RNAI AGENT TARGETING LRRK2 AND MEDICAL USE THEREOF

The present disclosure claims priority to Chinese Patent Application No. 202410292322.1 filed on Mar. 14, 2024 and Chinese Patent Application No. 202411444130.4 filed on Oct. 16, 2024, which are incorporated herein by reference in their entirety.

TECHNICAL FIELD

The present disclosure pertains to the field of biomedicine, and particularly relates to an RNAi agent targeting the leucine-rich repeat kinase 2 (LRRK2) gene, a composition, and pharmaceutical use thereof.

BACKGROUND

The LRRK2 gene, a member of the leucine-rich repeat kinase family, consists of 51 exons and encodes a protein having an ankyrin repeat region, a leucine-rich repeat (LRR) domain, a kinase domain, a DFG-like motif, a RAS domain, a GTPase domain, a MLK-like domain, and a WD40 domain. The LRRK2 gene is mainly expressed in the cytoplasm, is highly expressed in the lungs of healthy humans, and is lowly expressed in the brain.

Genome-wide association studies (GWAS) have shown that LRRK2 gene mutations are a powerful disease risk site and are associated with an increased risk of developing Parkinson's disease (PD). Increasing evidence demonstrates that the LRRK2 kinase is functionally enhanced in PD. Overactivation of the LRRK2 protein kinase is also present in neurons of idiopathic Parkinson's disease patients without mutations. The overactive LRRK2 protein kinase in neurons impairs vesicle trafficking and autophagy-lysosome functions, causing abnormal accumulation of α-synuclein and increasing the risk of PD.

LRRK2 mutation knock-in mice exhibit pathological changes in early PD, including increased susceptibility to nigrostriatal neurotransmission, development of motor and non-motor symptoms, mitochondrial and autophagy-lysosome defects, and synucleinopathy. In a mouse model of Parkinson's disease, ASO-mediated LRRK2 inhibition can prevent the formation of pathological synuclein α-syn inclusion bodies in dopamine neurons. In some preclinical studies and early clinical trials, LRRK2 inhibitors have exhibited neuroprotective effects. Therefore, there is a need for drugs that selectively and effectively inhibit or regulate LRRK2 gene expression, in order to effectively treat subjects with LRRK2-associated diseases. The objective of the present disclosure is to provide a method for treating such diseases.

SUMMARY

The present disclosure provides an RNAi agent targeting LRRK2.

In some embodiments, the present disclosure provides an RNAi agent comprising a sense strand and an antisense strand that form a double-stranded region, wherein the sense strand comprises at least 15 contiguous nucleotides differing from the nucleotide sequence set forth in any one of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64 by no more than 3 nucleotides, and the antisense strand comprises at least 15 contiguous nucleotides differing from the nucleotide sequence set forth in any one of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134 by no more than 3 nucleotides.

In some embodiments, the term "differing by no more than 3 nucleotides" means that the difference may be 0, 1, 2, or 3 nucleotides; in some embodiments, the term "at least 15 contiguous nucleotides" means that there is an identity of at least 15, 16, 17, 18, 19, 20, or 21 contiguous nucleotides; preferably, there is an identity of at least 17, 18, 19, 20, or 21 contiguous nucleotides.

In some embodiments, the antisense strand is at least partially complementary to a target sequence to mediate RNA interference. In some embodiments, there are no more than 5, 4, 3, 2, or 1 nucleotide mismatches between the antisense strand and the target sequence. In some embodiments, the antisense strand is fully reverse complementary to the target sequence.

In some embodiments, the sense strand is at least partially reverse complementary to the antisense strand to form a double-stranded region. In some embodiments, there are no more than 5, 4, 3, 2, or 1 nucleotide mismatches between the sense strand and the antisense strand. In some embodiments, the sense strand is fully reverse complementary to the antisense strand.

In some embodiments, the RNAi agent of the present disclosure comprises one or two blunt ends.

In some embodiments, the sense strand and/or the antisense strand of the RNAi agent of the present disclosure each independently comprise(s) 1 or 2 unpaired nucleotides. In some embodiments, the 3' end of the antisense strand comprises an overhang formed by unpaired nucleotides.

In some embodiments, the sense strand and the antisense strand each independently consist of 16 to 35, 16 to 34, 17 to 34, 17 to 33, 18 to 33, 18 to 32, 18 to 31, 18 to 30, 18 to 29, 18 to 28, 18 to 27, 18 to 26, 18 to 25, 18 to 24, 18 to 23, 19 to 25, 19 to 24, or 19 to 23 nucleotides. In some embodiments, the sense strand and the antisense strand each independently consist of 18, 19, 20, 21, 22, or 23 nucleotides.

In some embodiments, the sense strand and the antisense strand are identical or different in length; the sense strand is 19-23 nucleotides in length, and the antisense strand is 19-26 nucleotides in length. Therefore, the length ratio of the sense strand to the antisense

strand of the RNAi agent provided by the present disclosure may be 19/19, 19/20, 19/21, 19/22, 19/23, 19/24, 19/25, 19/26, 20/19, 20/20, 20/21, 20/22, 20/23, 20/24, 20/25, 20/26, 21/20, 21/21, 21/22, 21/23, 21/24, 21/25, 21/26, 22/20, 22/21, 22/22, 22/23, 22/24, 22/25, 22/26, 23/20, 23/21, 23/22, 23/23, 23/24, 23/25, or 23/26. In some embodiments, the length ratio of the sense strand to the antisense strand of the RNAi agent is 19/19, 19/21, 21/21, 21/23, 23/23, or 23/25. In some embodiments, the length ratio of the sense strand to the antisense strand is 19/21 or 21/23.

In some embodiments, the sense strand comprises at least 15 contiguous nucleotides and differs from any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64 by no more than 2 nucleotides; in some embodiments, by no more than one nucleotide; and in some embodiments, by one nucleotide.

In some embodiments, the antisense strand comprises at least 15 contiguous nucleotides and differs from the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134 by no more than 2 nucleotides; in some embodiments, by no more than one nucleotide; and in some embodiments, by one nucleotide.

In some embodiments, the sense strand comprises at least 15 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64. In some embodiments, the sense strand comprises at least 16 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64. In some embodiments, the sense strand comprises at least 17 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64. In some embodiments, the sense strand comprises at least 19 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64. In some embodiments, the sense strand comprises at least 18 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64.

In some embodiments, the antisense strand comprises at least 15 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134. In some embodiments, the antisense strand comprises at least 16 contiguous nucleotides of any one of the nucleotide sequences of

SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134. In some embodiments, the antisense strand comprises at least 17 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134. In some embodiments, the antisense strand comprises at least 18 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134. In some embodiments, the antisense strand comprises at least 19 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134. In some embodiments, the antisense strand comprises at least 20 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134. In some embodiments, the antisense strand comprises at least 21 contiguous nucleotides of any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134.

In some embodiments, the sense strand comprises or is selected from the group consisting of any one of the following nucleotide sequences: SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64.

In some embodiments, the antisense strand comprises or is selected from the group consisting of any one of the following nucleotide sequences: SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134.

In some embodiments, the RNAi agent comprises or is selected from the group consisting of any one of the following groups:

group 1): a sense strand set forth in SEQ ID NO: 4 and an antisense strand set forth in SEQ ID NO: 74;

group 2): a sense strand set forth in SEQ ID NO: 2 and an antisense strand set forth in SEQ ID NO: 72;

group 3): a sense strand set forth in SEQ ID NO: 3 and an antisense strand set forth in SEQ ID NO: 73, or a sense strand set forth in SEQ ID NO: 64 and an antisense strand set forth in SEQ ID NO: 134;

group 4): a sense strand set forth in SEQ ID NO: 1 and an antisense strand set forth in SEQ ID NO: 71;

group 5): a sense strand set forth in SEQ ID NO: 5 and an antisense strand set forth in

SEQ ID NO: 75;

group 6): a sense strand set forth in SEQ ID NO: 6 and an antisense strand set forth in SEQ ID NO: 76, or a sense strand set forth in SEQ ID NO: 26 and an antisense strand set forth in SEQ ID NO: 96;

group 7): a sense strand set forth in SEQ ID NO: 7 and an antisense strand set forth in SEQ ID NO: 77;

group 8): a sense strand set forth in SEQ ID NO: 8 and an antisense strand set forth in SEQ ID NO: 78;

group 9): a sense strand set forth in SEQ ID NO: 9 and an antisense strand set forth in SEQ ID NO: 79;

group 10): a sense strand set forth in SEQ ID NO: 10 and an antisense strand set forth in SEQ ID NO: 80, or a sense strand set forth in SEQ ID NO: 35 and an antisense strand set forth in SEQ ID NO: 105;

group 11): a sense strand set forth in SEQ ID NO: 11 and an antisense strand set forth in SEQ ID NO: 81; and

group 12): a sense strand set forth in SEQ ID NO: 12 and an antisense strand set forth in SEQ ID NO: 82, or a sense strand set forth in SEQ ID NO: 20 and an antisense strand set forth in SEQ ID NO: 90.

In some embodiments, at least one nucleotide in the sense strand and/or the antisense strand is a modified nucleotide. In some embodiments, all nucleotides in the sense strand and/or the antisense strand are modified nucleotides.

In some embodiments, the sense strand comprises three contiguous 2'-fluoro-modified nucleotides. In some embodiments, in the direction from the 5' end to the 3' end, the nucleotides at positions 7, 8, and 9 of the sense strand are 2'-fluoro-modified nucleotides, and the nucleotides at the remaining positions are non-fluoro-modified nucleotides; preferably, the non-fluoro-modified nucleotides are 2'-methoxy-modified nucleotides.

In some embodiments, the antisense strand comprises at least 5 2'-fluoro-modified nucleotides. In some embodiments, in the direction from the 5' end to the 3' end, each of the nucleotides at positions 2, 6, 12, 14, and 16 of the antisense strand is independently a 2'-fluoro-modified nucleotide.

In some embodiments, in the direction from the 5' end to the 3' end, each of the nucleotides at positions 2, 4, 6, 10, 12, 14, 16, and 18 of the antisense strand is independently a 2'-fluoro-modified nucleotide, and the nucleotides at the remaining positions are non-2'-fluoro-modified nucleotides. Preferably, the non-fluoro-modified nucleotides are 2'-methoxy-modified nucleotides.

In some embodiments, in the direction from the 5' end to the 3' end, each of the nucleotides at positions 2, 6, 12, 14, and 16 of the antisense strand is independently a 2'-fluoro-modified nucleotide, and the nucleotides at the remaining positions are non-2'-fluoro-modified nucleotides. Preferably, the non-fluoro-modified nucleotides are 2'-methoxy-modified nucleotides.

In some embodiments, at least one phosphodiester group in the sense strand and/or the antisense strand is a phosphodiester group with a modification group. The modification group causes the RNAi agent to have increased stability in a biological sample or environment. In some embodiments, the sense strand and/or the antisense strand comprise(s) a plurality of phosphodiester groups with modification groups. In some embodiments, the phosphodiester group with a modification group is present at one or more of the following positions selected from the group consisting of: between any two adjacent nucleotides of the 1st to the 3rd nucleotides of the 5' end and/or 3' end of the sense strand, and/or between any two adjacent nucleotides of the 1st to the 3rd nucleotides of the 5' end and/or 3' end of the antisense strand. In some embodiments, the phosphodiester group with a modification group is a phosphorothioate diester group. In some embodiments, the phosphorothioate diester group is present in at least one of the following positions:

between the 1st and 2nd nucleotides of the 5' end of the sense strand;
between the 2nd and 3rd nucleotides of the 5' end of the sense strand;
between the 1st and 2nd nucleotides of the 3' end of the sense strand;
between the 2nd and 3rd nucleotides of the 3' end of the sense strand;
between the 1st and 2nd nucleotides of the 5' end of the antisense strand;
between the 2nd and 3rd nucleotides of the 5' end of the antisense strand;
between the 1st and 2nd nucleotides of the 3' end of the antisense strand; and
between the 2nd and 3rd nucleotides of the 3' end of the antisense strand.

In some embodiments, the sense strand and/or the antisense strand comprise(s) a plurality of phosphorothioate diester groups, and the phosphorothioate diester groups are present:

between the 1st and 2nd nucleotides of the 5' end of the sense strand; and
between the 2nd and 3rd nucleotides of the 5' end of the sense strand; and
between the 1st and 2nd nucleotides of the 3' end of the sense strand; and
between the 2nd and 3rd nucleotides of the 3' end of the sense strand; and
between the 1st and 2nd nucleotides of the 5' end of the antisense strand; and
between the 2nd and 3rd nucleotides of the 5' end of the antisense strand; and
between the 1st and 2nd nucleotides of the 3' end of the antisense strand; and
between the 2nd and 3rd nucleotides of the 3' end of the antisense strand.

In some embodiments, the sense strand is selected from the group consisting of or comprises the nucleotide sequence set forth in any one of SEQ ID NO: 141 to SEQ ID NO: 164 and SEQ ID NO: 287 to SEQ ID NO: 290.

