was heated at 100°C under vigorous stirring for 3h. After complete disappearance of starting material, water (50 mL) was added. After extraction with CH_2Cl_2 twice, the combined organic layers were dried over $MgSO_4$ and the solvent was removed under reduced pressure. The crude material was purified by column chromatography to afford 2-chloro-6-methyl-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine **23** (1.68 g, 65%). MS (ESI) calcd for $C_{15}H_9ClF_3N_3$: 323.7; found: 325 [M+H].

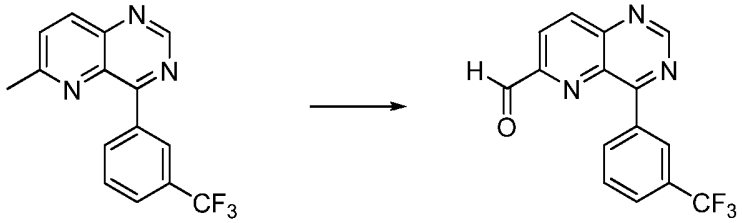
Step 4) Synthesis of 6-methyl-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine (24):



23 24

To a round-bottom flask were added palladium (100 mg) and isopropanol (10 mL). N_2 was bubbled in this mixture. A degassed ammonium formate solution in water (1.9 g, 30.6 mmol in 3 mL of water) was added, followed by 2-chloro-6-methyl-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine (**23**; 990 mg; 3.1 mmol). The above mixture was then stirred at r.t over night and filtered through celite. The filtrate was evaporated and the residue was purified by column chromatography to afford 6-methyl-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine **24** (418 mg, 47%). MS (ESI) calcd for $C_{15}H_{10}F_3N_3$: 289.3; found: 290 [M+H].

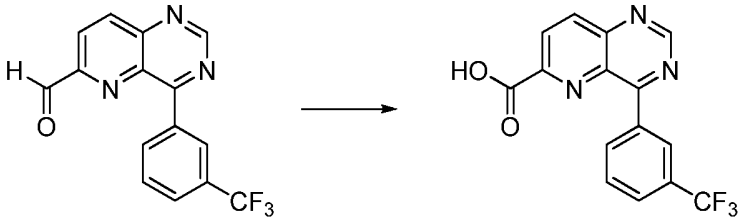
Step 5) Synthesis of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbaldehyde (25):



24 25

A mixture of 6-methyl-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine (**24**; 418 mg, 1.4 mmol) and SeO_2 (193 mg, 1.2 equiv) in dioxane (5 mL) was heated at 110°C for overnight, cooled, and the solids filtered off. The filtrate was concentrated and the residue purified by column chromatography on silica gel to afford 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbaldehyde **25** (238 mg, 54%) as a yellow solid. MS (ESI) calcd for $C_{15}H_8F_3N_3O$: 303.2; found: 304 [M+H].

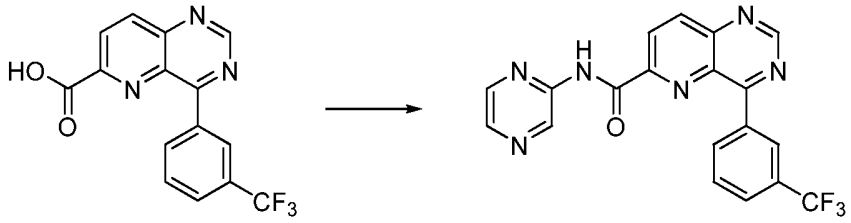
Step 6) Synthesis of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbaldehyde (18):



25 18

A mixture of 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbaldehyde (**25**; 238 mg, 0.76 mmol), sodium dihydrogen phosphate (245 mg, 1.57 mmol), 2-methyl-2-butene (1 mL) and sodium chlorite (141 mg, 1.57 mmol) is stirred at room temperature in 10 ml of a 1/1 tert-butanol/water mixture for 4 hours. The reaction medium is concentrated under reduced pressure to afford 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carbaldehyde **18** (245 mg, 97%) which was used directly in Example 8 without further purification. MS (ESI) calcd for $C_{15}H_8F_3N_3O_2$: 319.2; found: 320 [M+H].

Example 8. Preparation of N-(pyrazin-2-yl)-4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxamide (Compound 384):

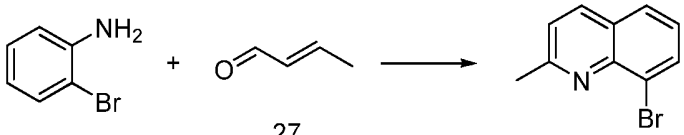


18 Compound 384

The carboxylic acid 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxylic acid (**18**; 31.9 mg, 0.1 mmol, 1 equiv), pyrazin-2-amine (9.51 mg, 0.1 mmol, 1 equiv), and *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (76.04 mg, 0.2 mmol, 2 equiv) were dissolved in *N,N*-dimethylformamide (0.1~0.2 mol/L), then added diisopropylethylamine (34.83 μL, 0.2 mmol, 2 equiv) and stirred at room temperature overnight. The reaction mixture was diluted with water and extracted with dichloromethane and the combined organic layers were washed with saturated sodium bicarbonate (1 × 2mL) and water and brine (2 × 2 mL), dried over anhydrous sodium sulfate and concentrated to give the crude product (Compound 379), which was purified by preparative thin layer chromatography. MS (ESI) calcd for $C_{19}H_{11}F_3N_6O$: 396.3; found: 397 [M+H]. This general procedure is used to produce any compound of the invention containing 4-(3-(trifluoromethyl)phenyl)pyrido[3,2-d]pyrimidine-6-carboxamide by using the appropriate amine component in place of pyrazin-2-amine.

Example 9. Preparation of 8-(3-(difluoromethyl)phenyl-*N*-(2-(morpholinomethyl)pyrimidin-4-yl)quinoline-2-carboxamide (Compound 373):

Step 1) Synthesis of 8-bromo-2-methylquinoline (28):

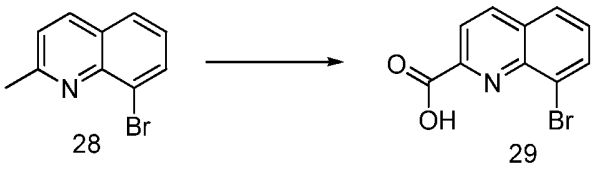


26 27 28

To a solution of 2-bromoaniline (**26**; 100 g, 0.58 mol) in 6N HCl (300 mL) was added crotonaldehyde (**27**; 81 g, 1.1 mol) at r.t. The mixture was stirred at 100°C overnight, cooled to room temperature, basified with ammonium hydroxide and extracted with ether. The organic phase was dried, concentrated and purified with silica gel column (petroleum ether:ethyl acetate 12:1 to 10:1) to give 8-bromo-

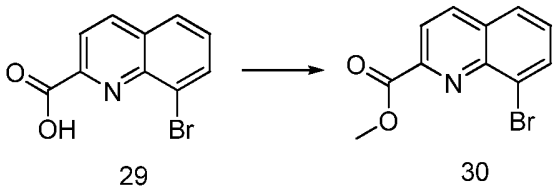
2-methylquinoline **28** as a white solid (59 g, yield 54%). MS (ESI) calcd for $C_{10}H_8BrN$: 222.1; found: 223 [M+H].

Step 2) Synthesis of 8-bromoquinoline-2-carboxylic acid (29):



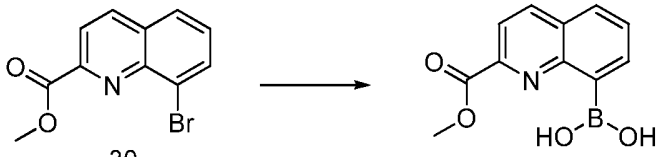
To a hot solution of selenium dioxide (100 g, 0.9 mol) in dioxane (1 L) was added 8-bromo-2-methylquinoline (**28**; 35.2 g, 0.16 mol) in one portion, and the mixture was heated under reflux for 3h, then filtered while hot. The mixture was concentrated and purified with silica gel column (pure dichloromethane) to give the aldehyde intermediate as a pale yellow solid (39g, yield 100%). The solution of $NaClO_2$ (108 g, 1.19 mol) and NaH_2PO_4 (108 g, 0.9 mol) in water(1 L) was added over 30 min to a mixture of the above aldehyde (27 g, 0.11 mol) in *t*-BuOH (700 mL) and 2-methylbut-2-ene (150 mL), then stirred at room temperature overnight. This mixture was concentrated and water (500 mL) was added. The precipitate was collected by filtration and dried to give 8-bromoquinoline-2-carboxylic acid **29** as a tan solid (25 g, 90% yield). MS calcd for $C_{10}H_6BrNO_2$: 250.96; found: 252 [M+1].

Step 3) Synthesis of methyl 8-bromoquinoline-2-carboxylate (30):



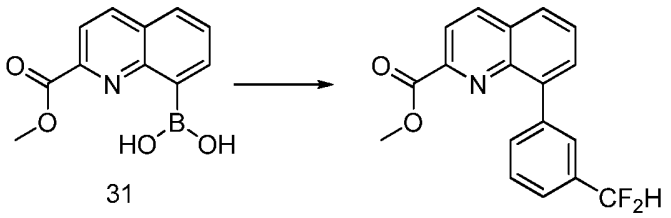
To a solution of 8-bromoquinoline-2-carboxylic acid (**29**; 7.1 g, 28 mol) in methanol (200 mL) was added concentrated sulfuric acid (18 mL). The reaction was heated under reflux overnight, concentrated and diluted with dichloromethane, basified with saturated $NaHCO_3$, dried, concentrated and purified with silica gel column (petroleum ether : ethyl acetate 6:1 to 3:1) to give 8-bromoquinoline-2-carboxylate **30** as a white solid (5.0 g, yield 67%). MS calcd for $C_{11}H_8BrNO_2$: 266.1; found: 267 [M+1].

Step 4) Synthesis of 2-(methoxycarbonyl)quinolin-8-ylboronic acid (31):



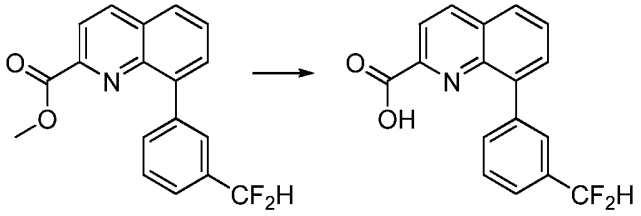
A mixture of 8-bromoquinoline-2-carboxylate (**30**; 33 g, 124 mmol), bis(pinacolato)diboron (47.3 g, 186 mmol), KOAc (36 g, 370 mmol) and Pd(dppf)Cl_2 (10 g) in DMSO (300 mL) was stirred under nitrogen at 60°C overnight. The reaction mixture was filtered and diluted with water (600 mL) and extracted with ethyl acetate (300 mL x 3). The combined organics were washed with water and brine, dried, concentrated and purified via silica gel chromatography (petroleum ether : ethyl acetate 3:1 to pure ethyl acetate) to give 2-(methoxycarbonyl)quinolin-8-ylboronic acid **31** as a white solid (14 g, yield 35%). MS calcd for $C_{11}H_{10}BNO_4$: 231.07; found: 232 [M+1].

Step 5) Synthesis of methyl-8-(3-(difluoromethyl)phenyl)quinoline-2-carboxylate(32):



A mixture of 2-(methoxycarbonyl)quinolin-8-ylboronic acid (**31**; 2.79 g, 12.1 mmol), 1-bromo-3-(difluoromethyl)benzene (2.5 g,12.1 mmol), K_2CO_3 (5.0 g, 36.3 mmol) and $PdCl_2$dppf (1.0 mg, 1.21 mmol) in dioxane (20.0mL) and water (2 mL) was stirred at 60°C overnight under nitrogen. The resulting mixture was extracted with dichloromethane from water. The organic layer was dried over sodium sulfate, concentrated and purified via silica gel column chromatography (petroleum ether : ethyl acetate 20:1 to 10:1) to give methyl-8-(3-(difluoromethyl)phenyl)quinoline-2-carboxylate **32** as a yellow solid (1.36 mg, 36%). MS calcd for $C_{18}H_{13}F_2NO_2$: 313.3; found: 314 [M+1].

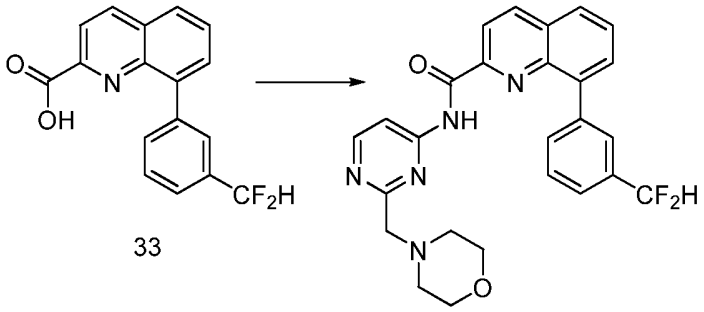
Step 6) Synthesis of 8-(3-(difluoromethyl)phenyl)quinoline-2-carboxylic acid (33):



32 33

Methyl-8-(3-(difluoromethyl)phenyl)quinoline-2-carboxylate (**32**; 1.36 g, 4.34 mmol) and LiOH·H_2O (730 mg, 17.36 mmol) in THF/H_2O (2:1, 20 ml) was stirred at room temp. for 2 h. The reaction mixture was then concentrated to remove the THF, more water (5 mL) was added and the pH was adjusted to 5. The acid was collected by filtration and dried to give 8-(3-(difluoromethyl)phenyl)quinoline-2-carboxylic acid **33** as a yellow solid (1.26 g, 97%).MS calcd for $C_{17}H_{11}F_2NO_2$: 299.08; found: 297.9 [M-1].

Step 7) Synthesis of 8-(3-(difluoromethyl)phenyl-N-(2-(morpholinomethyl)pyrimidin-4-yl)quinoline-2-carboxamide (Compound 373):

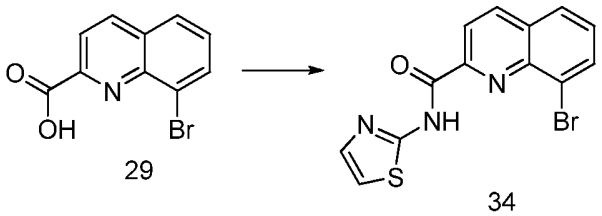


33 Compound 373

A mixture of 8-(3-(difluoromethyl)phenyl)quinoline-2-carboxylic acid (**33**; 50 mg, 0.17 mmol), 2-(morpholinomethyl)pyrimidin-4-amine (43 mg, 0.22 mmol), *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (129 mg, 0.34 mmol), diisopropylethylamine (66 mg, 0.51 mmol) in *N,N*-dimethylformamide (3 mL) was stirred at room temperature overnight. The reaction solution was poured into water (25 mL). The precipitate was filtered, dissolved in dichloromethane, dried, concentrated and purified via prep TLC (ethyl acetate/petroleum ether 1:1) to give 8-(3-(difluoromethyl)phenyl-*N*-(2-(morpholinomethyl)pyrimidin-4-yl)quinoline-2-carboxamide (Compound 368) (55 mg, 46%). MS calcd for $C_{26}H_{23}F_2N_5O_2$: 475.18; found: 476 [M+1].

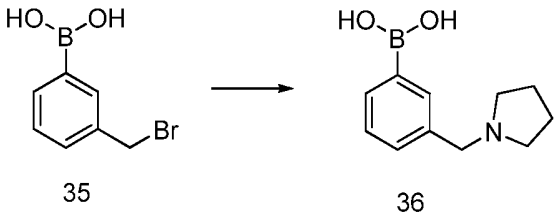
Example 10. Preparation of 8-(3-(pyrrolidin-1-ylmethyl)phenyl)-N-(thiazol-2-yl)quinoline-2-carboxamide (Compound 356):

Step 1) Synthesis of 8-bromo-N-(thiazol-2-yl)quinoline-2-carboxamide (34):



A mixture of 8-bromoquinoline-2-carboxylic acid (**29**; 758 mg, 3 mmol), thiazol-2-amine (405 mg, 4.1 mmol), *O*-(7-azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (2.28 g, 6 mmol), diisopropylethylamine (2.3 g, 17.8 mmol) in *N,N*-dimethylformamide (30 mL) was stirred at room temperature overnight. The reaction solution was poured into water (100 mL). The precipitate was filtered to give 8-bromo-N-(thiazol-2-yl)quinoline-2-carboxamide **34** as a yellow solid (632 mg, 63%). MS calcd for $C_{13}H_8BrN_3O_5$: 334.2; found: 335 [M+1].

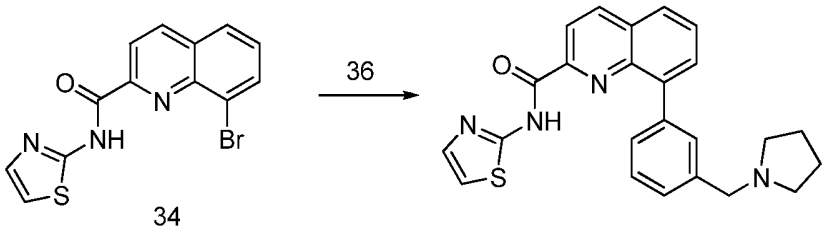
Step 2) Synthesis of 3-(pyrrolidin-1-ylmethyl)phenylboronic acid (36):



A mixture of 3-(bromomethyl)phenylboronic acid (**35**; 645 mg, 3 mmol) and pyrrolidine (0.5 mL) in THF (10 ml) was stirred at room temperature overnight. The mixture was filtered and the filtrate was evaporated. The residue was dissolved in EtOAc and washed with water, dried over $MgSO_4$, concentrated to give 3-(pyrrolidin-1-ylmethyl)phenylboronic acid **36** as a yellow solid (414 mg, 68%). MS calcd for $C_{11}H_{16}BNO_2$: 334.2; found: 335 [M+1].

The above procedure was followed using the appropriate amines to form 3-((dimethylamino)methyl)phenylboronic acid and 3-(morpholinomethyl)phenylboronic acid.

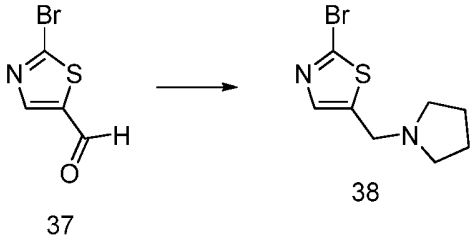
Step 3) Synthesis of 8-(3-(pyrrolidin-1-ylmethyl)phenyl)-N-(thiazol-2-yl)quinoline-2-carboxamide (Compound 356):



A mixture of 8-bromo-N-(thiazol-2-yl)quinoline-2-carboxamide (**34**; 100 mg, 0.30 mmol), 3-(pyrrolidin-1-ylmethyl)phenylboronic acid (**36**; 81 mg, 0.40 mmol), K_2CO_3 (134 mg, 0.97 mmol) and $PdCl_2$dppf (40 mg, 0.05 mmol) in dioxane/H_2O (5:1, 3.6 mL) was stirred at 85°C for 2.5 hours under nitrogen. The resulting mixture was evaporated, and the residue was dissolved in EtOAc, filtered and purified with TLC plate (CH_2Cl_2:MeOH=20:1) to give 8-(3-(pyrrolidin-1-ylmethyl)phenyl)-N-(thiazol-2-yl)quinoline-2-carboxamide (Compound 351) as a yellow powder (75 mg, 61%). MS calcd for $C_{24}H_{22}N_4OS$: 414.15; found: 415.0 [M+1].

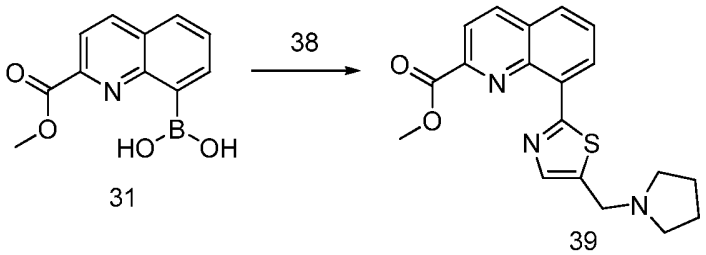
Example 11. Preparation of 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)-N-(thiazol-2-yl)quinoline-2-carboxamide (Compound 293):

Step 1) Synthesis of 2-bromo-5-(pyrrolidin-1-ylmethyl)thiazole (38):



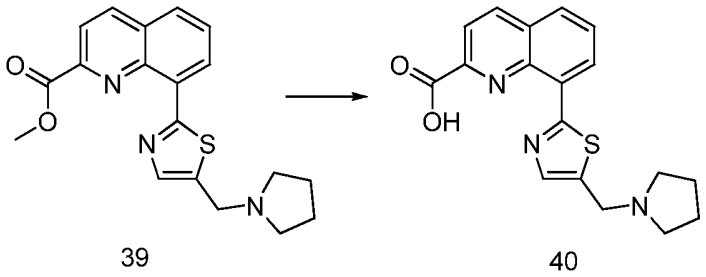
To a solution of 2-bromothiazole-5-carbaldehyde (**37**; 3.0 g, 15.6 mmol), pyrrolidine (1.77 g, 24.9 mmol) and acetic acid (1.5 g, 24.9 mmol) in MeOH (60 mL) was added $NaCNBH_3$ (1.56 g, 24.9 mmol) in portions over 30 min at room temperature, then stirred for 1 h. Ethyl acetate (100 mL) was added and the mixture was washed with water, dried over $MgSO_4$, and concentrated. The residue was purified by silica gel column (petroleum ether:ethyl acetate 3:1) to afford 2-bromo-5-(pyrrolidin-1-ylmethyl)thiazole **38** as a yellow oil (870 mg , 37%). MS calcd for $C_8H_{11}BrN_2S$: 247.2; found: 248 [M+1].

Step 2) Synthesis of methyl 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)quinoline-2-carboxylate (39):



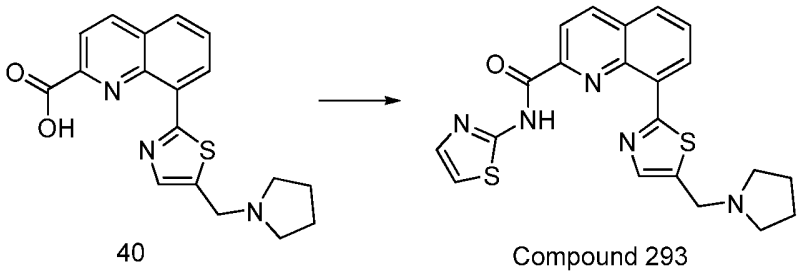
The mixture of 2-(methoxycarbonyl)quinolin-8-ylboronic acid (**31**; 512 mg, 1.56 mmol), 2-bromo-5-(pyrrolidin-1-ylmethyl)thiazole (**38**; 411 mg, 1.56 mmol), K_2CO_3 (646 mg, 4.68mmol) and $PdCl_2$dppf (125mg, 0.15 mmol) in dioxane/H_2O (5:1, 15 mL) was stirred at 60°C for 4 hours under nitrogen. The resulting mixture was extracted with dichloromethane from water. The organic layer was dried over Na_2SO_4, concentrated and purified with silica gel column (petroleum ether:ethyl acetate:triethylamine = 3:1:0.03 to ethyl acetate:triethylamine = 1000:5) to give methyl 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)quinoline-2-carboxylate **39** as a brown oil (402 mg, 73%). MS calcd for $C_{19}H_{19}N_3O_2S$: 353.4; found: 354 [M+1].

Step 3) Synthesis of 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)quinoline-2-carboxylic acid (40):



The mixture of methyl 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)quinoline-2-carboxylate (**39**; 402 mg, 1.14 mmol) and LiOH·H_2O (193 mg, 4.6 mmol) in THF/H_2O (4:1, 5 ml) was stirred at room temperature for 2 hours. The reaction mixture was extracted with dichloromethane and the pH of the water phase was adjusted to 6, lyophilized to give 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)quinoline-2-carboxylic acid **40** as a brown semi-solid (700 mg, 100%, mixed with inorganic salt). MS calcd for $C_{18}H_{17}N_3O_2S$: 339.4; found: 340 [M+1].

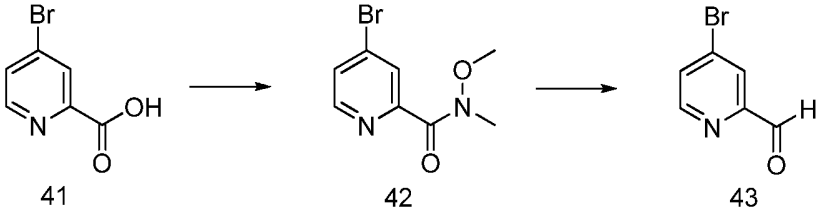
Step 4) Synthesis of 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)-N-(thiazol-2-yl)quinoline-2-carboxamide (Compound 293):



A mixture of 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)quinoline-2-carboxylic acid (**40**; 233 mg, crude, 0.34 mmol), thiazol-2-amine (68 mg, 0.68 mmol), *O*-(7-azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (230 mg, 0.68 mmol), diisopropylethylamine (440 mg, 3.4 mmol) in *N,N*-dimethylformamide (7 mL) was stirred at room temperature overnight. The reaction solution was poured into water (25 mL). The precipitate was filtered, dissolved in dichloromethane, dried, concentrated and triturated with ethyl acetate:petroleum ether (1:3), filtered and washed with ethyl acetate:petroleum ether (1:3) to give 8-(5-(pyrrolidin-1-ylmethyl)thiazol-2-yl)-N-(thiazol-2-yl)quinoline-2-carboxamide (Compound 288) as a white solid (8 mg, yield 6%). MS calcd for $C_{21}H_{19}N_5OS_2$: 421.10; found: 422 [M+1].

