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(54) **COMPOSITION FOR REGULATING
PRODUCTION OF INTERFERING
RIBONUCLEIC ACID**

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(57) **ABSTRACT**

Some embodiments of the present disclosure relate to one or more compositions that upregulate the production of one or more sequences of micro-interfering ribonucleic acid (miRNA). The sequences of miRNA may be complimentary to a sequence of target messenger RNA (mRNA) that encodes for translation of a target biomolecule and the miRNA can cause the target mRNA to be degraded or inactivated, thereby causing a decrease in bioavailability of the target biomolecule because it is degraded or inactivated by the miRNA, thereby decreasing the bioavailability of the target biomolecule within a subject that is administered the one or more compositions. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor, such as serotonin receptor 5HT1a, 5HT1b, 5HT1d, 5HT1e, 5HT1f, 5HT2a, 5HT2b, 5HT2c, 5HT3, 5HT4, 5HT6, or 5HT7.

Specification includes a Sequence Listing.

COMPOSITION FOR REGULATING PRODUCTION OF INTERFERING RIBONUCLEIC ACID

[0001] This application contains a Sequence Listing electronically submitted via Patent Center to the United States Patent and Trademark Office as an XML Document file entitled "A8149441US-Sequence Listing.xml" created on 2024 Feb. 12 and having a size of 110,545 bytes. The information contained in the Sequence Listing is incorporated by reference herein.

TECHNICAL FIELD

[0002] The present disclosure generally relates to compositions for regulating production of interfering ribonucleic acid (RNA). In particular, the present disclosure relates to compositions for regulating gene expression and therefore, the production of interfering RNA, that will suppress serotonin receptor expression.

BACKGROUND

[0003] Bioactive molecules, including complements and factors, are necessary for the homeostatic control of biological systems.

[0004] When bioactive molecules are over-expressed, under-expressed or mis-expressed, homeostasis is lost, and disease is often the result.

[0005] As such, it may be desirable to establish therapies, treatments and/or interventions that address when homeostasis and regulation of bioactive molecules is lost to prevent or treat the resulting disease.

SUMMARY

[0006] Some embodiments of the present disclosure relate to one or more compositions that upregulate the production of one or more sequences of micro-interfering ribonucleic acid (miRNA). The sequences of miRNA may be complementary to a sequence of target messenger RNA (mRNA) that encodes for translation of a target biomolecule and the miRNA can cause the target mRNA to be degraded or inactivated, thereby causing a decrease in bioavailability of the target biomolecule because it is degraded or inactivated by the miRNA, thereby decreasing the bioavailability of the target biomolecule within a subject that is administered the one or more compositions. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT1a. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT1b. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT1c. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT1d. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT1e. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT1f. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT2a. In some embodiments of the present disclosure, the target biomolecule is a serotonin

receptor such as serotonin receptor 5HT2b. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT2c. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT3. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT4. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT6. In some embodiments of the present disclosure, the target biomolecule is a serotonin receptor such as serotonin receptor 5HT7.

[0007] In some embodiments of the present disclosure the compositions comprise a plasmid of deoxyribonucleic acid (DNA) that includes one or more insert sequences of nucleic acids that encode for the production of miRNA and a backbone sequence of nucleic acids that facilitates introduction of the one or more insert sequences into one or more of a subject's cells where it is expressed and/or replicated. Expression of the one or more insert sequences by one or more cells of the subject results in an increased production of the miRNA and, therefore, decreased translation or production of the target biomolecule by one or more of the subject's cells.

[0008] Some embodiments of the present disclosure relate to compositions that upregulate the production of miRNA that degrades, or causes degradation of, or inactivates or causes the inactivation of, the target mRNA of the target biomolecule.

[0009] Some embodiments of the present disclosure relate to a recombinant plasmid (RP). In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 2. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT1a.

[0010] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 3. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT1b.

[0011] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 4. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT1d.

[0012] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 5. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT1e.

[0013] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 6. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT1f.

[0014] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 7. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT2a.

[0015] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 8. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT2b.

[0016] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 9. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT2c.

[0017] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 10. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT3.

[0018] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 11. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT4.

[0019] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 12. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT6.

[0020] Some embodiments of the present disclosure relate to a recombinant plasmid. In some embodiments of the present disclosure, the RP comprises a nucleotide sequence of SEQ ID NO. 1 and SEQ ID NO. 13. The RP comprises a nucleotide sequence encoding one or more nucleotide sequences encoding a miRNA sequence that targets the mRNA of serotonin receptor 5HT7.

[0021] Some embodiments of the present disclosure relate to a method of making a composition/target cell complex. The method comprising a step of administering a RP comprising SEQ ID NO. 1 and one of SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5, SEQ ID NO. 6, SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12, or SEQ ID NO. 13 to a target cell for forming the composition/target cell complex, wherein the composition/target cell complex causes the target cell to increase production of one or more sequences of miRNA that decreases production of a target biomolecule.

[0022] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin

receptor 5HT1a. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT1a, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0023] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT1b. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT1b, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0024] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT1d. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT1d, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0025] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT1e. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT1e, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0026] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT1f. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT1f, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0027] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT2a. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof,

that target and silence the mRNA of serotonin receptor 5HT2a, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0028] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT2b. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT2b, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0029] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT2c. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT2c, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0030] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT3. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT3 which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0031] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT4. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT4, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0032] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT6. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT6, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

[0033] Embodiments of the present disclosure relate to at least one approach for inducing endogenous production of one or more sequences of miRNA that target and silence the mRNA of a target biomolecule, for example serotonin receptor 5HT7. A first approach utilizes gene vectors containing nucleotide sequences for increasing the endogenous production of one or more sequences of miRNA, which are complete or partial sequences and/or combinations thereof, that target and silence the mRNA of serotonin receptor 5HT7, which can be administered to a subject to increase the subject's production of one or more sequences of the miRNA.