In some embodiments, the antisense strand is selected from the group consisting of or comprises the nucleotide sequence set forth in any one of SEQ ID NO: 214 to SEQ ID NO: 237 and SEQ ID NO: 291 to SEQ ID NO: 294.

In some embodiments, the RNAi agent further comprises one or more delivery groups, and the delivery group is linked to the sense strand and/or the antisense strand. In the

context of the present disclosure, the term “linked” includes covalent and non-covalent linkages. The delivery group is capable of delivering the RNAi agent of the present disclosure to a location where the LRRK2 gene expression is present.

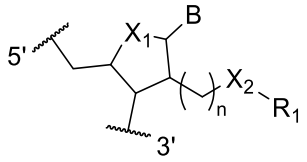
In some embodiments, the delivery group comprises a lipophilic group, and one or more lipophilic groups are linked to any one or more of the nucleotides in the sense strand or the antisense strand of the RNAi agent. In some embodiments, the one or more lipophilic groups are linked to the sense strand in the RNAi agent. In some embodiments, the one or more lipophilic groups are linked to the antisense strand in the RNAi agent. In some embodiments, at least one lipophilic group is linked to the sense strand in the RNAi agent, and at least one lipophilic group is linked to the antisense strand in the RNAi agent.

In some embodiments, the lipophilic group is linked to the base of the nucleotide. In some embodiments, the lipophilic group is linked to the sugar ring of the nucleotide. In some embodiments, the lipophilic group is linked to an internucleoside linking group between two adjacent nucleotides. In some embodiments, the lipophilic group comprises a saturated or unsaturated C_{4-30} hydrocarbon chain, and optionally a functional group selected from the group consisting of halogen, alkoxy, hydroxy, amine, carboxylic acid, sulfonate, phosphate, thiol, azide, and alkyne.

In some embodiments, the lipophilic group comprises a saturated or unsaturated C_{6-18} hydrocarbon chain. In some embodiments, the lipophilic group comprises a saturated or unsaturated C_{16} hydrocarbon chain.

In some embodiments, the RNAi agent comprises a lipophilic group, and the lipophilic group is n-hexadecyl.

In some embodiments, when the lipophilic group is covalently linked to the 2' position of the sugar ring of the nucleoside in the nucleotide, the nucleoside linked to the lipophilic group has a structure represented by formula (I):



formula (I), wherein:

X_1 is selected from the group consisting of O, S, N, and C atoms;

X_2 is selected from the group consisting of O and S atoms;

R_1 is selected from the group consisting of C_{10}-C_{30} linear alkyl, and C_{10}-C_{30} linear alkyl interrupted by one or more O or S atoms; optionally, the C_{10}-C_{30} linear alkyl may be substituted with one or more R_a, or optionally, C_{3-6} cycloalkyl forms between two adjacent carbon atoms of the C_{10}-C_{30} linear alkyl;

each R_a is independently selected from the group consisting of hydrogen, deuterium, halogen, hydroxy, cyano, alkyl, haloalkyl, alkoxy, cycloalkyl, and heterocycloalkyl;

n is 1;

B represents a base.

In some embodiments, the nucleoside represented by formula (I) has the structure of the nucleoside moiety of the compound represented by formula (I) or the pharmaceutically acceptable salt thereof described in PCT application WO2024125556A1, which is incorporated in the present disclosure by reference in its entirety.

In some embodiments, X_1 in formula (I) is selected from the group consisting of O and S atoms. In some embodiments, X_1 is an O atom.

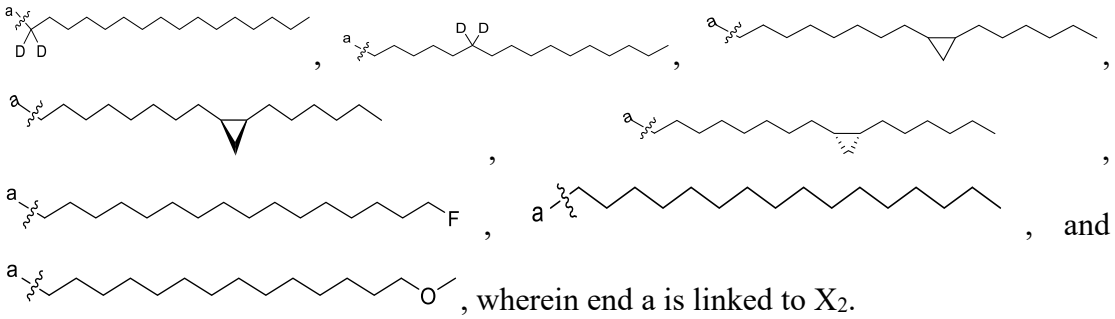
In some embodiments, X_2 in formula (I) is selected from the group consisting of O and S atoms. In some embodiments, X_2 is an O atom.

In some embodiments, R_1 in formula (I) is selected from the group consisting of C_{14}-C_{24} linear alkyl (e.g., C_{14}, C_{15}, C_{16}, C_{17}, C_{18}, C_{19}, C_{20}, C_{21}, C_{22}, C_{23}, or C_{24} linear alkyl), and C_{14}-C_{24} linear alkyl interrupted by one or more O or S atoms (e.g., C_{14}, C_{15}, C_{16}, C_{17}, C_{18}, C_{19}, C_{20}, C_{21}, C_{22}, C_{23}, or C_{24} linear alkyl).

In some embodiments, each R_a in formula (I) is independently selected from the group consisting of hydrogen, deuterium, halogen (e.g., fluorine, chlorine, or bromine), C_{1-6} alkyl (e.g., C_1, C_2, C_3, C_4, C_5, or C_6 alkyl, including but not limited to methyl, ethyl, and isopropyl), and C_{1-6} alkoxy (e.g., C_1 alkoxy, C_2 alkoxy, C_3 alkoxy, C_4 alkoxy, C_5 alkoxy, or C_6 alkoxy, including but not limited to methoxy, ethoxy, propoxy, and isopropoxy).

In some embodiments, each R_a in formula (I) is independently selected from the group consisting of hydrogen, deuterium, fluorine, methyl, and methoxy.

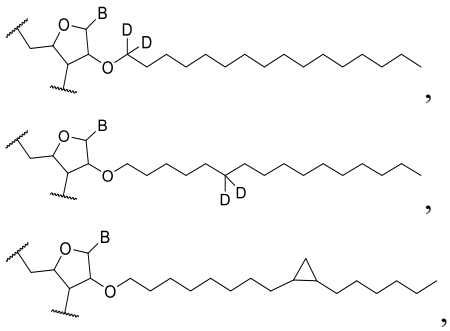
In some embodiments, R_1 in formula (I) is selected from the group consisting of:

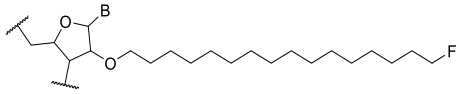
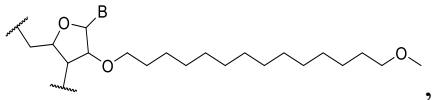
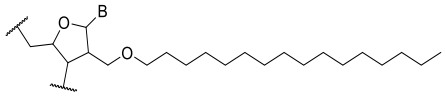


, wherein end a is linked to X_2.

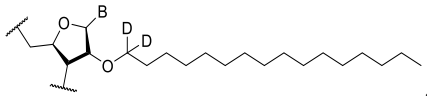
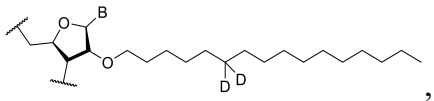
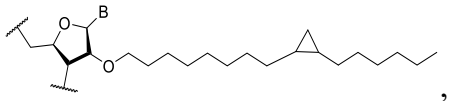
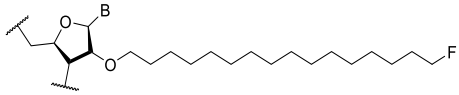
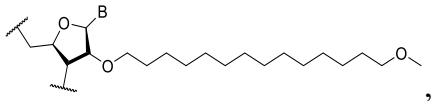
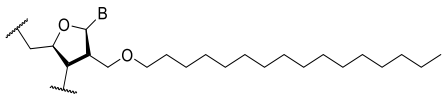
In some embodiments, B is selected from the group consisting of adenine, guanine, cytosine, uracil, and thymine.

In some embodiments, the nucleoside represented by formula (I) is selected from the group consisting of:

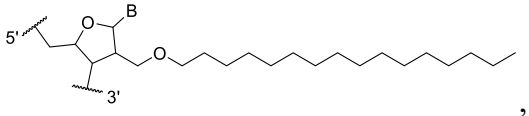



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in some embodiments, the nucleoside represented by formula (I) is selected from the group consisting of:


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In some embodiments, the nucleoside represented by formula (I) is:

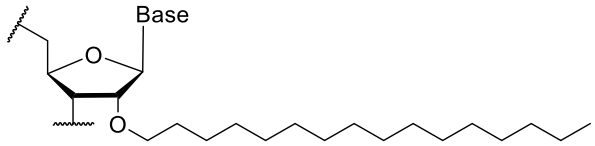

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wherein B represents a base.

In some embodiments, the nucleoside represented by formula (I) may be located at any position of the sense strand and/or the antisense strand in the RNAi agent. In some embodiments, the nucleoside represented by formula (I) is located in the sense strand. In some embodiments, the nucleoside represented by formula (I) is located at one or more of positions 1 to 8 from the 5' end or from the 3' end of the sense strand, for example, at position 1, position 2, position 3, position 4, position 5, position 6, position 7, or position 8. In some embodiments, the nucleoside represented by formula (I) is located at one or more of position 1, position 2, position 6, position 7, position 19, position 20, or position 21 from the 5' end of the sense strand.

In some embodiments, the nucleoside represented by formula (I) is located at one or more of position 2, position 7, or position 20 from the 5' end of the sense strand.

In some embodiments, the nucleoside linked to the lipophilic group has a structure represented by formula (II):



(II), wherein Base represents a base.

In some embodiments, the delivery group comprises a targeting ligand targeting the liver. In some embodiments, the targeting ligand binds to an asialoglycoprotein receptor (ASGPR). In some embodiments, the targeting ligand comprises a galactose cluster or a galactose derivative cluster, wherein the galactose derivative is selected from the group consisting of N-acetyl-galactosamine, N-trifluoroacetylgalactosamine, N-propionylgalactosamine, N-n-butyrylgalactosamine, and N-isobutyrylgalactosamine.

In some embodiments, the delivery group is linked to the 3' end of the sense strand of the RNAi agent.

In some embodiments, the delivery group is linked to an end of the RNAi agent by a phosphodiester group, a phosphorothioate diester group, or a phosphonic acid group. In some embodiments, the delivery group is linked to an end of the RNAi agent by a phosphodiester group.

In some embodiments, the delivery group is indirectly linked to an end of the RNAi agent by a phosphodiester group, a phosphorothioate diester group, or a phosphonic acid group. In some embodiments, the delivery group is indirectly linked to an end of the RNAi agent by a phosphodiester group.

In some embodiments, the delivery group is directly linked to an end of the RNAi agent by a phosphodiester group, a phosphorothioate diester group, or a phosphonic acid group. In some embodiments, the delivery group is directly linked to an end of the RNAi agent by a phosphodiester group.

In some embodiments, the delivery group is directly linked to the 3' end of the sense strand of the RNAi agent by a phosphodiester group or a phosphorothioate diester group. In some embodiments, the delivery group is directly linked to the 3' end of the sense strand of the RNAi agent by a phosphodiester group.

In another aspect, the present disclosure provides a pharmaceutical composition comprising the RNAi agent described herein, and one or more pharmaceutically acceptable carriers, for example, but not limited to, excipients, e.g., vehicles, diluents, and/or delivery systems (e.g., delivery polymers). Various drug delivery systems are known and can be used for the RNAi agent of the present disclosure, e.g., encapsulation in liposomes, microparticles, microcapsules, recombinant cells capable of expressing the RNAi agent, receptor-mediated endocytosis, and construction of a nucleic acid as part of a retroviral vector or other vectors.

In some embodiments, the pharmaceutical composition may further comprise a pharmaceutically acceptable auxiliary material and/or adjuvant, and the auxiliary material may be one or more of the various formulations or compounds conventionally

used in the art. For example, the pharmaceutically acceptable auxiliary material may include at least one of a pH buffer, a protective agent, and an osmotic pressure regulator. In some embodiments, a unit dose of the pharmaceutical composition is 0.001 mg-1000 mg.

In certain embodiments, the pharmaceutical composition comprises 0.01%-99.99% of the aforementioned RNAi agent based on the total weight of the composition. In certain embodiments, the pharmaceutical composition comprises 0.1%-99.9% of the aforementioned RNAi agent. In certain embodiments, the pharmaceutical composition comprises 0.5%-99.5% of the aforementioned RNAi agent. In certain embodiments, the pharmaceutical composition comprises 1%-99% of the aforementioned RNAi agent. In certain embodiments, the pharmaceutical composition comprises 2%-98% of the aforementioned RNAi agent.

