

[0056] **FIG. 2** provides exemplary ASO sequences and chemistries targeting human SYNGAP1 regRNAs. Light gray shading indicates 2'-MOE; * indicates 5Me-C; dark gray shading indicates LNA; dark gray line indicates phosphodiester bond (PO); white indicates DNA. FIG. 2 discloses SEQ ID NOS 592-701, 742-851, 544-568, 4853, 4854, 569-594, 4855-4901, 702-719, 4902, 720-728, 4903-4913, 729-735, 4914-4917, 736-738, 4918, 739-741 and 4919, respectively, in order of appearance.

[0057] **FIG. 3** shows that SYNGAP1 regRNAs RR86 and RR93 were detected in HEK293 and SK-N-AS cells, as well as human brain samples via RNA capture seq and qPCR.

[0058] **FIGs. 4A** and **4B** show SYNGAP1 mRNA levels in HEK293 cells (**FIG. 4A**) and SK-N-AS cells (**FIG. 4B**) after treatment with the indicated SYNGAP1 regRNA targeting ASOs, or a gapmer non-targeting control (NTC) ASO (CO-1588), a steric NTC ASO (CO-1589), or untreated control ("UTC" or "No ASO"). **FIGs. 4C** and **4D** show a dose dependent increase of SYNGAP1 mRNA levels in HEK293 cells (**FIG. 4C**) and SK-N-AS cells (**FIG. 4D**) after treatment with the indicated SYNGAP1 regRNA targeting ASOs, as compared to cells treated with a gapmer NTC ASO (control; CO-1588). **FIGs. 4E** and **4F** show a dose dependent increase of SYNGAP1 mRNA levels in HEK293 cells (**FIG. 4E**) and SK-N-AS cells (**FIG. 4F**) after treatment with the indicated ASOs, as compared to cells treated with a gapmer NTC ASO (control; CO-1588).

[0059] **FIG. 5** shows SYNGAP1 mRNA levels in SK-N-AS cells and HEK293 cells after treatment with the indicated SYNGAP1 regRNA targeting ASOs, or a gapmer non-targeting control (NTC) ASO (CO-1588), a steric NTC ASO (CO-1589), or untreated control ("UTC").

[0060] **FIG. 6** shows a dose dependent upregulation of SYNGAP1 mRNA levels in both SK-N-AS and HEK293 cells after treatment with the indicated SYNGAP1 regRNA targeting ASOs, as compared to untreated control ("UTC") or cells treated with a gapmer NTC ASO (control; CO-1588).

[0061] **FIG. 7** shows SYNGAP1 mRNA levels in neurons differentiated from human induced pluripotent stem cells after treatment with the indicated concentrations of SYNGAP1 regRNA targeting ASOs or a gapmer NTC ASO (CO-1588; control).

DETAILED DESCRIPTION

[0062] The present disclosure provides antisense oligonucleotides (ASOs) targeting regulatory RNAs, such as promoter-associated RNAs (paRNAs) and enhancer RNAs (eRNAs), and methods using these ASOs to regulate gene expression. These methods are useful for modulating the levels of gene products, for example, modulating expression levels of SYNGAP1, to thereby treat SYNGAP1-related disorder (e.g., diseases associated with SYNGAP1 mutations), such as mental retardation, autosomal dominant 5 (MRD5) and SYNGAP1-related non-syndromic intellectual disability (NSID) or other disease or disorders.

[0063] Various aspects of the compositions and methods described in the present application are set forth in the sections below.

I. Definitions

[0064] To facilitate an understanding of the present application, a number of terms and phrases are defined below.

[0065] The terms “a” and “an” as used herein mean “one or more” and include the plural unless the context is inappropriate.

[0066] As used herein, the terms “SYNGAP1” or “synaptic Ras GTPase activating protein 1” refer to the gene of NCBI Gene ID: 8831 or Hugo Gene Nomenclature Committee (HGNC) ID: 11497 when used in reference to the human gene, or the protein of UniProt Accession No. Q96PV0 (human) when used in reference to a human version of the protein, and to the gene of NCBI Gene ID: 240057 when used in reference to the mouse gene, or to the protein of UniProt Accession No. J3QQ18 (mouse), when used in reference to a mouse version of the protein, and related isoforms and orthologs of the foregoing. SYNGAP1 is a protein of the post-synaptic density (PSD) of glutamatergic neurons that interacts with PSD95 and SAP102, and is capable of positively or negatively regulating the density of N-Methyl-D-aspartic acid (NMDA) and α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors at the glutamatergic synapses, and also negatively regulates small G protein signaling downstream of glutamate receptor activation (see, e.g., Jeyabalan et al. (2016) *Front. Cell Neurosci.* 10: 32, incorporated herein by reference). In some embodiments, a SYNGAP1 protein comprises an isoform of SYNGAP1 (e.g., an N-terminus isoform A, B, and C and/or a C-terminus isoform α 1 (α 1), α 2 (α 2), β (β), or γ (γ) of human SYNGAP1. In some embodiments, a SYNGAP1 protein comprises an isoform selected from SYNGAP1 A α 1, SYNGAP1 A α 2, SYNGAP1 A β ,