DETAILED DESCRIPTION

[0034] Unless defined otherwise, all technical and scientific terms used therein have the meanings that would be commonly understood by one of skill in the art in the context of the present description. Although any methods and materials similar or equivalent to those described therein can also be used in the practice or testing of the present disclosure, the preferred methods and materials are now described. All publications mentioned therein are incorporated therein by reference to disclose and describe the methods and/or materials in connection with which the publications are cited.

[0035] As used therein, the singular forms "a", "an", and "the" include plural references unless the context clearly dictates otherwise. For example, reference to "a composition" includes one or more compositions and reference to "a subject" or "the subject" includes one or more subjects.

[0036] As used therein, the terms "about" or "approximately" refer to within about 25%, preferably within about 20%, preferably within about 15%, preferably within about 10%, preferably within about 5% of a given value or range. It is understood that such a variation is always included in any given value provided therein, whether or not it is specifically referred to.

[0037] As used therein, the term "ameliorate" refers to improve and/or to make better and/or to make more satisfactory.

[0038] As used therein, the term "cell" refers to a single cell as well as a plurality of cells or a population of the same cell type or different cell types. Administering a composition to a cell includes in vivo, in vitro and ex vivo administrations and/or combinations thereof.

[0039] As used therein, the term "complex" refers to an association, either direct or indirect, between one or more particles of a composition and one or more target cells. This association results in a change in the metabolism of the target cell. As used therein, the phrase "change in metabolism" refers to an increase or a decrease in the one or more target cells' production of one or more proteins, and/or any post-translational modifications of one or more proteins.

[0040] As used therein, the term "composition" refers to a substance that, when administered to a subject, causes one or more chemical reactions and/or one or more physical reactions and/or one or more physiological reactions and/or one or more biological reactions in the subject. In some embodiments of the present disclosure, the composition is a plasmid vector.

[0041] As used therein, the term "endogenous" refers to the production and/or modification of a molecule that originates within a subject.

[0042] As used therein, the term "exogenous" refers to a molecule that is within a subject but that did not originate

within the subject. As used therein, the terms “production”, “producing” and “produce” refer to the synthesis and/or replication of DNA, the transcription of one or more sequences of RNA, the translation of one or more amino acid sequences, the post-translational modifications of an amino acid sequence, and/or the production of one or more regulatory molecules that can influence the production and/or functionality of an effector molecule or an effector cell. For clarity, “production” is also used therein to refer to the functionality of a regulatory molecule, unless the context reasonably indicates otherwise.

[0043] As used therein, the term “subject” refers to any therapeutic target that receives the composition. The subject can be a vertebrate, for example, a mammal including a human. The term “subject” does not denote a particular age or sex. The term “subject” also refers to one or more cells of an organism, an in vitro culture of one or more tissue types, an in vitro culture of one or more cell types, ex vivo preparations, and/or a sample of biological materials such as tissue, and/or biological fluids.

[0044] As used therein, the term “target biomolecule” refers to a serotonin receptor that is found within a subject. A biomolecule may be endogenous or exogenous to a subject and when bioavailable the biomolecule may inhibit or stimulate a biological process within the subject.

[0045] As used therein, the term “target cell” refers to one or more cells and/or cell types that are deleteriously affected, either directly or indirectly, by a dysregulated biomolecule. The term “target cell” also refers to cells that are not deleteriously affected but that are the cells in which it is desired that the composition interacts.

[0046] As used therein, the term “therapeutically effective amount” refers to the amount of the composition used that is of sufficient quantity to ameliorate, treat and/or inhibit one or more of a disease, disorder or a symptom thereof. The “therapeutically effective amount” will vary depending on the composition used, the route of administration of the composition and the severity of the disease, disorder or symptom thereof. The subject’s age, weight and genetic make-up may also influence the amount of the composition that will be a therapeutically effective amount.

[0047] As used therein, the terms “treat”, “treatment” and “treating” refer to obtaining a desired pharmacologic and/or physiologic effect. The effect may be prophylactic in terms of completely or partially preventing an occurrence of a disease, disorder or symptom thereof and/or the effect may be therapeutic in providing a partial or complete amelioration or inhibition of a disease, disorder, or symptom thereof. Additionally, the term “treatment” refers to any treatment of a disease, disorder, or symptom thereof in a subject and includes: (a) preventing the disease from occurring in a subject which may be predisposed to the disease but has not yet been diagnosed as having it; (b) inhibiting the disease, i.e., arresting its development; and (c) ameliorating the disease.

[0048] As used therein, the terms “unit dosage form” and “unit dose” refer to a physically discrete unit that is suitable as a unitary dose for patients. Each unit contains a predetermined quantity of the composition and optionally, one or more suitable pharmaceutically acceptable carriers, one or more excipients, one or more additional active ingredients, or combinations thereof. The amount of composition within each unit is a therapeutically effective amount.

[0049] Where a range of values is provided therein, it is understood that each intervening value, to the tenth of the unit of the lower limit unless the context clearly dictates otherwise, between the upper and lower limit of that range and any other stated or intervening value in that stated range, is encompassed within the disclosure. The upper and lower limits of these smaller ranges may independently be included in the smaller ranges, and are also, encompassed within the disclosure, subject to any specifically excluded limit in the stated range. Where the stated range includes one or both of the limits, ranges excluding either or both of those included limits are also included in the disclosure.