Example 12. Preparation of 4-bromo-2-(pyrrolidin-1-ylmethyl)pyridine:

Step 1) Synthesis of 4-bromopicolinaldehyde (43):

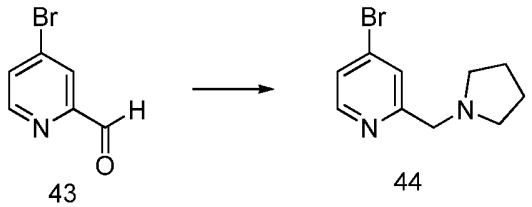


To the solution of 4-bromopicolinic acid (**41**; 6.1 g, 30 mmol), methoxymethylamine HCl salt (3.92 g, 40 mmol) and N-methylmorpholine (12.2 g, 120 mmol) in dichloromethane (120 ml) was added 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (7.68 g, 40 mmol) in portions at 0°C, the reaction mixture was warmed up to r.t. overnight. The reaction mixture was extracted with dichloromethane and washed with water and brine. The organic

solution was dried over Na_2SO_4 and concentrated to give crude 4-bromo-N-methoxy-N-methylpicolinamide **42** (5.84 g, yield 79%).

4-bromo-N-methoxy-N-methylpicolinamide (**42**; 5.84 g, 23.8 mmol) was dissolved in anhydrous THF (100 ml) and cooled to -78° C and 1.0 M lithium aluminum hydride in THF (14.3 ml, 14.3 mmol) was added via syringe and then the resulting mixture was stirred for 1 hour. 1 M NaOH (20 ml) and water (20 ml) was added carefully to the reaction mixture, and then the resulting solution was stirred for 30 minutes. Ethyl acetate and water was added, the organic phase was washed with water and brine, dried over Na_2SO_4 and concentrated to give 4-bromopicolinaldehyde **43** as a yellow oil (3.95 g, yield: 89%). MS calcd for C_6H_4BrNO: 186; found: 187 [M+1].

Step 2) Synthesis of 4-bromo-2-(pyrrolidin-1-ylmethyl)pyridine (44):

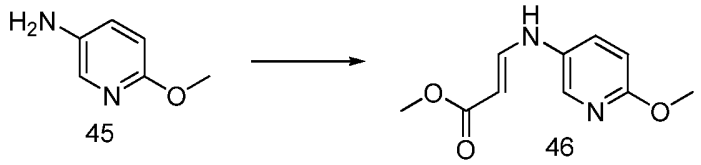


4-Bromo-2-(pyrrolidin-1-ylmethyl)pyridine **44** was prepared following a similar procedure as that of 2-bromo-5-(pyrrolidin-1-ylmethyl)thiazole **38** above using 4-bromopicolinaldehyde (1.35 g, 30%). MS calcd for $C_{10}H_{13}BrN2$: 240; found: 241 [M+1].

The above procedures were followed using the appropriate aldehydes in place of 4-bromopicolinaldehyde **43** to form analogous bromo-(pyrrolidin-1-ylmethyl)pyridines.

Example 13. Preparation of *N*-(pyridine-3-yl)-8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxamide (Compound 401):

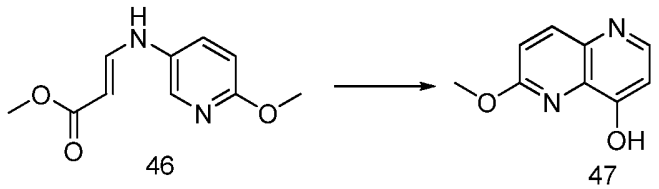
Step 1) Preparation of (E)-methyl 3-(6-methoxypyridin-3-ylamino)acrylate (46):



A mixture of 6-methoxypyridin-3-amine (**45**; 66.0 g, 532 mmol) and methyl propiolate (54.0 g, 640 mmol) in methanol (150 mL) was stirred at 80 °C (oil bath)

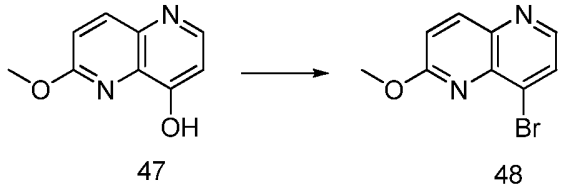
for 24h. The reaction mixture was cooled to room temperature and filtered. The filter cake was washed with methanol (20 mL × 3) to give (*E*)-methyl 3-(6-methoxypyridin-3-ylamino)acrylate **46** (84.0 g, 403 mmol, 76%) as a white solid. MS calcd for $C_{10}H_{12}N_2O_3$: 208.2; found: 209 [M+1].

Step 2) Synthesis of 6-methoxy-1,5-naphthyridin-4-ol (47):



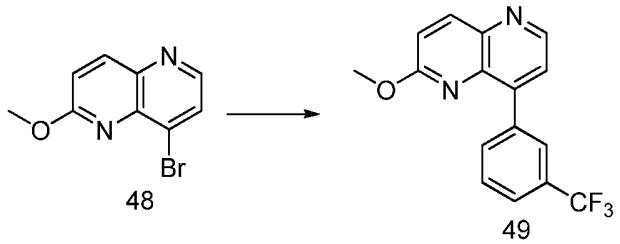
To a hot solvent (250°C) of diphenyl ether (150 mL) was added 3-(6-methoxypyridin-3-ylamino)acrylate (**46**; 20.0 g, 96 mmol) portionwise and the mixture stirred for 10-20 min. to give 6-methoxy-1,5-naphthyridin-4-ol **47** (6.0 g, 33.9 mmol, 35%) as a grey solid. MS calcd for $C_9H_8N_2O_2$: 176.2; found: 177 [M+1].

Step 3) Synthesis of 8-bromo-2-methoxy-1,5-naphthyridine (48):



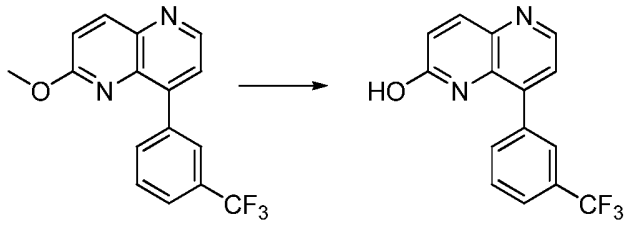
To a mixture of *N,N*-dimethylformamide (26.4 g, 0.36 mol) in acetonitrile (350 mL) was added PBr_3 (51.0 g, 0.188 mol) at room temperature. The mixture was stirred at 90°C for 30 min. Then 6-methoxy-1,5-naphthyridin-4-ol (**47**; 22.0 g, 0.124 mol) was added and the mixture was stirred for 30 min. After cooling to room temperature, the solvent was removed in vacuo and adjusted with saturated $NaHCO_3$ to pH=10. The precipitate was filtered and the filter cake was washed with water and dried in vacuo to give 8-bromo-2-methoxy-1,5-naphthyridine **48** (20.0 g, 0.083 mol, 67%) as a white solid. MS calcd for $C_9H_7BrN_2O$: 239.1; found: 240 [M+1].

Step 4) Synthesis of 2-methoxy-8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine (49):



The mixture of 8-bromo-2-methoxy-1,5-naphthyridine (**48**; 11.0 g, 45.98 mmol), 3-(trifluoromethyl) boronic acid and $PdCl_2$dppf (3.85 g, 4.6 mmol) in dioxane/water 10:1 (110 mL) was stirred under nitrogen at 60°C for 6 h. The mixture was diluted with dichloromethane (200 ml) and washed with water. The combined organic phase was dried over Na_2SO_4, concentrated and purified with silica gel column (petroleum ether:ethyl acetate 5:1) to give 2-methoxy-8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine **49** as a yellow solid (9.73 g, 70%). MS calcd for $C_{16}H_{11}F_3N_2O$: 304.3; found: 305 [M+1].

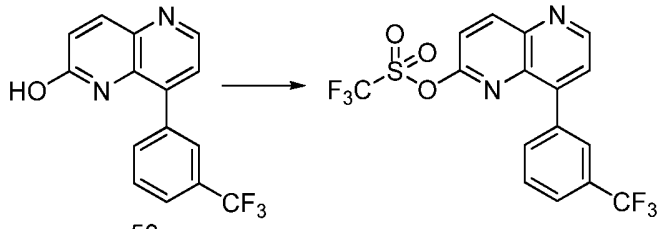
Step 5) Synthesis of 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridin-2-ol (50):



49 50

A solution of 2-methoxy-8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine (**49**; 1.0 g, 3.29 mmol) in HCl (6M, 40 mL) was heated at reflux for 2 h. The reaction mixture was cooled to 0°C and the pH was adjusted to 7 with 50% NaOH solution. The mixture was filtered, and the solid was washed with water. The solid was then dissolved in dichloromethane, dried and concentrated to give 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridin-2-ol **50** as a white solid (930mg, 98%). MS calcd for $C_{15}H_9F_3N_2O$: 290.2; found: 291 [M+1].

Step 6) Synthesis of 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridin-2-yl trifluoromethanesulfonate (51):

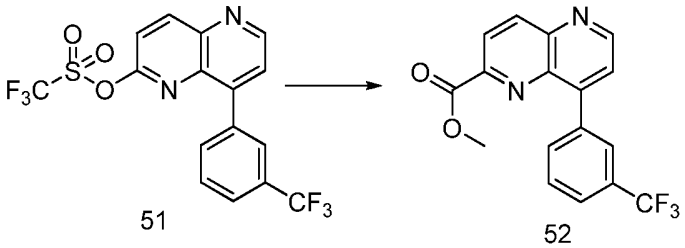


50 51

To a solution of 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridin-2-ol (**50**; 930 mg, 3.2 mmol) in pyridine (15 ml) was added trifluoromethanesulfonic anhydride (1.35 g, 4.8 mmol) dropwise at 0°C. The reaction mixture was warmed to room temperature slowly, then stirred for 4h. The reaction was quenched with water

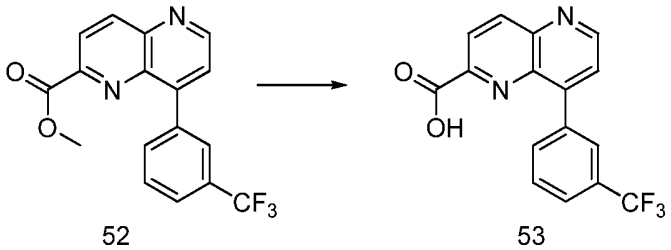
(10 mL) and poured slowly into sat. $NaHCO_3$ (50 ml). The mixture was extracted with ethyl acetate, washed with water, dried over anhydrous sodium sulfate, concentrated and purified via silica gel column chromatography (petroleum ether : ethyl acetate 20:1 to 10:1) to give 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridin-2-yl trifluoromethanesulfonate **51** as a yellow oil (1.2 g, 89%). MS calcd for $C_{16}H_8F_6N_2O_3S$: 422.3; found: 423 [M+1].

Step 7) Synthesis of methyl 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxylate (52):



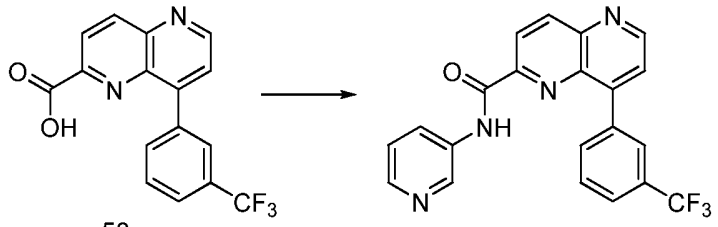
8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridin-2-yl trifluoromethanesulfonate (**51**; 211 mg, 0.5 mmol) and $PdCl_2$dppf (42 mg, 0.05 mmol) were put under nitrogen via three vacuum/N_2 cycles. Et_3N (0.14 ml, 1.03 mmol), *N,N*-dimethylformamide (4 mL), MeOH (2 mL) were added via septum and syringe. The flask was connected to a nitrogen balloon, followed by exchange with a carbon monoxide balloon. The mixture was heated under a carbon monoxide atmosphere to 80°C for 24h. The mixture was diluted with ethyl acetate, washed with water and brine, dried over anhydrous sodium sulfate, concentrated, and the residue purified with prep. TLC (petroleum ether : ethyl acetate 2:1) to give methyl 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxylate **52** as a yellow solid (71 mg, 43%). MS calcd for $C_{17}H_{11}F_3N_2O_2$: 332.3; found: 333 [M+1].

Step 8) Synthesis of 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxylic acid (53):



To a mixture of methyl 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxylate (**52**; 2.1 g, 6.34 mmol) in THF/water 10:1 (15 ml) was added $LiOH{\cdot}H_2O$ (533 mg, 12.7 mmol). The reaction mixture was stirred at room temperature overnight. The solvent was evaporated, water was added to the mixture, and the byproducts were extracted with diethyl ether. The water layer was adjusted with acetic acid to pH=6 to form a white precipitate. The mixture was filtered to collect the solid. The solid was then dissolved with ethyl acetate/dichloromethane (30 ml), dried over anhydrous sodium sulfate, and concentrated to give 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxylic acid **53** as a white solid (1.71 g, 85%). MS calcd for $C_{16}H_9F_3N_2O_2$: 318.06; found: 317.0 (M-1).

Step 9) Synthesis of N-(pyridine-3-yl)-8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxamide (Compound 401):



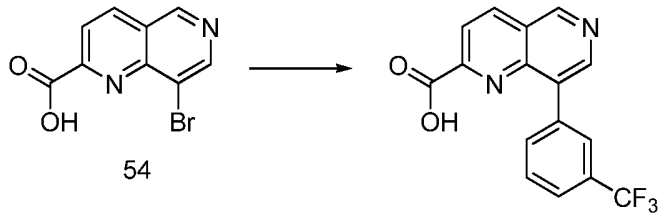
53 Compound 401

A mixture of 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxylic acid (**53**; 50 mg, 0.16 mmol), 3-aminopyridine (16 mg, 0.17 mmol), *O*-(7-Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (122 mg, 0.32 mmol), and diisopropylethylamine (62 mg, 0.48 mmol) in *N,N*-dimethylformamide (3 mL) was stirred at room temperature overnight. The mixture was poured into water and filtered to get a yellow solid. The solid was dissolved in ethyl acetate, washed with water and brine, dried over anhydrous sodium sulfate, and concentrated to give a yellow solid. The solid was triturated with petroleum ether:diethyl ether (4:1) and filtered to give *N*-(pyridine-3-yl)-8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxamide (Compound 396; 50.3 mg, 79.8%). MS calcd for $C_{21}H_{13}F_3N_4O$: 394.10; found: 395.1[M+1].

This general procedure is used to produce any compound of the invention containing 8-(3-(trifluoromethyl)phenyl)-1,5-naphthyridine-2-carboxamide, by using the appropriate amine component in place of 3-aminopyridine.

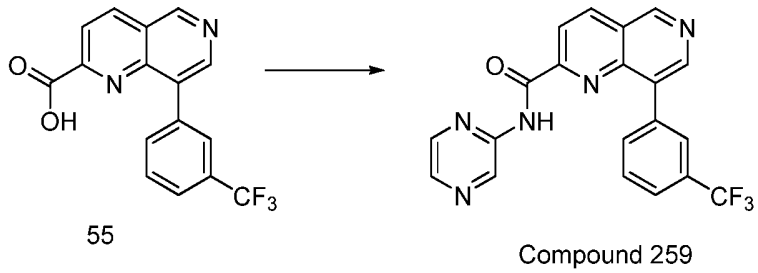
Example 14. Preparation of *N*-(pyrazin-2-yl)-8-(3-(trifluoromethyl)phenyl)-1,6-naphthyridine-2-carboxamide (Compound 259):

Step 1) Synthesis of 8-(3-(trifluoromethyl)phenyl)1,6-naphthyridine-2-carboxylic acid (55):



In a 5 mL microwaveable vial, a mixture of 8-bromo-1,6-naphthyridine-2-carboxylic acid (**54**; 200 mg, 0.79 mmol), 3-(trifluoromethyl)phenylboronic acid (180 mg, 0.95 mmol), potassium phosphate (503 mg, 2.4 mmol), and dicyclohexyl(2',6'-dimethoxybiphenyl-2-yl)phosphine (26 mg, 0.06 mmol) in 1,4-dioxane (3.0 mL) and water (0.3 mL) was sparged with nitrogen for 3 minutes. Tris(dibenzylideneacetone)dipalladium(0) (29 mg, 0.03 mmol) was added, and the mixture was sparged with nitrogen another 2 minutes, then sealed and heated in the microwave to 125 °C for 1.5 h. Water (20 mL) and ethyl acetate (20 mL) were added, and the layers separated. The aqueous layer was adjusted to pH 4 by addition of drops of 5 N HCl, after which time a precipitate formed. The mixture was filtered, and the solid was washed with water, ethyl acetate and ether. The solid was dried under vacuum to give 8-(3-(trifluoromethyl)phenyl)1,6-naphthyridine-2-carboxylic acid **55** as a gray solid (181 mg, 72%). MS (ESI) calcd for $C_{16}H_9F_3N_2O_2$: 318.06; found: 319 [M+H].

Step 2) Synthesis of N-(pyrazin-2-yl)-8-(3-(trifluoromethyl)phenyl)-1,6-naphthyridine-2-carboxamide (Compound 259):

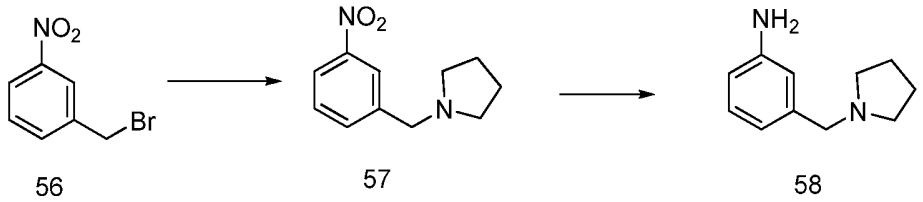


To a solution of 8-(3-(trifluoromethyl)phenyl)1,6-naphthyridine-2-carboxylic acid (**55**; 36 mg, 0.11 mmol) in *N, N*-dimethylformamide (0.9 mL) was added *O*-(7-

Azabenzotriazol-1-yl)-*N,N,N',N'*-tetramethyluronium hexafluorophosphate (64 mg, 0.17 mmol) and diisopropylethylamine (0.08 mL, 0.45 mmol). Aminopyrazine (11 mg, 0.11 mmol) was dissolved in *N, N*-dimethylformamide (1.0 mL) and added to the reaction mixture. The reaction was warmed to 50 °C under nitrogen atmosphere. After 4 h, aqueous sodium bicarbonate (5 mL) was added and the mixture was allowed to stir for 10 min. Water was added (20 mL), and the mixture was extracted with dichloromethane (3 x 20 mL). The combined organics were washed with brine, dried with sodium sulfate, filtered, and concentrated. The crude material was purified by prep HPLC (0.1% trifluoroacetic acid in water, with 10-80% acetonitrile). The product obtained after lyophilization was *N*-(pyrazin-2-yl)-8-(3-(trifluoromethyl)phenyl)-1,6-naphthyridine-2-carboxamide (Compound 254; 8.4 mg, 19%). MS (ESI) calcd for $C_{20}H_{12}F_3N_5O$: 395.10; found: 396 [M+H].

This general procedure is used to produce other compounds of the invention containing 8-(3-(trifluoromethyl)phenyl)-1,6-naphthyridine-2-carboxamide by using the appropriate amine component in place of aminopyrazine.

Example 15. Preparation of 3-(pyrrolidin-1-ylmethyl)aniline (58):

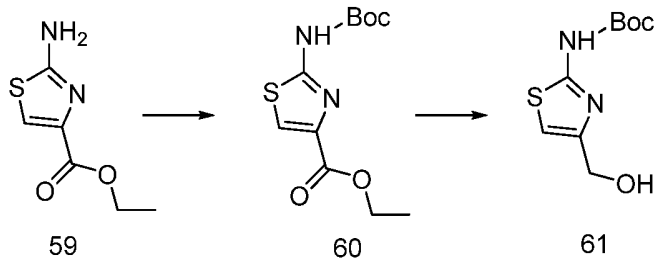


1-(Bromomethyl)-3-nitrobenzene (**56**; 5 g, 23.1 mmol) was taken up in 100 mL of anhydrous THF along with pyrrolidine (2.3 mL, 27.72 mmol) and K_2CO_3 (4.8 g, 34.6 mmol). The reaction mixture was stirred at room temperature for 18 h and then filtered. The filtrate was concentrated under reduced pressure to afford 1-(3-nitrobenzyl)pyrrolidine **57**. This material was taken up in 100 mL of absolute EtOH and 10% Pd on C (300 mg) was added. The resulting reaction mixture was stirred at room temperature under 1 atm of hydrogen for 18 h. The mixture was then filtered through a pad of Celite and the filtrate was concentrated under reduced pressure to afford 2.81 g of 3-(pyrrolidin-1-ylmethyl)aniline **58** (70%). MS (ESI) calcd for $C_{11}H_{16}N_2$: 176.3; found: 177 [M+H].

4-(pyrrolidin-1-ylmethyl)aniline was prepared similarly to the above procedure by using 1-(Bromomethyl)-4-nitrobenzene as starting material in place of 1-(Bromomethyl)-3-nitrobenzene **56**.

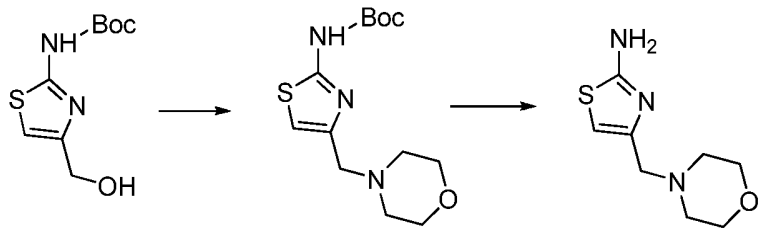
Example 16. Preparation of 4-(morpholinomethyl)thiazol-2-amine (63):

Step 1) Synthesis of tert-butyl 4-(hydroxymethyl)thiazol-2-ylcarbamate (61):



Ethyl 2-aminothiazole-4-carboxylate (**59**; 10.0 g, 58.1 mmol) was taken up in 150 mL of anhydrous THF along with di-tert-butyl carbonate (Boc_2O, 12.67 g, 58.1 mmol) along with 10 mg of 4-(dimethyl)aminopyridine (DMAP). The reaction mixture was stirred at 50°C for 4 h and then at room temperature for 18 h. It was then concentrated under reduced pressure to obtain a thick oil. Pentane was added and the resulting crystalline materials were collected by filtration and dried to afford 10.5 g of ethyl 2-(tert-butoxycarbonylamino)thiazole-4-carboxylate **60**. This material (**60**; 10.5 g, 38.5 mmol) was dissolved in 300 mL of anhydrous THF and cooled in Dry Ice-acetonitrile bath. A solution of 1 M Super Hydride™ in THF (85 mL) was then added over a period of 10 min. The resulting reaction mixture was stirred at -45°C for 2 h. Another portion of 1 M Super Hydride™ in THF (35 mL) was then added and the reaction mixture was stirred for an additional 2 h at -45°C. The reaction was quenched at -45°C by the addition of 50 mL of brine. Upon warming to room temperature, the reaction mixture was concentrated under reduced pressure. The resulting mixture was extracted with EtOAc. The combined organic layers were washed with brine, dried (Na_2SO_4) and concentrated under reduced pressure. The resulting residue was purified by chromatography to afford 6.39 g of tert-butyl 4-(hydroxymethyl)thiazol-2-ylcarbamate **61** (72%). MS (ESI) calcd for $C_9H_{14}N_2O_3S$: 230.3; found: 231 [M+H].

Step 2) Synthesis of 4-(morpholinomethyl)thiazol-2-amine (63):

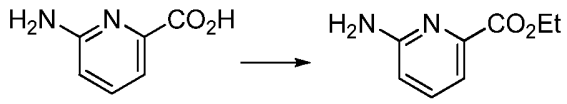


61 62 63

tert-Butyl 4-(hydroxymethyl)thiazol-2-ylcarbamate (**61**; 2.0 g, 8.7 mmol) was taken up in 25 mL of CH_2Cl_2 along with Et_3N (1.82 mL, 13.05 mmol) and cooled to 0 °C. Methanesulfonyl chloride (0.85 mL, 10.88 mmol) was added and the resulting reaction mixture was stirred at 0 °C for 60 min. Morpholine (3.0 mL, 35 mmol) was then added and the reaction mixture was stirred at room temperature for 18 h. The reaction mixture was concentrated under reduced pressure. The resulting residue was taken up in EtOAc and washed with dilute aqueous $NaHCO_3$, brine, dried (Na_2SO_4) and concentrated under reduced pressure. This material was purified by filtering through a short column of silica gel. The filtrate was concentrated to afford 1.88 g of tert-butyl 4-(morpholinomethyl)thiazol-2-ylcarbamate **62**. The Boc group was removed by treating tert-butyl 4-(morpholinomethyl)thiazol-2-ylcarbamate **62** with 20 mL of 25% TFA in CH_2Cl_2 for 18 h at room temperature. After all the solvent had been removed by concentrating and drying under high vacuum, the resulting residue was treated with a mixture of pentane/EtOAc to afford 2.17 g 4-(morpholinomethyl)thiazol-2-amine **63** as a white solid. MS (ESI) calcd for $C_8H_{13}N_3OS$: 199.3; found: 200 [M+H].