SYNGAP1 A γ , SYNGAP1 B α 1, SYNGAP1 B α 2, SYNGAP1 B β , SYNGAP1 B γ , SYNGAP1 C α 1, SYNGAP1 C α 2, SYNGAP1 C β , SYNGAP1 C γ , or any combination of the foregoing isoforms.

[0067] As used herein, the terms “regulatory RNA” and “regRNA” are used interchangeably to refer to a noncoding RNA transcribed from a regulatory element of a gene (*e.g.*, a protein-coding gene), wherein the gene is not the noncoding RNA itself. Exemplary regulatory elements include but are not limited to promoters, enhancers, super-enhancers, and natural antisense transcripts. A noncoding RNA transcribed from a promoter, in the antisense direction, is also called “promoter RNA” or “paRNA.” A noncoding RNA transcribed from an enhancer or super-enhancer, in either the sense direction or the anti-sense direction, is also called “enhancer RNA” or “eRNA.”

[0068] As used herein, the term “nascent RNA” refers to an RNA that is still being transcribed or has just been transcribed by RNA polymerase and remains tethered to the DNA from which it is transcribed. An RNA that has dissociated from the DNA from which it is transcribed is also called an “untethered RNA.”

[0069] As used herein, the term “antisense oligonucleotide” or “ASO” refers to a single-stranded oligonucleotide having a nucleotide sequence that hybridizes with a target nucleic acid under suitable conditions or a conjugate comprising such single-stranded oligonucleotide. In some embodiments, the disclosure encompasses pharmaceutically acceptable salts of any of the ASOs described herein. Suitable pharmaceutically acceptable salts include, but are not limited to, sodium, potassium, calcium, and magnesium salts. In some embodiments, the ASOs provided herein are lyophilized and isolated as salts (*e.g.*, sodium salts).

[0070] As used herein, in some embodiments, the stability of a regRNA is reversely correlated with the degradation rate of the regRNA. In some embodiments, where an ASO increases the stability of a regRNA, it reduces the degradation rate of the regRNA. In some embodiments, where an ASO decreases the stability of a regRNA, it increases the degradation rate of the regRNA. In some embodiments, the degradation rate of a regRNA can be measured by blocking synthesis of new regRNA and assessing the half-life of the existing regRNA.

[0071] As used herein, the terms “subject” and “patient” refer to an organism to be treated by the methods and compositions described herein. Such organisms preferably include, but are not

limited to, mammals (*e.g.*, rodents (*e.g.*, mice), primates, simians, equines, bovines, porcines, canines, felines, and the like), and more preferably include humans.

[0072] As used herein, the term “effective amount” refers to the amount of a compound (*e.g.*, a compound of the present application) sufficient to effect beneficial or desired results. An effective amount can be administered in one or more administrations, applications or dosages and is not intended to be limited to a particular formulation or administration route. As used herein, the term “treating” includes any effect, *e.g.*, lessening, reducing, modulating, ameliorating or eliminating, that results in the improvement of the condition, disease, disorder, and the like, or ameliorating a symptom thereof.

[0073] As used herein, the term “pharmaceutical composition” refers to the combination of an active agent with a carrier, inert or active, making the composition especially suitable for diagnostic or therapeutic use *in vivo* or *ex vivo*.

[0074] As used herein, the term “pharmaceutically acceptable carrier” refers to any of the standard pharmaceutical carriers, such as a phosphate buffered saline solution, water, emulsions (*e.g.*, such as an oil/water or water/oil emulsions), and various types of wetting agents. The compositions also can include stabilizers and preservatives. For examples of carriers, stabilizers and adjuvants, *see e.g.*, Martin, *Remington's Pharmaceutical Sciences*, 15th Ed., Mack Publ. Co., Easton, PA (1975).

[0075] Throughout the description, where compositions are described as having, including, or comprising specific components, or where processes and methods are described as having, including, or comprising specific steps, it is contemplated that, additionally, there are compositions described in the present application that consist essentially of, or consist of, the recited components, and that there are processes and methods according to the present application that consist essentially of, or consist of, the recited processing steps.