In certain embodiments, the pharmaceutical composition comprises 0.01%-99.99% of the pharmaceutically acceptable carrier based on the total weight of the composition. In certain embodiments, the pharmaceutical composition comprises 0.1%-99.9% of the pharmaceutically acceptable carrier. In certain embodiments, the pharmaceutical composition comprises 0.5%-99.5% of the pharmaceutically acceptable carrier. In certain embodiments, the pharmaceutical composition comprises 1%-99% of the pharmaceutically acceptable carrier. In certain embodiments, the pharmaceutical composition comprises 2%-98% of the pharmaceutically acceptable carrier.

In some embodiments, when the RNAi agent or the pharmaceutical composition of the present disclosure is in contact with a target gene-expressing cell, the RNAi agent or the pharmaceutical composition of the present disclosure inhibits the expression of the target gene by at least 5%, at least 10%, at least 15%, at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99%, as measured by, for example, psiCHECK activity screening and luciferase reporter gene assay, other methods such as PCR or branched DNA (bDNA)-based methods, or protein-based methods such as immunofluorescence assay, e.g., western blot or flow cytometry.

In some embodiments, when the RNAi agent or the pharmaceutical composition of the present disclosure is in contact with a target gene-expressing cell, the RNAi agent or the pharmaceutical composition of the present disclosure results in a remaining expression percentage of the target gene mRNA of no greater than 99%, no greater than 95%, no greater than 90%, no greater than 85%, no greater than 80%, no greater than 75%, no greater than 70%, no greater than 65%, no greater than 60%, no greater than 55%, no greater than 50%, no greater than 45%, no greater than 40%, no greater than 35%, no greater than 30%, no greater than 25%, no greater than 20%, no greater than 15%, or no greater than 10%, as measured by, for example, psiCHECK activity

screening and luciferase reporter gene assay, other methods such as PCR or branched DNA (bDNA)-based methods, or protein-based methods such as immunofluorescence assay, e.g., western blot or flow cytometry.

In some embodiments, when the RNAi agent or the pharmaceutical composition of the present disclosure is in contact with a target gene-expressing cell, the RNAi agent, while retaining on-target activity, reduces off-target activity by at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, or at least 75%, as measured by, for example, psiCHECK activity screening and luciferase reporter gene assay, other methods such as PCR or branched DNA (bDNA)-based methods, or protein-based methods such as immunofluorescence assay, e.g., western blot or flow cytometry.

In some embodiments, when the RNAi agent or the pharmaceutical composition of the present disclosure is in contact with a target gene-expressing cell, the RNAi agent, while reducing on-target activity by at most 20%, at most 19%, at most 15%, at most 10%, at most 5%, or more than 1%, reduces off-target activity by at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, or at least 75%, as measured by, for example, psiCHECK activity screening and luciferase reporter gene assay, other methods such as PCR or branched DNA (bDNA)-based methods, or protein-based methods such as immunofluorescence assay, e.g., western blot or flow cytometry.

In some embodiments, when the RNAi agent or the pharmaceutical composition of the present disclosure is in contact with a target gene-expressing cell, the RNAi agent, while increasing on-target activity by at least 1%, at least 5%, at least 10%, at least 15%, at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, or at least 80%, reduces off-target activity by at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, or at least 75%, as measured by, for example, psiCHECK activity screening and luciferase reporter gene assay, other methods such as PCR or branched DNA (bDNA)-based methods, or protein-based methods such as immunofluorescence assay, e.g., western blot or flow cytometry.

In another aspect, the present disclosure provides a method for reducing LRRK2 expression, comprising administering to a subject an effective amount or effective dose of the RNAi agent and/or the pharmaceutical composition of the present disclosure.

In another aspect, the present disclosure provides a method for treating and/or preventing a disease associated with LRRK2 gene expression in a subject, comprising administering to the subject an effective amount or effective dose of the RNAi agent and/or the pharmaceutical composition of the present disclosure.

In another aspect, the present disclosure provides use of the RNAi agent and/or the pharmaceutical composition of the present disclosure in the manufacture of a

medicament for treating and/or preventing an associated disease, wherein the associated disease is a disease associated with LRRK2 gene expression.

In another aspect, the present disclosure provides use of the RNAi agent and/or the pharmaceutical composition of the present disclosure in the manufacture of a medicament for inhibiting LRRK2 expression.

In some embodiments, the disease is a disease associated with LRRK2. In some embodiments, the disease is a neurodegenerative disease. In some embodiments, the neurodegenerative disease may be a familial disorder or a sporadic disorder, including Parkinson's disease, amyotrophic lateral sclerosis (ALS), Alzheimer's disease, Huntington's disease, schizophrenia, progressive myoclonic epilepsy (Unver-Richt-Lundberg Lafora disease), Hallervorden-Spatz disease, and the like. In some embodiments, the disease is Parkinson's disease.

In another aspect, the present disclosure discloses a method for delivering an RNAi agent inhibiting LRRK2 expression and/or replication *in vivo*, and the method comprises administering to a subject an effective amount or effective dose of the RNAi agent and/or the pharmaceutical composition of the present disclosure.

The RNAi agent or the pharmaceutical composition and the methods disclosed herein can reduce the level of a target mRNA in a cell, a cell population, a cell population, a tissue, or a subject, comprising administering to the subject a therapeutically effective amount of the RNAi agent or the pharmaceutical composition described in the present disclosure. The RNAi agent is linked to the delivery group, thereby inhibiting the expression of the target mRNA in the subject.

In some embodiments, the subject has been identified as having pathological up-regulation of the target gene in the targeted cell or tissue prior to administration of the RNAi agent and/or the pharmaceutical composition of the present disclosure.

The subject described in the present disclosure refers to a subject having (or suspected to have or susceptible to) a disease or disorder that would benefit from reduction or inhibition of target mRNA expression.

The RNAi agent and/or the pharmaceutical composition of the present disclosure may be delivered by topical administration (e.g., direct injection, implantation, or topical administration), systemic administration, or through subcutaneous, intravenous, intraperitoneal, or parenteral routes, including intracranial (e.g., intraventricular, intraparenchymal, and intrathecal), intramuscular, transdermal, airway (aerosol), nasal, oral, rectal, or topical (including buccal and sublingual) administration and any other suitable mode of administration commonly used in the art.

In an optional embodiment, the pharmaceutical composition provided by the present disclosure may be administered by injection, e.g., intravenous, intramuscular, intradermal, subcutaneous, intraduodenal, or intraperitoneal injection.

In another aspect, the present disclosure provides a cell comprising the RNAi agent of the present disclosure. The cell cannot develop into an animal or plant individual.

In another aspect, the present disclosure provides a kit comprising the RNAi agent and/or the pharmaceutical composition of the present disclosure. In an optional embodiment, after the delivery group is linked to the RNAi agent, the RNAi agent can be packaged in the kit in the form of a conjugate.

The present disclosure further provides a method for silencing an mRNA of a target gene in a cell, and the method comprises a step of introducing into the cell the RNAi agent and/or the pharmaceutical composition of the present disclosure.

The present disclosure further provides a method for silencing a target gene or mRNA of the target gene in a cell *in vivo* or *in vitro*, and the method comprises a step of introducing into the cell the RNAi agent and/or the pharmaceutical composition of the present disclosure.

The present disclosure further provides a method for inhibiting the expression of a target gene or mRNA of the target gene, and the method comprises administering to a subject in need thereof an effective amount or effective dose of the RNAi agent and/or the pharmaceutical composition according to the present disclosure.

In some embodiments, the effective amount or effective dose of the RNAi agent and/or the pharmaceutical composition is about 0.001 mg/kg body weight to about 200 mg/kg body weight, about 0.01 mg/kg body weight to about 100 mg/kg body weight, or about 0.5 mg/kg body weight to about 50 mg/kg body weight.

In some embodiments, the target gene is the LRRK2 gene, and the target mRNA is an mRNA expressed by the target gene.

The present disclosure further provides a method for preparing an RNAi agent, comprising: synthesizing the RNAi agent described in the present disclosure.

The pharmaceutically acceptable salts of the compounds described in the present disclosure are selected from the group consisting of inorganic salts and organic salts. The compounds described in the present disclosure can react with acidic or basic substances to form corresponding salts. In the context of the present disclosure, the compound comprises the RNAi agent of the present disclosure.

In another aspect, where the configuration is not specified, the compounds of the present disclosure may have particular geometric or stereoisomeric forms. The present disclosure contemplates all such compounds, including cis and trans isomers, (-)- and (+)-enantiomers, (R)- and (S)-enantiomers, diastereomers, (D)-isomer, (L)-isomer, and racemic mixtures and other mixtures thereof, such as enantiomerically or diastereomerically enriched mixtures, all of which fall within the scope of the present disclosure. Additional asymmetric carbon atoms may be present in substituents such as an alkyl group. All such isomers and mixtures thereof are included within the scope of the present disclosure.

In addition, when the configuration is not specified, the compounds and intermediates of the present disclosure can also be present in different tautomeric forms, and all such forms fall within the scope of the present disclosure. The term “tautomer” or

"tautomeric form" refers to structural isomers of different energies that can interconvert via a low-energy barrier.

The compounds of the present disclosure may be asymmetric, for example, having one or more stereoisomers. Unless otherwise specified, all stereoisomers are included, for example, enantiomers and diastereomers. The compounds of the present disclosure containing asymmetric carbon atoms may be separated in an optically active pure form or in a racemic form. The optically active pure form may be isolated from a racemic mixture or synthesized using chiral starting materials or chiral reagents.

Optically active (R)- and (S)-isomers, and D- and L-isomers may be prepared by chiral synthesis, using chiral reagents, or other conventional techniques. If one enantiomer of a certain compound of the present disclosure is desired, it may be prepared by asymmetric synthesis or derivatization with a chiral auxiliary, wherein the resulting mixture of diastereomers is separated, and the auxiliary group is cleaved to provide the pure desired enantiomer. Alternatively, when the molecule contains a basic functional group (e.g., amino) or an acidic functional group (e.g., carboxyl), salts of diastereomers are formed with an appropriate optically active acid or base, diastereomeric resolution is then performed by conventional methods well-known in the art, and pure enantiomers are then recovered. In addition, the separation of enantiomers and diastereomers is generally accomplished by chromatography using a chiral stationary phase, optionally in combination with chemical derivatization (e.g., carbamate formation from amines).

The present disclosure also includes some isotopically labeled compounds of the present disclosure that are identical to those recited herein but have one or more atoms replaced with an atom having an atomic mass or mass number different from the atomic mass or mass number usually found in nature. Examples of isotopes that can be incorporated into the compounds of the present disclosure include isotopes of hydrogen, carbon, nitrogen, oxygen, phosphorus, sulfur, fluorine, iodine, and chlorine, such as ^{2}H, ^{3}H, ^{11}C, ^{13}C, ^{14}C, ^{13}N, ^{15}N, ^{15}O, ^{17}O, ^{18}O, ^{31}P, ^{32}P, ^{35}S, ^{18}F, ^{123}I, ^{125}I, and ^{36}Cl.

Where the configurations are not specified, in the chemical structures of the compounds of the present disclosure, the bond "" indicates an unspecified configuration; that is, if chiral isomers exist in the chemical structures, the bond "" may be "" or "", or includes both the configurations "" and "" simultaneously. Although all of the structural formulas of the present disclosure are drawn as certain isomeric forms for the sake of simplicity, the present disclosure may include all isomers, such as tautomers, rotamers, geometric isomers, diastereomers, racemates, and enantiomers. In the chemical structures of the compounds of the present disclosure, the bond "" does not specify a configuration; that is, the configuration of the bond "" may be an E configuration or a Z configuration, or includes both the E configuration and the Z configuration simultaneously.

WO2023274395A1 is incorporated in the present disclosure by reference in its entirety.

Terms and Definitions

In order to facilitate the understanding of the present disclosure, some technical and scientific terms are specifically defined below. Unless otherwise specifically defined herein, all other technical and scientific terms used herein have the meanings generally understood by those of ordinary skill in the art to which the present disclosure pertains.

As used herein, "RNAi agent" comprises an RNA or RNA-like (e.g., chemically modified RNA) oligonucleotide molecule that is capable of degrading or inhibiting (e.g., degrading or inhibiting under appropriate conditions) translation of a messenger RNA (mRNA) transcript of a target mRNA in a sequence-specific manner. The RNAi agent used herein may act through an RNA interference mechanism (i.e., inducing RNA interference through interaction with the RNA interference pathway machinery (RNA-induced silencing complex or RISC) of mammalian cells) or through any alternative mechanism or pathway. Although the term RNAi agent as used herein is believed to act primarily through an RNA interference mechanism, the disclosed RNAi agent is not bound or limited by any particular pathway or mechanism of action. The RNAi agent disclosed herein consists of a sense strand and an antisense strand, and includes, but is not limited to, short (or small) interfering RNAs (siRNAs). The antisense strand of the RNAi agent described herein is at least partially complementary to the targeted mRNA. The RNAi agent may comprise one or more modified nucleotides and/or one or more non-phosphodiester linkages.