Example 17. Preparation of 6-(pyrrolidin-1-ylmethyl)pyridin-2-amine (70):

Step 1) Synthesis of ethyl 6-aminopicolinate (65):

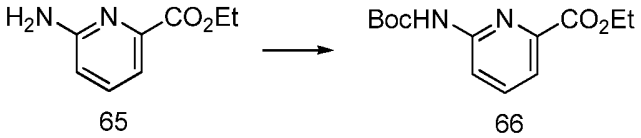


64 65

To a solution of 2-amino-6-pyridinecarboxylic acid (**64**; 6.0 g, 43.5 mmol) in ethanol (150 mL) was added $SOCl_2$ (12.0 g, 101 mmol) at 0°C. The resulting reaction mixture was stirred under reflux for 12 h. Upon cooling to room temperature, the reaction mixture was concentrated under reduced pressure. Enough saturated aqueous Na_2CO_3 solution was added to adjust the pH = 9. The mixture was concentrated under reduced pressure and dichloromethane (150 mL) was added to

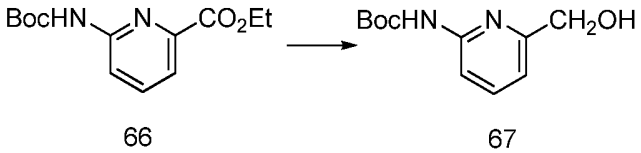
the resulting residue. The mixture was stirred vigorously at room temperature for 30 min and then filtered. The filtrate was concentrated under reduced pressure to afford ethyl 6-aminopicolinate **65** (5.5 g, 76%). MS (ESI) calcd for $C_8H_{10}N_2O_2$: 166.2; found: 167 [M+H].

Step 2) Synthesis of ethyl 6-(tert-butoxycarbonylamino)picolinate (66):



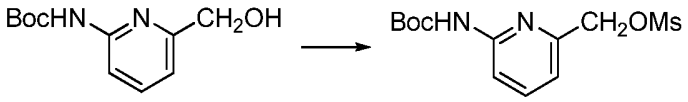
To a solution of ethyl 6-aminopicolinate (**65**; 5.5 g, 33 mmol) in *t*-BuOH (120 mL) and acetone (40 mL) was added DMAP (0.08g, 0.66 mmol) and di-t-butyl dicarbonate (10.8 g, 49.5 mmol). The reaction mixture was stirred at room temperature for 18 h. The solvent was removed by concentration under reduced pressure and a mixture of hexane/dichloromethane (180 mL, 3:1) was added. The resulting mixture was cooled to -20°C for 2 h. The resulting solids were collected by filtration and dried to afford ethyl 6-(tert-butoxycarbonylamino)picolinate **66** (11.0 g, 91%). MS (ESI) calcd for $C_{13}H_{18}N_2O_4$: 266.3; found: 267 [M+H].

Step 3) Synthesis of tert-butyl 6-(hydroxymethyl)pyridin-2-ylcarbamate (67):



To a stirred solution of ethyl 6-(tert-butoxycarbonylamino)picolinate (**66**; 11.0 g, 33 mmol) in THF (120 mL) under nitrogen was added $LiAlH_4$ (3.80 g, 100 mmol) in THF (60 mL) over a period of 30 min at 0°C. The reaction mixture was stirred at 0°C for 6 h and carefully quenched by the addition of water (2.0 mL) and 10% NaOH solution (4.0 mL) at 0°C. The reaction mixture was filtered and the filtrate was dried (Na_2SO_4) and concentrated under reduced pressure. The resulting residue purified by chromatography (1:1 petroleum ether:ethyl acetate) to afford tert-butyl 6-(hydroxymethyl)pyridin-2-ylcarbamate **67** (3.0 g, 41%). MS (ESI) calcd for $C_{11}H_{16}N_2O_3$: 224.3; found: 225 [M+H].

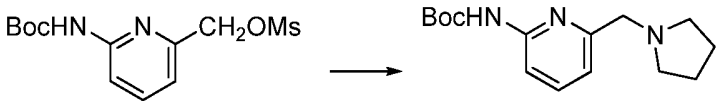
Step 4) Synthesis of (6-(tert-butoxycarbonylamino)pyridin-2-yl)methyl methanesulfonate (68):



67 68

To a solution of tert-butyl 6-(hydroxymethyl)pyridin-2-ylcarbamate (**67**; 3.0 g, 13.4 mmol) and DIPEA (5.0 g, 40 mmol) in acetonitrile (30 mL) was added MsCl (2.0 g, 17.4 mmol) over a period of 30 min at 0°C and the mixture was stirred for 2 h at room temperature. The reaction was quenched by adding saturated aqueous $NaHCO_3$ and extracted with ethyl acetate (3 × 60 mL). The combined organic layers were washed with brine, dried (Na_2SO_4) and concentrated under reduced pressure to afford essentially quantitative yield of crude (6-(tert-butoxycarbonylamino)pyridin-2-yl)methyl methanesulfonate **68**. MS (ESI) calcd for $C_{12}H_{18}N_2O_5S$: 302.3; found: 303 [M+H].

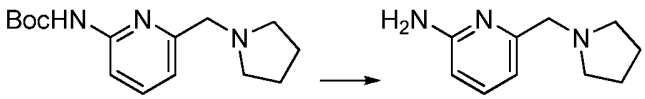
Step 5) Synthesis of tert-butyl 6-(pyrrolidin-1-ylmethyl)pyridin-2-ylcarbamate (69):



68 69

A mixture containing (6-(tert-butoxycarbonylamino)pyridin-2-yl)methyl methanesulfonate (**68**; 1.30 g, 3.2 mmol), pyrrolidine (0.46 g, 6.4 mmol) and K_2CO_3 (1.30 g, 9.6 mmol) in acetonitrile (15 mL) was stirred at room temperature for 12 h. Saturated aqueous $NaHCO_3$ was added and the mixture was concentrated under reduced pressure. The resulting aqueous layer was extracted with EtOAc. The combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure to afford tert-butyl 6-(pyrrolidin-1-ylmethyl)pyridin-2-ylcarbamate **69** (0.75 g, 2.7 mmol, 62% for two steps). MS (ESI) calcd for $C_{15}H_{23}N_3O_5$: 277.4; found: 278 [M+H].

Step 6) Synthesis of 6-(pyrrolidin-1-ylmethyl)pyridin-2-amine (70):

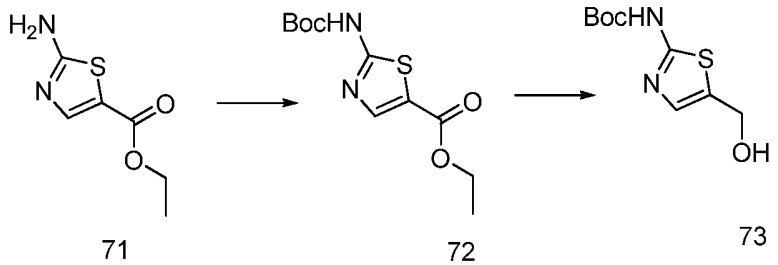


69 70

To a solution of tert-butyl 6-(pyrrolidin-1-ylmethyl)pyridin-2-ylcarbamate (**69**; 750 mg, 2.7 mmol) in dichloromethane (10 mL) was added TFA (4.0 mL) at room temperature. The resulting reaction mixture was stirred at room temperature for 6 h and then concentrated under reduced pressure. Enough saturated aqueous Na_2CO_3 was added to the resulting residue to adjust the pH = 9. The mixture was then extracted with ethyl acetate (3×25 mL). The combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure to afford 6-(pyrrolidin-1-ylmethyl)pyridin-2-amine **70** (440 mg, 92%). MS (ESI) calcd for $C_{10}H_{15}N_3$: 177.2; found: 178 [M+H].

Example 18. Preparation of 5-(pyrrolidin-1-ylmethyl)thiazol-2-amine (75):

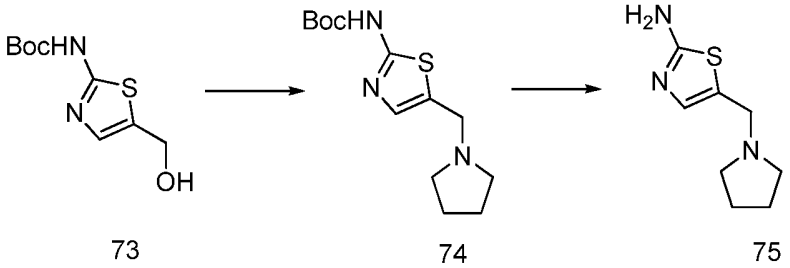
Step 1) Synthesis of tert-butyl 5-(hydroxymethyl)thiazol-2-ylcarbamate (73):



Ethyl 2-aminothiazole-5-carboxylate (**71**; 10.0 g, 58.1 mmol) was taken up in 150 mL of anhydrous THF along with di-tert-butyl carbonate (12.67 g, 58.1 mmol) along with 10 mg of 4-(dimethyl)aminopyridine. The reaction mixture was stirred at 50°C for 4 h and then at room temperature for 18 h. It was then concentrated under reduced pressure to obtain a thick oil. Pentane was added and the resulting crystalline materials were collected by filtration and dried to afford 10.5 g of ethyl 2-(tert-butoxycarbonylamino)thiazole-5-carboxylate **72**. This material (10.5 g, 38.5 mmol) was dissolved in 300 mL of anhydrous THF and cooled to -78 °C. A solution of 1 M Super Hydride™ in THF (85 mL) was then added over a period of 10 min. The resulting reaction mixture was stirred at -45°C for 2 h. Another portion of 1 M Super Hydride™ in THF (35 mL) was then added and the reaction mixture was stirred for an additional 2 h at -45°C. The reaction was quenched at -45°C by the addition of 50 mL of brine. Upon warming to room temperature, the reaction mixture was concentrated under reduced pressure. The resulting mixture was extracted with EtOAc. The combined organic layers were washed with brine, dried (Na_2SO_4) and concentrated under reduced pressure. The resulting residue was

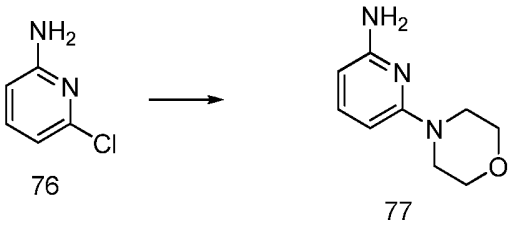
purified by silica gel chromatography to afford tert-butyl 5-(hydroxymethyl)thiazol-2-ylcarbamate **73**. MS (ESI) calcd for $C_9H_{14}N_2O_3S$ (m/z): 230.07, found 231 [M+H].

Step 2) Synthesis of 5-(pyrrolidin-1-ylmethyl)thiazol-2-amine (75):



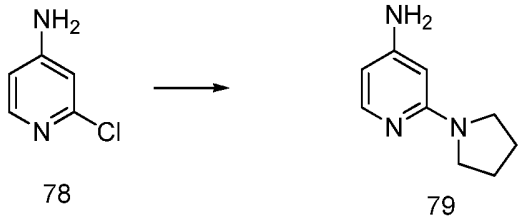
tert-butyl 5-(hydroxymethyl)thiazol-2-ylcarbamate (**73**; 2.0 g, 8.7 mmol) was taken up in CH_2Cl_2 (25 mL) along with Et_3N (1.82 mL, 13.05 mmol) and cooled to 0°C. Methanesulfonyl chloride (0.85 mL, 10.88 mmol) was added and the resulting reaction mixture was stirred at 0°C for 60 min. Pyrrolidine (2.87 mL, 35.0 mmol) was then added and the reaction mixture was stirred at room temperature for 18 h. The reaction mixture was concentrated under reduced pressure. The resulting residue was taken up in EtOAc and washed with dilute aqueous $NaHCO_3$, brine, dried (Na_2SO_4) and concentrated under reduced pressure. This material was purified by filtering through a short column of silica gel. The filtrate was concentrated to afford 1.8 g of tert-butyl 5-(pyrrolidin-1-ylmethyl)thiazol-2-ylcarbamate **74**. The Boc group was removed by treating tert-butyl 5-(pyrrolidin-1-ylmethyl)thiazol-2-ylcarbamate with 20 mL of 25% TFA in CH_2Cl_2 for 18 h at room temperature. After all the solvent had been removed by concentrating and drying under high vacuum, the resulting residue was treated with a mixture of pentane/EtOAc to afford 5-(pyrrolidin-1-ylmethyl)thiazol-2-amine **75** as a white solid (2.17 g). MS (ESI) calcd for $C_8H_{13}N_3S$ (m/z): 183.08 found, 184 [M+H].

Example 19. Preparation of 6-morpholinopyridin-2-amine (77):



A mixture containing 4-chloro-2-aminopyridine (**76**; 26g g, 0.20 mol), K_2CO_3 (0.40 mol) and morpholine (0.6 mol) in DMSO (150 mL) was stirred at 190°C for 10 h. Upon cooling to room temperature, the reaction mixture was diluted with water (300 mL) and the resulting mixture was extracted with ethyl acetate (4 x 150 mL). The combined organic layers were washed with water (3 x 25 mL), dried (Na_2SO_4) and concentrated under reduced pressure. The resulting residue was purified by chromatography (petroleum ether:ethyl acetate = 10:1) to afford 6-morpholinopyridin-2-amine **77** (17 g, 47%)as a white solid. MS (ESI) calcd for $C_9H_{13}N_3O$ (m/z): 179.2 found, 180 [M+H].

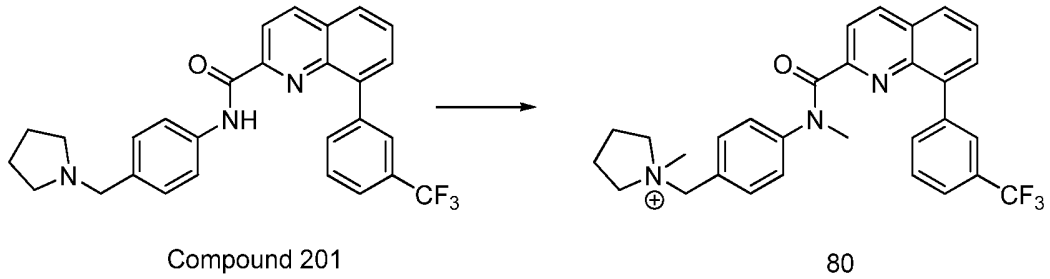
Example 20. Preparation of 2-(pyrrolidin-1-yl)pyridin-4-amine (79):



2-Chloropyridin-4-amine **78** was subjected to the same reaction conditions described above for the preparation of 6-morpholinopyridin-2-amine. Pyrrolidine was used as the amine component instead of morpholine.

Example 21. Preparation of *N*-methyl-*N*-(4-(pyrrolidin-1-ylmethyl)phenyl)-8-(3-(trifluoromethyl) phenylquinoline-2-carboxamide (Compound 265):

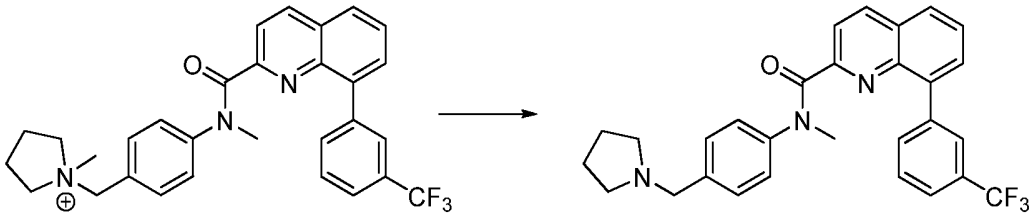
Step 1) Synthesis of 1-methyl-1-(4-(N-methyl-8-(3-(trifluoromethyl)phenyl) quinoline-2-carboxamido)benzyl)pyrrolidinium (80):



N-(4-(pyrrolidin-1-ylmethyl)phenyl)-8-(3-(trifluoromethyl)phenyl)quinoline-2-carboxamide (Compound 201; 50 mg, 0.1 mmol), was dissolved in toluene (3.3 mL) and cooled to 0°C under nitrogen atmosphere. Potassium hexamethyldisilazide (0.5 M, 1.05 mL, 0.53 mmol) was added and the reaction was allowed to warm to room temperature for 15 min, followed by re-cooling to 0°C. Iodomethane (0.03

mL, 0.53 mmol) was added, and after 5 min. the reaction was again warmed to room temperature. After 3 h, water was added (20 mL), and the mixture was extracted with dichloromethane (3 x 20 mL). The combined organics were washed with brine, dried with sodium sulfate, filtered and concentrated to give 60 mg of a mixture of 1-methyl-1-(4-(*N*-methyl-8-(3-(trifluoromethyl)phenyl) quinoline-2-carboxamido)benzyl)pyrrolidinium **80** and *N*-methyl-*N*-(4-(pyrrolidin-1-ylmethyl)phenyl)-8-(3-(trifluoromethyl) phenylquinoline-2-carboxamide.

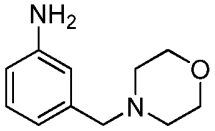
Step 2) Synthesis of N-methyl-N-(4-(pyrrolidin-1-ylmethyl)phenyl)-8-(3-(trifluoromethyl) phenylquinoline-2-carboxamide (Compound 265):



80 Compound 265

The crude mixture from above (0.1 mmol) was dissolved in neat pyrrolidine (0.8 mL, 10 mmol). The mixture was stirred at 70°C overnight, then sealed and heated in the microwave to 110°C for 1 h. Water was added (20 mL), and the mixture was extracted with dichloromethane (5 x 10 mL). The combined organics were washed with brine, dried with sodium sulfate, filtered and concentrated. The crude material was purified by prep. HPLC using 15-85% acetonitrile/water with 0.1% trifluoroacetic acid. The TFA salt was exchanged for the HCl salt, and the material was lyophilized to give *N*-methyl-*N*-(4-(pyrrolidin-1-ylmethyl)phenyl)-8-(3-(trifluoromethyl) phenylquinoline-2-carboxamide (Compound 260; 15 mg, 27%). MS (ESI) calcd for $C_{29}H_{26}F_3N_3O$: 489.2; found: 490 [M+1].

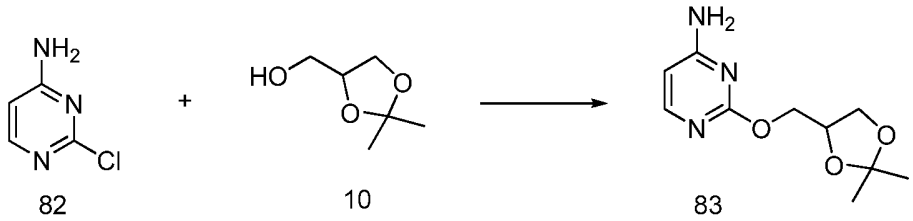
Synthesis of 3-(morpholinomethyl)aniline (81):



81

3-(morpholinomethyl)aniline **81** was prepared by a procedure similar to that reported in *J. Med. Chem.* **1990**, *33(1),* 327-36.

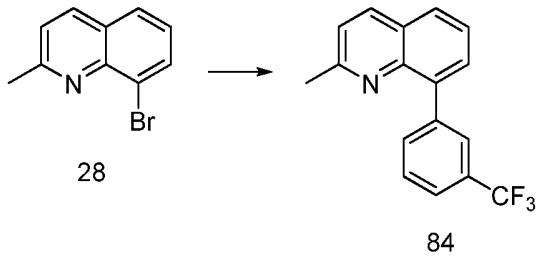
Example 22. Preparation of 2-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)pyrimidin-4-amine (83):



To a solution of solketal (**10**; 34.4 g, 260 mmol) in THF (150 mL) was added NaH (10.4 g, 260 mmol) at room temperature and the mixture stirred for 1h. 2-chloro-4-aminopyrimidine (**82**; 15.0 g, 115 mmol) was then added, and the mixture was stirred at 70°C for 48 h. The reaction mixture was concentrated and the crude residue was purified by flash chromatography (dichloromethane:methanol = 15:1 – 10:1) to give 2-((2,2-dimethyl-1,3-dioxolan-4-yl)methoxy)pyrimidin-4-amine **83** (18.2 g, 70 % yield) as an oil. MS (ESI) calcd for $C_{10}H_{15}N_3O_3$: 225.2; found: 226 [M+1].

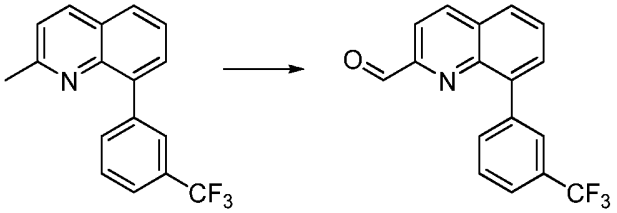
Example 23. Preparation of *N*-((8-(3-(trifluoromethyl)phenyl)quinolin-2-yl)methyl)pyrimidin-4-amine (Compound 388):

Step 1) Synthesis of 2-methyl-8-(3-(trifluoromethyl)phenyl)quinoline (84):



8-bromo-2-methylquinoline (**28**; 5.0 g, 22 mmol) was taken up in dioxane (50 mL) and water (15mL) along with 3-(trifluoromethoxy)phenylboronic acid (4.7 g, 25 mmol), Cs_2CO_3 (22 g, 67 mmol) and $Pd(dppf)Cl_2$ (938mg, 1mmol). The reaction mixture was stirred at 80^0C under nitrogen atmosphere for 3 h. The solid was filtered. The filtration was then diluted with water and extracted with ethyl acetate. The combined organic layers were washed with brine, dried over sodium sulfate then concentrated under reduced pressure. The residue was purified by column chromatography to afford 2-methyl-8-(3-(trifluoromethyl)phenyl)quinoline **84** (6.3 g , 97%). MS (ESI) calcd for $C_{17}H_{12}F_3N$: 287.3; found: 288 [M+1].

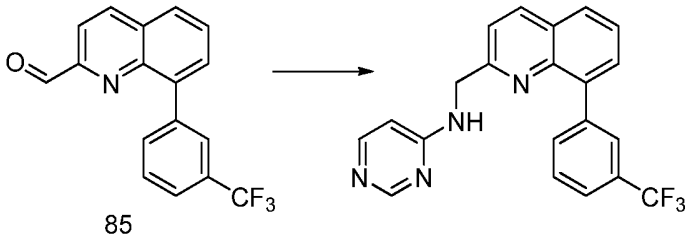
Step 2) Synthesis of 8-(3-(trifluoromethyl)phenyl)quinoline-2-carbaldehyde (85):



84 85

A mixture of 2-methyl-8-(3-(trifluoromethyl)phenyl)quinoline (**84**; 1.0 g) and SeO_2 (2.33 g) in dioxane (10 mL) was refluxed for 2 h. The solvent was removed and the residue was purified by column chromatography to afford 8-(3-(trifluoromethyl)phenyl)quinoline-2-carbaldehyde **85** (1.8 g, 82%). MS (ESI) calcd for $C_{17}H_{10}F_3NO$: 301.3; found: 302 [M+1].

Step 3) Synthesis of N-((8-(3-(trifluoromethyl)phenyl)quinolin-2-yl)methyl)pyrimidin-4-amine (Compound 388):



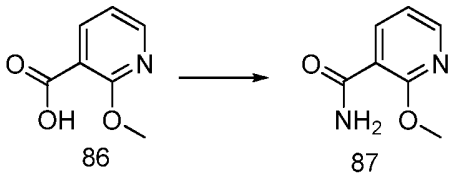
85 Compound 388

8-(3-(trifluoromethyl)phenyl)quinoline-2-carbaldehyde (**85**; 300 mg) and pyrinmidine-4-ylamine (114 mg) was dissolved in dimethylformamide (3 mL). Then $Ti(O\text{-}iPr)_4$ (876 mg) was added to the solution. The reaction mixture was stirred at room temperature under nitrogen atmosphere overnight. $NaBH_4$ (45 mg) was added to the mixture. The mixture was stirred at room temperature for another 2 h. The mixture was then diluted with aq. NH_4Cl and extracted with ethyl acetate. The combined organic layers were washed with brine, dried over sodium sulfate and then concentrated under reduced pressure. The residue was purified by preparative TLC to afford *N*-((8-(3-(trifluoromethyl)phenyl)quinolin-2-yl)methyl)pyrimidin-4-amine (Compound 383) (76 mg, 20%). MS (ESI) calcd for $C_{21}H_{15}F_3N_4$: 380.4; found: 381 [M+1].

This general procedure was used to synthesize other (8-(3-(trifluoromethyl)phenyl)quinolin-2-yl derivatives in Table 1 and can be used for any other (8-(3-(trifluoromethyl)phenyl)quinolin-2-yl methyl derivatives of the invention by substituting the appropriate amine for pyrinmidine-4-ylamine.

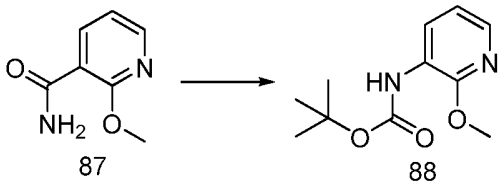
Example 24. Preparation of 8-Bromo-1,7-Naphthyridine-2-Carboxylic Acid (91)

Step 1) Preparation of 2-methoxynicotinamide (87):



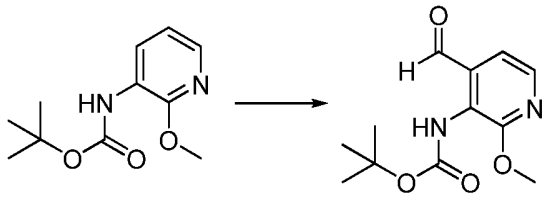
2-methoxynicotinamide **87** is prepared in a manner similar to the procedures in *Bioorg. Med. Chem. Lett.* **2007**, *17*, 2031 and *J. Org. Chem.* **1975**, *40*, 3554. To 2-methoxynicotinic acid (**86**; 49 mmol) suspended dichloromethane (60 mL) and DMF (3 drops) is added oxalyl chloride (16.6 mL, 186 mmol) at room temperature. The mixture is stirred at room temperature for 1 h, then concentrated and redissolved in petroleum ether (60 mL). This mixture is then added at -35°C to 250 mL of anhydrous acetonitrile that has been saturated with ammonia at -30 °C. After addition, the reaction is warmed to room temperature and allowed to stir 10 min. The mixture is concentrated and the residue taken up in hot ethyl acetate. This is filtered through diatomaceous earth, concentrated and followed by recrystallization from ethyl acetate to produce 2-methoxynicotinamide **87**.