[0050] In some embodiments of the present disclosure, a composition is a recombinant plasmid (RP) for introducing genetic material, such as one or more nucleotide sequences, into a target cell for reproduction or transcription of an insert that comprises one or more nucleotide sequences that are carried within the RP. In some embodiments of the present disclosure, the RP is delivered without a carrier, by a viral vector, by a protein coat, or by a lipid vesicle. In some embodiments of the present disclosure, the vector is an adeno-associated virus (AAV) vector.

[0051] In some embodiments of the present disclosure, the insert comprises one or more nucleotide sequences that encode for production of at least one sequence of miRNA that decreases the production of target biomolecules. The miRNA may, directly or indirectly, bind to and degrade the target mRNA or otherwise inactivate the target mRNA so that less or none of the target-biomolecule protein is produced.

[0052] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT1a.

[0053] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT1b.

[0054] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT1d.

[0055] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT1e.

[0056] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT1f.

[0057] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT2a.

[0058] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT2b.

[0059] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT2c.

[0060] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT3.

[0061] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT4.

[0062] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT6.

[0063] In some embodiments of the present disclosure, the target biomolecule is serotonin receptor 5HT7.

[0064] In some embodiments of the present disclosure, the insert comprises one or more nucleotide sequences that each encode for one or more miRNA sequences that may be complimentary to and degrade, or cause degradation of, mRNA of the target biomolecule.

[0065] Some embodiments of the present disclosure relate to a composition that can be administered to a subject with a condition that results, directly or indirectly, from the production of a dysregulated biomolecule. When a therapeutically effective amount of the composition is adminis-

tered to the subject, the subject may change production and/or functionality of one or more biomolecules.

[0066] In some embodiments of the present disclosure, the subject may respond to receiving the therapeutic amount of the composition by changing production and/or functionality of one or more intermediary molecules by changing production of one or more DNA sequences, one or more RNA sequences, and/or one or more proteins that regulate the levels and/or functionality of the one or more intermediary molecules. The one or more intermediary molecules regulate the subject's levels and/or functionality of the one or more biomolecules.

[0067] In some embodiments of the present disclosure, administering a therapeutic amount of the composition to a subject upregulates the production, functionality or both one or more sequences of miRNA that each target the mRNA of one or more target biomolecules. In some embodiments of the present disclosure, there are one, two, three, four, five, or six miRNA sequences that each are complimentary to and degrade, or cause degradation of, one biomolecule, such as the mRNA of serotonin receptor 5HT1a, serotonin receptor 5HT1b, serotonin receptor 5HT1d, serotonin receptor 5HT1e, serotonin receptor 5HT1f, serotonin receptor 5HT2a, serotonin receptor 5HT2b, serotonin receptor 5HT2c, serotonin receptor 5HT3, serotonin receptor 5HT4, serotonin receptor 5HT6, or serotonin receptor 5HT7. In some embodiments of the present disclosure, the composition may comprise multiple copies of the same nucleotide sequence of miRNA.

[0068] In some embodiments of the present disclosure, the composition is an RP that may be used for gene therapy. The gene therapy is useful for increasing the subject's endogenous production of one or more sequences of miRNA that target the mRNA of a target biomolecule. For example, the RP can contain one or more nucleotide sequences that cause increased production of one or more nucleotide sequences that cause an increased production of one or more miRNA sequences that are each complimentary to and degrade, or cause degradation of, or inactivate, or cause inactivation of, one biomolecule, such as serotonin receptor 5HT1a, serotonin receptor 5HT1b, serotonin receptor 5HT1d, serotonin receptor 5HT1e, serotonin receptor 5HT1f, serotonin receptor 5HT2a, serotonin receptor 5HT2b, serotonin receptor 5HT2c, serotonin receptor 5HT3, serotonin receptor 5HT4, serotonin receptor 5HT6, or serotonin receptor 5HT7.

[0069] In some embodiments of the present disclosure, the delivery vehicle of the RP used for gene therapy may be a vector that is comprised of a virus that can be enveloped, or not (unenveloped), replication effective or not (replication ineffective), or combinations thereof. In some embodiments of the present disclosure, the vector is a virus that is not enveloped and not replication effective. In some embodi-

ments of the present disclosure, the vector is a virus of the Parvoviridae family. In some embodiments of the present disclosure, the vector is a virus of the genus Dependoparvovirus. In some embodiments of the present disclosure, the vector is an adeno-associated virus (AAV). In some embodiments of the present disclosure, the vector is a recombinant AAV. In some embodiments of the present disclosure, the vector is a recombinant AAV6.2FF.

[0070] In some embodiments of the present disclosure, the delivery vehicle of the RP used for gene therapy may be a protein coat.

[0071] In some embodiments of the present disclosure, the delivery vehicle of the RP used for gene therapy may be a lipid vesicle.

[0072] The embodiments of the present disclosure also relate to administering a therapeutically effective amount of the composition. In some embodiments of the present disclosure, the therapeutically effective amount of the composition that is administered to a patient is between about 10 and about 1×10^{16} TCID₅₀/kg (50% tissue culture infective dose per kilogram of the patient's body mass). In some embodiments of the present disclosure, the therapeutically effective amount of the composition that is administered to the patient is about 1×10^{13} TCID₅₀/kg. In some embodiments of the present disclosure, the therapeutically effective amount of the composition that is administered to a patient is measured in TPC/kg (total particle count of the composition per kilogram of the patient's body mass). In some embodiments the therapeutically effective amount of the composition is between about 10 and about 1×10^{16} TCP/kg.