Unless otherwise specified, in the context of the present disclosure, the terms "leucine-rich repeat kinase 2" and "LRRK2" are used interchangeably in the present disclosure. LRRK2 includes, but is not limited to, human LRRK2, cynomolgus monkey LRRK2, mouse LRRK2, and rat LRRK2, and their amino acids, complete coding sequences, and mRNA sequences are readily accessible using publicly available databases, e.g., GenBank, UniProt, OMIM, and the Macaca genome project website.

The term "LRRK2" also refers to naturally occurring DNA sequence variations of the LRRK2 gene, such as a single nucleotide polymorphism (SNP) in the LRRK2 gene. Exemplary SNPs may be found in the dbSNP database.

The term "target sequence" refers to a contiguous portion of the nucleotide sequence of an mRNA molecule formed during the transcription of LRRK2, including mRNA that is a product of RNA processing of a primary transcription product. The targeted portion in the target sequence should be long enough to serve as a substrate for iRNA-directed cleavage. In one embodiment, the target sequence is within the protein-coding region of LRRK2. As used herein, in the context of RNA-mediated gene silencing, the sense strand (also referred to as SS or SS strand) refers to a strand comprising a sequence identical or substantially identical to a target mRNA sequence; the antisense strand (also referred to as AS or AS strand) refers to a strand comprising a sequence complementary to a target mRNA sequence.

In the context describing the sense strand of the RNAi agent described herein, the term

"at least 15 contiguous nucleotides differing from the nucleotide sequence set forth in SEQ ID NO: 1 by no more than 3 nucleotides" is intended to mean that the sense strand of the RNAi agent described herein comprises at least 15 contiguous nucleotides of the nucleotide sequence set forth in SEQ ID NO: 1, or a sequence differing from at least 15 contiguous nucleotides of the nucleotide sequence set forth in SEQ ID NO: 1 by no more than 3 nucleotides (optionally by no more than 2 nucleotides; optionally by one nucleotide). Other similar descriptions in the context of the present disclosure should also be understood similarly. The term "differing" described herein does not include the case of nucleotides containing different modifications; that is, nucleotides containing identical bases but different modifications do not belong to the nucleotides with the difference of the present disclosure. Other similar descriptions of the sense strand and/or the antisense strand in the context of the present disclosure should also be understood in the same way.

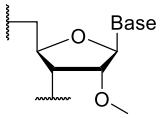
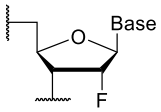
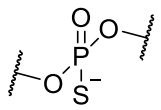
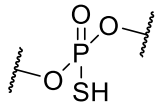
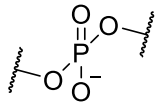
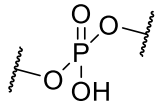
In the present disclosure, the "5' region", "5' end", and "5' terminus" of the sense or antisense strand are used interchangeably. For example, the nucleotides at positions 2 to 8 of the 5' region of the antisense strand may also be replaced with the nucleotides at positions 2 to 8 of the 5' end of the antisense strand. Likewise, the "3' region", "3' terminus", and "3' end" of the sense or antisense strand are also used interchangeably.

Unless otherwise specified, in the context of the present disclosure, "G", "C", "A", "T", and "U" represent nucleotides, and comprise the bases of guanine, cytosine, adenine, thymidine, and uracil, respectively. It is well-known to those skilled in the art that the mutual replacement between bases T and U does not significantly affect the properties of the RNAi agent sequence. In the sequences of the present disclosure, U can be arbitrarily replaced with T, and the sequences after the replacement also fall within the protection scope of the present disclosure. In the sequences of the present disclosure, for the same nucleic acid strand, the direction from the 5' end to the 3' end is a direction from the left to the right; the lowercase letter m means that the nucleoside adjacent to the letter m on the left side is a 2'-methoxy-modified nucleoside; the lowercase letter f means that the nucleoside adjacent to the letter f on the left side is a 2'-fluoro-modified nucleoside; the lowercase letter s means that the two nucleosides adjacent to the letter s are linked by a phosphorothioate diester group. Unless otherwise specified, two nucleosides are linked by a phosphodiester group. Unless otherwise specified, the "RNAi agent", "nucleotide", "compound", "chemical modification", "oligonucleotide", "double-stranded RNAi inhibitor molecule", "siRNA", "dsRNA", "nucleic acid", and "RNAi" of the present disclosure can each independently be present in the form of a salt or mixed salt or in a non-salt form (e.g., a free acid or free base). When present in the form of a salt or mixed salt, it may be a pharmaceutically acceptable salt. The term "pharmaceutically acceptable salt" includes pharmaceutically acceptable acid addition salts and pharmaceutically acceptable base addition salts. When it is present in the form of a salt, part of the groups may be ionized to form anions/cations; for example,

phosphodiester groups and phosphorothioate diester groups may be present in the form of anions. Unless otherwise specified, the structures in the form of a salt corresponding to the following structures also fall within the protection scope of the present disclosure. Unless otherwise specified, the 3' position of the first nucleotide of the 3' end of each strand is hydroxy; the 5' position of the first nucleotide of the 5' end of each strand is hydroxy.

The modifications and linking groups described above separately have the structures shown in the table below, where Base represents a base at the corresponding position:

Table 1

Structure	Name
	2'-Methoxy-modified nucleoside
	2'-Fluoro-modified nucleoside
	Anion form of phosphorothioate diester group
	Phosphorothioate diester group
	Anion form of phosphodiester group
	Phosphodiester group

The term “lipophilic group” or “lipophilic moiety” broadly refers to any compound or chemical moiety that has an affinity for lipids. One way to characterize the lipophilicity of a lipophilic moiety is by the octanol-water partition coefficient, $\log K_{ow}$, where K_{ow} is the ratio of the concentration of a chemical substance in the octanol phase to its concentration in the aqueous phase in a biphasic system in equilibrium. In principle, a chemical substance is lipophilic when its $\log K_{ow}$ exceeds 0. Typically, a lipophilic moiety has a $\log K_{ow}$ of greater than 1, greater than 1.5, greater than 2, greater than 3, greater than 4, greater than 5, or greater than 10; for example, 6-aminohexanol has a $\log K_{ow}$ of about 0.7, and cholesteryl N-(hexan-6-ol)carbamate has a $\log K_{ow}$ of 10.7. The lipophilicity of a molecule may vary relative to the functional groups it carries. For example, adding a hydroxy group or an amine group to an end of a lipophilic moiety

may increase or decrease the partition coefficient (e.g., $\log K_{ow}$) value of the lipophilic moiety. For example, a lipophilic moiety may be an aliphatic, cyclic such as alicyclic, or polycyclic such as polyalicyclic compounds, such as steroids (e.g., sterols) or linear or branched aliphatic hydrocarbons. A lipophilic moiety may generally contain a hydrocarbon chain, and the hydrocarbon chain may be cyclic or acyclic. The hydrocarbon chain may contain various substituents and/or one or more heteroatoms, such as oxygen or sulfur atoms. Such lipophilic aliphatic moieties include, but are not limited to, saturated or unsaturated C_4-C_{30} hydrocarbons (e.g., C_{10}-C_{30} hydrocarbons), saturated or unsaturated fatty acids, waxes (e.g., monohydric alcohol esters of fatty acids and fatty diamides), terpenes (e.g., C_{10} terpenes, C_{15} sesquiterpenes, C_{20} diterpenes, C_{30} triterpenes, and C_{40} tetraterpenes), and other polyalicyclic hydrocarbons; for example, a lipophilic moiety may be an optionally substituted $C_{10\text{-}30}$ linear alkyl group; for example, a lipophilic moiety may be an optionally substituted $C_{14\text{-}24}$ linear alkyl group.

As used herein, the terms "complementary" and "reverse complementary" are used interchangeably and have the meaning well-known to those skilled in the art; that is, in a double-stranded nucleic acid molecule, the bases of one strand are paired with the bases of the other strand in a complementary manner. In DNA, the purine base adenine is always paired with the pyrimidine base thymine (or uracil in RNA), and the purine base guanine is always paired with the pyrimidine base cytosine. Each base pair comprises a purine and a pyrimidine. When adenines of one strand are always paired with thymines (or uracils) of another strand, and guanines are always paired with cytosines, the two strands are considered complementary to each other, and the sequences of the strands can be deduced from the sequences of their complementary strands. Accordingly, "mismatch" in the art means that in a double-stranded nucleic acid, the bases at the corresponding positions are not paired in a complementary manner.

As used herein, the term "inhibit" is used interchangeably with "decrease", "silence", "down-regulate", "repress", and other similar terms, and includes any level of inhibition. Inhibition can be assessed in terms of a decrease in the absolute or relative level of one or more of these variables relative to a control level. The control level can be any type of control level used in the art, such as a pre-dose baseline level or a level determined from an untreated or control (e.g., buffer-only control or inert agent control) treated subject, cell, or sample. For example, the remaining mRNA expression level can be used to characterize the degree of inhibition of target gene expression by the RNAi agent; for example, the remaining expression level of mRNA is not greater than 99%, not greater than 95%, not greater than 90%, not greater than 85%, not greater than 80%, not greater than 75%, not greater than 70%, not greater than 65%, not greater than 60%, not greater than 55%, not greater than 50%, not greater than 45%, not greater than 40%, not greater than 35%, not greater than 30%, not greater than 25%, not greater than 20%, not greater than 15%, or not greater than 10%. The inhibition rate of target gene

expression can be measured using Dual-Glo® Luciferase Assay System: the Firefly chemiluminescence value (Fir) and the Renilla chemiluminescence value (Ren) are each read, and the relative value Ratio = Ren/Fir is calculated; in the present disclosure, the ratio of the remaining mRNA expression level (or residual activity%) = Ratio (RNAi agent-treated group)/Ratio (RNAi agent-free control group), and the inhibition rate (%) = 100% - remaining mRNA expression level (%).

The term "pharmaceutically acceptable salt" includes pharmaceutically acceptable acid addition salts and pharmaceutically acceptable base addition salts. As an example, the pharmaceutically acceptable salt of the RNAi agent of the present disclosure is a sodium salt.

"Pharmaceutically acceptable acid addition salt" refers to salts that are capable of retaining the biological effectiveness of free bases without having any undesirable effects and that are formed with inorganic or organic acids. Inorganic acid salts include, but are not limited to, hydrochlorides, hydrobromides, sulfates, nitrates, phosphates, etc.; organic acid salts include, but are not limited to, formates, acetates, 2,2-dichloroacetates, trifluoroacetates, propionates, caproates, caprylates, caprates, undecenates, glycolates, gluconates, lactates, sebacates, adipates, glutarates, malonates, oxalates, maleates, succinates, fumarates, tartrates, citrates, palmitates, stearates, oleates, cinnamates, laurates, malates, glutamates, pyroglutamates, aspartates, benzoates, mesylates, benzenesulfonates, p-toluenesulfonates, alginates, ascorbates, salicylates, 4-aminosalicylates, napadisylates, etc. These salts can be prepared using methods known in the art.

"Pharmaceutically acceptable base addition salt" refers to salts that are capable of retaining the biological effectiveness of free acids without having any undesirable effects and that are formed with inorganic bases or organic bases. Salts derived from inorganic bases include, but are not limited to, sodium salts, potassium salts, lithium salts, ammonium salts, calcium salts, magnesium salts, iron salts, zinc salts, copper salts, manganese salts, aluminum salts, etc. The inorganic salts in some embodiments are ammonium salts, sodium salts, potassium salts, calcium salts, and magnesium salts. Salts derived from organic bases include, but are not limited to, salts of the following: primary, secondary, and tertiary amines, substituted amines including naturally occurring substituted amines, cyclic amines, and basic ion exchange resins, such as ammonia, isopropylamine, trimethylamine, diethylamine, triethylamine, tripropylamine, ethanolamine, diethanolamine, triethanolamine, dimethylethanolamine, 2-dimethylaminoethanol, 2-diethylaminoethanol, dicyclohexylamine, lysine, arginine, histidine, caffeine, procaine, choline, betaine, ethylenediamine, glucosamine, methylglucamine, theobromine, purine, piperazine, piperidine, N-ethylpiperidine, polyamine resins, etc. The organic bases in some embodiments include isopropylamine, diethylamine, ethanolamine, trimethylamine, dicyclohexylamine, choline, and caffeine. These salts can be prepared using methods known in the art.

"Effective amount" or "effective dose" refers to the amount of a drug, an RNAi agent, a compound, or a pharmaceutical composition necessary to obtain any one or more beneficial or desired therapeutic results. For prophylactic use, the beneficial or desired results include elimination or reduction of risk, reduction of severity, or delay of the onset of a disorder, including the biochemistry, histology, and/or behavioral symptoms of the disorder, complications thereof, and intermediate pathological phenotypes that appear during the progression of the disorder. For therapeutic applications, the beneficial or desired results include clinical results, such as reducing the incidence of various disorders related to the target gene, target mRNA, or target protein of the present disclosure or ameliorating one or more symptoms of the disorders, reducing the dose of other agents required to treat the disorders, enhancing the therapeutic effect of another agent, and/or delaying the progression of disorders related to the target gene, target mRNA, or target protein of the present disclosure in patients.

As used herein, "patient", "subject", and "individual" are used interchangeably and include human or non-human animals, e.g., mammals, e.g., humans or monkeys.