Step 2) Preparation of tert-butyl 2-methoxypyridin-3-ylcarbamate (88):



Tert-butyl 2-methoxypyridin-3-ylcarbamate **88** is prepared in a manner similar to the procedure in *J. Med. Chem.* **1988**, *31*, 2136. To a suspension of 2-methoxynicotinamide (**87**; 10 mmol) in anhydrous *tert*-butyl alcohol (25 mL) is added lead tetraacetate (4.44 g, 10 mmol) under nitrogen atmosphere. The reaction mixture is heated to reflux for 2 h, then cooled and filtered through diatomaceous earth. The filtrate is concentrated and the residue dissolved in diethyl ether. The solution is washed with saturated aqueous sodium bicarbonate and brine, then dried with sodium sulfate, filtered, and concentrated to give the desired product **88** which can be purified by recrystallization.

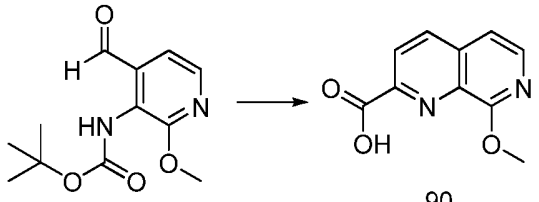
Step 3) Preparation of tert-butyl 4-formyl-2-methoxypyridin-3-ylcarbamate (89):



88 89

Tert-butyl 4-formyl-2-methoxypyridin-3-ylcarbamate **89** is prepared in a manner similar to the procedure in *J. Med. Chem.* **1988**, *31*, 2136. A solution of 2-methoxypyridin-3-ylcarbamate (**88**; 100 mmol) in dry THF (350 mL) is cooled to -78 °C, followed by dropwise addition of *tert*-butyllithium (120 mL, 2 M in pentane, 240 mmol) at a rate such that the temperature does not exceed -65 °C. The reaction is stirred at -78 °C for an additional 15 min., then at -20 °C for 1.5 h. Dry *N*-formylpiperidine (300 mmol) is added while maintaining the temperature below -15 °C, then the reaction is allowed to stir at room temperature overnight. The reaction is then cooled to 0 °C and quenched by addition of 1 N HCl to bring the pH to 2. Solid sodium carbonate is added to adjust the pH to 7. The solution is extracted with ethyl acetate and the combined organic layers washed with water and brine, dried with sodium sulfate, filtered and concentrated. The product **89** can be purified by silica gel column chromatography.

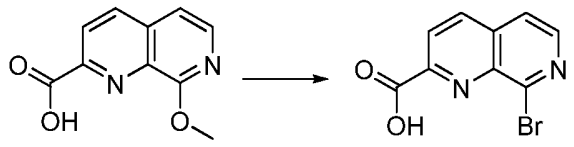
Step 4) Preparation of 8-methoxy-1,7-naphthyridine-2-carboxylic acid (90):



89 90

8-methoxy-1,7-naphthyridine-2-carboxylic acid **90** is prepared in a manner similar to the procedure in *Tetrahedron Lett.* **2000**, *41*, 8053. The Boc group of *yert*-butyl 4-formyl-2-methoxypyridin-3-ylcarbamate **89** is removed using trifluoroacetic acid in dichloromethane, followed by an aldol condensation with sodium pyruvate in sodium hydroxide/water to produce the desired product **90**.

Step 5) Preparation of 8-bromo-1,7-naphthyridine-2-carboxylic acid (91):



90 91

8-bromo-1,7-naphthyridine-2-carboxylic acid **91** is prepared in a manner similar to the procedure in *Bioorg. Med. Chem. Lett.* **2002**, *12*, 233. 8-methoxy-1,7-naphthyridine-2-carboxylic acid **90** is dissolved in DMF and treated with PBr_3 at 100 °C for 30 min to produce the desired product **91**.

8-bromo-1,7-naphthyridine-2-carboxylic acid **91** is derivatized with the appropriate R^2 group at the 8-position to produce a carboxylic acid intermediate that may be combined with an appropriate amine to produce compounds of the invention containing in a 8-substituted-1,7-naphthyridine-2-carboxamide.

Example 25. Biological activity

A mass spectrometry based assay was used to identify modulators of SIRT1 activity. The mass spectrometry based assay utilizes a peptide having 20 amino acid residues as follows: Ac-EE-K(biotin)-GQSTSSHSK(Ac)NleSTEG–K(5TMR)-EE-NH_2 (SEQ ID NO: 1) wherein K(Ac) is an acetylated lysine residue and Nle is a norleucine. The peptide is labeled with the fluorophore 5TMR (excitation 540 nm/emission 580 nm) at the C-terminus. The sequence of the peptide substrate is based on p53 with several modifications. In addition, the methionine residue naturally present in the sequence was replaced with the norleucine because the methionine may be susceptible to oxidation during synthesis and purification.

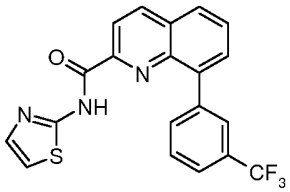
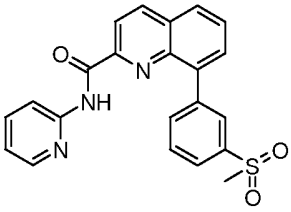
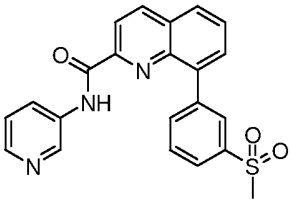
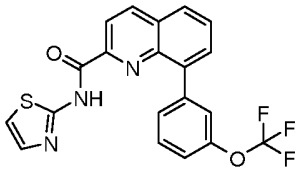
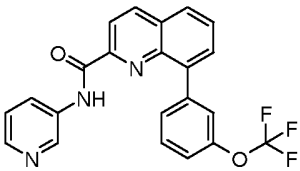
The mass spectrometry assay is conducted as follows: 0.5 μM peptide substrate and 120 μM βNAD^+ is incubated with 10 nM SIRT1 for 25 minutes at 25°C in a reaction buffer (50 mM Tris-acetate pH 8, 137 mM NaCl, 2.7 mM KCl, 1 mM $MgCl_2$, 5 mM DTT, 0.05% BSA). Test compounds may be added to the reaction as described above. The SirT1 gene is cloned into a T7-promoter containing vector and transformed into BL21(DE3). After the 25 minute incubation with SIRT1, 10 μL of 10% formic acid is added to stop the reaction. Reactions are sealed and frozen for later mass spec analysis. Determination of the mass of the substrate

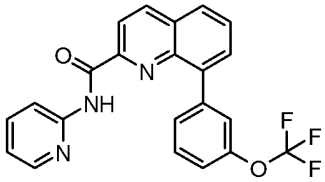
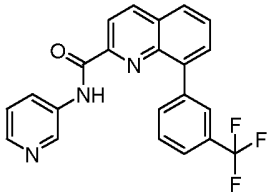
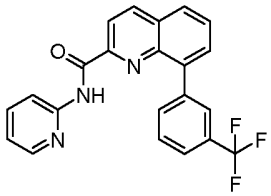
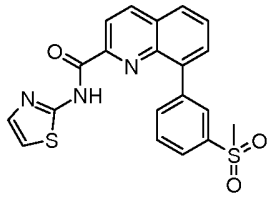
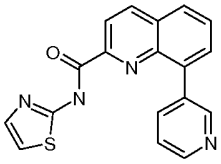
peptide allows for precise determination of the degree of acetylation (i.e. starting material) as compared to deacetylated peptide (product).

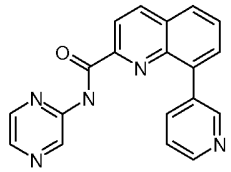
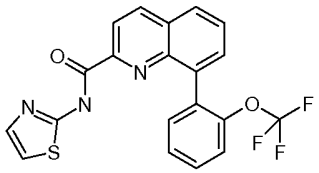
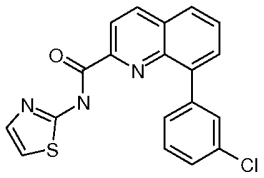
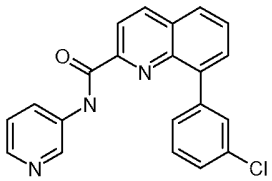
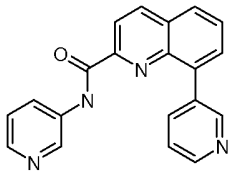
For the above assay, SIRT1 protein was expressed and purified as follows. The SirT1 gene was cloned into a T7-promoter containing vector and transformed into BL21(DE3). The protein was expressed by induction with 1 mM IPTG as an N-terminal His-tag fusion protein at 18°C overnight and harvested at 30,000 x g. Cells were lysed with lysozyme in lysis buffer (50 mM Tris-HCl, 2 mM Tris[2-carboxyethyl] phosphine (TCEP), 10 μM $ZnCl_2$, 200 mM NaCl) and further treated with sonication for 10 min for complete lysis. The protein was purified over a Ni-NTA column (Amersham) and fractions containing pure protein were pooled, concentrated and run over a sizing column (Sephadex S200 26/60 global). The peak containing soluble protein was collected and run on an Ion-exchange column (MonoQ). Gradient elution (200 mM - 500 mM NaCl) yielded pure protein. This protein was concentrated and dialyzed against dialysis buffer (20 mM Tris-HCl, 2 mM TCEP) overnight. The protein was aliquoted and frozen at -80°C until further use.

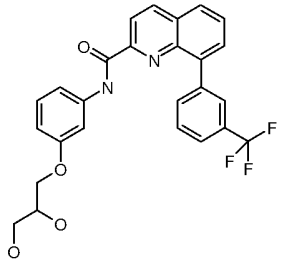
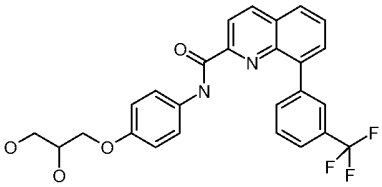
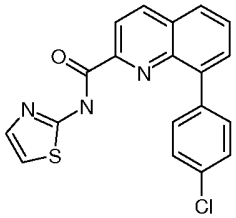
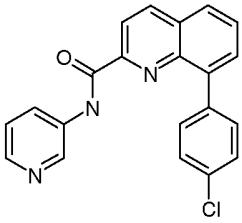
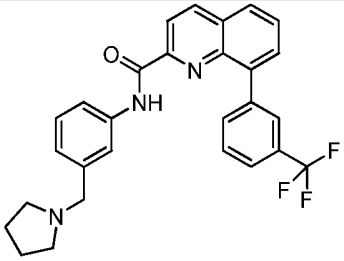
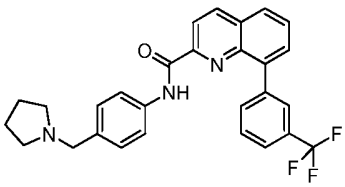
Sirtuin modulating compounds that activated SIRT1 were identified using the assay described above and are shown below in Table 1. The $EC_{1.5}$ values represent the concentration of test compounds that result in 150% activation of SIRT1. The $EC_{1.5}$ values for the activating compounds are represented A ($EC_{1.5} \leq 1$ uM), B ($EC_{1.5}$ >1 and ≤15 uM), or C ($EC_{1.5}$ >15 uM). The percent maximum fold activation is represented by A (Fold activation ≥350%), B (Fold Activation ≥150% and < 350%), or C (Fold Activation <150%).

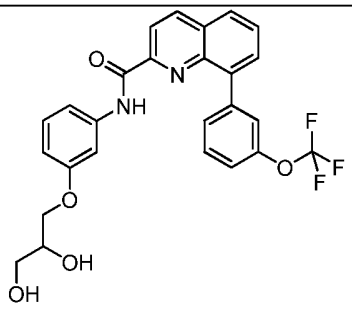
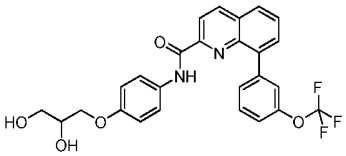
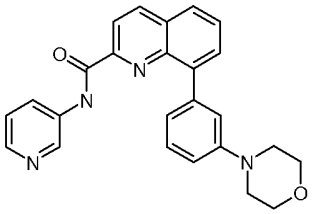
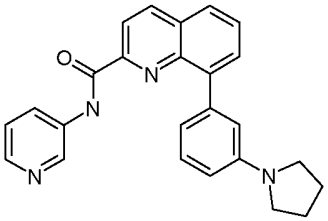
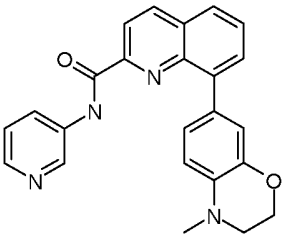
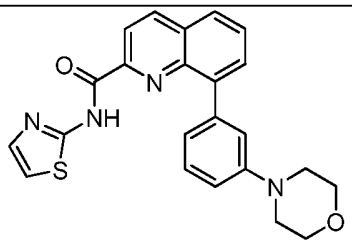
Table 1.

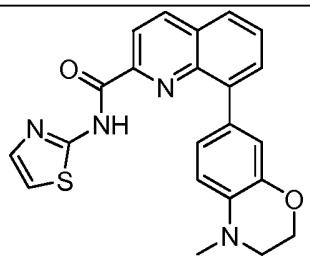
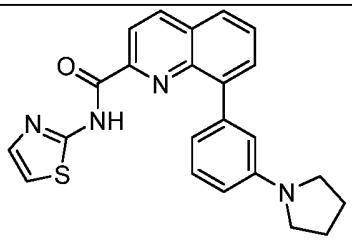
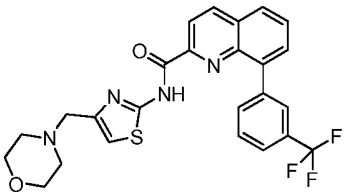
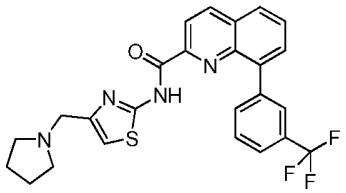
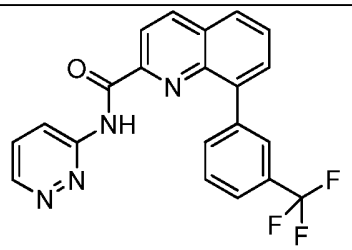
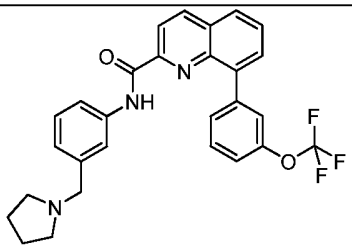
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
101	400		A	A
102	404		B	A
103	404		B	B
104	416		B	B
105	410		A	B

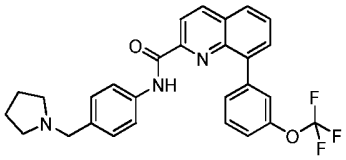
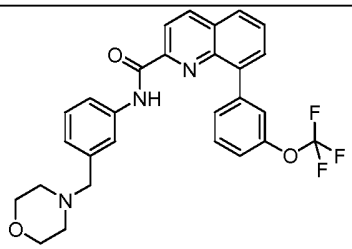
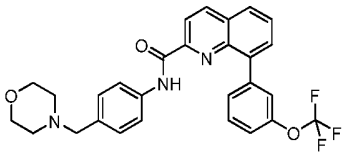
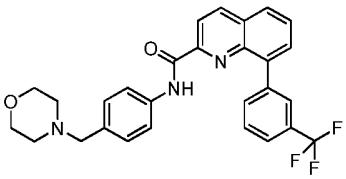
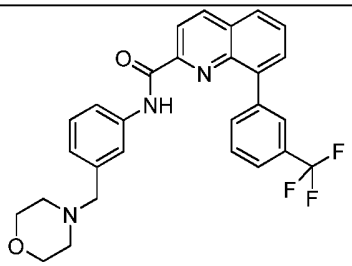
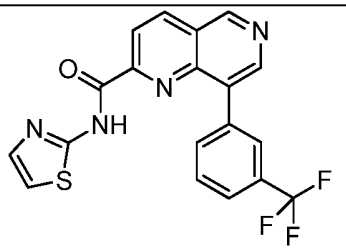
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
106	410		B	B
107	394		A	A
108	394		A	B
109	410		B	B
110	333		B	C

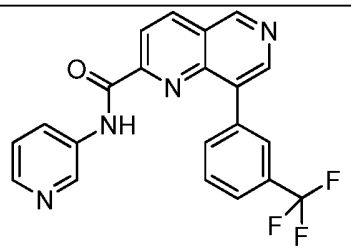
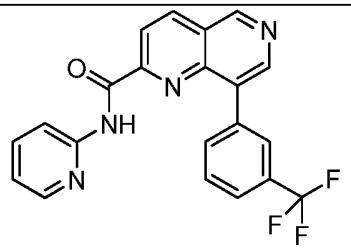
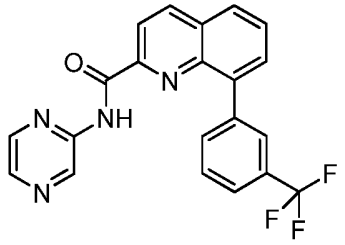
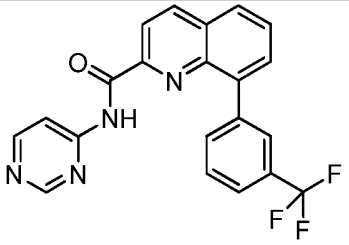
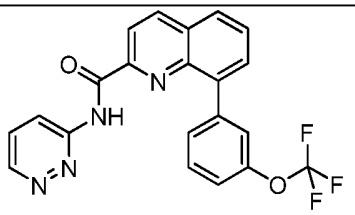
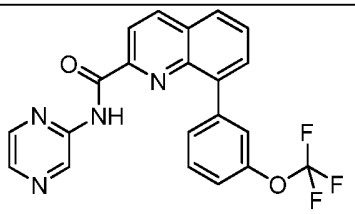
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
111	328		C	
112	416		C	
113	366		A	B
114	360		B	B
115	327		C	

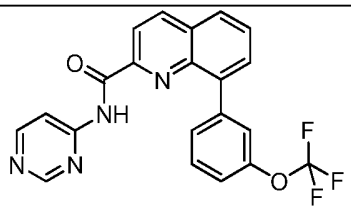
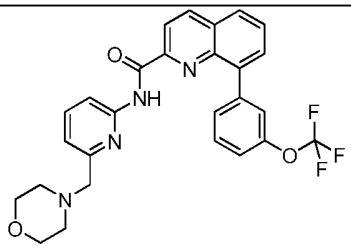
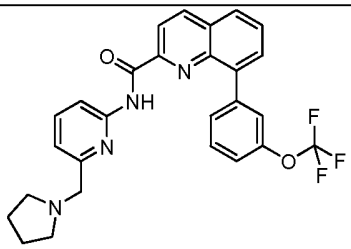
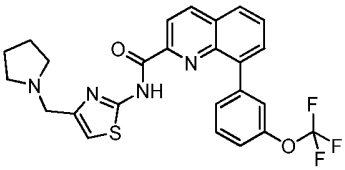
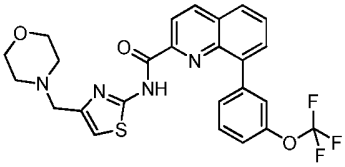
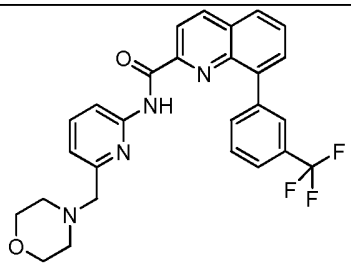
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
116	483		A	A
117	483		A	A
118	366		C	
119	360		C	
200	476		A	A
201	476		A	A

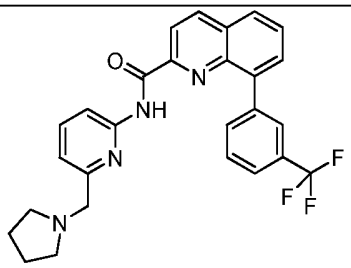
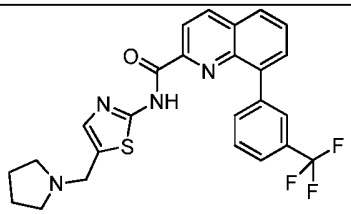
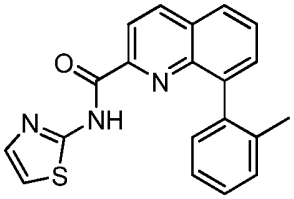
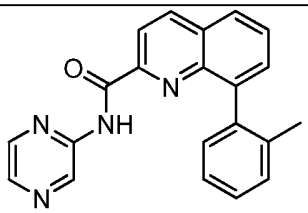
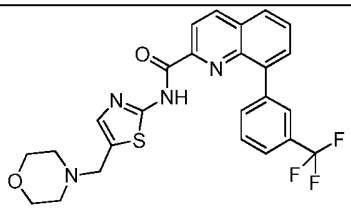
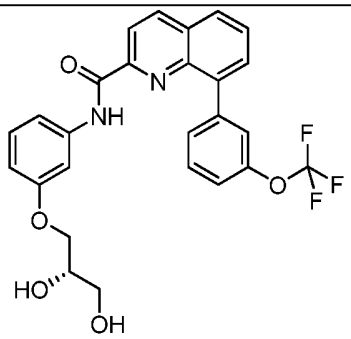
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
202	499		A	A
203	499		A	A
204	411		A	A
205	395		A	B
206	397		B	B
207	417		A	B

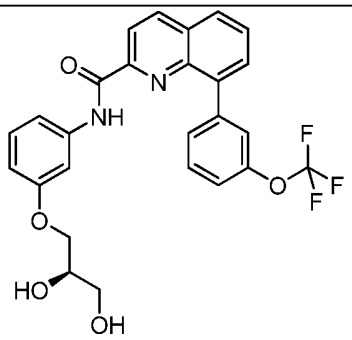
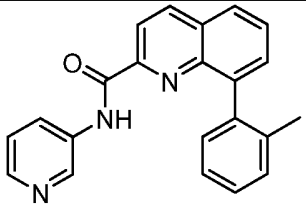
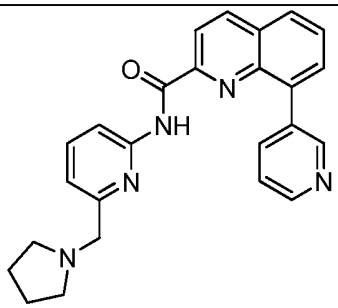
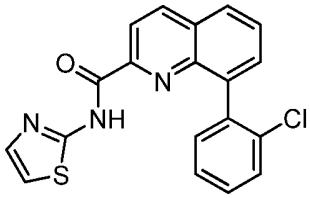
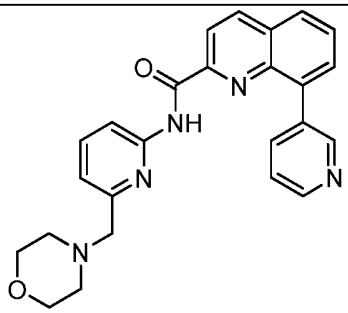
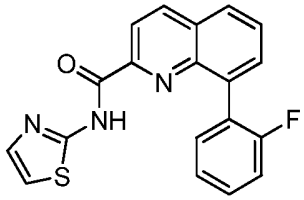
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
208	403		A	B
209	401		C	B
210	499		A	A
211	483		A	B
212	395		A	B
213	492		A	A

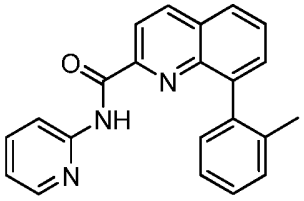
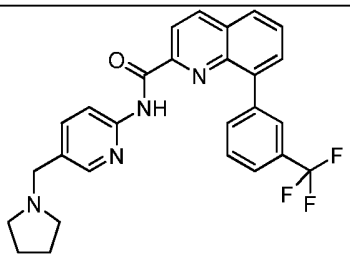
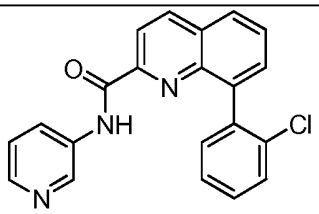
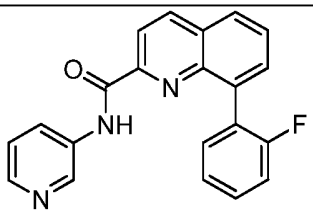
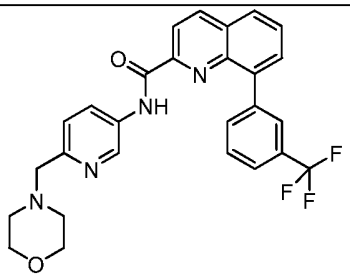
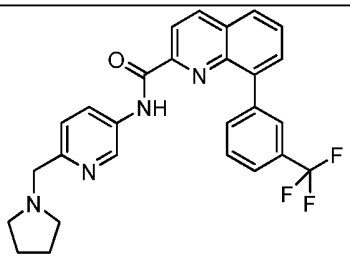
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
214	492		A	A
215	508		A	A
216	508		A	A
217	492		A	A
218	492		A	A
219	401		A	A