[0076] As a general matter, compositions specifying a percentage are by weight unless otherwise specified. Further, if a variable is not accompanied by a definition, then the previous definition of the variable controls.

II. Antisense Oligonucleotides

[0077] In some embodiments, the antisense oligonucleotides (ASO) disclosed herein hybridize with or target a regRNA (*e.g.*, an eRNA, a paRNA, or a NAT) transcribed from a regulatory element of a SYNGAP1 gene, also referred to herein as a “SYNGAP1 regRNA”. It is

understood that NATs, eRNAs, and paRNAs are regRNAs modulating (e.g., facilitating or upregulating) gene expression (**FIG. 1**). In some embodiments, the SYNGAP1 regRNA is a human SYNGAP1 regRNA. In some embodiments, the SYNGAP1 regRNA is a mouse SYNGAP1 regRNA. In certain embodiments, the SYNGAP1 regRNA is an eRNA. In certain embodiments, the SYNGAP1 regRNA is a paRNA. In certain embodiments, the SYNGAP1 regRNA is a NAT. In certain embodiments, the SYNGAP1 regRNA is not a polyadenylated RNA.

[0078] eRNAs can be identified using methods known in the art, such as Assay for Transposase-Accessible Chromatin using sequencing (ATAC-seq), global run-on sequencing, precision run-on sequencing, cap analysis gene expression, and histone modification analysis (see, e.g., Sartorelli & Lauberth, Nat. Struct. Mol. Biol. (2020) 27:521–28; PCT Application Publication No. WO2013/177248). paRNAs are RNAs transcribed from promoters of target genes in the antisense direction (transcripts in the sense direction are mRNAs of the target genes). They can be identified by similar methods, taking into account of their specific location and orientation. The nucleotide sequences of exemplary human and mouse SYNGAP1 regRNAs are provided in **Table 1** below. Any of these human and mouse SYNGAP1 regRNAs are contemplated as a target regRNA of an ASO disclosed herein.

Table 1. Exemplary regRNAs

SEQ ID NO	Name	Sequence
1	RR86_v1 human regRNA	TAGTAGGAGGGCACAGGCCAGGGTGTCTCGCAACGGGCGTCCCGGGGAGCTGCTGAAGT CCTCCCCGCTGCAGAGGACCGAGAGGCAGGGCCCGGAGAGAACAGCCCGGTCCGAGGGGG TGGGCCGTCAGCCGCCCGGAAAGCTGCGTTTCCCGGTGATTAAGTGTAGCACCCGCCCCC GGTAGGTCCTGAGAGGGGCGAGATTAGTTGGGTACTTACCATGTGGGCCTGGCACTTGCCC ACATCACTTTGTGTCTTCCACCGGCCCTGCGAGGGTGGGGGTGCCGTCCCTCAGTTTCCA GCGGGGAAACGGGCTGGAAGCGTCACTCAGCTAATAGTAGTGGACCTGGGACTGGAACCT CAGATTTGCTAAGGAACCAGGCAGGTTCTTTCTGTTTTCTGCACGGGAGCATTTTAACCA GCAATTAAAGAAACCTTATTTCTAGGTAATTTTAAAAAGA
2	RR87 human regRNA	GCCGCCCCCTCACCTGCCAATGATGCTCTTGATCTGGGAATCCTTCTCTGCCTGCTGCTGCC TTAGCCTCTTCTCACTCTGCTCCAGTCGGGCCTGATACTGCATCAGGATTTTGCTGGTTTG TTCTTCTGGGACAGCAGCCTCCGCTCATACTCTTCCAGCTTCCGGTTGGACATGTGCAGC CGCTCTTTCA
3	RR88 human regRNA	GATCCCTGCAAATGTAATCAAACCTAAGAAGAGGCCAGCCATGGTATCTCATTCCTGTAATC CCAGCACTTTGGGAGGCCGAGGTGGGTGGATTGCCTGAGGTGAGGATTCGAGCCAGCCTG GCCAACATAGTGAAACCCCGTCTCTAATAAAAAATACAAAAACAAATTAGCTGGGCATAGTG GCACGTGCCTGTAATCCAGCTACTCAGGAGGCTGAGGCAGGAGAATCACTTGAACCTGGA AGGCGGAGGTTGCAGTGAGCTGAGCTCATGCCATTGCACTCCAAGCCTGGGAAACAAGAGC GATATTCCATCTCAAAAAAAGAGGCCATACTGAATTAGAGTGGACCCCTTAACCCACCTTA