[0073] Some embodiments of the present disclosure relate to an adeno-associated virus (AAV) genome consisting of a RP that when operable inside a target cell will cause the target cell to produce a miRNA sequence that downregulates production of a biomolecule, with examples being serotonin receptor 5HT1a, serotonin receptor 5HT1b, serotonin receptor 5HT1d, serotonin receptor 5HT1e, serotonin receptor 5HT1f, serotonin receptor 5HT2a, serotonin receptor 5HT2b, serotonin receptor 5HT2c, serotonin receptor 5HT3, serotonin receptor 5HT4, serotonin receptor 5HT6, or serotonin receptor 5HT7. The RP is comprised of AAV2 inverted terminal repeats (ITRs), a composite CASI promoter, a human growth hormone (HGH) signal peptide followed by a miRNA expression cassette containing up to six different miRNAs targeting the mRNA of serotonin receptor 5HT1a, serotonin receptor 5HT1b, serotonin receptor 5HT1d, serotonin receptor 5HT1e, serotonin receptor 5HT1f, serotonin receptor 5HT2a, serotonin receptor 5HT2b, serotonin receptor 5HT2c, serotonin receptor 5HT3, serotonin receptor 5HT4, serotonin receptor 5HT6, or serotonin receptor 5HT7, followed by a Woodchuck Hepatitis Virus post-transcriptional regulatory element (WPRE) and a Simian virus 40 (SV40) polyadenylation (polyA) signal.

SEQ ID NO. 1 (backbone sequence No. 1):

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TTGTGGCCCGTTGTGAGGCAACGTGGCGTGGTGTGCACTGTGTTTGTGACGCAACCCCC
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-continued

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GATTTTGCGGATTTGCGCTATTGGTTAAAAATGAGCTGATTTAACAAAAATTTAACGC
GAATTTTAACAAAATATTAACGTTTACAATTTAAATATTTGCTTATACAATCTTCCGTGTT
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AGAGACCTCTCAAAAATAGCTACCTCTCCGGCATGAATTTATCAGCTAGAACGTTGAA
TATCATATTGATGGTGATTGACTGTCTCCGGCTTTCTCACCCTTTGAATCTTTACCT
ACACATTACTCAGGCATTGCATTTAAAAATATATGAGGGTTCTAAAAATTTTATCCTTGC
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GATTTAGCTTTATGCTCTGAGGCTTTATTGCTTAATTTTGCTAATCTTTGCCTTGCCCTG
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TATTTACACCCGATATGGTGCACTCTCAGTACAATCTGCTCTGATGCCGCATAGTTAAG
CCAGCCCCGACACCCGCCAACCCGCTGACGCGCCCTGACGGGCTTGTCTGCTCCCGC
ATCCGCTTACAGACAAGCTGTGACCGTCTCCGGGAGCTGCATGTGTACAGAGTTTTTCACC
GTCATCACCGAAACGCGGAGACGAAAGGGCTCGTGATACGCCATTTTTTATAGGTTAA
TGTCATGATAAATAGGTTTCTTAGACGTGAGGTGGCACTTTTCGGGGAATGTGCGCGG
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ACCCTGATAAATGCTTCAATAATATTGAAAAAGGAAGAGTATGAGTATTCAACATTTCCG
TGTCGCCCTTATTCCCTTTTTTGCGGCATTTTGCTTCTCTGTTTTTGCTCACCCAGAAAC
GCTGGTGAAAGTAAAAGATGCTGAAGATCAGTTGGGTGCACGAGTGGGTACATCGAACT
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GAGCACTTTTAAAGTCTGCTATGTGGCGCGGTATTATCCCGTATTGACGCCGGGCAAGA
GCAACTCGGTGCGCCGCATACACTATTCTCAGAATGACTTGGTTGAGTACTCACCAGTCAC
AGAAAAGCATCTTACGGATGGCATGACAGTAAGAGAATTATGCAGTGCTGCCATAACCAT
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CGCTTTTTTGACACAACATGGGGGATCATGTAACGCGCTTGATCGTTGGGAACCGGAGCT
GAATGAAGCCATACCAACGACGAGCGTGACACCACGATGCCGTAGCAATGGCAACAAC
GTTGCGCAAACTATTAAGTGGCAACTACTTACTCTAGCTTCCCGGCAACAATTAATAGA
CTGGATGGAGGCGGATAAAGTTGCAGGACCCTTCTGCGCTCGGCCCTTCCGGCTGGCTG
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GGGGCCAGATGGTAAGCCCTCCCGTATCGTAGTTATCTACACGACGGGAGTCAGGCAAC
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GGGAAACGCTTGGTATCTTTATAGTCTGTCGGGTTTCGCCACCTCTGACTGAGCGTCG
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AACGACCGAGCGCAGCGAGTCAGTGAGCGAGGAAGCGGAAGAGCGCCCAATACGCAAACC
GCCTCTCCCCGCGCGTTGGCCGATTCAATTAATGCAGCAGCTGCGCGCTCGCTCGCTCACT
GAGGCCGCCCGGGCAAGCCCGGCGTCGGGCGACCTTTGGTCGCCCGGCTCAGTGAGC
GAGCGAGCGCGCAGAGAGGAGTGGCCAACTCCATCACTAGGGGTTCTTGTAGTTAATG
ATTAACCCGCCATGCTACTTATCTACGTAGCCATGCTCTAGGACATTGATTATTGACTAG
TGGAGTTCCGCGTTACATAACTTACGTTAAATGGCCGCTGGCTGACCGCCCAACGACC
CCCCCCCATTGACGTCAATAATGACGTATGTTCCCATAGTAACGCCAATAGGGACTTTCC
ATTGACGTCAATGGGTGGAGTATTTACGGTAACTGCCCACTTGGCAGTACATCAAGTGT
ATCATATGCCAAGTACGCCCCCTATTGACGTCAATGACGGTAAATGGCCCGCTTGGCATT