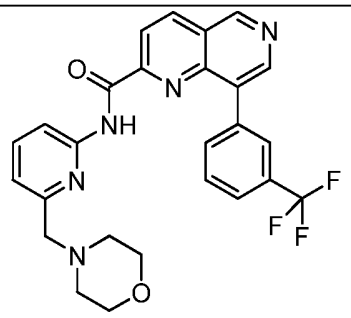
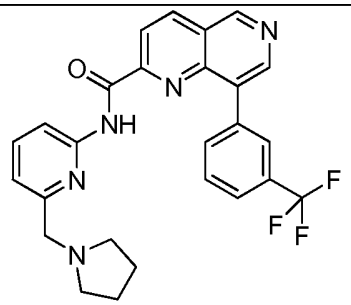
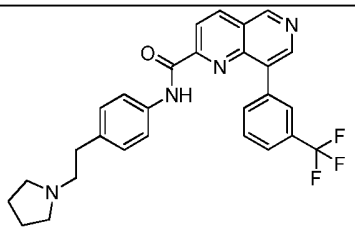
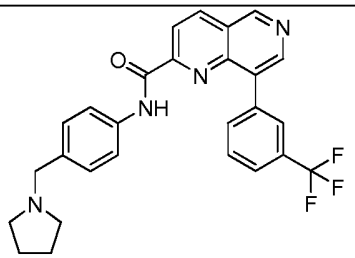
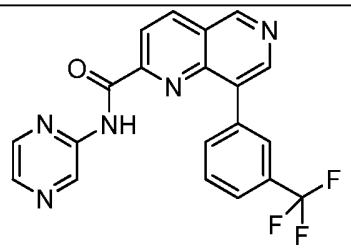
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
220	395		A	A
221	395		A	B
222	395		A	A
223	395		A	A
224	411		A	B
225	411		A	B

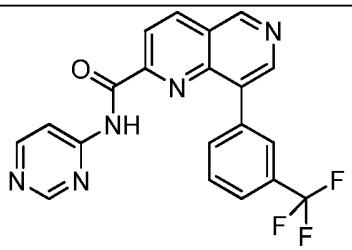
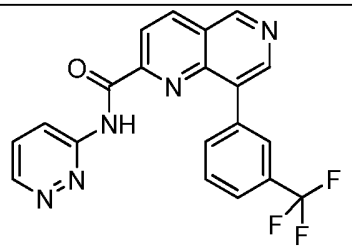
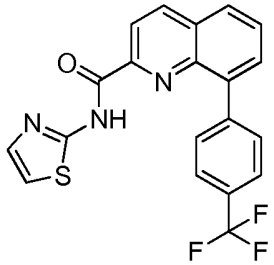
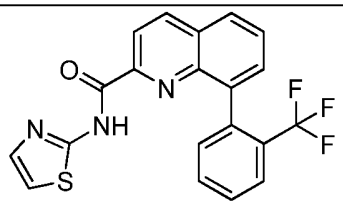
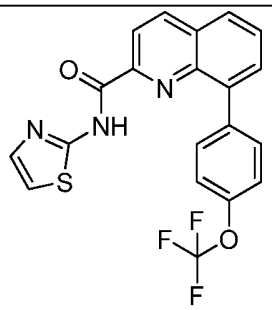
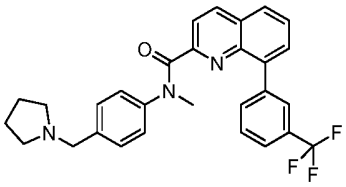
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
226	411		A	B
227	509		A	A
228	493		A	A
229	499		B	B
230	515		A	A
231	493		A	A

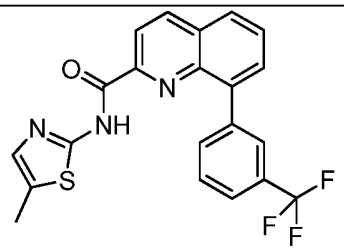
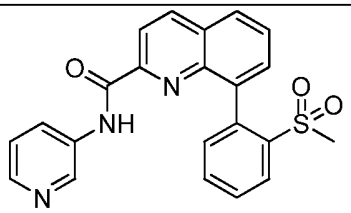
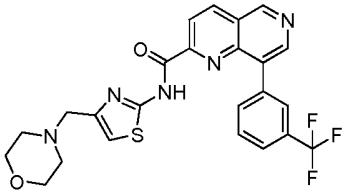
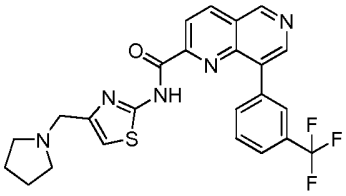
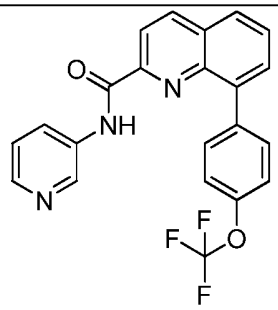
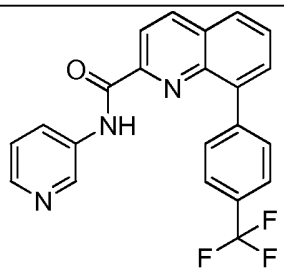
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
232	477		A	A
233	483		B	B
234	346		C	
235	341		C	C
236	499		A	A
237	499		A	A

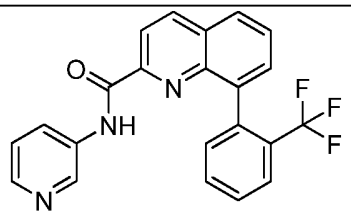
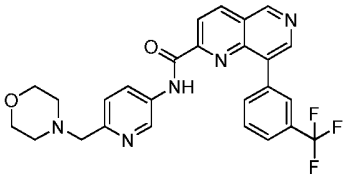
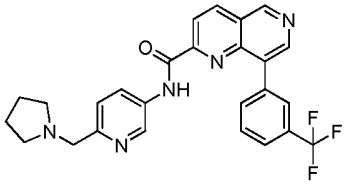
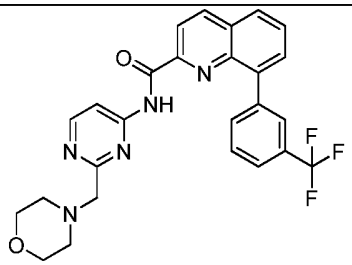
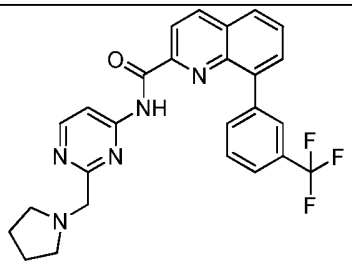
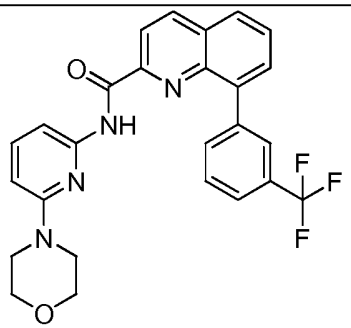
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
238	499		A	A
239	340		B	B
240	410		B	B
241	366		C	
245	426		A	A
246	350		C	

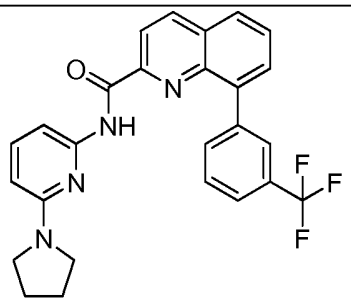
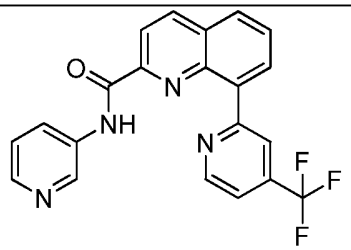
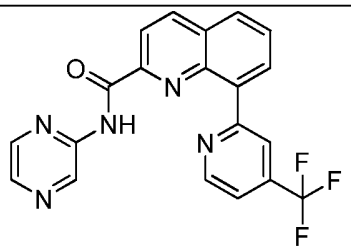
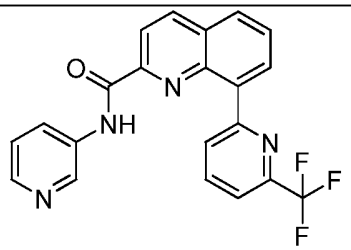
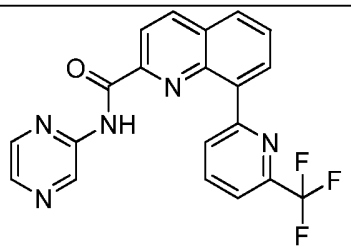
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
249	340		C	
250	477		A	A
251	360		B	B
252	344		B	B
253	493		A	A
254	477		A	A

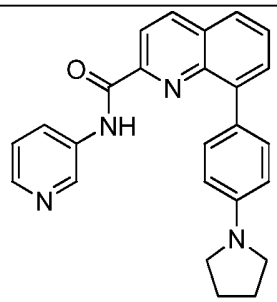
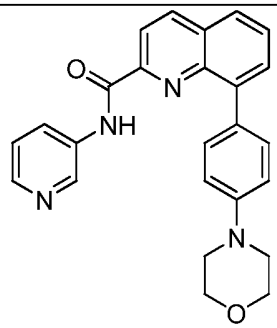
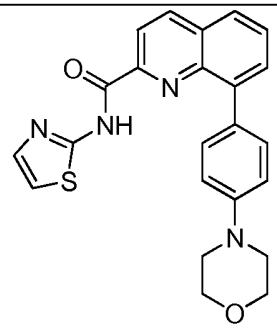
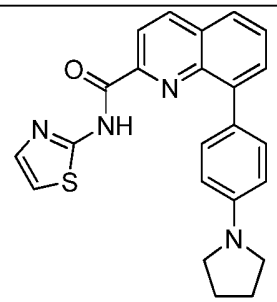
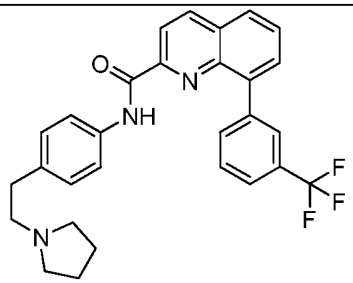
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
255	494		A	A
256	478		A	A
257	491		A	A
258	477		A	A
259	396		A	B

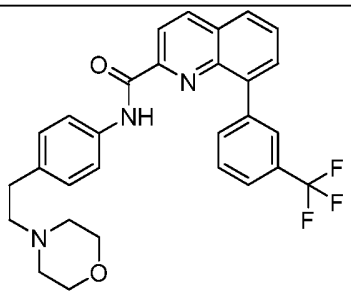
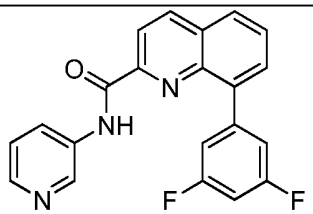
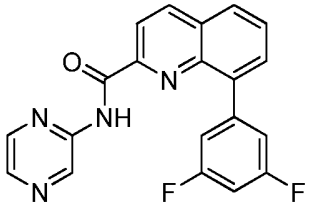
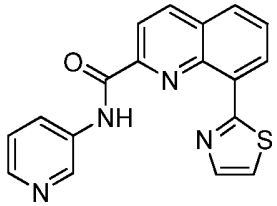
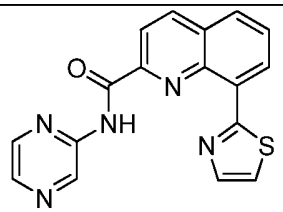
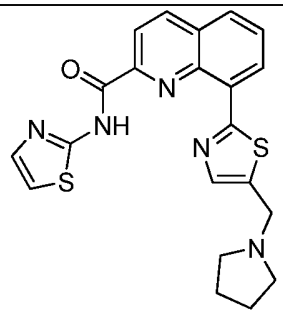
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
260	396		A	A
261	396		A	A
262	400		C	
263	400		C	
264	416		C	
265	490		B	B

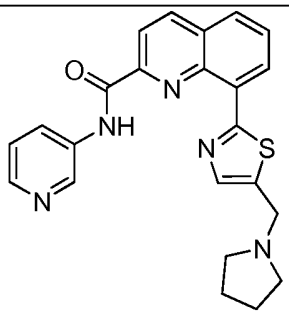
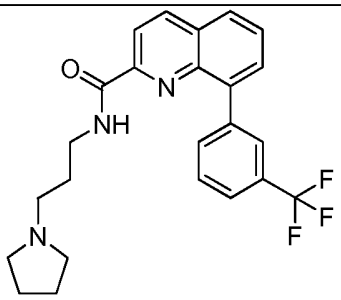
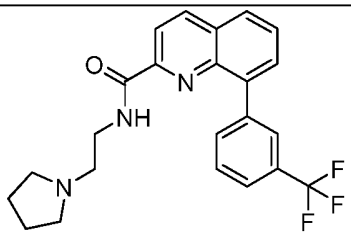
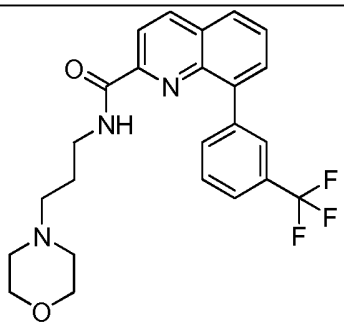
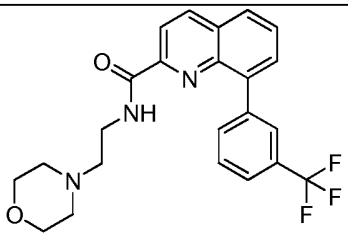
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
266	414		A	B
267	404		C	
268	500		A	A
269	484		A	A
270	410		C	
271	394		C	

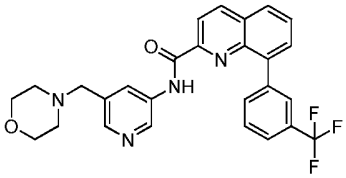
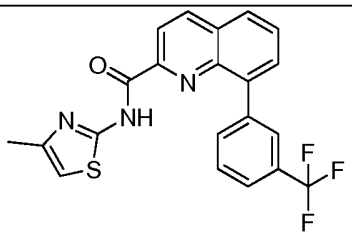
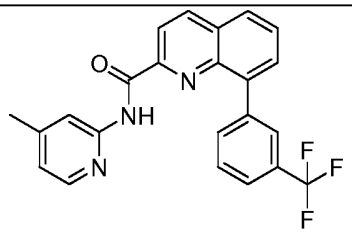
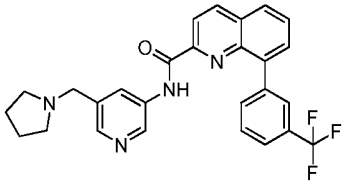
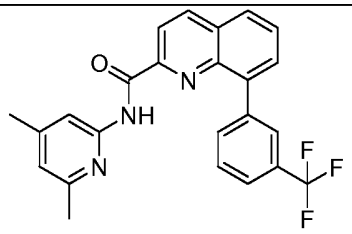
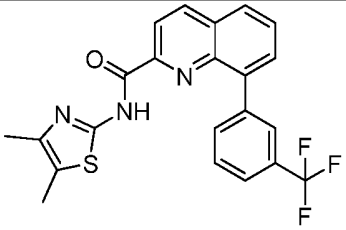
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
272	394		B	B
273	494		A	A
274	478		A	A
275	494		A	A
276	478		A	A
277	479		A	A

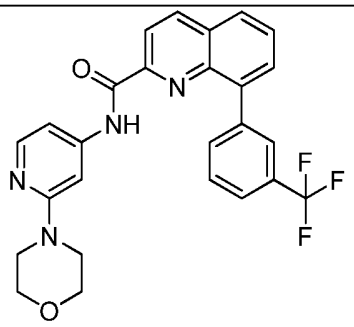
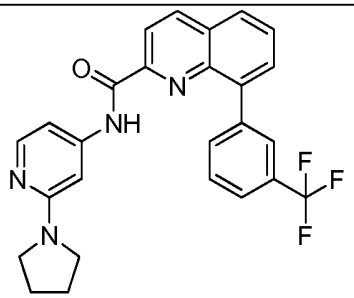
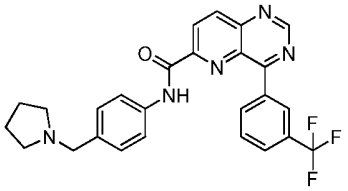
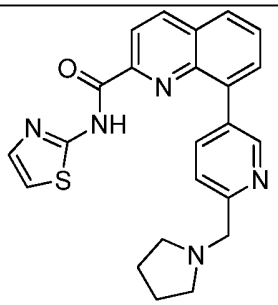
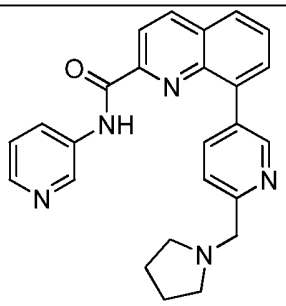
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
278	463		A	B
279	395		A	A
280	396		B	A
281	395		B	B
282	396		C	

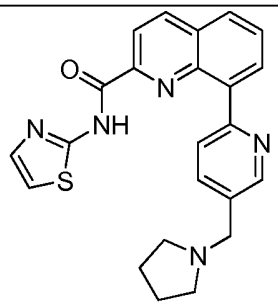
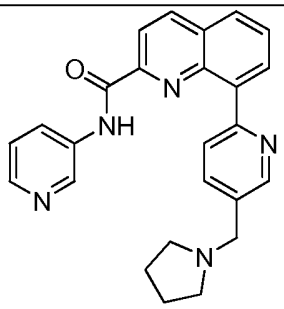
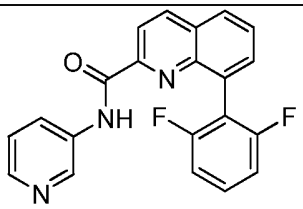
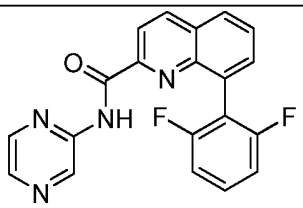
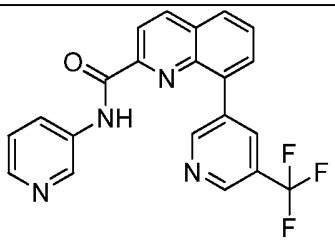
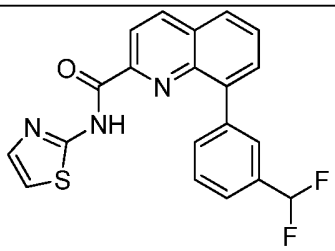
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
283	395		C	
284	411		C	
285	417		B	B
286	401		C	
287	490		A	A

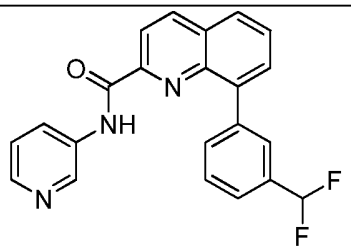
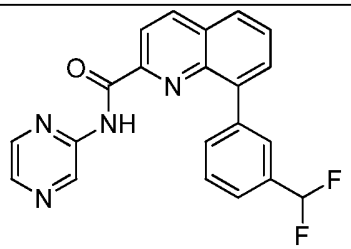
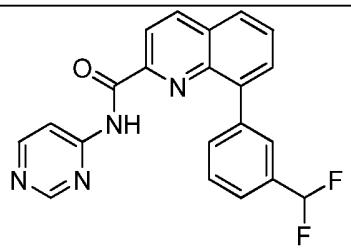
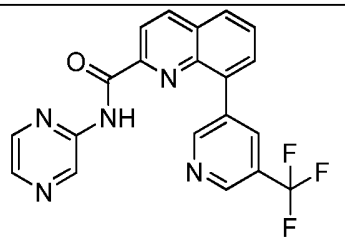
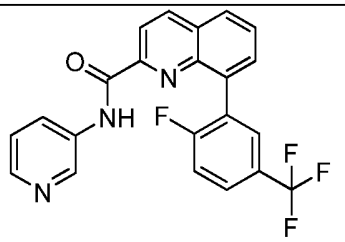
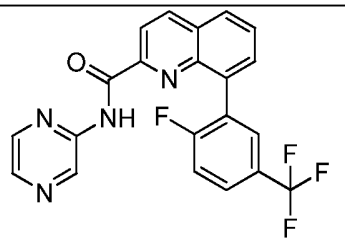
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
288	506		A	A
289	362		C	
290	363		B	B
291	333		B	B
292	334		B	B
293	422		B	A

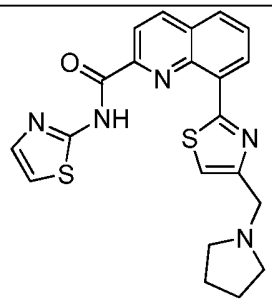
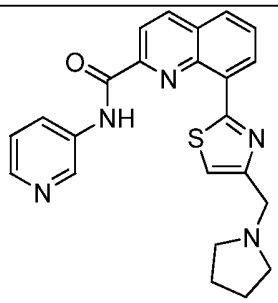
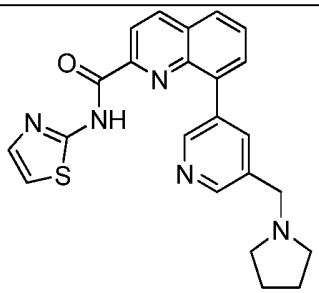
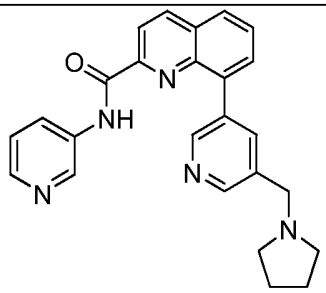
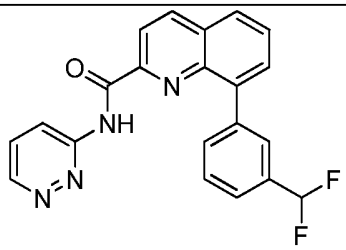
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
294	416		B	B
295	428		B	B
296	414		B	B
297	444		B	A
298	430		B	A

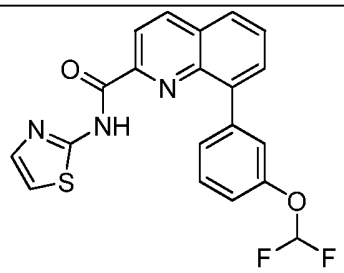
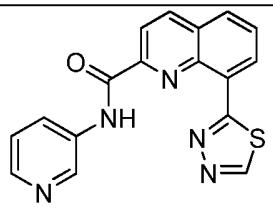
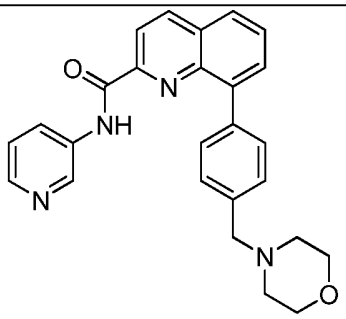
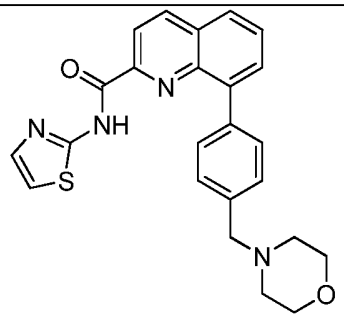
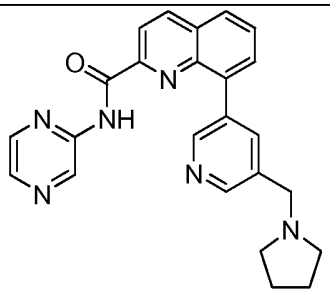
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
299	493		A	A
300	414		A	B
301	408		A	A
302	477		A	A
303	422		A	A
304	428		A	B

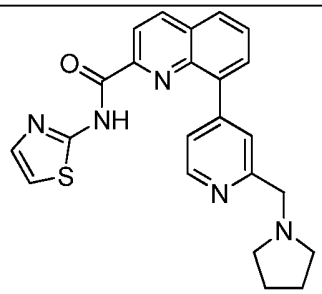
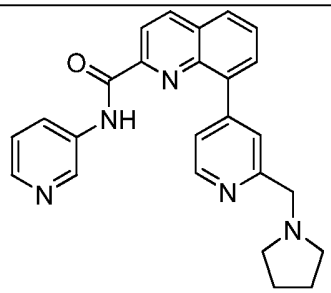
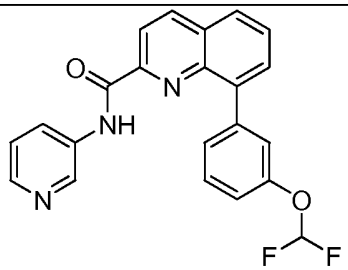
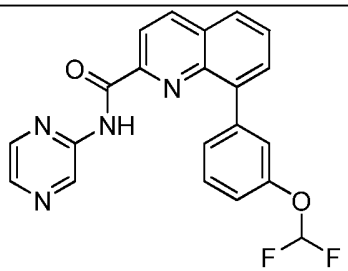
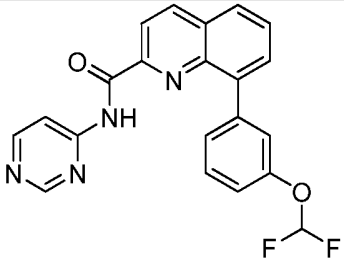
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
305	479		A	A
306	463		A	A
307	478		A	A
308	416		C	B
309	410		B	B