		AGCAAGGTTCTCCAGACCCATGTTAATCTTATTCGAACCCTGGACTTGTAGTTTGGGAGAC CCAAAACAGAGACCTAGACCCAAAGAAAAAGATGAAGCCAATATTAGAAAAGTCCCCAGGC CAGAATTAAATTTTAGAATCTAGCTAGAGGTAAGAAACCCACATTCTGATCTCAAAGAAA CTGCAAACCTGGGGATGAAGGAGGAGACATTCGGCTGACCCAAAGATGAAGATGACCACAT AGAGATCTAAAATACATTACATCTGAAAGATGTGAGTTAGTGACCGATTGCGAGGTTCAAT TTCTGAAACAACCTCCCCAGGTAACCCAGACCCTAGGAAGGGCCCAAATATTGGACCCCCA CCCCACAGGACCCAAATATTGGACCCCCAACCCCTCACCTACACACACAA
--	--	---

[0079] The present disclosure describes ASOs that may be used to increase expression of the target gene *SYNGAP1* (e.g., human *SYNGAP1* or murine *SYNGAP1*). Without wishing to be bound by theory, this increased gene expression may be due to increasing the amount or stability of the targeted SYNGAP1 regRNA, or interference with regRNA-associated repressors that inhibit the expression of the gene to thereby increase *SYNGAP1* gene expression. These ASOs are different from the ASOs previously described that were designed to inhibit eRNAs (see, e.g., PCT Application Publication Nos. WO2013/177248 and WO2017/075406). Without wishing to be bound by theory, it is hypothesized that the ASOs' ability to upregulate SYNGAP1 gene expression is attributable to the selection of a target sequence in the regRNA and/or the chemical modifications of the ASOs.

[0080] Increased *SYNGAP1* gene expression can be at least about 0.1%, 0.5%, 1%, 2%, 3%, 4%, 5%, 6%, 7%, 8%, 9%, 10%, 15%, 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 100%, 105%, 110%, 115%, 120%, 125%, 130%, 135%, 140%, 145%, 150%, 155%, 160%, 165%, 170%, 175%, 180%, 185%, 190%, 195%, 200%, 205%, 210%, 215%, 220%, 225%, 230%, 235%, 240%, 245%, 250%, 255%, 260%, 265%, 270%, 275%, 280%, 285%, 290%, 295%, 300%, 350%, 400%, 450%, 500%, 550%, 600%, 650%, 700%, 750%, 800%, 850%, 900%, 950%, or 1000% or more increase in expression as compared to baseline gene expression, gene expression prior to treatment, or gene expression after treatment with a control ASO. Increased *SYNGAP1* gene expression can be at least about 0.1-fold, 0.5-fold, 1-fold, 1.5-fold, 2-fold, 2.5-fold, 3-fold, 3.5-fold, 4-fold, 4.5-fold, 5-fold, 5.5-fold, 6-fold, 6.5-fold, 7-fold, 7.5-fold, 8-fold, 8.5-fold, 9-fold, 9.5-fold, 10-fold or more increase in expression as compared to baseline gene expression, gene expression prior to treatment, or gene expression after treatment with a control ASO.

[0081] ASOs that hybridize to (e.g., are complementary to) a portion of any of the regulatory RNAs provided herein (e.g., as described in Table 1 above), are contemplated by the present

disclosure. In some embodiments, the regulatory RNA has a nucleotide sequence of any one of SEQ ID NO: 1, 2, 3, 4, 5, 6, and 7.

Sequences of ASOs

[0082] In certain embodiments, an ASO disclosed herein is complementary to a sequence in a SYNGAP1 regRNA (e.g., a SYNGAP1 regRNA provided in **Table 1**) that is no more than 300, 250, 200, 150, 100, 50, 40, 30, 20, 10, 8, 5, or 1 nucleotide(s) from the 5' or 3' end of the SYNGAP1 regRNA. In certain embodiments, the ASO disclosed herein is complementary to a sequence in the SYNGAP1 regRNA that is no more than 300, 250, 200, 150, 100, 50, 40, 30, 20, or 10 nucleotides from the 5' end of the SYNGAP1 regRNA (*i.e.*, the 5' most nucleotide of the regRNA sequence forming a duplex with the ASO is no more than 300, 250, 200, 150, 100, 50, 40, 30, 20, 10, 8, 5, or 1 nucleotide(s) from the 5' end of the SYNGAP1 regRNA). In certain embodiments, the ASO disclosed herein is complementary to a sequence in the SYNGAP1 regRNA that is no more than 300, 250, 200, 150, 100, 50, 40, 30, 20, 10, 8, 5, or 1 nucleotide(s) nucleotides from the 3' end of the SYNGAP1 regRNA (*i.e.*, the 3' most nucleotide of the regRNA sequence forming a duplex with the ASO is no more than 300, 250, 200, 150, 100, 50, 40, 30, 20, or 10 nucleotides from the 3' end of the SYNGAP1 regRNA). In some embodiments, provided herein are ASOs comprising a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of a portion of a SYNGAP1 regRNA provided herein (e.g., a regRNA comprising or consisting a portion of or the full length nucleotide sequence provided in any one of SEQ ID NOs: 1, 2, 3, 4, 5, 6, and 7). In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of a SYNGAP1 regRNA identified herein as RR86_v1 (SEQ ID NO: 1). In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of a 3' portion of SEQ ID NO: 1 (e.g., nucleotides 185-467, 186-467, 187-467, 188-467, 189-467, 190-467, 191-467, 192-467, 193-467, 194-467, 195-467, 196-467, 197-467, 198-467, 199-467, 200-467, 201-467, 202-467, 203-467, 204-467, 205-467, 210-467, 215-467, or 220-467 of SEQ ID