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 CCTCCCCACCCCAATTTTGTATTTATTTATTTTAAATTATTTTGTGCAGCGATGGGGG
 CGGGGGGGGGGGGGCGCGCCAGGCGGGCGGGCGGGCGAGGGCGGGCGGGGCGGGG
 GAGGCGGAGAGGTGCGGCGGCAGCCAATCAGAGCGGCGCGCTCCGAAAGTTTCTTTTAT
 GGCGAGGCGGCGGCGGCGGCGCTATAAAAAGCGAAGCGCGGCGGGCGGGAGTCGC
 TGGCGCGTGCCTTCGCCCCGTGCCCGCTCCGCCGCGCCTCGCGCCGCCCGCCCGGCT
 CTGACTGACCGGTTACTAAAACAGGTAAGTCCGGCCTCCGCGCCGGGTTTGGCGCCTC
 CCGCGGGCGCCCCCTCTCACGGCGAGCGCTGCCACGTGACAGGAAGGCGCAGCGAGC
 GTCTGTATCCTTCGCCCGGACGCTCAGGACAGCGGCCCGCTGCTCATAAGACTCGGCCT
 TAGAACCACCATATCAGCAGAAGGACATTTTAGGACGGGACTTGGGTGACTCTAGGGCAC
 TGGTTTTCTTTCCAGAGAGCGGAACAGGCGAGGAAAAGTAGTCCCTTCTCGCGATTCTG
 CGGAGGGATCTCCGTGGGGCGGTGAACGCCGATGATGCCTCTACTAACCATGTTTCATGTT
 TTCTTTTTTTTCTACAGGTCTGGGTGACGAACAGGGTACC 3'

SEQ ID NO. 2 (miRNA expression cassette No. 2-serotonin receptor 5HT1a):
 5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
 CTTGCTGAAGGCTGTATGCTGATCAATCGGATTGCGGTAATCGCGTTTTGGCCTCTGACT
 GACGCGATTACCGATCCGATTGATCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
 CAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGATCTTTGCTAAATTGGT
 GCACGCGTTTTGGCCTCTGACTGACGCGTGCAACATTAGCAAAGATCAGGACACAAGGCC
 TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
 TGCTGACTTCAATCACAATTCAGCGCGTTTTTGGCCTCTGACTGACGCGCTGGAAGTG
 ATTGAAGTCAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
 AAT 3'

SEQ ID NO. 3 (miRNA expression cassette No. 3-serotonin receptor 5HT1b):
 5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
 CTTGCTGAAGGCTGTATGCTGTAATCTTTCGCTGGCTGCAGTTCGTTTTGGCCTCTGACT
 GACGAACTGCAGCGCGAAAGATTACAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
 CAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTGTTAATGCTGATGTCA
 CGCTGCGTTTTGGCCTCTGACTGACGCGAGCTGACAGCATTAAACAGGACACAAGGCC
 TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
 TGCTGTTACCTGGTTAACACATACACCGTTTTTGGCCTCTGACTGACGGTGTATGTGAAC
 CAGGTGAACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
 AAT 3'

SEQ ID NO. 4 (miRNA expression cassette No. 4-serotonin receptor 5HT1d):
 5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
 CTTGCTGAAGGCTGTATGCTGATTTCTTCTGTGCGCTTTCGCCGTTTTGGCCTCTGACT
 GACGGCGAAGCGCAGGAAGAAATCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA

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CAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGAGAATAATCAGATCAGC
ACGCTCGTTTTGGCCTCTGACTGACGAGCGTGCTGCTGATTATTCTCAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
TGCTGTAATCAGGCTGAATTCAGATAGCGTTTTGGCCTCTGACTGACGCTATCTGAACAG
CCTGATTACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
AAT 3'

SEQ ID NO. 5 (miRNA expression cassette No. 5-serotonin receptor 5HT1e):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGATAATCACCGCTGCAGGTTTCAGCGTTTTGGCCTCTGACT
GACGCTGAACCTGGCGGTGATTATCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
CAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTTCAATCGCGTATTGGT
AATCGCGTTTTGGCCTCTGACTGACGCGATTACCAACGCGATTGAACAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
TGCTGTGATCATGCTGAAAATGGTGCACGTTTTGGCCTCTGACTGACGTGCACCATTCAG
CATGATCACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
AAT 3'

SEQ ID NO. 6 (miRNA expression cassette No. 6-serotonin receptor 5HT1f):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGAGGTAATATCCTGACGCTCAGCCGTTTTGGCCTCTGACT
GACGGCTGAGCGTGGATATTACCTCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
CAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTACAGAATCAGATAATC
AGCGCCGTTTTGGCCTCTGACTGACGGCGCTGATTCTGATTCTGTACAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
TGCTGTATGTTTAAAAATTCGCTGCGCGTTTTGGCCTCTGACTGACGCGCAGCGAATTT
AAACATGACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
AAT 3'

SEQ ID NO. 7 (miRNA expression cassette No. 7-serotonin receptor 5HT2a):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGATGAATCGGGTTGTCTGAATCGCGTTTTGGCCTCTGACT
GACGCGATTCAGAACCCGATTTCATCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
CAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGAACACTTTGCTATATCA
TCCTGCGTTTTGGCCTCTGACTGACGCGAGGATGATAGCAAAGTGTTCAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
TGCTGTTCTGTTCTGTTAAGCTAATGCTCGTTTTGGCCTCTGACTGACGAGCATTAGCAAC
GAACAGAACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
AAT 3'

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SEQ ID NO. 8 (miRNA expression cassette No. 8-serotonin receptor 5HT2b):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGGAGCATTAGCAATGCGAACAGAAGTTTGGCCTCTGACT
GACTTCTGTTCTGTGCTAATGCTCCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
CAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGAAACATAATGGATTAG
CAGCGCGTTTTGGCCTCTGACTGACGCGCTGCTGACCATTATGTTTCAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
TGCTGTTATCTTTGCGAAGCTGCCATCCGTTTGGCCTCTGACTGACGGATGGCAGCCGC
AAAGATAACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
AAT 3'