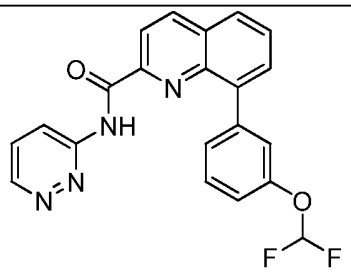
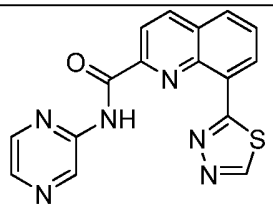
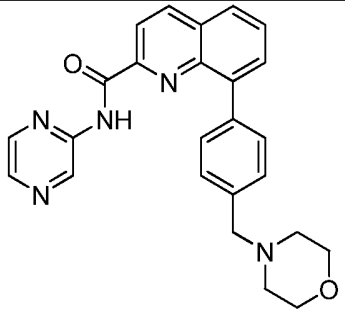
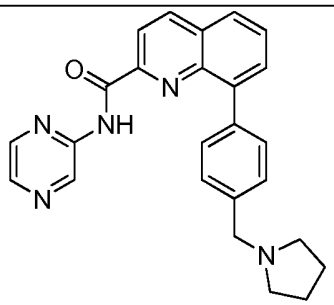
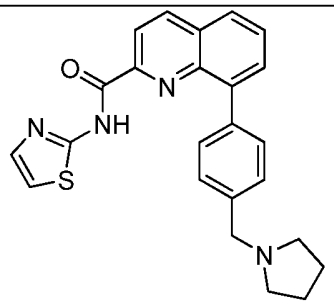
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
310	416		C	C
311	410		B	B
312	362		B	B
313	363		B	B
314	395		C	
315	382		A	B

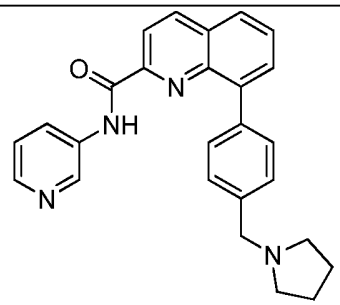
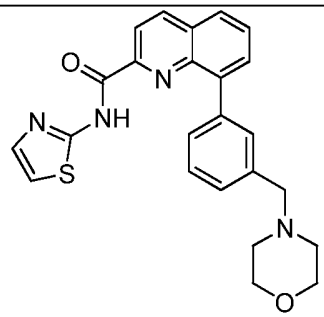
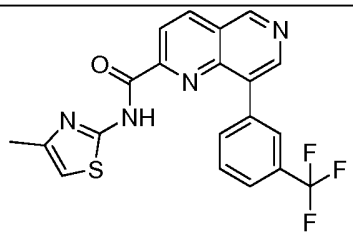
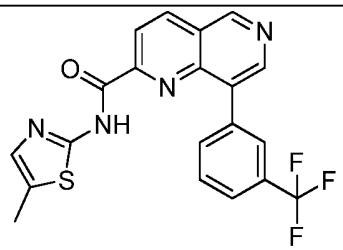
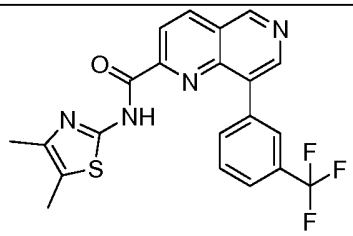
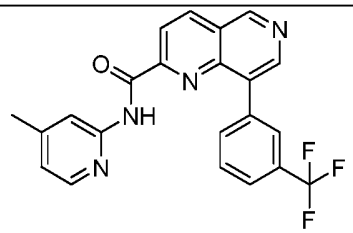
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
316	376		A	A
317	377		A	B
318	377		C	
319	396		A	A
320	412		A	B
321	413		A	A

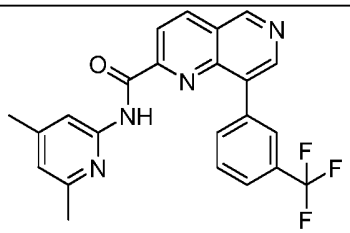
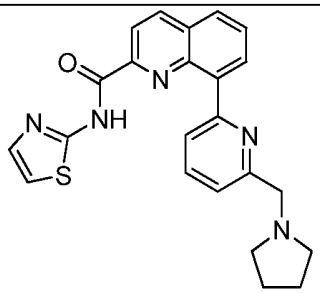
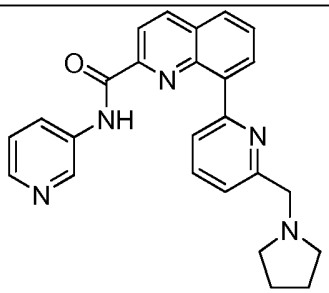
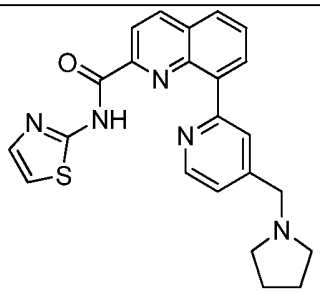
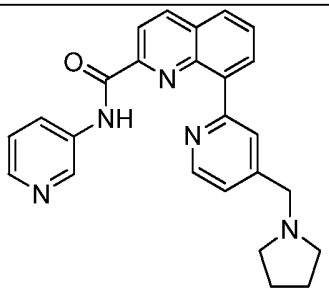
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
322	422		B	B
323	416		B	B
324	416		B	B
325	410		B	B
326	377		A	B

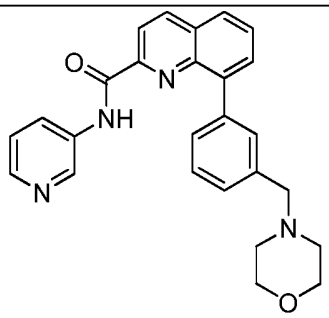
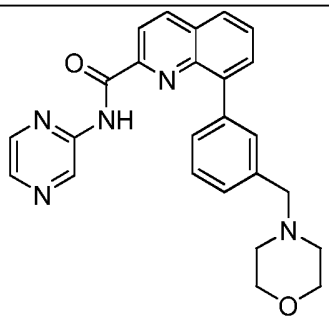
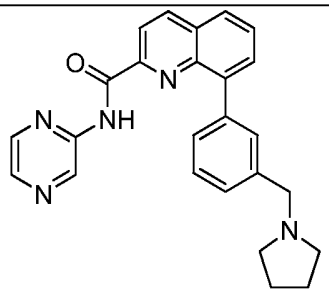
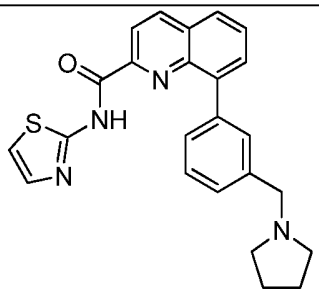
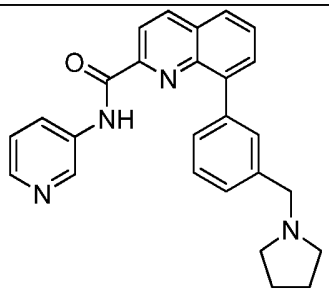
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
327	398		B	B
328	334		C	B
329	424		B	B
330	431		B	B
331	411		B	B

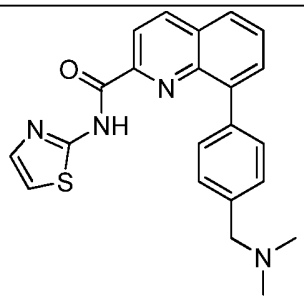
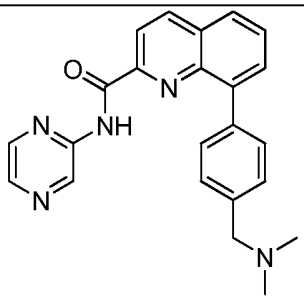
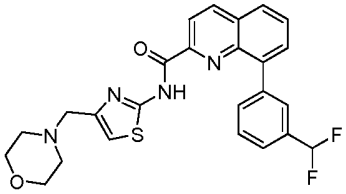
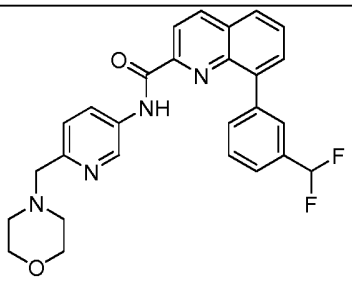
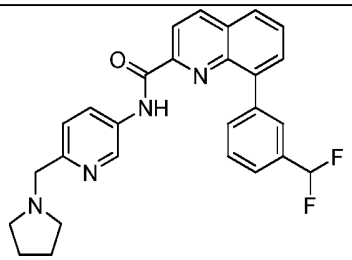
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
332	416		B	B
333	410		B	B
334	392		A	B
335	393		B	B
336	393		B	B

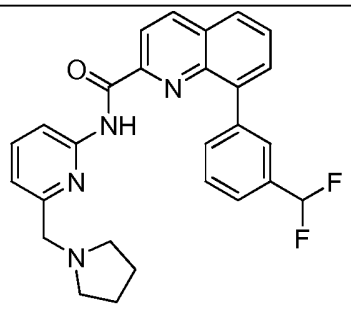
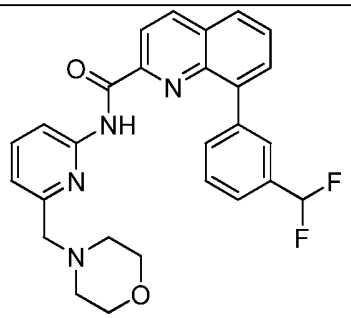
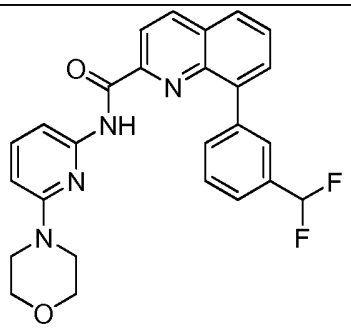
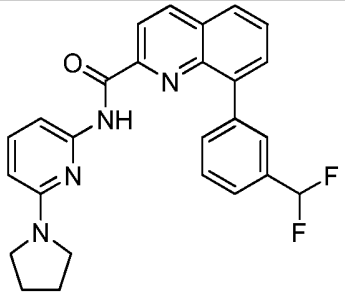
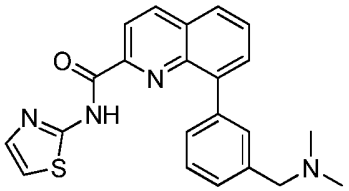
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
337	393		A	B
338	335		C	
339	426		B	B
340	410		B	B
341	415		B	B

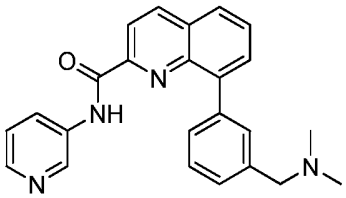
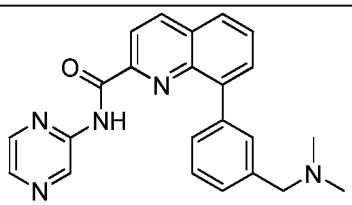
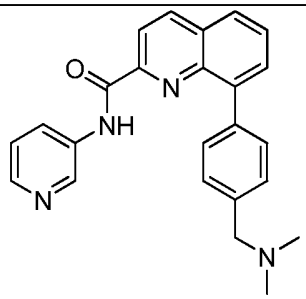
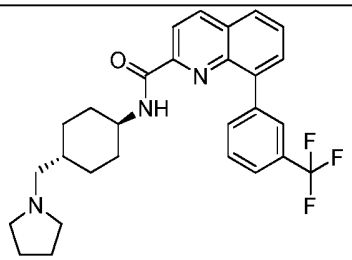
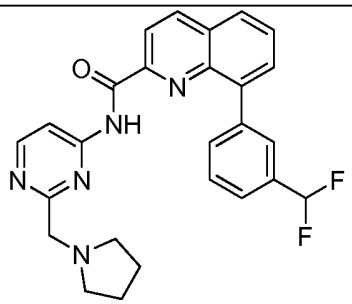
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
342	409		C	B
343	431		B	A
344	415		A	B
345	415		A	B
346	429		A	B
347	409		A	B

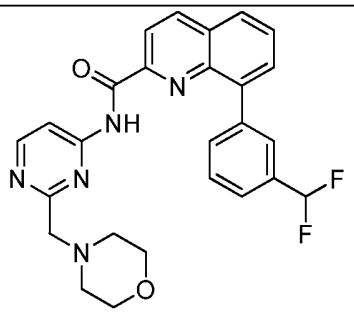
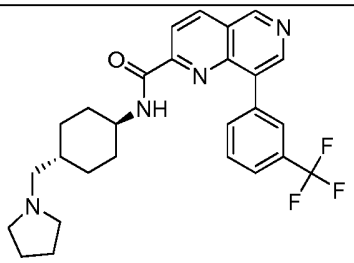
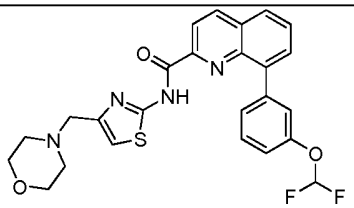
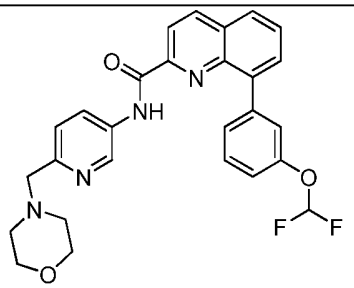
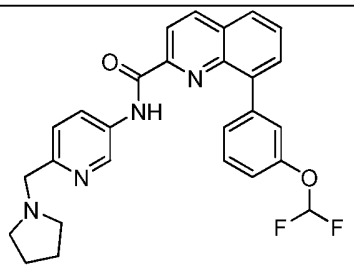
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
348	423		A	A
349	416		C	B
350	410		C	
351	416		C	B
352	410		C	

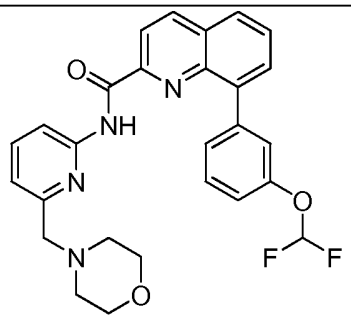
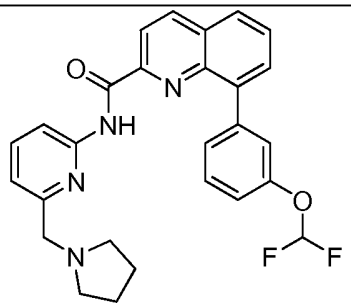
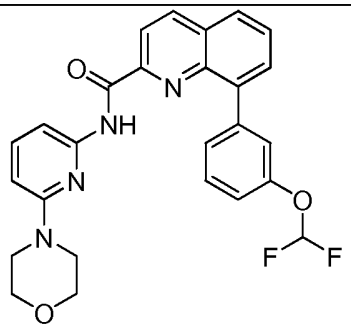
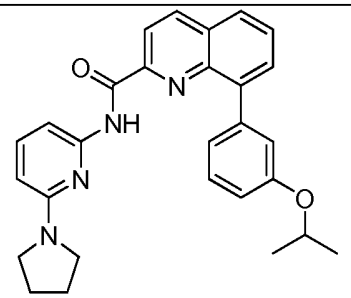
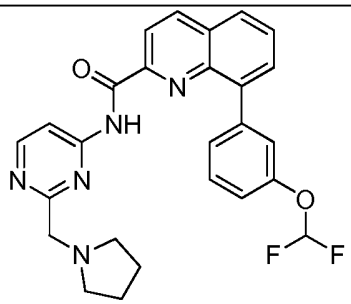
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
353	425		B	B
354	426		A	A
355	410		B	A
356	415		B	B
357	409		B	B

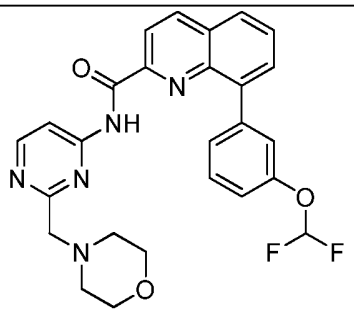
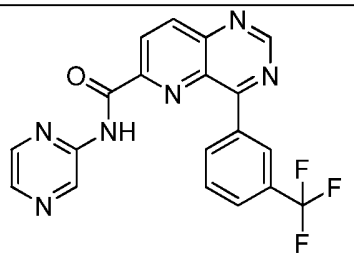
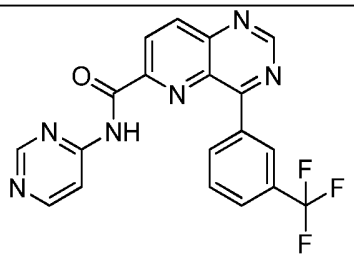
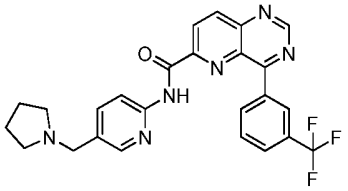
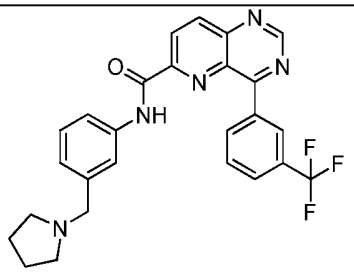
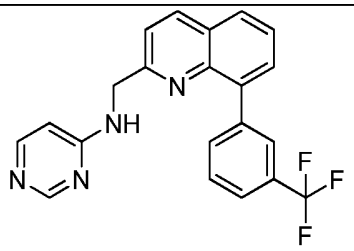
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
358	389		B	B
359	384		B	B
360	481		A	A
361	475		A	A
362	459		A	A

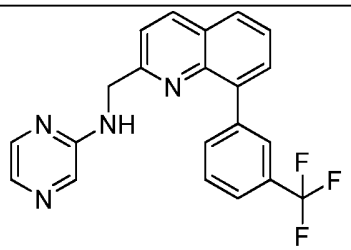
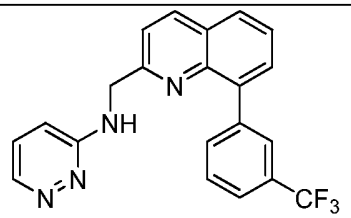
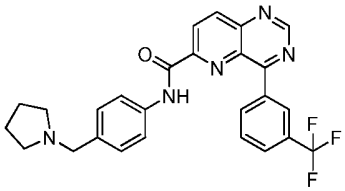
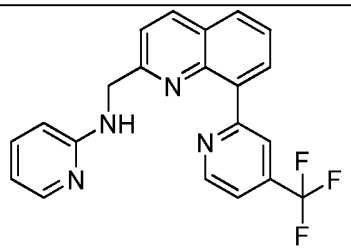
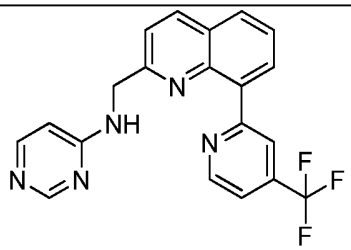
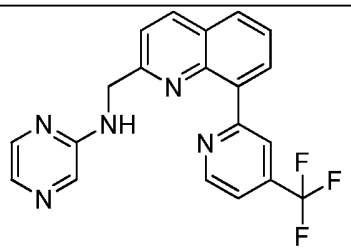
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
363	459		A	A
364	475		A	A
365	461		B	A
366	445		A	B
367	389		B	A

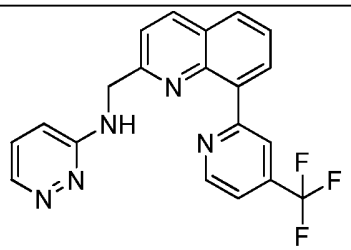
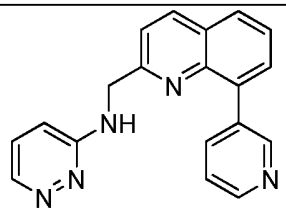
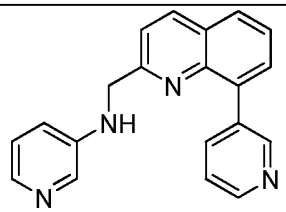
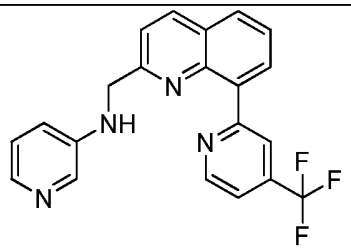
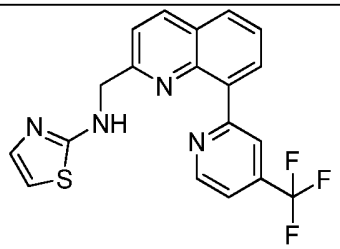
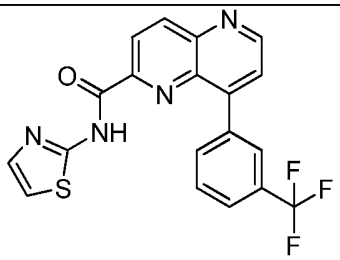
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
368	383		B	A
369	384		B	A
370	383		B	B
371	482		B	B
372	460		A	A

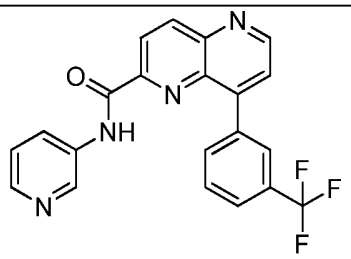
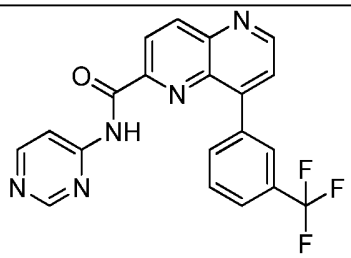
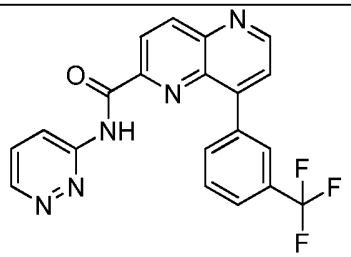
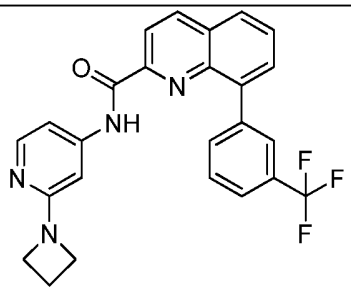
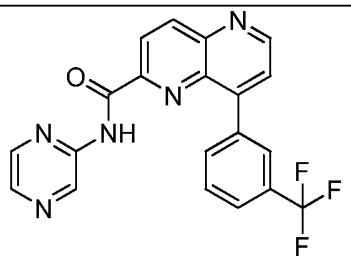
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
373	476		A	A
374	483		A	B
375	497		A	A
376	491		A	A
377	475		A	A

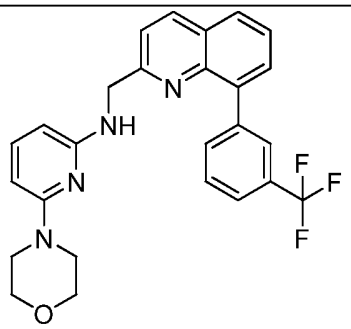
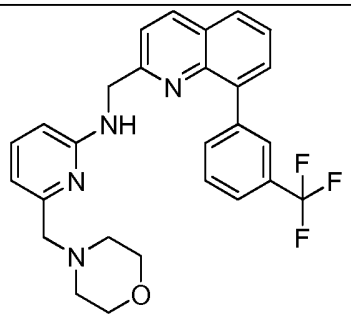
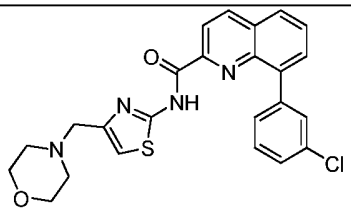
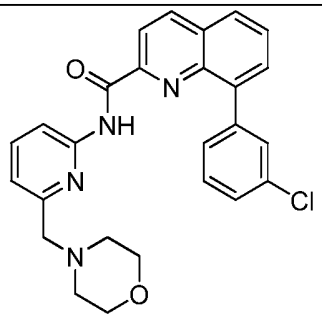
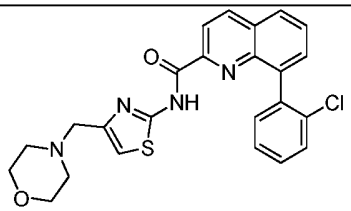
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
378	491		A	A
379	475		A	A
380	477		A	A
381	461		A	B
382	476		A	A