The RNAi agent provided by the present disclosure can be obtained using a preparation method conventional in the art (e.g., solid-phase synthesis and liquid-phase synthesis). Solid-phase synthesis has been commercially available as a customization service. A modified nucleotide group can be introduced into the RNAi agent described in the present disclosure using a nucleoside monomer with a corresponding modification. Methods of preparing a nucleoside monomer with a corresponding modification and introducing a modified nucleotide group into an RNAi agent are also well known to those skilled in the art.

The term "chemical modification" or "modification" includes all changes made to a nucleotide by chemical means, such as the addition or removal of a chemical moiety, or the substitution of one chemical moiety for another.

The term "base" encompasses any known DNA and RNA bases and base analogs such as purines or pyrimidines, and also includes the natural compounds adenine, thymine, guanine, cytosine, uracil, inosine, and natural analogs.

The terms "blunt" and "blunt-ended" are used interchangeably and mean that there are no unpaired nucleotides or nucleotide analogs at a given end of an RNAi agent, i.e., no nucleotide overhangs. In most cases, an RNAi agent whose ends are both blunt-ended will be double-stranded over its entire length.

The terms "about" and "approximately" mean that a numerical value is within an acceptable margin of error for the specific value determined by those of ordinary skill in the art, and the numerical value depends in part on how the value is measured or determined (i.e., the limitations of the measurement system). For example, "about" may mean a standard deviation within 1 or greater than 1. Alternatively, "about" or "substantially comprise" may mean a range of at most 20%, e.g., a change of between 1% and 15%, between 1% and 10%, between 1% and 5%, between 0.5% and 5%, or

between 0.5% and 1%. In the present disclosure, every instance where a number or numerical range is preceded by the term "about" also includes an embodiment of the given number. Unless otherwise specified, when a specific value is provided in the present application and claims, the meaning of "about" or "substantially comprise" should be assumed to be within an acceptable margin of error for that specific value.

Unless otherwise specified, "optionally" or "optional" means that the event or circumstance subsequently described may, but does not necessarily, occur, and this description includes instances where the event or circumstance occurs or does not occur. For example, "optionally, R1 and R2 are directly linked to form a ring" means that R1 and R2 being directly linked to form a ring may occur, but does not necessarily exist, and this description includes an instance where R1 and R2 are directly linked to form a ring and an instance where R1 and R2 do not form a ring.

In the chemical structural formulas of the present disclosure, "" or  or  may be linked to any group or groups in accordance with the scope of the invention described herein.

The term "link", when referring to a relationship between two molecules, means that the two molecules are linked by a covalent bond or that the two molecules are associated via a non-covalent bond (e.g., a hydrogen bond or an ionic bond), and includes direct linkage and indirect linkage.

The term "directly linked" means that a first compound or group is linked to a second compound or group without any atom or group of atoms interposed between.

The term "indirectly linked" means that a first compound or group is linked to a second compound or group by an intermediate group, a compound, or a molecule (e.g., a linking group).

The term "substituted" means that any one or more hydrogen atoms on the specified atom (usually a carbon, oxygen, or nitrogen atom) are replaced with any group as defined herein, provided that the normal valency of the specified atom is not exceeded and that the substitution results in a stable compound. Non-limiting examples of substituents include C1-C6 alkyl, C2-C6 alkenyl, C2-C6 alkynyl, cyano, hydroxy, oxo, carboxyl, cycloalkyl, cycloalkenyl, heterocyclyl, heteroaryl, aryl, ketone, alkoxycarbonyl, aryloxycarbonyl, heteroaryloxycarbonyl, or halogen (e.g., F, Cl, Br, or I). When the substituent is a ketone or oxo (i.e., =O), two (2) hydrogens on the atom are replaced.

"Substituted with one or more..." refers to substitution with a single substituent or multiple substituents. In the case of substitution with multiple substituents, there may be a plurality of identical substituents, or one or a group of a plurality of different substituents.

DETAILED DESCRIPTION

The present disclosure is further described below with reference to examples, but these

examples are not intended to limit the scope of the present disclosure. In the examples of the present disclosure, experimental methods without specific conditions indicated were generally conducted under conventional conditions, such as those outlined in Antibodies: A Laboratory Manual and Molecular Cloning: A Laboratory Manual by Cold Spring Harbor Laboratory, or under conditions recommended by the manufacturers of the starting materials or commercial products. Where the specific source of a reagent is not indicated, the reagent may be obtained from any supplier of molecular biology reagents at a quality/purity level for molecular biology applications. Unless otherwise specified, all reagents used in the following examples are commercially available.

Example 1. Design of LRRK2 RNAi Agents

The sequences of the sense strands and antisense strands in the RNAi agents of the present disclosure are shown in Table 2 below. With the human LRRK2 gene (NM_198578.4) as a target gene, unmodified sense/antisense strands of 19/21nt were designed in accordance with the general rules for active double-stranded oligonucleotides. The sequences of the modified RNAi agents of the present disclosure are shown in Table 3.

Table 2. The sequences of unmodified RNAi agents targeting the LRRK2 gene

Double strand No.	SEQ ID NO:	Unmodified SS strand (5'-3')	SEQ ID NO:	Unmodified AS strand (5'-3')
TJR104830	1	UGAAGAAGUUGAUAGUCAA	71	UUGACUAUCAACUUCUUCAGA
TJR104725	2	GUCACUUCUGUGCAAAUUA	72	UAAUUUGCACAGAAGUGACCA
TJR104831	3	ACCAAGAAAUUCAGUGUCA	73	UGACACUGAAUUUCUUGGUCA
TJR104832	4	UCACUUCUGUGCAAAUUAA	74	UUAAUUUGCACAGAAGUGACC
TJR104833	5	GAAGAAGUUGAUAGUCAGA	75	UCUGACUAUCAACUUCUUCAG
TJR104834	6	GAAGAUUGGAGAUGAAGAA	76	UUCUUCAUCUCCAAUCUUCUC
TJR104835	7	CUUCUGUGCAAAUUAAUAA	77	UUAUUAAUUUGCACAGAAGUG
TJR104836	8	GUUGGUCACUUCUGUGCAA	78	UUGCACAGAAGUGACCAACCC

TJR104837	9	AGUUGAUAGUCAGGCUGAA	79	UUCAGCCUGACUAUCAACUUC
TJR104838	10	AGAAAUUCAGUGUCUGGGA	80	UCCCAGACACUGAAUUUCUUG
TJR104839	11	GUUAAUUUUUGAUGCCAUA	81	UAUGGCAUCAAAAAUUAACAU
TJR104840	12	CACUUCUGUGCAAAUUAAA	82	UUUAAUUUGCACAGAAGUGAC
TJR104723	13	CUGAAGAAGUUGAUAGUCA	83	UGACUAUCAACUUCUUCAGAG
TJR104724	14	GGUCACUUCUGUGCAAAUU	84	AAUUUGCACAGAAGUGACCAA
TJR104726	15	UGAAGAAGUUGAUAGUCAG	85	CUGACUAUCAACUUCUUCAGA
TJR104727	16	AGAAGAUUGGAGAUGAAGA	86	UCUUCAUCUCCAAUCUUCUCA
TJR104728	17	UGAGAAGAUUGGAGAUGAA	87	UUCAUCUCCAAUCUUCUCAUG
	18	AGAAGUUGAUAGUCAGGCU	88	AGCCUGACUAUCAACUUCUUC
	19	GAAGUUGAUAGUCAGGCUA	89	UAGCCUGACUAUCAACUUCUU
	20	CACUUCUGUGCAAAUUAAU	90	AUUAAUUUGCACAGAAGUGAC
	21	CUGUGCAAAUUAAUAGAAA	91	UUUCUAUUAAUUUGCACAGAA
	22	GAAAUGAUUGGGAAGUCCU	92	AGGACUUCCCAAUCAUUUCCA
	23	AAAUGAUUGGGAAGUCCUU	93	AAGGACUUCCCAAUCAUUUCC
	24	UGAGAAGAUUGGAGAUGAA	94	UUCAUCUCCAAUCUUCUCAUG
	25	AGAAGAUUGGAGAUGAAGA	95	UCUUCAUCUCCAAUCUUCUCA

	26	GAAGAUUGGAGAUGAAGAU	96	AUCUUCAUCUCCAAUCUUCUC
	27	AGAUGAAGAUGGCCAUUUA	97	UAAAUGGCCAUCUUCAUCUCC
	28	UGAAGAUGGCCAUUUCCCA	98	UGGGAAAUGGCCAUCUUCAUC
	29	CAGGUUCAUUGGAAAUCCU	99	AGGAUUUCCAAUGAACCUGUU
	30	GAUGUAUCCAGAUGACCAA	100	UUGGUCAUCUGGAUACAUCUG
	31	GUAUCCAGAUGACCAAGAA	101	UUCUUGGUCAUCUGGAUACAU
	32	UAUCCAGAUGACCAAGAAA	102	UUUCUUGGUCAUCUGGAUACA
	33	CCAAGAAAUUCAGUGUCUA	103	UAGACACUGAAUUUCUUGGUC
	34	CAAGAAAUUCAGUGUCUGA	104	UCAGACACUGAAUUUCUUGGU
	35	AGAAAUUCAGUGUCUGGGU	105	ACCCAGACACUGAAUUUCUUG
	36	GAAAUUCAGUGUCUGGGUU	106	AACCCAGACACUGAAUUUCUU
	37	ACUGAAAGAGAUUCCUCCU	107	AGGAGGAAUCUCUUUCAGUUU
	38	GAAAGAGAUUCCUCCUGAA	108	UUCAGGAGGAAUCUCUUUCAG
	39	CCAAAGACAUCAUAAGGUU	109	AACCUUAUGAUGUCUUUGGCU
	40	CAAAGACAUCAUAAGGUUU	110	AAACCUUAUGAUGUCUUUGGC
	41	GCAGGUCGUGAGGAAUUCU	111	AGAAUUCCUCACGACCUGCAA
	42	GAAGCCUUGGCUCUUCAAU	112	AUUGAAGAGCCAAGGCUUCAU

	43	AUUUCAAGAUCC GAGAUCA	113	UGAUCUCGGAUCUU GAAAUUA
	44	CCAAGUGGCUUU GUAAAAU	114	AUUUUACAAAGCCA CUUGGGU
	45	CAAGUGGCUUUG UAAAAUA	115	UAUUUUACAAAGCC ACUUGGG
	46	GUGGCUUUGUAA AAUCAUA	116	UAUGAUUUUACAAA GCCACUU
	47	UGACCACAGGCC UGUGAUA	117	UAUCACAGGCCUGU GGUCAGA
	48	GAAGUGGCUGUG AAGAUUU	118	AAAUCUUCACAGCC ACUUCUU
	49	GUGGCUGUGAAG AUUUUUA	119	UAAAAAUCUUCACA GCCACUU
	50	GGCUGUGAAGAU UUUUAAU	120	AUUAAAAAUCUUCA CAGCCAC
	51	CAGGACAAAGCC AGCCUCA	121	UGAGGCUGGCUUUG UCCUGCU
	52	GGACAAAGCCAG CCUCACU	122	AGUGAGGCUGGCUU UGUCCUG
	53	GACAAAGCCAGC CUCACUA	123	UAGUGAGGCUGGCU UUGUCCU
	54	GUUUGAAAGAAA AUCCUCA	124	UGAGGAUUUUCUUU CAAACAC
	55	CUGCCCAGGUCU UUGACAU	125	AUGUCAAAGACCUG GGCAGAA
	56	UGCCCAGGUCUU UGACAUU	126	AAUGUCAAAGACCU GGGCAGA
	57	GCCCAGGUCUUU GACAUUU	127	AAAUGUCAAAGACC UGGGCAG
	58	AGAAAAACACAU UGAAGUA	128	UACUUCAAUGUGUU UUUCUAA
	59	GAAAAACACAUU GAAGUGA	129	UCACUUCAAUGUGU UUUUCUA

	60	AAAACACAUUGAAGUGAGA	130	UCUCACUUCAAUGUGUUUUUC
	61	ACUUCUGUGCAAAUUAAUA	131	UAUUAAUUUGCACAGAAGUGA
	62	UUCUGUGCAAAUUAAUAGA	132	UCUAUUAAUUUGCACAGAAGU
	63	UCUGUGCAAAUUAAUAGAA	133	UUCUAUUAAUUUGCACAGAAG
	64	ACCAAGAAAUUCAGUGUCU	134	AGACACUGAAUUUCUUGGUCA
	65	UAAACUGAAAGAGAUUCCU	135	AGGAAUCUCUUUCAGUUUAUU
	66	AAACUGAAAGAGAUUCCUA	136	UAGGAAUCUCUUUCAGUUUAU
	67	AAAGACAUCAUAAGGUUUA	137	UAAACCUUAUGAUGUCUUUGG
	68	UGGCUGUGAAGAUUUUUAA	138	UUAAAAAUCUUCACAGCCACU
	69	CUGUGAAGAUUUUUAAUAA	139	UUAUUAAAAAUCUUCACAGCC
	70	UGUGAAGAUUUUUAAUAAA	140	UUUAUUAAAAAUCUUCACAGC