SEQ ID NO. 9 (miRNA expression cassette No. 9-serotonin receptor 5HT2c):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGGCTCCTCCACTTGGTGGTTTGGTTTGGCCTCTGACTGA
CGCGCAACATTCTGGTGATTACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACA
AATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTCATAATCGTATTTGGTG
CGCGCTTTTGGCCTCTGACTGACGCCGACCAAAGCGATTATGACAGGACACAAGGCCTG
TTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATG
CTGTTCTGATCCTGAAGTTCGGGTTGTTTTGGCCTCTGACTGACGAACCCGAACCAGGA
TCAGAACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAA
T 3'

SEQ ID NO. 10 (miRNA expression cassette No. 10-serotonin receptor 5HT3):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGAAATCTCCGGTGGTTCCACTGCGTTTTGGCCTCTGACT
GACGCACTGGAACCCGGAAGATTTAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
CAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGATATCCTGAATATGGTA
TGCAGCGTTTTGGCCTCTGACTGACGCTGCATACCATTAGGATATCAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA
TGCTGTTTAAAGCTCAAACGCGTTCGCCGTTTTGGCCTCTGACTGACGGCGAACGCGTGA
GCTTTAAACAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG
AAT 3'

SEQ ID NO. 11 (miRNA expression cassette No. 11-serotonin receptor 5HT4):
5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG
CTTGCTGAAGGCTGTATGCTGTAATAAAGGCTCGGGAATACCCGTTTTGGCCTCTGACT
GACGGTGATTCGACCTTTATTACAGGACACAAGGCCTGTTACTAGCACTCACATGGAA
CAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTAATACGCCAGATCACC
ATCAGCGTTTTGGCCTCTGACTGACGCTGATGGTGTGGCGTATTACAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA

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TGCTGATACAGAAACGAAGGTTTCAGGCCGTTTGGCCTCTGACTGACGGCCTGAACCCGT

TTCTGTATCAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG

AAT ' 5'

SEQ ID NO. 12 (miRNA expression cassette No. 12-serotonin receptor 5HT6):

5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG

CTTGCTGAAGGCTGTATGCTGTGAGATCGCTGTGGTAAACAGGCGTTTGGCCTCTGACT

GACGCCCTGTTTACCAGCGATCTGACAGGACACAAGGCCTGTTACTAGCACTCACATGGAA

CAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGAGAATCAGATCAGATAG

CGATCCGTTTGGCCTCTGACTGACGGATCGCTATCTGCTGATTCTCAGGACACAAGGCC

TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA

TGCTGAAACATGCCAACAGCAGAATGCCGTTTGGCCTCTGACTGACGGCATTCTGCTGG

GCATGTTTCAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG

AAT 3'

SEQ ID NO. 13 (miRNA expression cassette No. 13-serotonin receptor 5HT7):

5' GCCACCATGGCCACCGGCTCTCGCACAGCCTGCTGCTGGCTTTCGGACTGCTGTGC

CTGCCTTGGCTCCAGGAGGGCTCCGCCGCTAGCATCGATACCGTCGCTATGTGCTGGAGG

CTTGCTGAAGGCTGTATGCTGACAATCAGATATGGTTGCTCGGCGTTTGGCCTCTGACT

GACGCCGAGCAACTATCTGATTGTCAGGACACAAGGCCTGTTACTAGCACTCACATGGAA

CAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTTTCAATGCATCGTT

CAGCGCGTTTGGCCTCTGACTGACGCGCTGAACGGCATTGTGAAACAGGACACAAGGCC

TGTTACTAGCACTCACATGGAACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTA

TGCTGACAATAATGCCAACAGGGTGGTCGTTTGGCCTCTGACTGACGACCCCTGGGC

ATTATTGTCAGGACACAAGGCCTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAG

AAT 3'