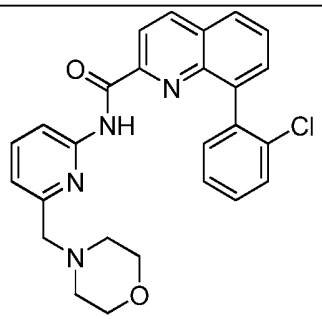
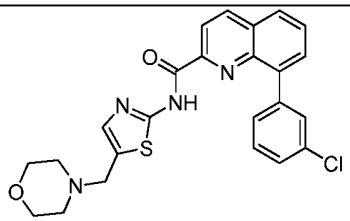
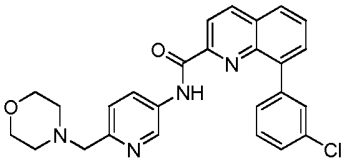
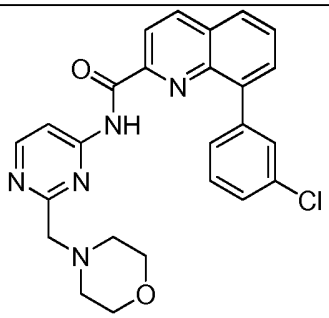
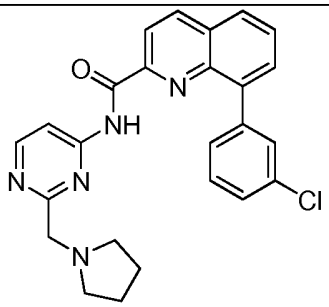
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
383	492		A	A
384	397		A	B
385	397		C	B
386	479		B	A
387	478		A	A
388	381		B	B

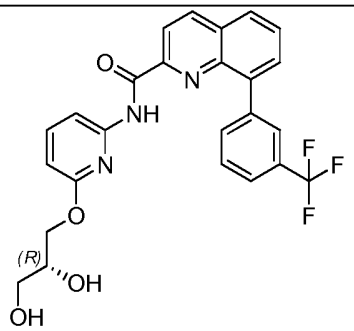
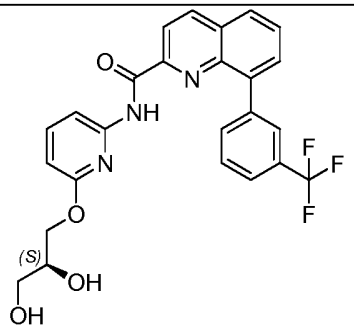
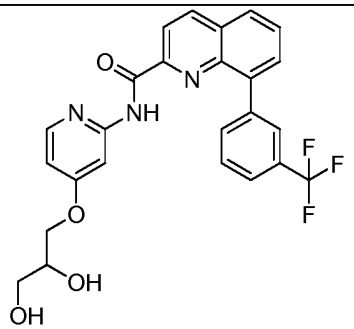
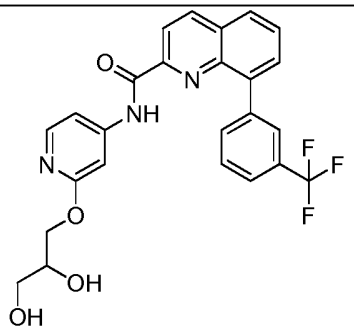
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
389	381		B	B
390	381		B	B
391	478		A	A
392	381		B	B
393	382		C	B
394	382		B	B

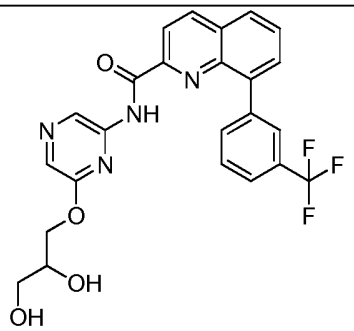
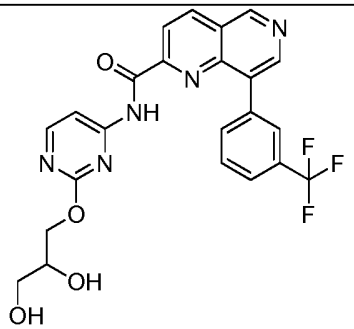
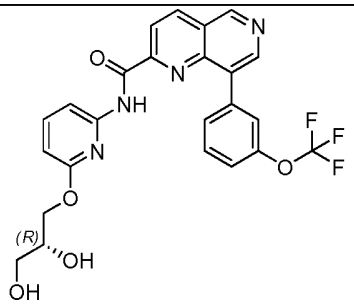
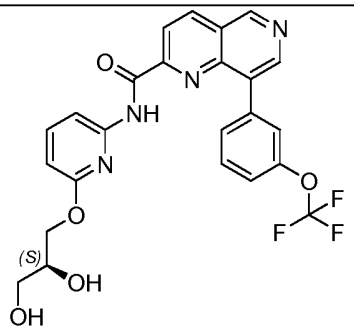
CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
395	382		B	B
396	314		C	
397	313		C	
398	381		C	
399	387		B	B
400	401		C	

CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
401	395		A	A
402	396		A	A
403	396		A	B
404	449		A	A
405	395		A	A

CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
406	465		C	C
407	479		B	B
408	465		A	A
409	459		A	A
410	465		B	B

CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
411	459		B	A
412	465		A	B
413	459		A	A
414	460		A	A
415	444		A	A

CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
416	484		A	A
417	484		A	A
418	484		A	B
419	484		A	A

CPD. #	[M+H]+	STRUCTURE	$EC_{1.5}$ μM	% FOLD ACT.
420	485		A	A
421	486		A	A
422	501		A	A
423	501		A	A

Example 26. Cell based Assay

In order to further characterize the biological effects of the sirtuin modulators, representative compounds were assayed for their effect on LPS-induced inflammatory cytokine production. Tumor necrosis factor (TNF-α) is a cytokine

involved in systemic inflammation and is a member of a group of cytokines that stimulate the acute phase immune reaction. The primary role of TNF-α is in the regulation of immune cells. TNF-α is also able to induce apoptotic cell death and to induce inflammation. Dysregulation of TNF-α production has been implicated in a variety of human diseases, as well as cancer. Sirtuin modulators identified by their ability to activate SIRT1 in the in vitro assay described above were tested for their ability to inhibit LPS-induced TNF-α production in macrophages.

Materials and Methods for the MsTNF-α Assay

Raw Macrophage 264.7 cells were obtained from the American Tissue Culture Collection (ATCC Deposit No. TIB-71). Approximately 4 x 10^4 cells were seeded into microtiter plate wells and incubated for 17-18 hours at 37°C and 5.0% CO_2 prior to each assay. The Raw264.7 Macrophage Growth Culture Media (Assay Media) consists of DMEM Media supplemented with 10% low endotoxin FBS (fetal bovine serum), 100 units/ml penicillin, and 100 μg/ml streptomycin. Media was stored at 4°C and warmed to 37°C prior to use. For each SIRT1 activator assayed, 8 doses were tested, typically ranging from 5 mM to 0.008 mM (8 μM) in order to determine the IC_{50} value for each (i.e., the concentration of compound required to inhibit the LPS-induced production of TNF-α by 50%). The SIRT1 activator was added to the Raw Macrophage 264.7 test cells and incubated for 2 hours at 37°C and 5.0% CO_2 prior to the addition of LPS. As a positive control, a TACE inhibitor compound was added to a control sample at a concentration that completely inhibits TNF-α secretion (100% inhibition control). As a negative control, the solvent DMSO was added to another control sample (0% inhibition control). Lipopolysaccharide (LPS) (EMD Biosciences) was added to a final assay concentration of 100 ng/ml LPS and the cells were incubated for 1 hour at 37°C and 5.0% CO_2. At the end of the incubation, the supernatant was removed and TNF levels quantified by a standard ELISA sandwich assay as described below.

TNF-α is secreted from mouse RAW 264.7 macrophages (a mouse leukaemic monocyte macrophage cell line) when the cells are exposed to Lipopolysaccharide (LPS). The ability to test compounds to inhibit the secretion of

MsTNF-α was measured in a solid phase sandwich Enzyme Linked-Immuno-Sorbent Assay (ELISA). Following treatment with test compounds and LPS, cell supernatants were harvested for use in ELISA plates. A polyclonal antibody specific for MsTNF-α was coated onto the wells of the microtiter strips. Samples, including standards of known MsTNF-α content, control specimens and unknowns were pipetted into these wells, followed by the addition of a biotinylated monoclonal second antibody. During the first incubation, the MsTNF-α antigen binds simultaneously to the immobilized (capture) antibody (on one site), and to the solution phase biotinylated antibody (on a second site). After removal of excess secondary antibody, Streptavidin-Peroxidase was added. This binds to the biotinylated antibody to complete the four-member sandwich. Unbound enzyme was removed by washing, and then a peroxidase substrate solution was added, which is processed by the bound enzyme to produce color. The colorimetric read-out was measured by the Spectramax M5 Plate Reader. The intensity of the produced absorbance is directly proportional to the concentration of MsTNF-α present in the original specimen. The amount of Ms-TNF-α secreted in the samples can thus be interpolated and calculated from a standard curve using recombinant mouse TNF-α, and the percent inhibition is then calculated based on the TACE and DMSO control wells.

The MsTNF-α Assay measures the amount of MsTNF-α that is secreted by cells, in the presence of LPS and a known SIRT1 activator. Using this assay one can assess the ability of a test compound to inhibit MsTNF-α secretion in a dose-dependent manner. An IC_{50} value was determined for each compound tested.

The results of the TNF-α Raw Macrophage assay are shown below in Table 2. Values expressed as "≥40 μM" indicate an actual IC_{50} value of greater than or equal to 40 μM for that test compound.

Table 2.

Compound No.	TNF α IC_{50} (μM)
107	35.68
112	≥ 40
115	≥ 40
116	36.77
117	≥ 40
119	≥ 40
200	9.1
201	5.42
202	3.42
203	≥ 40
204	16.52
210	≥ 40
212	≥ 40
213	9.72
214	12.57
217	19.16
218	29.24
219	≥ 40
221	≥ 40
222	≥ 40
223	≥ 40
224	≥ 40
225	≥ 40
227	≥ 40
228	15.31
230	≥ 40
231	≥ 40
232	13.52
237	10.45
238	2.87
245	17.2
246	22.35
249	≥ 40
253	37.57
254	11.55
255	16.41
256	18.19
257	1.76
258	4.39
259	19.32

260	19.6
261	≥ 40
262	≥ 40
268	39.02
269	20.65
270	≥ 40
273	39.08
274	24.64
275	5.78
276	38.2
277	≥ 40
278	≥ 40
279	≥ 40
287	10.83
288	≥ 40
299	37.49
300	≥ 40
301	≥ 40
302	13.67
303	≥ 40
305	≥ 40
306	≥ 40
315	≥ 40
317	≥ 40
319	≥ 40
326	≥ 40
337	36.86
344	≥ 40
345	7.5
346	≥ 40
347	≥ 40
348	≥ 40
355	22.6
360	15.76
361	33.82
362	8.56
363	15.9
364	≥ 40
370	6.48
371	5.34
372	8
373	≥ 40
374	7.67
375	≥ 40
377	7.31
378	≥ 40

379	1.88
380	≥ 40
381	≥ 40
382	7.79
383	≥ 40
401	≥ 40
402	≥ 40
404	≥ 40
405	≥ 40
408	24.58
409	11.9
412	7.44
413	12.06
414	≥ 40
415	10.41
416	≥ 40
417	≥ 40
418	≥ 40
419	≥ 40
420	≥ 40

EQUIVALENTS

The present invention provides among other things sirtuin-activating compounds and methods of use thereof. While specific embodiments of the subject invention have been discussed, the above specification is illustrative and not restrictive. Many variations of the invention will become apparent to those skilled in the art upon review of this specification. The full scope of the invention should be determined by reference to the claims, along with their full scope of equivalents, and the specification, along with such variations.

INCORPORATION BY REFERENCE

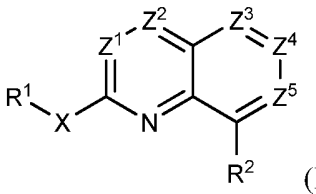
All publications and patents mentioned herein, including those items listed below, are hereby incorporated by reference in their entirety as if each individual publication or patent was specifically and individually indicated to be incorporated by reference. In case of conflict, the present application, including any definitions herein, will control.

Also incorporated by reference in their entirety are any polynucleotide and polypeptide sequences which reference an accession number correlating to an entry in a public database, such as those maintained by The Institute for Genomic

Research (TIGR) (www.tigr.org) and/or the National Center for Biotechnology Information (NCBI) (www.ncbi.nlm.nih.gov).

CLAIMS:

1. A compound represented by structural formula (I):



(I),

or a salt thereof, wherein:

each of Z^1, Z^2, Z^3, Z^4, and Z^5 are independently selected from N and CR, wherein:

each of Z^3, Z^4 and Z^5 is independently CR, or

at least one of Z^1 or Z^2 is N and no more than two of Z^1-Z^5 are simultaneously N, or

two of Z^3, Z^4 and Z^5 are N and each other of Z^1-Z^5 is independently CR;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2,

-NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl), and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), $S(=O)_2$, and O, and

X is selected from $-NR^6$-C(=O)-†, $-NR^6$-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-$(CR^4R^5)_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1;

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

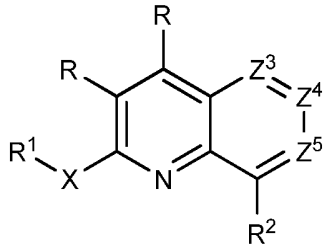
each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl.

2. The compound of claim 1, wherein each of Z^3, Z^4 and Z^5 is CR.

3. The compound of claim 1 or 2, wherein X is selected from $-NR^6$-C(=O)-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, -NR^6-C(=O)-CR^4R^5-†, -NR^6-S(=O)$_2$-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -NR^6-C(=O)-NR^6-†, -C(=O)-NR^6-†, -C(=O)-NR^6-$(CR^4R^5)_{1-3}$-†, -NR^6-C(=O)-CR^4R^5-O-†, -NR^6-C(=O)-O-†,-CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-† and -C(=NR^6)-NR^6-†.

4. The compound of claim 3, wherein X is -C(O)-NR^6-†.

5. A compound represented by structural formula (II):



(II),

or a salt thereof, wherein:

each of Z^3, Z^4, and Z^5 are independently selected from N and CR, wherein only one of Z^3, Z^4, and Z^5 is N, wherein:

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when X is not -C(=O)-NH-†, R^1 is also optionally substituted with =O and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom

with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle other than piperazine, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, and

X is selected from -NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†,

-C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -CR^{4}R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^{4}R^5-†, -CR^{4}R^5-C(=S)-NR^6-†, -NH-S(O)-CR^{4}R^5-†, -CR^{4}R^5-S(O)-NH-†, -NR^6-S(O)$_2$-CR^{4}R^5-†, -CR^{4}R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^{4}R^5-†, -CR^{4}R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^{4}R^5-†, -NR^6-C(=O)-CR^{4}R^5-†, -CR^{4}R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^{4}R^5-NR^6-†, and -NR^6-C(=O)-CR^{4}R^5-O-†, and when Z^3 or Z^5 is N, X is also selected from: -C(=O)-NR^6-(CR^{4}R^5)$_{1-3}$-†,

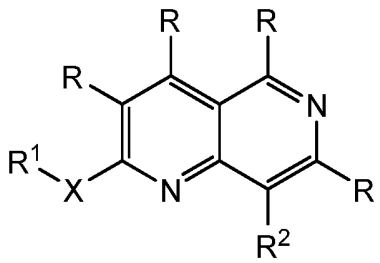
wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C$_1$-C$_4$ alkyl, and halo-substituted C$_1$-C$_4$ alkyl; and

each R^6 is independently selected from hydrogen, C$_1$-C$_4$ alkyl, and halo-substituted C$_1$-C$_4$ alkyl.

6. The compound of claim 5, represented by structural formula (IV):



(IV),

or a salt thereof, wherein:

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C$_1$-C$_2$ alkyl, -O-(C$_1$-C$_2$) fluoro-substituted alkyl, -S-(C$_1$-C$_2$) fluoro-substituted alkyl, C$_1$-C$_4$ alkyl, -O-(C$_1$-C$_4$) alkyl, -S-(C$_1$-C$_4$) alkyl, C$_3$-C$_7$ cycloalkyl, -(C$_1$-C$_2$) alkyl-N(R^3)(R^3), hydroxy-substituted C$_1$-C$_4$ alkoxy, -(C$_1$-C$_4$)-O- saturated heterocycle, -O-(C$_1$-C$_3$) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C$_1$-C$_4$ alkyl, hydroxy-substituted C$_1$-C$_4$ alkoxy, =O, C$_3$-C$_7$ cycloalkyl, fluoro-substituted C$_1$-C$_2$ alkyl, -O-R^3, -S-R^3, -(C$_1$-C$_4$ alkyl)-N(R^3)(R^3), -N(R^3)(R^3),

-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$ and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl),and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O, and

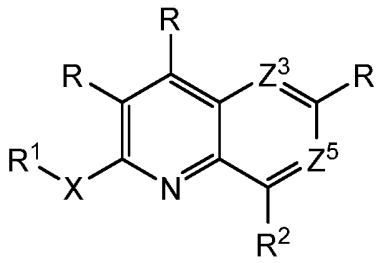
X is selected from –NR^6-C(=S)-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† and when R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, X is additionally selected from -C(=S)-NR^6-† and -NR^6-C(=O)-NR^6-†, and when R^1 is optionally substituted cycloalkyl or saturated heterocyclyl X is additionally selected from –NR^6-C(=O)-†, wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

7. The compound of claim 5, represented by structural formula (VI):



(VI),

or a salt thereof, wherein:

one of Z^3 or Z^5 is N and the other is CR;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O-saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro-or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro- substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl;

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†,

-NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-$S(O)_2$-CR^4R^5-†, -CR^4R^5-$S(O)_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-†, and when R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, X is additionally selected from -C(=O)-NR^6-†, -C(=S)-NR^6-† and -NR^6-C(=O)-NR^6-†, wherein:

† represents where X is bound to R^1; and

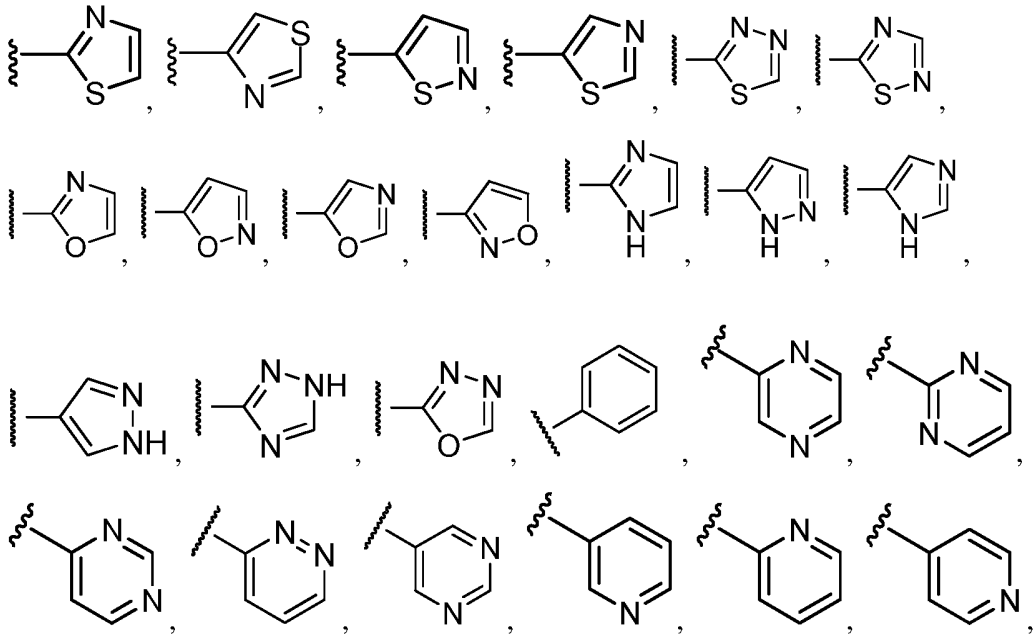
each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

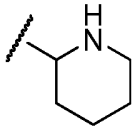
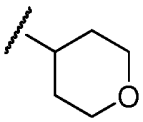
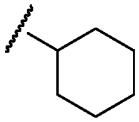
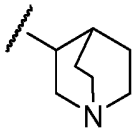
each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

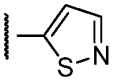
8. The compound of any of claims 5 to 7, wherein X is selected from -NR^6-C(=O)-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, -NR^6-C(=O)-CR^4R^5-†, -NR^6-$S(=O)_2$-†, -NR^6-$S(=O)_2$-CR^4R^5-†, -NR^6-C(=O)-NR^6-†, -C(=O)-NR^6-†, -NR^6-C(=O)-CR^4R^5-O-†, -NR^6-C(=O)-O-†, -CR^4R^5-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-† and -C(=NR^6)-NR^6-†.

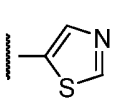
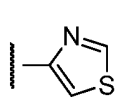
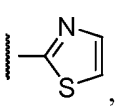
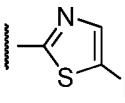
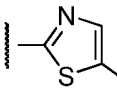
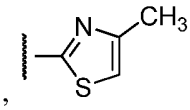
9. The compound of any of claims 1 to 8, wherein R^1 is optionally substituted aryl, heteroaryl or saturated heterocyclyl, and X is -C(=O)-NR^6-†.

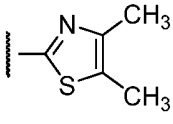
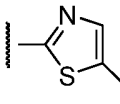
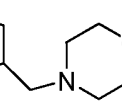
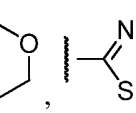
10. The compound of any one of claims 1 to 9, wherein R^1 is selected from:

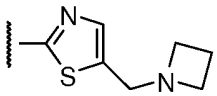
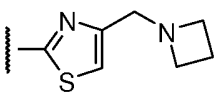
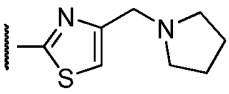
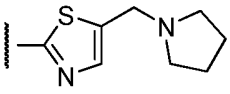


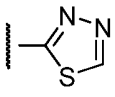
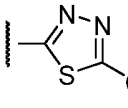
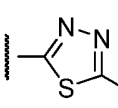
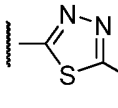
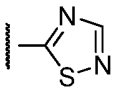
 ,  ,  and  , wherein R^1 is optionally substituted with one or two substituents independently selected from halo, C_1-C_4 alkyl, -(C_1-C_4 alkyl)-N(R^3)(R^3), =O, -N(R^3)(R^3), and -O-R^3.