Table 3. The sequences of modified RNAi agents targeting the LRRK2 gene

Double strand No.	SEQ ID NO:	SS strand (5'-3')	SEQ ID NO:	AS strand (5'-3')
TJR104711	141	UmsGmsAmAmGmAmAfGfUfUmGmAmUmAmGmUmCmsAmsAm	214	UmsUfsGmAfCmUfAmUmCmAfAmCfUmUfCmUfUmCfAmsGmsAm
TJR104712	142	GmsUmsCmAmCmUmUfCfUfGmUmGmCmAmAmAmUmsUmsAm	215	UmsAfsAmUfUmUfGmCmAmCfAmGfAmAfGmUfGmAfCmsCmsAm
TJR104713	143	AmsCmsCmAmAmGmAfAfAfUmUmCmAmGmUm	216	UmsGfsAmCfAmCfUmGmAmAfUmUfUmCfUmUfGmGf

		GmUmsCmsAm		UmsCmsAm
TJR104714	144	UmsCmsAmCmUmUmCfUfGfUmGmCmAmAmAmUmUmsAmsAm	217	UmsUfsAmAfUmUfUmGmCmAfCmAfGmAfAmGfUmGfAmsCmsCm
TJR104715	145	GmsAmsAmGmAmAmGfUfUfGmAmUmAmGmUmCmAmsGmsAm	218	UmsCfsUmGfAmCfUmAmUmCfAmAfCmUfUmCfUmUfCmsAmsGm
TJR104716	146	GmsAmsAmGmAmUmUfGfGfAmGmAmUmGmAmAmGmsAmsAm	219	UmsUfsCmUfUmCfAmUmCmUfCmCfAmAfUmCfUmUfCmsUmsCm
TJR104717	147	CmsUmsUmCmUmGmUfGfCfAmAmAmUmUmAmAmUmsAmsAm	220	UmsUfsAmUfUmAfAmUmUmUfGmCfAmCfAmGfAmAfGmsUmsGm
TJR104718	148	GmsUmsUmGmGmUmCfAfCfUmUmCmUmGmUmGmCmsAmsAm	221	UmsUfsGmCfAmCfAmGmAmAfGmUfGmAfCmCfAmAfCmsCmsCm
TJR104719	149	AmsGmsUmUmGmAmUfAfGfUmCmAmGmGmCmUmGmsAmsAm	222	UmsUfsCmAfGmCfCmUmGmAfCmUfAmUfCmAfAmCfUmsUmsCm
TJR104720	150	AmsGmsAmAmAmUmUfCfAfGmUmGmUmCmUmGmGmsGmsAm	223	UmsCfsCmCfAmGfAmCmAmCfUmGfAmAfUmUfUmCfUmsUmsGm
TJR104721	151	GmsUmsUmAmAmUmUfUfUfUmGmAmUmGmCmCmAmsUmsAm	224	UmsAfsUmGfGmCfAmUmCmAfAmAfAmAfUmUfAmAfCmsAmsUm
TJR104722	152	CmsAmsCmUmUmCmUfGfUfGmCmAmAmAmUmUmAmsAmsAm	225	UmsUfsUmAfAmUfUmUmGmCfAmCfAmGfAmAfGmUfGmsAmsCm
TJR101840	153	UmsGmsAmAmGmAmAfGfUfUmGmAmUmAmGmUmCmsAmsAm	226	UmsUfsGmAmCmUfAmUmCmAmAmCfUmUfCmUfUmCmAmsGmsAm
TJR101841	154	GmsAmsAmGmAmAmGfUfUfGmAmUmAmGmUmCmAmsGmsAm	227	UmsCfsUmGmAmCfUmAmUmCmAmAfCmUfUmCfUmUmCmsAmsGm
TJR10	155	AmsGmsUmUmGmAmUf	228	UmsUfsCmAmGmCfCmUm

1844		AfGfUmCmAmGmGmCmUmGmsAmsAm		GmAmCmUfAmUfCmAfAmCmUmsUmsCm
TJR101845	156	GmsUmsUmGmGmUmCfAfCfUmUmCmUmGmUmGmCmsAmsAm	229	UmsUfsGmCmAmCfAmGmAmAmGmUfGmAfCmCfAmAmCmsCmsCm
TJR101846	157	GmsUmsCmAmCmUmUfCfUfGmUmGmCmAmAmAmUmsUmsAm	230	UmsAfsAmUmUmUfGmCmAmCmAmGfAmAfGmUfGmAmCmsCmsAm
TJR101847	158	UmsCmsAmCmUmUmCfUfGfUmGmCmAmAmAmUmUmsAmsAm	231	UmsUfsAmAmUmUfUmGmCmAmCmAfGmAfAmGfUmGmAmsCmsCm
TJR101848	159	CmsAmsCmUmUmCmUfGfUfGmCmAmAmAmUmUmAmsAmsUm	232	AmsUfsUmAmAmUfUmUmGmCmAmCfAmGfAmAfGmUmGmsAmsCm
TJR101849	160	CmsUmsUmCmUmGmUfGfCfAmAmAmUmUmAmAmUmsAmsAm	233	UmsUfsAmUmUmAfAmUmUmUmGmCfAmCfAmGfAmAmGmsUmsGm
TJR101853	161	GmsUmsUmAmAmUmUfUfUfUmGmAmUmGmCmCmAmsUmsAm	234	UmsAfsUmGmGmCfAmUmCmAmAmAfAmAfUmUfAmAmCmsAmsUm
TJR101856	162	GmsAmsAmGmAmUmUfGfGfAmGmAmUmGmAmAmGmsAmsUm	235	AmsUfsCmUmUmCfAmUmCmUmCmCfAmAfUmCfUmUmCmsUmsCm
TJR102519	163	AmsCmsCmAmAmGmAfAfAfUmUmCmAmGmUmGmUmsCmsUm	236	AmsGfsAmCfAmCfUmGmAmAfUmUfUmCfUmUfGmGfUmsCmsAm
TJR101865	164	AmsGmsAmAmAmUmUfCfAfGmUmGmUmCmUmGmGmsGmsUm	237	AmsCfsCmCmAmGfAmCmAmCmUmGfAmAfUmUfUmCmUmsUmsGm
TJR101842	165	AmsGmsAmAmGmUmUfGfAfUmAmGmUmCmAmGmGmsCmsUm	238	AmsGfsCmCmUmGfAmCmUmAmUmCfAmAfCmUfUmCmUmsUmsCm
TJR101843	166	GmsAmsAmGmUmUmGfAfUfAmGmUmCmAmGmGmCmsUmsAm	239	UmsAfsGmCmCmUfGmAmCmUmAmUfCmAfAmCfUmUmCmsUmsUm

TJR101850	167	CmsUmsGmUmGmCmAfAfAfUmUmAmAmUmAmGmAmsAmsAm	240	UmsUfsUmCmUmAfUmUmAmAmUmUfUmGfCmAfCmAmGmsAmsAm
TJR101851	168	GmsAmsAmAmUmGmAfUfUfGmGmGmAmAmGmUmCmsCmsUm	241	AmsGfsGmAmCmUfUmCmCmCmAmAfUmCfAmUfUmUmCmsCmsAm
TJR101852	169	AmsAmsAmUmGmAmUfUfGfGmGmAmAmGmUmCmCmsUmsUm	242	AmsAfsGmGmAmCfUmUmCmCmCmAfAmUfCmAfUmUmUmsCmsCm
TJR101854	170	UmsGmsAmGmAmAmGfAfUfUmGmGmAmGmAmUmGmsAmsAm	243	UmsUfsCmAmUmCfUmCmCmAmAmUfCmUfUmCfUmCmAmsUmsGm
TJR101855	171	AmsGmsAmAmGmAmUfUfGfGmAmGmAmUmGmAmAmsGmsAm	244	UmsCfsUmUmCmAfUmCmUmCmCmAfAmUfCmUfUmCmUmsCmsAm
TJR101857	172	AmsGmsAmUmGmAmAfGfAfUmGmGmCmCmAmUmUmsUmsAm	245	UmsAfsAmAmUmGfGmCmCmAmUmCfUmUfCmAfUmCmUmsCmsCm
TJR101858	173	UmsGmsAmAmGmAmUfGfGfCmCmAmUmUmUmCmCmsCmsAm	246	UmsGfsGmGmAmAfAmUmGmGmCmCfAmUfCmUfUmCmAmsUmsCm
TJR101859	174	CmsAmsGmGmUmUmCfAfUfUmGmGmAmAmAmUmCmsCmsUm	247	AmsGfsGmAmUmUfUmCmCmAmAmUfGmAfAmCfCmUmGmsUmsUm
TJR101860	175	GmsAmsUmGmUmAmUfCfCfAmGmAmUmGmAmCmCmsAmsAm	248	UmsUfsGmGmUmCfAmUmCmUmGmGfAmUfAmCfAmUmCmsUmsGm
TJR101861	176	GmsUmsAmUmCmCmAfGfAfUmGmAmCmCmAmAmGmsAmsAm	249	UmsUfsCmUmUmGfGmUmCmAmUmCfUmGfGmAfUmAmCmsAmsUm
TJR101862	177	UmsAmsUmCmCmAmGfAfUfGmAmCmCmAmAmGmAmsAmsAm	250	UmsUfsUmCmUmUfGmGmUmCmAmUfCmUfGmGfAmUmAmsCmsAm
TJR101863	178	CmsCmsAmAmGmAmAfAfUfUmCmAmGmUmGmUmCmsUmsAm	251	UmsAfsGmAmCmAfCmUmGmAmAmUfUmUfCmUfUmGmGmsUmsCm

TJR101864	179	CmsAmsAmGmAmAmAfUfUfCmAmGmUmGmUmCmUmsGmsAm	252	UmsCfsAmGmAmCfAmCmUmGmAmAfUmUfUmCfUmUmGmsGmsUm
TJR101866	180	GmsAmsAmAmUmUmCfAfGfUmGmUmCmUmGmGmGmsUmsUm	253	AmsAfsCmCmCmAfGmAmCmAmCmUfGmAfAmUfUmUmCmsUmsUm
TJR101867	181	AmsCmsUmGmAmAmAfGfAfGmAmUmUmCmCmUmCmsCmsUm	254	AmsGfsGmAmGmGfAmAmUmCmUmCfUmUfUmCfAmGmUmsUmsUm
TJR101868	182	GmsAmsAmAmGmAmGfAfUfUmCmCmUmCmCmUmGmsAmsAm	255	UmsUfsCmAmGmGfAmGmGmAmAmUfCmUfCmUfUmUmCmsAmsGm
TJR101869	183	CmsCmsAmAmAmGmAfCfAfUmCmAmUmAmAmGmGmsUmsUm	256	AmsAfsCmCmUmUfAmUmGmAmUmGfUmCfUmUfUmGmGmsCmsUm
TJR101870	184	CmsAmsAmAmGmAmCfAfUfCmAmUmAmAmGmGmGmUmsUmsUm	257	AmsAfsAmCmCmUfUmAmUmGmAmUfGmUfCmUfUmUmGmsGmsCm
TJR101871	185	GmsCmsAmGmGmUmCfGfUfGmAmGmGmAmAmUmUmsCmsUm	258	AmsGfsAmAmUmUfCmCmUmCmAmCfGmAfCmCfUmGmCmsAmsAm
TJR101872	186	GmsAmsAmGmCmCmUfUfGfGmCmUmCmUmUmCmAmsAmsUm	259	AmsUfsUmGmAmAfGmAmGmCmCmAfAmGfGmCfUmUmCmsAmsUm
TJR101873	187	AmsUmsUmUmCmAmAfGfAfUmCmCmGmAmGmAmUmsCmsAm	260	UmsGfsAmUmCmUfCmGmGmAmUmCfUmUfGmAfAmAmUmsUmsAm
TJR101874	188	CmsCmsAmAmGmUmGfGfCfUmUmUmGmUmAmAmAmsAmsUm	261	AmsUfsUmUmUmAfCmAmAmAmGmCfCmAfCmUfUmGmGmsGmsUm
TJR101875	189	CmsAmsAmGmUmGmGfCfUfUmUmGmUmAmAmAmAmsUmsAm	262	UmsAfsUmUmUmUfAmCmAmAmAmGfCmCfAmCfUmUmGmsGmsGm
TJR101876	190	GmsUmsGmGmCmUmUfUfGfUmAmAmAmAmUmCmAmsUmsAm	263	UmsAfsUmGmAmUfUmUmUmAmCmAfAmAfGmCfCmAmCmsUmsUm