SEQ ID NO. 14 = SEQ ID NO. 1 + SEQ ID NO. 2

5' AATCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGGTATTCTTAACATATGTT

GCTCCTTTTACGCTATGTGGATACGCTGCTTAAATGCCTTTGTATCATGCTATTGCTTCC

CGTATGGCTTTTCAATTTCTCCTCTGTATAAATCCTGGTTGCTGTCTCTTTATGAGGAG

TTGTGGCCCGTTGTGAGCAACGTGGCGTGGTGTGCACTGTGTTTGTGACGCAACCCCC

ACTGGTTGGGGCATTGCCACCACCTGTCAGCTCCTTCCGGGACTTTCGCTTTCCCCCTC

CCTATTGCCACGGCGGAATCATCGCCGCTGCCTTGCCCGCTGCTGGACAGGGGCTCGG

CTGTTGGGCACTGACAATTCGGTGGTGTGTCGGGAAATCATCGTCCTTTCTTGGCTG

CTCGCCTGTGTTGCCACCTGGATTCTGCGCGGACGTCCTTCTGCTACGTCCTTTCGGCC

CTCAATCCAGCGACCTTCCTTCCCGCGGCTGCTGCGGCTCTGCGGCTCTTCCGCGT

CTTCGCCTTCGCCCTCAGACGAGTCGGATCTCCCTTTGGGCCGCTCCCCGCTAAGCTT

ATCGATACCGTCGAGATCTAATTTGTTTATGTCAGCTTATAATGGTTACAAATAAGCAA

TAGCATCACAATTTCAAAATAAAGCATTTTTTCACTGCATTCTAGTTGTGGTTTGTG

CAAACTCATCAATGTATCTTATCATGTCTGGATCTCGACCTCGACTAGAGCATGGCTACG

TAGATAAGTAGCATGGCGGTTAATCATTAACACAAGGAACCCCTAGTGATGGAGTTGG

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CCACTCCCTCTCTGCGCGCTCGCTCGCTCACTGAGGCCGGGCGACCAAGGTCGCCCAGC
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CGCTCTGATTCTAACGAGGAAAGCACGTTATACGTGCTCGTCAAAGCAACCATAGTACGC
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ACACATTACTCAGGCATTGCATTTAAATATATGAGGGTTCTAAAAATTTTATCCTTGC
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GGATCTCAACAGCGGTAAGATCCTTGAGAGTTTTCGCCCCGAAGACGTTTCCAATGAT
GAGCACTTTTAAAGTCTGCTATGTGGCGCGGTATTATCCCGTATTGACGCCGGGCAAGA
GCAACTCGGTGCGCGCATACACTATTCTCAGAATGACTTGGTTGAGTACTCACCAGTCAC
AGAAAAGCATCTTACGGATGGCATGACAGTAAGAGAATTATGCAGTGCTGCCATAACCAT
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CGCTTTTTTGCAACATGGGGGATCATGTAACGCGCTTGATCGTTGGGAACCGGAGCT

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GAATGAAGCCATACCAAACGACGAGCGTGACACCACGATGCCTGTAGCAATGGCAACAAC
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GGGGCCAGATGGTAAGCCCTCCCGTATCGTAGTTATCTACACGACGGGAGTCAGGCAAC
TATGGATGAACGAAATAGACAGATCGCTGAGATAGGTGCCTCACTGATTAAGCATTGGTA
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CGATAAGTCGTCTTACCAGGTGGACTCAAGACGATAGTTACCGGATAAGGCGCAGCG
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TCGCTATTACCATGGTCGAGGTGAGCCCCACGTTCTGCTTCACTCTCCCCATCTCCCCC
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GGCGAGGCGGCGGCGGCGGCGCTATAAAAAGCGAAGCGCGGCGGGCGGGAGTCGC
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SEQ ID NO. 15 = SEQ ID NO. 1 + SEQ ID NO. 3

5' AATCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGGTATTCTTAACATATGTT
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CTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAAT 3'

SEQ ID NO. 16 = SEQ ID NO. 1 + SEQ ID NO. 4

5' AATCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGGTATTCTTAACATGTT
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CTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAAT 3'

SEQ ID NO. 17 = SEQ ID NO. 1 + SEQ ID NO. 5
5' AATCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGGTATTCTTAACATGTT
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SEQ ID NO. 18 = SEQ ID NO. 1 + SEQ ID NO. 6

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TGCGCGTTTTGGCCTCTGACTGACGCGCAGCGAATTAAACATGACAGGACACAAGGCCT
GTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAAT 3'

SEQ ID NO. 19 = SEQ ID NO. 1 + SEQ ID NO. 7

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SEQ ID NO. 20 = SEQ ID NO. 1 + SEQ ID NO. 8
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SEQ ID NO. 21 = SEQ ID NO. 1 + SEQ ID NO. 9

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TGATTCTGTGGATAACCGTATTACCGCCTTTGAGTGAGCTGATACCGCTCGCCGAGCCG
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GAGGCCGCCCGGGCAAAGCCCGGGCGTCGGGCGACCTTTGGTCGCCCCGCCCTCAGTGAGC
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ACAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGTTTAAAGCTCAAACGC
GTTTCGCCGTTTTGGCCTCTGACTGACGGCGAACGCGTGAGCTTTAAACAGGACACAAGGC
CTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAAT 3'

SEQ ID NO. 23 = SEQ ID NO. 1 + SEQ ID NO. 11

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CTGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAAT 3'

SEQ ID NO. 24 = SEQ ID NO. 1 + SEQ ID NO. 12
5' AATCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGGTATTCTTAACATGTT
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CAGATCGCTGTGGTAAACAGGCGTTTGGCCTCTGACTGACGCCTGTTTACCAGCGATCT
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CAAAATGGCCTCTAGCCTGGAGGCTTGCTGAAGGCTGTATGCTGAAACATGCCAACAGCAG
AATGCCGTTTTGGCCTCTGACTGACGGCATTCTGCTGGGCATGTTTCAGGACACAAGGCC
TGTTACTAGCACTCACATGGAACAAATGGCCTCTCTAGAAT 3'

SEQ ID NO. 25 = SEQ ID NO. 1 + SEQ ID NO. 13

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[0074] As will be appreciated by those skilled in the art, because the recombinant plasmid is a circular vector, the one or more sequences of the miRNA expression cassettes may be connected at the 3' end of SEQ ID NO. 1, as shown in SEQ ID NO. 14, SEQ ID NO. 15, SEQ ID NO. 16, SEQ ID NO. 17, SEQ ID NO. 18, SEQ ID NO. 19, SEQ ID NO. 20, SEQ ID NO. 21, SEQ ID NO. 22, SEQ ID NO. 23, SEQ ID NO. 24 and SEQ ID NO. 25, or at the 5' end of SEQ ID NO. 1.

[0075] As will be appreciated by those skilled in the art, a perfect match of nucleotides with each of the miRNA expression cassette sequences is not necessary in order to have the desired result of decreased bioavailability of the target biomolecule as a result of the target cell producing the miRNA sequence that will bind to and degrade the mRNA of the target biomolecule. In some embodiments of the present disclosure, about 80% to about 100% nucleotide sequence matching with each of the miRNA expression cassettes causes the desired result. In some embodiments of the present disclosure, about 85% to about 100% nucleotide sequence matching with each of the miRNA expression cassettes causes the desired result. In some embodiments of the present disclosure, about 90% to about 100% nucleotide sequence matching with each of the miRNA expression cassettes causes the desired result. In some embodiments of the present disclosure, about 95% to about 100% nucleotide

sequence matching with each of the miRNA expression cassettes causes the desired result.