NO: 1). In some embodiments, provided herein are ASOs that do not comprise or consist of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of a 5' portion of SEQ ID NO: 1 (e.g., nucleotides 1-184, 1-183, 1-182, 1-181, 1-180, 1-179, 1-178, 1-177, 1-176, 1-175, 1-174, 1-173, 1-172, 1-171, 1-170, 1-169, 1-168, 1-167, 1-166, 1-165, 1-164, 1-163, 1-162, or 1-161 of SEQ ID NO: 1).

[0083] In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of the SYNGAP1 regRNA identified herein as RR87 (SEQ ID NO: 2).

[0084] In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of the SYNGAP1 regRNA identified herein as RR88 (SEQ ID NO: 3).

[0085] In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of the SYNGAP1 regRNA identified herein as RR93_v1 (SEQ ID NO: 4).

[0086] In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of the SYNGAP1 regRNA identified herein as RR86_v2 (SEQ ID NO: 5).

[0087] In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of the SYNGAP1 regRNA identified herein as RR93_v2 (SEQ ID NO: 6).

[0088] In some embodiments, provided herein are ASOs comprising or consisting of a nucleotide sequence that is complementary to at least 8 nucleotides (e.g., 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides) of the SYNGAP1 regRNA identified herein as RR121 (SEQ ID NO: 7).

[0089] In certain embodiments, the ASO is no more than 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 50, 60, 70, 80, 90, or 100 nucleotides in length. In certain embodiments, the ASO is at least 8, 10, 15, 20, 25, 30, 40, 50, 60, 70, 80, 90, or 100 nucleotides in length. In certain embodiments, the ASO is at least 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, or 20 nucleotides in length. In certain embodiments, the ASO is 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 nucleotides in length.

[0090] In certain embodiments, the ASO is designed to lack a stable secondary structure formed within itself or between each other, thereby increasing the amount of the ASO in a single-stranded form ready to hybridize with the SYNGAP1 regRNA. Methods to predict secondary structures are known in the art (see, *e.g.*, Seetin and Mathews, *Methods Mol. Biol.* (2012) 905:99-122; Zhao *et al.*, *PLoS Comput. Biol.* (2021) 17(8):e1009291) and web-based programs (*e.g.*, RNAfold) are available to public users.

[0091] For example, ASOs have been designed to target a human SYNGAP1 regRNA (*e.g.*, an eRNA, a NAT or a paRNA). The nucleotide sequences of some of these ASOs are provided in **Table 2** below. In some embodiments, an ASO of the disclosure comprises or consists of a nucleotide sequence provided in any one of **Tables 2-4**. In some embodiments, an ASO of the disclosure comprises or consists of a nucleotide sequence and/or a chemistry modification as provided in **Table 2**. Any chemical modification or combination of chemical modifications described herein can be applied to any ASO sequence provided herein (*e.g.*, in **Table 2, 3, or 4**). In some embodiments, an ASO comprises or consists of a nucleotide sequence and/or a chemistry modification of any one of SEQ ID NOs: 10-4852. In some embodiments, an ASO comprises or consists of a nucleotide sequence and/or a chemistry modification of any one of SEQ ID NOs: 10-1003, 1004-2961, or 2962-4852.

[0092] Additional ASO sequences that target SYNGAP1 regRNAs RR86_v1, RR86_v2, RR93_v1, and RR93_v2 are provided in **Tables 3 and 4**. In some embodiments, an ASO of the disclosure comprises or consists of a nucleotide sequence selected from any one of the ASOs provided in **Tables 2-4**. In some embodiments, the ASO comprises or consists of a nucleotide sequence as set for in any one of SEQ ID NOs: 1004-2961 or 2962-4852.