TJR101877	191	UmsGmsAmCmCmAmCfAfGfGmCmCmUmGmUmGmAmsUmsAm	264	UmsAfsUmCmAmCfAmGmGmCmCmUfGmUfGmGfUmCmAmsGmsAm
TJR101878	192	GmsAmsAmGmUmGmGfCfUfGmUmGmAmAmGmAmUmsUmsUm	265	AmsAfsAmUmCmUfUmCmAmCmAmGfCmCfAmCfUmUmCmsUmsUm
TJR101879	193	GmsUmsGmGmCmUmGfUfGfAmAmGmAmUmUmUmUmsUmsAm	266	UmsAfsAmAmAmAfUmCmUmUmCmAfCmAfGmCfCmAmCmsUmsUm
TJR101880	194	GmsGmsCmUmGmUmGfAfAfGmAmUmUmUmUmUmAmsAmsUm	267	AmsUfsUmAmAmAfAmAmUmCmUmUfCmAfCmAfGmCmCmsAmsCm
TJR101881	195	CmsAmsGmGmAmCmAfAfAfGmCmCmAmGmCmCmUmsCmsAm	268	UmsGfsAmGmGmCfUmGmGmCmUmUfUmGfUmCfCmUmGmsCmsUm
TJR101882	196	GmsGmsAmCmAmAmAfGfCfCmAmGmCmCmUmCmAmsCmsUm	269	AmsGfsUmGmAmGfGmCmUmGmGmCfUmUfUmGfUmCmCmsUmsGm
TJR101883	197	GmsAmsCmAmAmAmGfCfCfAmGmCmCmUmCmAmCmsUmsAm	270	UmsAfsGmUmGmAfGmGmCmUmGmGfCmUfUmUfGmUmCmsCmsUm
TJR101884	198	GmsUmsUmUmGmAmAfAfGfAmAmAmAmUmCmCmUmsCmsAm	271	UmsGfsAmGmGmAfUmUmUmUmCmUfUmUfCmAfAmAmCmsAmsCm
TJR101885	199	CmsUmsGmCmCmCmAfGfGfUmCmUmUmUmGmAmCmsAmsUm	272	AmsUfsGmUmCmAfAmAmGmAmCmCfUmGfGmGfCmAmGmsAmsAm
TJR101886	200	UmsGmsCmCmCmAmGfGfUfCmUmUmUmGmAmCmAmsUmsUm	273	AmsAfsUmGmUmCfAmAmAmGmAmCfCmUfGmGfGmCmAmsGmsAm
TJR101887	201	GmsCmsCmCmAmGmGfUfCfUmUmUmGmAmCmAmUmsUmsUm	274	AmsAfsAmUmGmUfCmAmAmAmGmAfCmCfUmGfGmGmCmsAmsGm
TJR101888	202	AmsGmsAmAmAmAmAfCfAfCmAmUmUmGmAmAmGmsUmsAm	275	UmsAfsCmUmUmCfAmAmUmGmUmGfUmUfUmUfUmCmUmsAmsAm

TJR101889	203	GmsAmsAmAmAmAmCfAfCfAmUmUmGmAmAmGmUmsGmsAm	276	UmsCfsAmCmUmUfCmAmAmUmGmUfGmUfUmUfUmUmCmsUmsAm
TJR101890	204	AmsAmsAmAmCmAmCfAfUfUmGmAmAmGmUmGmAmsGmsAm	277	UmsCfsUmCmAmCfUmUmCmAmAmUfGmUfGmUfUmUmUmsUmsCm
TJR102516	205	AmsCmsUmUmCmUmGfUfGfCmAmAmAmUmUmAmAmsUmsAm	278	UmsAfsUmUfAmAfUmUmUmGfCmAfCmAfGmAfAmGfUmsGmsAm
TJR102517	206	UmsUmsCmUmGmUmGfCfAfAmAmUmUmAmAmUmAmsGmsAm	279	UmsCfsUmAfUmUfAmAmUmUfUmGfCmAfCmAfGmAfAmsGmsUm
TJR102518	207	UmsCmsUmGmUmGmCfAfAfAmUmUmAmAmUmAmGmsAmsAm	280	UmsUfsCmUfAmUfUmAmAmUfUmUfGmCfAmCfAmGfAmsAmsGm
TJR102520	208	UmsAmsAmAmCmUmGfAfAfAmGmAmGmAmUmUmCmsCmsUm	281	AmsGfsGmAfAmUfCmUmCmUfUmUfCmAfGmUfUmUfAmsUmsUm
TJR102521	209	AmsAmsAmCmUmGmAfAfAfGmAmGmAmUmUmCmCmsUmsAm	282	UmsAfsGmGfAmAfUmCmUmCfUmUfUmCfAmGfUmUfUmsAmsUm
TJR102522	210	AmsAmsAmGmAmCmAfUfCfAmUmAmAmGmGmUmUmsUmsAm	283	UmsAfsAmAfCmCfUmUmAmUfGmAfUmGfUmCfUmUfUmsGmsGm
TJR102523	211	UmsGmsGmCmUmGmUfGfAfAmGmAmUmUmUmUmUmsAmsAm	284	UmsUfsAmAfAmAfAmUmCmUfUmCfAmCfAmGfCmCfAmsCmsUm
TJR102524	212	CmsUmsGmUmGmAmAfGfAfUmUmUmUmUmAmAmUmsAmsAm	285	UmsUfsAmUfUmAfAmAmAmAfUmCfUmUfCmAfCmAfGmsCmsCm
TJR102525	213	UmsGmsUmGmAmAmGfAfUfUmUmUmUmAmAmUmAmsAmsAm	286	UmsUfsUmAfUmUfAmAmAmAfAmUfCmUfUmCfAmCfAmsGmsCm
TJR104841	287	AmsCmsCmAmAmGmAfAfAfUmUmCmAmGmUmGmUmsCmsAm	291	UmsGfsAmCmAmCfUmGmAmAmUmUfUmCfUmUfGmGmUmsCmsAm

TJR104842	288	GmsAmsAmGmAmUmUfGfGfAmGmAmUmGmAmAmGmsAmsAm	292	UmsUfsCmUmUmCfAmUmCmUmCmCfAmAfUmCfUmUmCmsUmsCm
TJR104843	289	AmsGmsAmAmAmUmUfCfAfGmUmGmUmCmUmGmGmsGmsAm	293	UmsCfsCmCmAmGfAmCmAmCmUmGfAmAfUmUfUmCmUmsUmsGm
TJR104844	290	CmsAmsCmUmUmCmUfGfUfGmCmAmAmAmUmUmAmsAmsAm	294	UmsUfsUmAmAmUfUmUmGmCmAmCfAmGfAmAfGmUmGmsAmsCm

In Table 2 and Table 3, in the same nucleic acid sequence, in the direction from the 5' end to the 3' end (from left to right), G, C, A, and U represent nucleosides having guanine, cytosine, adenine, and uracil, respectively, and the sequences are shown in the direction from the 5' end to the 3' end (from left to right); the lowercase letter m means that the nucleoside adjacent to the letter m on the left side is a 2'-methoxy-modified nucleoside; the lowercase letter f means that the nucleoside adjacent to the letter f on the left side is a 2'-fluoro-modified nucleoside; the lowercase letter s means that the two nucleosides adjacent to the letter s are linked by a phosphorothioate diester group. Unless otherwise specified, two nucleosides are linked by a phosphodiester group. Unless otherwise specified, the 3' position of the first nucleotide of the 3' end of each strand is hydroxy; the 5' position of the first nucleotide of the 5' end of each strand is hydroxy.

The structures of the 2'-methoxy-modified nucleoside, the 2'-fluoro-modified nucleoside, the phosphorothioate diester group, and the phosphodiester group are shown in Table 1. When the RNAi agent of the present disclosure is present in the form of a salt, for example, in the form of a sodium salt, the structures of salt forms corresponding to the structures in Table 1 also fall within the protection scope of the present disclosure.

Example 2. Synthesis of RNAi Agents of Present Disclosure

The synthesis of RNAi agents of the present disclosure does not differ from the conventional phosphoramidite solid-phase synthesis method. The synthesis process is briefly described below: Unmodified nucleoside phosphoramidite monomers or nucleoside phosphoramidite monomers having modifications at corresponding positions in the sequence were linked one by one according to a synthesis program on a Dr. Oligo48 synthesizer (Biolytic), with a universal CPG support as a start. The nucleoside phosphoramidite monomers used in the examples of the present disclosure were purchased from Shanghai Hongene and Suzhou Genepharma. 5-Ethylthio-1H-tetrazole (ETT) was used as an activator (0.6 M in acetonitrile), a 0.22 M solution of

PADS in acetonitrile and 3-methylpyridine (1:1 by volume) (Shanghai Lingjiang) was used as a sulfurizing agent, and an aqueous iodopyridine solution (Shanghai Lingjiang) was used as an oxidant.

After solid-phase synthesis was completed, the oligoribonucleotides were cleaved from the solid support by soaking in a solution of 28% ammonia water and ethanol (3:1) at 50 °C for 16 h. Centrifugation was then performed, and the supernatant was transferred to another centrifuge tube. After the supernatant was concentrated to dryness by evaporation, the residue was purified by C18 reversed-phase chromatography with 0.1 M TEAA and acetonitrile as the mobile phase. The target oligonucleotides were collected, then lyophilized, identified as the target products by LC-MS, and quantified by UV (260 nm).

The single-stranded oligonucleotides obtained were complementarily paired in an equimolar ratio and annealed. The final RNAi agents of the present disclosure were dissolved in 1× PBS, and the concentration of the solutions was adjusted to the concentration required for the experiment so that they were ready for use.

Example 3. Activity of RNAi Agents in Endogenous Cells at Single Concentration Point

RNAi agents were subjected to a molecular-level simulation of endogenous cell activity screening using a single concentration.

A549 cells were cultured in an RPMI1640 culture medium (Gibco) (containing 10% FBS) at 37 °C with 5% CO_2. 24 h prior to transfection, the A549 cells were seeded into a 24-well plate at a density of 5×10^4 cells per well, with each well containing 500 μL of the culture medium.

The cells were transfected with the RNAi agents using an RNAi MAX transfection reagent (ThermoFisher, 13778150) according to the instructions. A single concentration point of 20 nM was set for the RNAi agents. After 48 h of transfection, total cell RNA extraction was performed using a high-throughput cell RNA extraction kit (ThermoFisher, A27828), and RNA reverse transcription (Takara, RR037B) and quantitative real-time PCR detection (ThermoFisher, 4444557) were performed to determine the mRNA level of human LRRK2. The mRNA level of human LRRK2 was corrected based on the GAPDH internal reference gene level.

Results analysis method:

After the Q-PCR detection experiment was completed, the corresponding Ct values were obtained based on the threshold automatically set by the system. The expression of a certain gene can be relatively quantified by comparing the Ct values. Comparing Ct refers to calculating the differences in gene expression through the differences from the Ct value of the internal reference gene, which is also referred to as $2^{-\triangle\triangle Ct}$, where $\triangle\triangle Ct$ = [(Ct target gene in experimental group - Ct internal reference in experimental group) - (Ct target gene in control group - Ct internal reference in control group)].

Inhibition rate (%) = (1 - remaining level of target gene expression) × 100%.

The results are expressed relative to the remaining percentage of human LRRK2 expression in cells treated with the RNAi agents. The inhibition rate results are shown in Table 4.

Table 4. The single-point activity results of the RNAi agents of the present disclosure in A549 cells

Compound No.	Remaining percentage of target gene mRNA expression (mean)	
	20 nM	STDEV (%)
TJR101840	13.83	0.25
TJR101841	13.86	1.49
TJR101842	17.13	0.08
TJR101843	21.04	0.22
TJR101844	18.93	3.93
TJR101845	10.56	1.36
TJR101846	14.74	1.21
TJR101847	12.92	0.02
TJR101848	15.46	0.07
TJR101849	13.40	0.85
TJR101850	19.44	2.43
TJR101851	40.71	2.07
TJR101852	20.58	3.38
TJR101853	18.70	2.02
TJR101854	13.28	3.02
TJR101855	12.10	0.81
TJR101856	12.72	0.19
TJR101857	26.17	3.92
TJR101858	27.64	0.11
TJR101859	28.95	2.92
TJR101860	31.62	1.58
TJR101861	28.14	7.81
TJR101862	14.15	0.52

TJR101863	18.10	1.61
TJR101864	18.48	0.43
TJR101865	12.41	1.65
TJR101866	19.16	0.18
TJR101867	30.62	0.74
TJR101868	32.84	4.46
TJR101869	39.62	1.99
TJR101870	24.82	1.27
TJR101871	28.72	2.43
TJR101872	35.01	2.03
TJR101873	36.15	0.27
TJR101874	17.07	1.28
TJR101875	35.11	2.78
TJR101876	39.85	3.89
TJR101877	44.09	5.21
TJR101879	42.14	7.99
TJR101880	44.17	1.65
TJR101886	48.60	2.26
TJR101890	34.66	4.13
TJR102516	22.87	1.79
TJR102517	23.29	4.08
TJR102518	21.24	1.72
TJR102519	17.91	0.32
TJR102520	42.65	1.39
TJR102521	45.46	4.80
TJR102522	45.48	3.07
TJR102523	39.07	3.69
TJR102524	46.65	4.97
TJR102525	32.65	6.32

Example 4. psiCHECK On-Target Activity of RNAi Agents in Human Embryonic Kidney Cells (HEK293A)

RNAi agents were subjected to an *in vitro* molecular-level simulation of on-target activity screening in HEK293A cells using 9 concentration gradients.