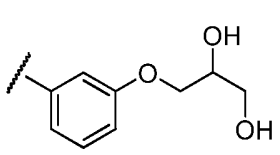
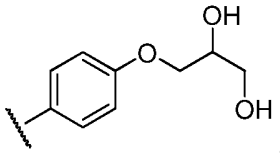
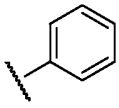
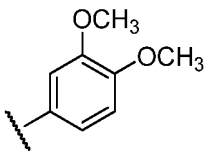
11. The compound of claim 10, wherein R^1 is selected from:  ,

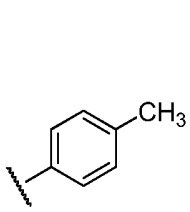
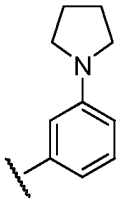
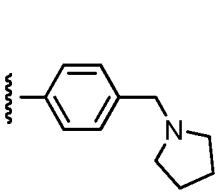
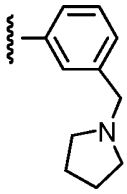
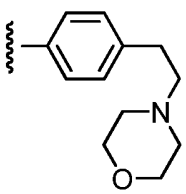
 ,  ,  ,  ,  ,  ,

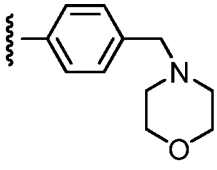
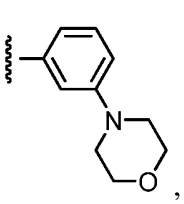
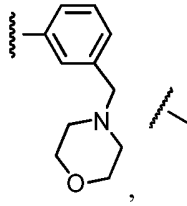
 ,  ,  ,  ,

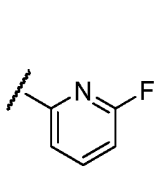
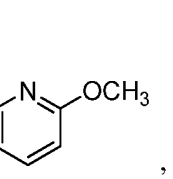
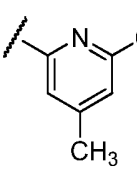
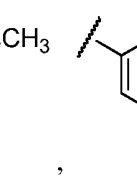
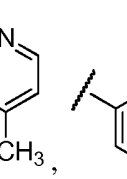
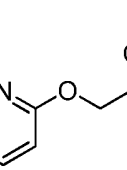
 ,  ,  ,  ,

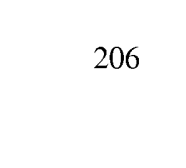
 ,  ,  ,  ,  ,

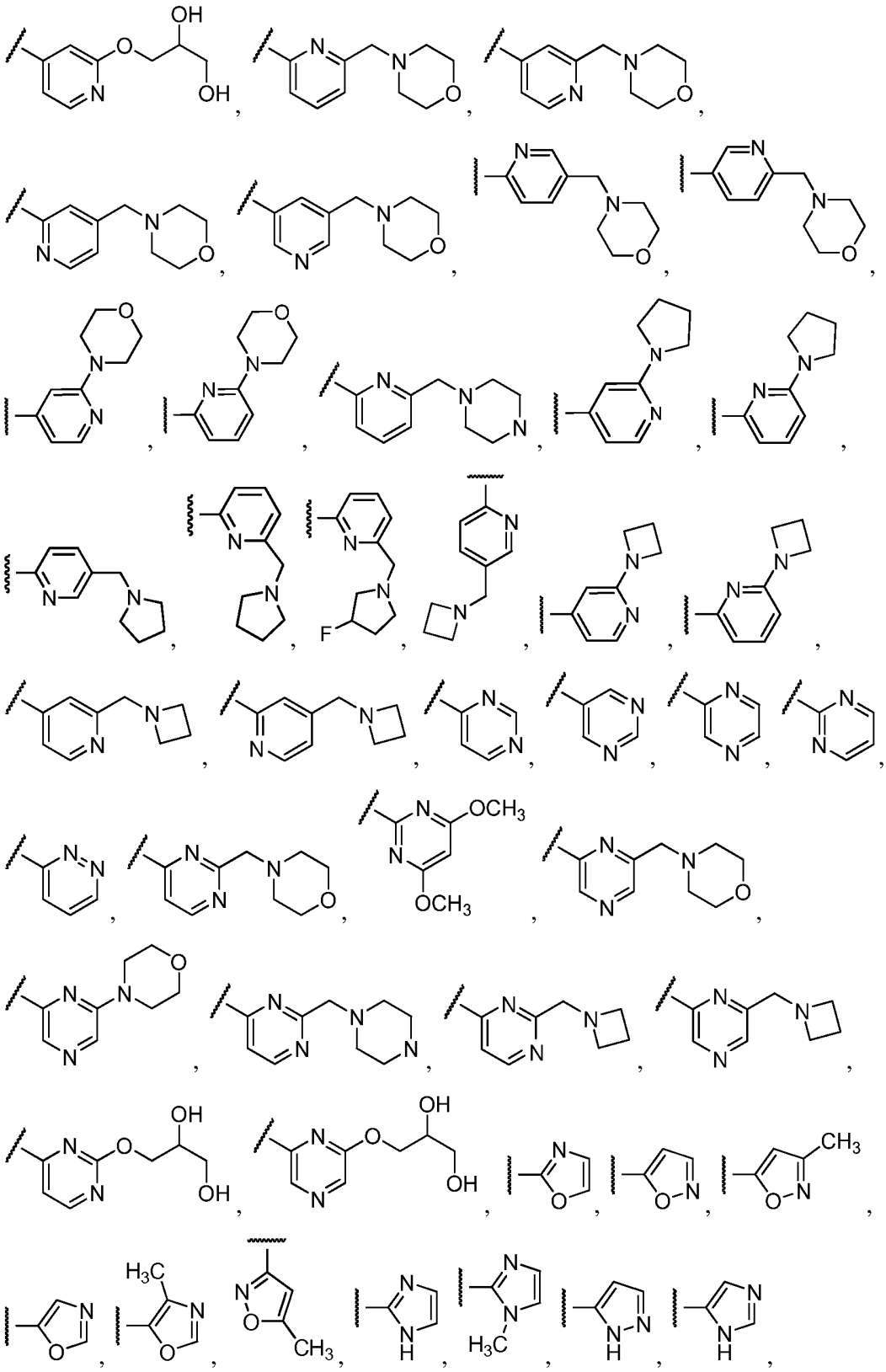
 ,  ,  ,  ,

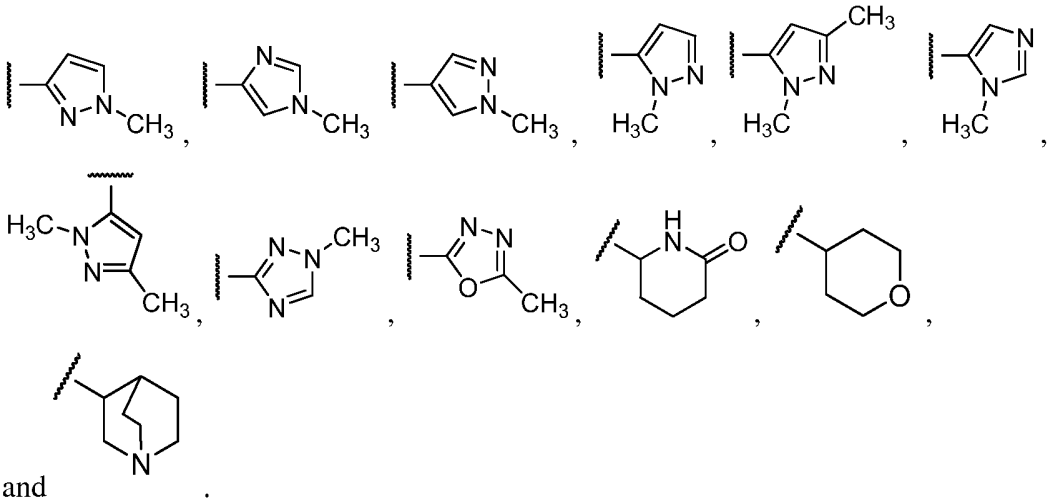
 ,  ,  ,  ,  ,

 ,  ,  ,

 ,  ,  ,  ,  ,  ,

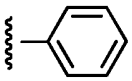
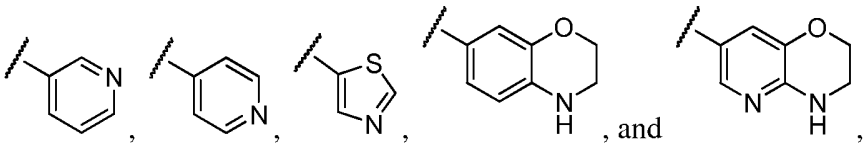
, ,  , ,



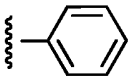


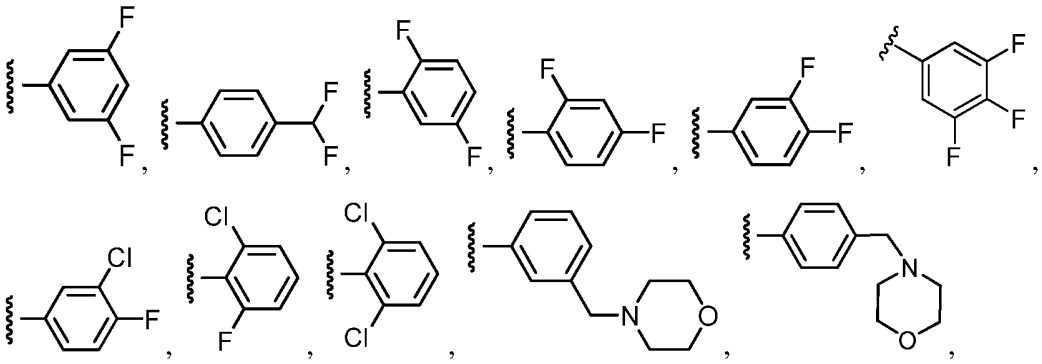
and .

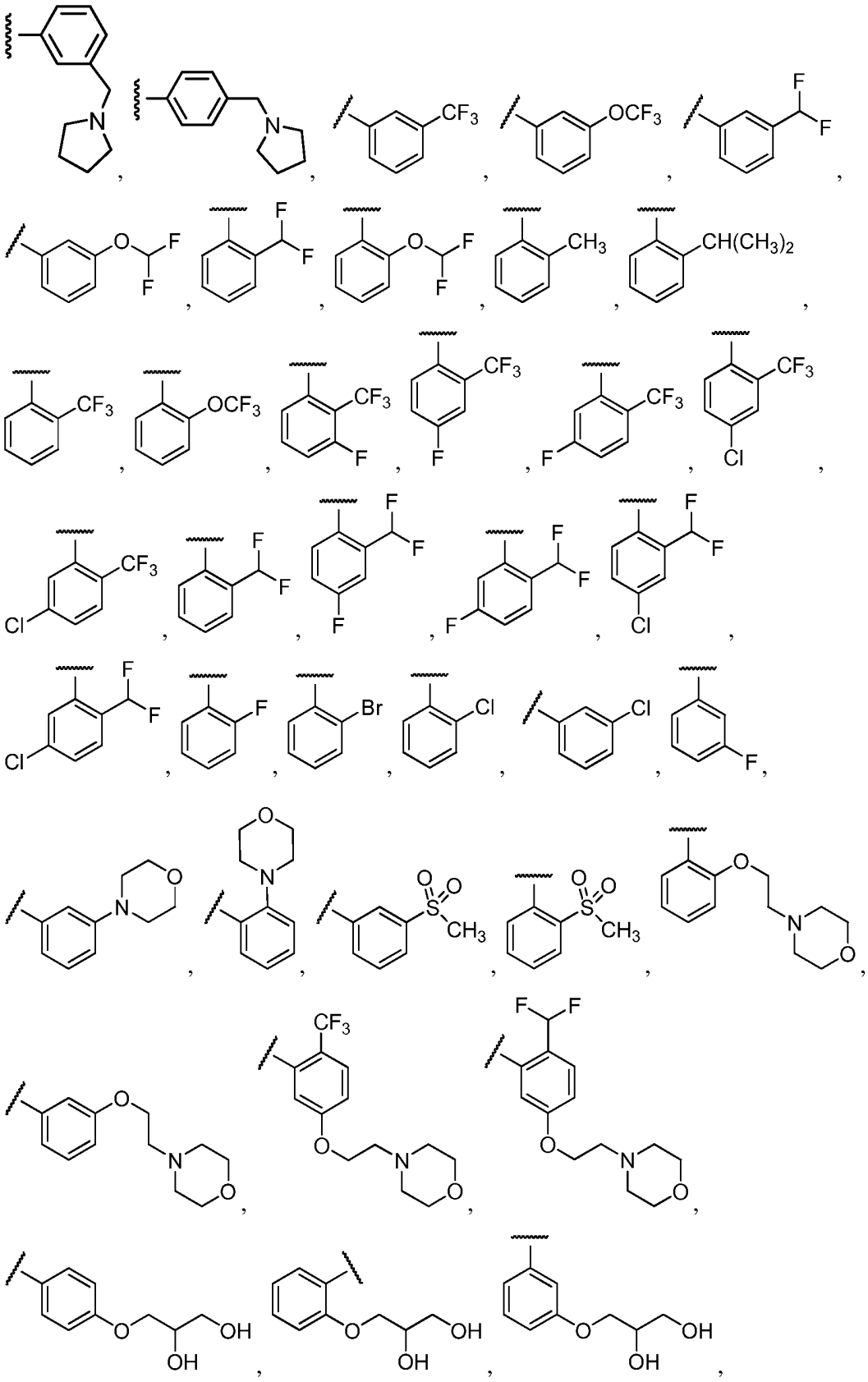
12. The compound of any one of claims 1 to 11, wherein R^2 is selected from optionally substituted aryl and optionally substituted heteroaryl.

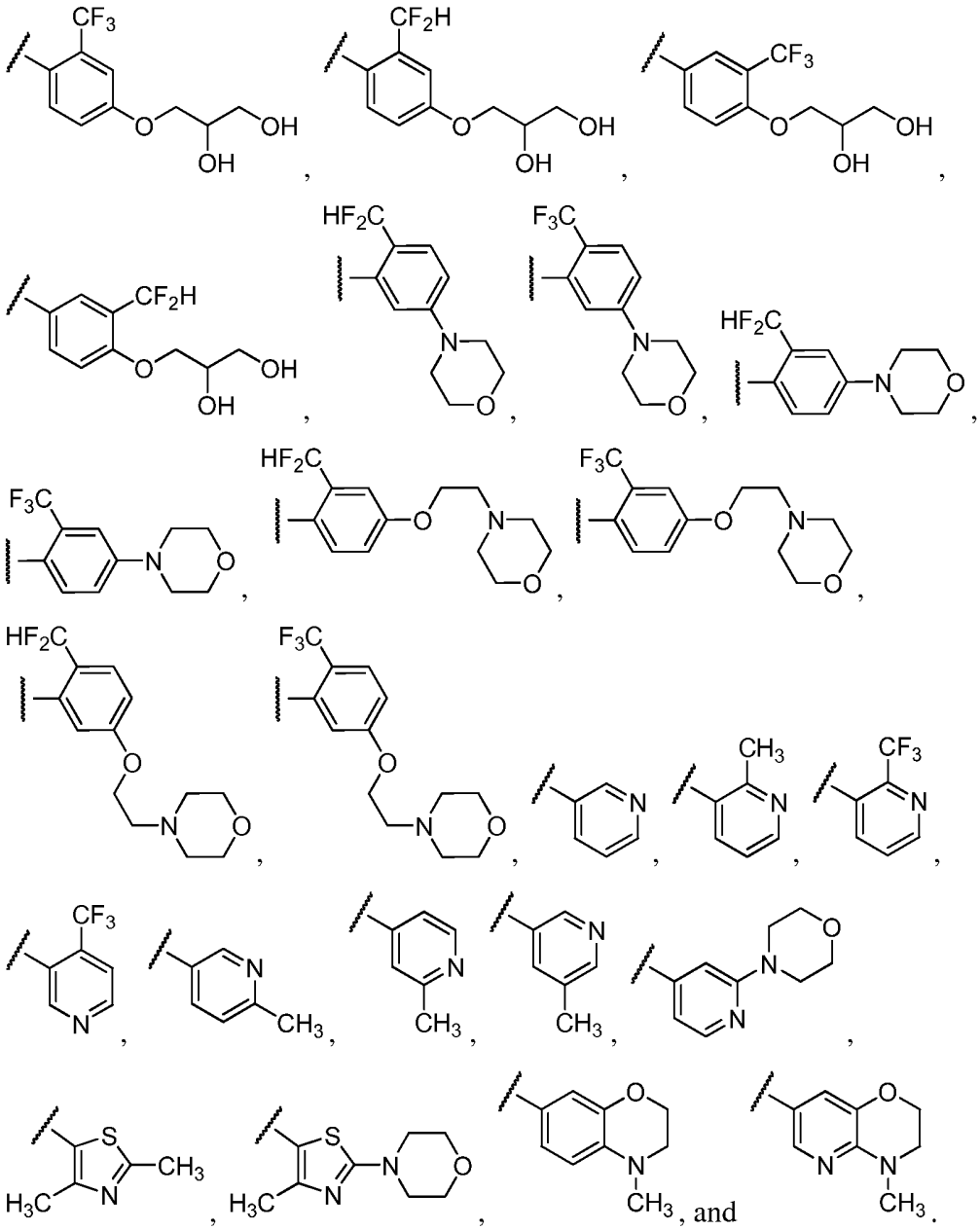
13. The compound of claim 12, wherein R^2 is selected from:  ,  , and , wherein R^2 is optionally substituted with one or more groups independently selected from halo, C_1-C_4 alkyl, -(C_1-C_4 alkyl)-N(R^3)(R^3), C_1-C_2 fluoro-substituted alkyl, -O-R^3, -SO_2-R^3, -N(R^3)(R^3), and -O-(C_1-C_4 alkyl)-N(R^3)(R^3).

14. The compound of claim 12, wherein R^2 is meta-substituted relative to the attachment of R^2 to the rest of the compound, and wherein R^2 is optionally further substituted.

15. The compound of claim 14, wherein R^2 is selected from:  ,



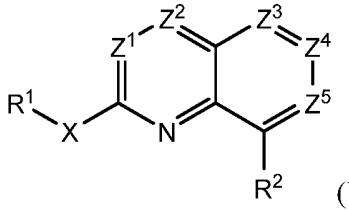




16. A pyrogen-free pharmaceutical composition comprising a compound of any of claims 1 to 15, or a pharmaceutically acceptable salt thereof and a pharmaceutically acceptable carrier.

17. The pharmaceutical composition of claim 16, further comprising an additional active agent.

18. A method of increasing sirtuin-1 activity in a cell comprising the step of contacting the cell with a compound represented by Structural Formula (VII):



(VII),

or a salt thereof, wherein:

each of Z^1-Z^5 is independently selected from N and CR, wherein no more than two of Z^1-Z^5 are simultaneously N;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O- saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen, and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NH-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†,

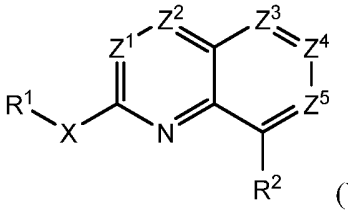
-NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

19. A method for treating a subject suffering from or susceptible to insulin resistance, a metabolic syndrome, diabetes, or complications thereof, or for increasing insulin sensitivity in a subject, comprising administering to the subject in need thereof a compound represented by Structural Formula (VIII):



(VIII),

or a salt thereof, wherein:

each of Z^1-Z^5 is independently selected from N and CR, wherein no more than two of Z^1-Z^5 are simultaneously N;

each R is independently selected from hydrogen, halo, -OH, -C≡N, fluoro-substituted C_1-C_2 alkyl, -O-(C_1-C_2) fluoro-substituted alkyl, -S-(C_1-C_2) fluoro-substituted alkyl, C_1-C_4 alkyl, -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl and C_3-C_7 cycloalkyl, -(C_1-C_2) alkyl-N(R^3)(R^3), hydroxy-substituted C_1-C_4 alkoxy, -(C_1-C_4)-O- saturated heterocycle, -O-(C_1-C_3) alkyl-N(R^3)(R^3), and -N(R^3)(R^3);

R^1 is selected from a carbocycle and a heterocycle, wherein R^1 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, hydroxy-substituted C_1-C_4 alkoxy, =O, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, -O-R^3, -S-R^3, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), and -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), and when R^1 is phenyl, R^1 is

also optionally substituted with -(aryl), -(heterocycle), O-(heterocycle), -O-(carbocycle), methylenedioxy, fluoro-substituted-methylenedioxy, ethylenedioxy, or fluoro-substituted-ethylenedioxy, wherein:

any aryl, cycloalkyl, carbocycle, saturated heterocycle, or heterocycle substituent of R^1 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from -OH, -C_1-C_4 alkyl, fluoro, fluoro- or chloro-substituted C_1-C_4 alkyl, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; and

any heterocycle or saturated heterocycle substituent of R^1 is optionally and independently substituted at any substitutable nitrogen atom with C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_4 alkyl or –(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

R^2 is selected from a carbocycle and a heterocycle, wherein R^2 is optionally substituted with one or more substitutents independently selected from halo, -C≡N, C_1-C_4 alkyl, C_3-C_7 cycloalkyl, fluoro-substituted C_1-C_2 alkyl, hydroxy-substituted C_1-C_4 alkoxy, -O-R^3, -S-R^3, -SO_2-R^3, =O, -(C_1-C_4 alkyl)-N(R^3)(R^3), -N(R^3)(R^3), -O-(C_1-C_4 alkyl)-N(R^3)(R^3), -(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl)-N(R^3)(R^3), -C(O)-N(R^3)(R^3), -(C_1-C_4 alkyl)-C(O)-N(R^3)(R^3), -O-phenyl, phenyl, -SO_2-(C_1-C_4 alkyl)-, and a second heterocycle, and when R^2 is phenyl, R^2 is also optionally substituted with -O-(second heterocycle), -O-(C_3-C_7 cycloalkyl), methylenedioxy, fluoro-substituted methylenedioxy, ethylenedioxy, or fluoro-substituted ethylenedioxy, wherein:

any phenyl, saturated heterocycle, second heterocycle or cycloalkyl substituent of R^2 is optionally substituted at any substitutable carbon atom with one or more substituents independently selected from halo, -C≡N, C_1-C_4 alkyl, fluoro- or chloro-substituted C_1-C_2 alkyl, -O-(fluoro-substituted C_1-C_2 alkyl), -O-(C_1-C_4) alkyl, -S-(C_1-C_4) alkyl, -S-(fluoro-substituted C_1-C_2 alkyl), -NH-(C_1-C_4) alkyl, and -N-(C_1-C_4)$_2$ alkyl; and

any second heterocycle or saturated heterocycle substituent of R^2 is optionally and independently substituted at any substitutable nitrogen atom

with C_1-C_4 alkyl fluoro- or chloro-substituted C_1-C_4 alkyl or–(C_1-C_4 alkyl)-O-(C_1-C_4 alkyl);

each R^3 is independently selected from hydrogen, and -C_1-C_4 alkyl, wherein the alkyl is optionally substituted with one or more of -OH, fluoro, -NH_2, -NH(C_1-C_4 alkyl), -N(C_1-C_4 alkyl)$_2$, -NH($CH_2CH_2OCH_3$), or -N($CH_2CH_2OCH_3$)$_2$; or

two R^3 are taken together with the nitrogen atom to which they are bound to form a 4- to 8-membered saturated heterocycle optionally comprising one additional heteroatom selected from N, S, S(=O), S(=O)$_2$, and O; and

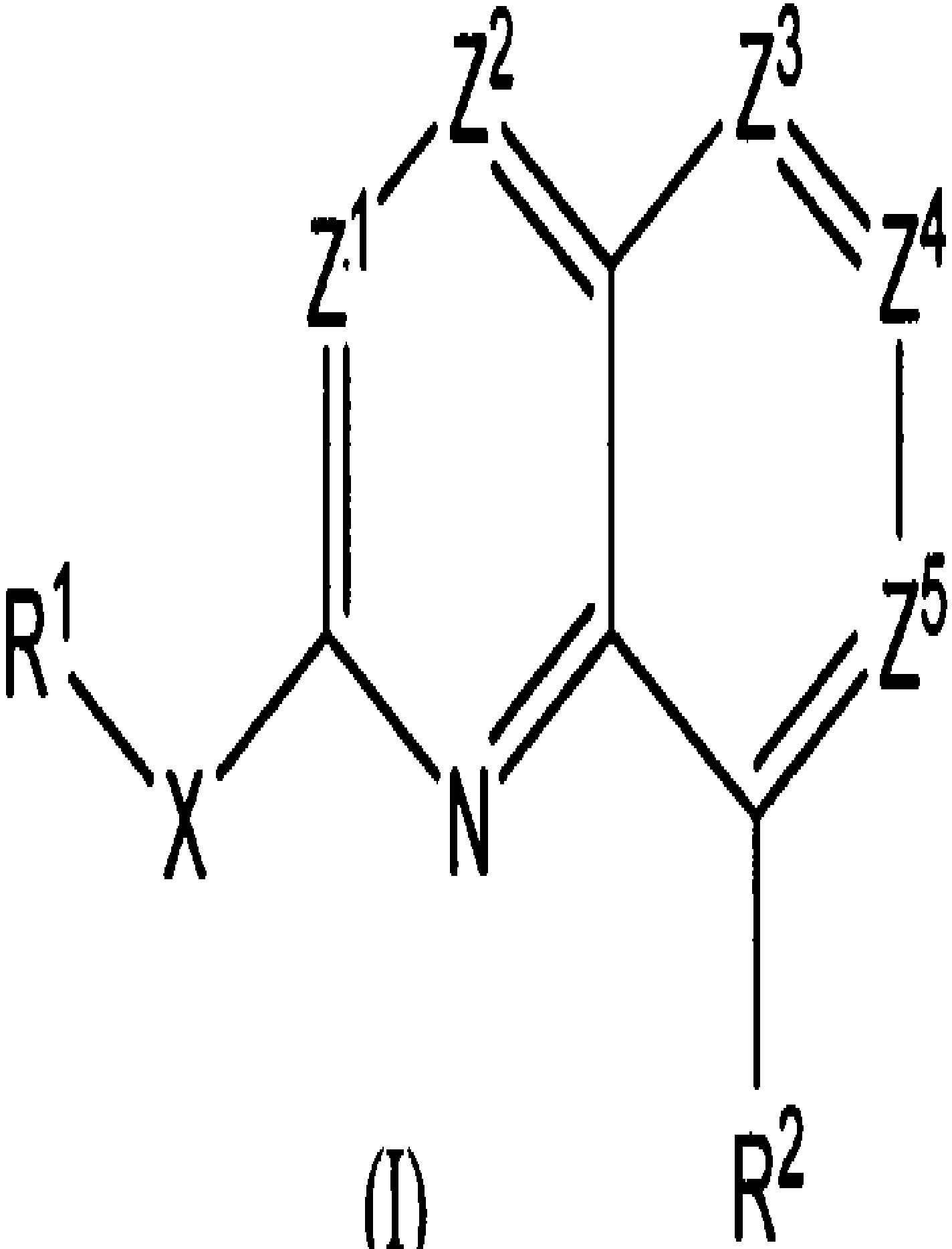
X is selected from –NR^6-C(=O)-†, -NR^6-C(=S)-†, -C(=O)-NR^6-†, -C(=S)-NR^6-†, -NH-S(=O)-†, -S(=O)-NH-†, -S(=O)$_2$-NR^6-†, -NR^6-S(=O)$_2$-†, -NR^6-S(O)$_2$-NR^6-†, -NR^6-C(=O)-O-†, -O-C(=O)NR^6-†, -NR^6-C(=O)-NR^6-†, -NR^6-NR^6-†, -O-NH-†, -NH-O-†, -NR^6-CR^4R^5-†, -CR^4R^5-NR^6-†, -NH-C(=NR^6)-†, -C(=NR^6)-NR^6-†, -NR^6-C(=NR^6)-NR^6-†, -C(=O)-NR^6-(CR^4R^5)$_{1-3}$-†, -CR^4R^5-NR^6-C(O)-†, -NR^6-C(=S)-CR^4R^5-†, -CR^4R^5-C(=S)-NR^6-†, -NH-S(O)-CR^4R^5-†, -CR^4R^5-S(O)-NH-†, -NR^6-S(=O)$_2$-CR^4R^5-†, -CR^4R^5-S(O)$_2$-NR^6-†, -NH-C(=O)-O-CR^4R^5-†, -CR^4R^5-O-C(=O)-NH-†, -NH-C(=O)-NR^6-CR^4R^5-†, -NR^6-C(=O)-CR^4R^5-†, -CR^4R^5-NH-C(=O)-O-†, -NR^6-C(=O)-CR^4R^5-NR^6-†, and -NR^6-C(=O)-CR^4R^5-O-† wherein:

† represents where X is bound to R^1; and

each R^4 and R^5 is independently selected from hydrogen, halo, C_1-C_4 alkyl, and halo-substituted C_1-C_4 alkyl; and

each R^6 is independently selected from hydrogen, C_1-C_4 alkyl, halo-substituted C_1-C_4 alkyl.

20. The method of claim 19, further comprising co-administering to the patient in need thereof an additional therapeutic agent.]



(I)