HEK293A cells were cultured at 37 °C with 5% CO_2 in a DMEM high-glucose culture medium containing 10% fetal bovine serum. 24 h prior to transfection, the HEK293A cells were seeded into a 96-well plate at a density of 1×10^4 cells per well, with each well containing 100 μL of the culture medium.

The cells were co-transfected with the RNAi agents and corresponding plasmids using Lipofectamine2000 (ThermoFisher, 11668019) according to the instructions, with 0.3 μL of Lipofectamine2000 used per well. The amount of plasmids for transfection was 20 ng per well. For on-target sequence plasmids, a total of 9 concentration points were set for the RNAi agents; the highest concentration point final concentration was 10 nM, and a 5-fold serial dilution was performed; the lowest concentration point final concentration was 0.0000256 nM. 24 h after transfection, the on-target levels were determined using a Dual-Luciferase Reporter Assay System (Promega, E2940). The results are shown in Table 5. The results show that the RNAi agents of the present disclosure exhibited good target inhibitory activity.

Table 5. psiCHECK on-target activity of the RNAi agents of the present disclosure in HEK293A cells

Double strand No.	IC50 (nM)
TJR104830	0.0035
TJR104725	0.0069
TJR104831	0.0139
TJR104832	0.0067
TJR104833	0.0041
TJR104834	0.1218
TJR104835	0.0158
TJR104836	0.0062
TJR104837	0.0042
TJR104838	0.0322
TJR104840	0.0210
TJR104711	0.0044
TJR104712	0.0035
TJR104713	0.0150

TJR104714	0.0046
TJR104715	0.0055
TJR104716	0.0850
TJR104717	0.0031
TJR104718	0.0171
TJR104719	0.0148
TJR104720	0.0396
TJR104721	0.0222
TJR104722	0.0224
TJR104841	0.025
TJR104842	0.144
TJR104843	0.065
TJR104844	0.021

Example 5. Endogenous Cell Activity of RNAi Agents

RNAi agents were subjected to an *in vitro* molecular-level simulation of endogenous cell activity screening in A549 cells using 7 concentration gradients. The dsRNAs were subjected to a 5-fold serial dilution from an initial concentration of 20 nM to form 7 concentration points (20 nM, 4 nM, 0.8 nM, 0.16 nM, 0.032 nM, 0.0064 nM, and 0.00128 nM) using an RNAi MAX transfection reagent (ThermoFisher, 13778150). A549 cells were cultured in an RPMI1640 culture medium (Gibco) containing 10% FBS at 37 °C with 5% CO_2. After an RNAi agent was added to a 96-well plate, the A549 cells were seeded at a density of 1.4×10^4 cells/well (with each well containing 90 μL of the culture medium). After 24 h of transfection, total RNA extraction was performed using a high-throughput cell RNA extraction kit (GeneOn BioTech, MNTR/FX96), and RNA reverse transcription (Takara, RR037B) and quantitative real-time PCR detection (ThermoFisher, 4444557) were performed to determine the mRNA level of human LRRK2. The expression level was corrected using GAPDH as an internal reference gene.

Reference was made to Example 3 for the analysis of results. The dsRNA sequences were subjected to *in vitro* molecular-level activity screening in A549 cells using 7 concentration gradients. The results are expressed relative to the remaining percentage of human LRRK2 expression in cells treated with the dsRNAs. The inhibition rate results are shown in Table 6.

Table 6. The endogenous activity of the RNAi agents of the present disclosure in A549 cells

Double strand No.	IC_{50} (nM)
TJR104725	0.0374
TJR104832	0.0239
TJR104724	0.1825
TJR104834	0.0940
TJR104728	>20

CLAIMS

1. An RNAi agent, comprising a sense strand and an antisense strand that form a double-stranded region, wherein

the sense strand comprises at least 15 contiguous nucleotides differing from the nucleotide sequence set forth in any one of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64 by no more than 3 nucleotides;

the antisense strand comprises at least 15 contiguous nucleotides differing from the nucleotide sequence set forth in any one of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134 by no more than 3 nucleotides.

2. The RNAi agent according to claim 1, wherein:

the sense strand comprises at least 17 contiguous nucleotides differing from any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64 by no more than 3 nucleotides;

the antisense strand comprises at least 17 contiguous nucleotides differing from the nucleotide sequence set forth in any one of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134 by no more than 3 nucleotides;

preferably, the sense strand comprises at least 18 contiguous nucleotides differing from any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64 by no more than 3 nucleotides, preferably by no more than one nucleotide; and/or

preferably, the antisense strand comprises at least 20 contiguous nucleotides differing from any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO: 96, SEQ ID NO: 105, and SEQ ID NO: 134 by no more than 3 nucleotides, preferably by no more than one nucleotide;

more preferably, the sense strand comprises at least 19 contiguous nucleotides differing from any one of the nucleotide sequences of SEQ ID NO: 4, SEQ ID NO: 1 to SEQ ID NO: 3, SEQ ID NO: 5 to SEQ ID NO: 12, SEQ ID NO: 20, SEQ ID NO: 26, SEQ ID NO: 35, and SEQ ID NO: 64 by no more than 3 nucleotides, preferably by no more than one nucleotide; and/or

more preferably, the antisense strand comprises at least 21 contiguous nucleotides differing from any one of the nucleotide sequences of SEQ ID NO: 74, SEQ ID NO: 71 to SEQ ID NO: 73, SEQ ID NO: 75 to SEQ ID NO: 82, SEQ ID NO: 90, SEQ ID NO:

96, SEQ ID NO: 105, and SEQ ID NO: 134 by no more than 3 nucleotides, preferably by no more than one nucleotide.

3. The RNAi agent according to claim 1 or 2, comprising the sense strand and the antisense strand shown in any one of the following groups:
group 1): a sense strand set forth in SEQ ID NO: 4 and an antisense strand set forth in SEQ ID NO: 74;
group 2): a sense strand set forth in SEQ ID NO: 2 and an antisense strand set forth in SEQ ID NO: 72;
group 3): a sense strand set forth in SEQ ID NO: 3 and an antisense strand set forth in SEQ ID NO: 73, or a sense strand set forth in SEQ ID NO: 64 and an antisense strand set forth in SEQ ID NO: 134;
group 4): a sense strand set forth in SEQ ID NO: 1 and an antisense strand set forth in SEQ ID NO: 71;
group 5): a sense strand set forth in SEQ ID NO: 5 and an antisense strand set forth in SEQ ID NO: 75;
group 6): a sense strand set forth in SEQ ID NO: 6 and an antisense strand set forth in SEQ ID NO: 76, or a sense strand set forth in SEQ ID NO: 26 and an antisense strand set forth in SEQ ID NO: 96;
group 7): a sense strand set forth in SEQ ID NO: 7 and an antisense strand set forth in SEQ ID NO: 77;
group 8): a sense strand set forth in SEQ ID NO: 8 and an antisense strand set forth in SEQ ID NO: 78;
group 9): a sense strand set forth in SEQ ID NO: 9 and an antisense strand set forth in SEQ ID NO: 79;
group 10): a sense strand set forth in SEQ ID NO: 10 and an antisense strand set forth in SEQ ID NO: 80, or a sense strand set forth in SEQ ID NO: 35 and an antisense strand set forth in SEQ ID NO: 105;
group 11): a sense strand set forth in SEQ ID NO: 11 and an antisense strand set forth in SEQ ID NO: 81; and
group 12): a sense strand set forth in SEQ ID NO: 12 and an antisense strand set forth in SEQ ID NO: 82, or a sense strand set forth in SEQ ID NO: 20 and an antisense strand set forth in SEQ ID NO: 90.

4. The RNAi agent according to any one of claims 1-3, wherein at least one nucleotide in the sense strand and/or the antisense strand is a modified nucleotide.

5. The RNAi agent according to claim 4, wherein:
three contiguous nucleotides in the sense strand are 2'-fluoro-modified nucleotides, preferably, in the direction from the 5' end to the 3' end, each of the nucleotides at

positions 7, 8, and 9 of the sense strand is independently a 2'-fluoro-modified nucleotide; and/or

in the direction from the 5' end to the 3' end, each of the nucleotides at positions 2, 6, 12, 14, and 16 of the antisense strand is independently a 2'-fluoro-modified nucleotide, or the nucleotide at position 2, 4, 6, 10, 12, 14, 16, or 18 of the antisense strand is independently a 2'-fluoro-modified nucleotide;

the nucleotides at the remaining positions of the sense strand and the antisense strand are 2'-methoxy-modified nucleotides.

6. The RNAi agent according to any one of claims 1-5, wherein at least one phosphodiester group in the sense strand and/or the antisense strand is a phosphodiester group with a modification group, preferably a phosphorothioate diester group.

7. The RNAi agent according to claim 6, wherein the phosphodiester group with a modification group is present in at least one of the following positions:

between the 1st and 2nd nucleotides of the 5' end of the sense strand;

between the 2nd and 3rd nucleotides of the 5' end of the sense strand;

between the 1st and 2nd nucleotides of the 3' end of the sense strand;

between the 2nd and 3rd nucleotides of the 3' end of the sense strand;

between the 1st and 2nd nucleotides of the 5' end of the antisense strand;

between the 2nd and 3rd nucleotides of the 5' end of the antisense strand;

between the 1st and 2nd nucleotides of the 3' end of the antisense strand; and

between the 2nd and 3rd nucleotides of the 3' end of the antisense strand;

preferably, the sense strand and/or the antisense strand comprise(s) a plurality of phosphorothioate diester groups, and the phosphorothioate diester groups are present:

between the 1st and 2nd nucleotides of the 5' end of the sense strand; and

between the 2nd and 3rd nucleotides of the 5' end of the sense strand; and

between the 1st and 2nd nucleotides of the 3' end of the sense strand; and

between the 2nd and 3rd nucleotides of the 3' end of the sense strand; and

between the 1st and 2nd nucleotides of the 5' end of the antisense strand; and

between the 2nd and 3rd nucleotides of the 5' end of the antisense strand; and

between the 1st and 2nd nucleotides of the 3' end of the antisense strand; and

between the 2nd and 3rd nucleotides of the 3' end of the antisense strand.

8. The RNAi agent according to any one of claims 1-7, wherein:

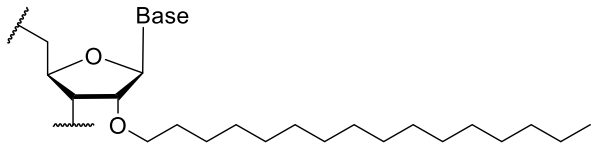
the sense strand comprises the nucleotide sequence set forth in any one of SEQ ID NO: 141 to SEQ ID NO: 164 and SEQ ID NO: 287 to SEQ ID NO: 290; and/or

the antisense strand comprises the nucleotide sequence set forth in any one of SEQ ID NO: 214 to SEQ ID NO: 237 and SEQ ID NO: 291 to SEQ ID NO: 294.

9. The RNAi agent according to any one of claims 1-8, wherein the sense strand and/or the antisense strand further comprise(s) one or more lipophilic groups;
preferably, the lipophilic group comprises a saturated or unsaturated C16 hydrocarbon chain;
more preferably, the RNAi agent comprises a lipophilic group, and the lipophilic group is n-hexadecyl.

10. The RNAi agent according to claim 9, wherein the lipophilic group is linked to the sense strand;
preferably, the lipophilic group is linked to the nucleoside at position 1, position 2, position 6, position 7, position 19, position 20, or position 21 from the 5' end of the sense strand;
more preferably, the lipophilic group is linked to the nucleoside at position 2, position 7, or position 20 from the 5' end of the sense strand.

11. The RNAi agent according to claim 9 or 10, wherein the nucleoside linked to the lipophilic group has a structure represented by formula (II):



(II), wherein Base represents a base.

12. A pharmaceutical composition, comprising:
the RNAi agent according to any one of claims 1-11, and
a pharmaceutically acceptable carrier.

13. A cell, comprising the RNAi agent according to any one of claims 1-11.

14. A kit, comprising the RNAi agent according to any one of claims 1-11 and/or the pharmaceutical composition according to claim 12.

15. A method for reducing LRRK2 gene expression, comprising administering to a subject an effective amount or effective dose of the RNAi agent according to any one of claims 1-11 and/or the pharmaceutical composition according to claim 12.

16. A method for treating and/or preventing a disease in a subject, comprising administering to the subject an effective amount or effective dose of the RNAi agent according to any one of claims 1-11 and/or the pharmaceutical composition according to claim 12, wherein
preferably, the disease is a neurodegenerative disease;
more preferably, the neurodegenerative disease is selected from the group consisting of:

Parkinson's disease, amyotrophic lateral sclerosis, Alzheimer's disease, Huntington's disease, schizophrenia, progressive myoclonic epilepsy, and Hallervorden-Spatz disease.

17. A method for delivering an RNAi agent inhibiting LRRK2 expression and/or replication *in vivo*, comprising administering to a subject an effective amount or effective dose of the RNAi agent according to any one of claims 1-11 and/or the pharmaceutical composition according to claim 12.

18. A method for preparing an RNAi agent, comprising: synthesizing the RNAi agent according to any one of claims 1-11.