Example 1—Expression Cassette

[0076] Expression cassettes for expressing miRNA were synthesized. The synthesized miRNA expression cassettes were cloned into the pAVA-00200 plasmid backbone containing the CASI promoter, multiple cloning site (MCS), Woodchuck Hepatitis Virus post-transcriptional regulatory element (WPRE), and Simian virus 40 (SV40) polyadenylation (polyA) sequence, all flanked by the AAV2 inverted terminal repeats (ITR). pAVA-00200 was cut with the restriction enzymes KpnI and XbaI in the MCS and separated on a 1% agarose gel. The band of interest was excised and purified using a gel extraction kit. Each miRNA expression cassette was amplified by polymerase chain reaction (PCR) using Taq polymerase and the PCR products were gel purified and the bands of interest were also excised and purified using a gel extraction kit. These PCR products contained the miRNA expression cassettes in addition to 15 base pair 5' and 3' overhangs that aligned with the ends of the linearized pAVA-00200 backbone. Using in-fusion cloning, the amplified miRNA expression cassettes were integrated with the pAVA-00200 backbone via homologous recombination. The resulting RP contained the following: 5' ITR, CASI promoter, miRNA expression cassette, WPRE, SV40 polyA and ITR 3'.

SEQUENCE LISTING

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gctgaaggct gtatgctgat aatcacgct gcaggttcag cgttttggcc tctgactgac 180
gctgaacctg gcggtgatta tcaggacaca aggcctgtta ctgacctca catggaacaa 240
atggcctcta gcctggaggc ttgctgaagg ctgtatgctg ttcaatcgcg tattggtaat 300
cgcgttttgg cctctgactg acgcgattac caacgcgatt gaacaggaca caaggcctgt 360
tactagcact cacatggaac aaatggcctc tagcctggag gcttgcgtgaa ggctgtatgc 420
tgtgatcatg ctgaaaatgg tgcacgtttt ggccctctgac tgacgtgcac cattcagcat 480
gatcacagga cacaaggcct gttactagca ctcacatgga acaaatggcc tctctagaat 540

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SEQ ID NO: 6      moltype = DNA length = 540
FEATURE          Location/Qualifiers
source           1..540
                 mol_type = other DNA
                 organism = synthetic construct

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SEQUENCE: 6
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gctgaaggct gtatgctgat gtaatacct gacgctcagc cgttttggcc tctgactgac 180
ggctgagcgt gatattacc tcaggacaca aggcctgtta ctgacctca catggaacaa 240
atggcctcta gcctggaggc ttgctgaagg ctgtatgctg tacagaatca gataatcagc 300
gcggttttgg cctctgactg acggcctga ttctgattct gtacaggaca caaggcctgt 360
tactagcact cacatggaac aaatggcctc tagcctggag gcttgcgtgaa ggctgtatgc 420
tgtcatgttt aaaaattcgc tgcgcgtttt ggccctctgac tgacgcgcag cgaatttaaa 480
catgacagga cacaaggcct gttactagca ctcacatgga acaaatggcc tctctagaat 540

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SEQ ID NO: 7      moltype = DNA length = 540
FEATURE          Location/Qualifiers
source           1..540
                 mol_type = other DNA
                 organism = synthetic construct

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SEQUENCE: 7
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gctgaaggct gtatgctgat gaatcgggtt gtctgaatcg cgttttggcc tctgactgac 180
cgatttcaga acccgattca tcaggacaca aggcctgtta ctgacctca catggaacaa 240
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tgcgttttgg cctctgactg acgcaggatg atagcaaaat gtacaggaca caaggcctgt 360
tactagcact cacatggaac aaatggcctc tagcctggag gcttgcgtgaa ggctgtatgc 420
tgttctgttc gtttaagctaa tgctcgtttt ggccctctgac tgacgagcat tagcaacgaa 480
cagaacagga cacaaggcct gttactagca ctcacatgga acaaatggcc tctctagaat 540

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SEQ ID NO: 8      moltype = DNA length = 540
FEATURE          Location/Qualifiers
source           1..540
                 mol_type = other DNA
                 organism = synthetic construct

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SEQUENCE: 8
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gctgaaggct gtatgctgga gcattagcaa tgcgaacaga agttttggcc tctgactgac 180
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SEQ ID NO: 9      moltype = DNA length = 538
FEATURE          Location/Qualifiers
source           1..538
                 mol_type = other DNA
                 organism = synthetic construct

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SEQUENCE: 9
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gctgaaggct gtatgctggc tcctccactt ggtggtttgg ttttggcctc tgactgacgc 180
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ttctgatcct gaagtctggg ttgcgttttg cctctgactg acgaaccoga accaggatca 480
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SEQ ID NO: 10     moltype = DNA length = 540
FEATURE          Location/Qualifiers

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source                1..540
                      mol_type = other DNA
                      organism = synthetic construct

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gctgaaggct gtatgtctgaa atcttcgggt ggttccactg cgttttggcc tctgactgac 180
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tgtttaaagc tcaaacgcgt tcgccgtttt ggccctctgac tgacggcgaa cgcgtgagct 480
ttaaacagga cacaaggcct gttactagca ctccatagga acaaatggcc tctctagaat 540

SEQ ID NO: 11        moltype = DNA length = 540
FEATURE              Location/Qualifiers
source                1..540
                      mol_type = other DNA
                      organism = synthetic construct

SEQUENCE: 11
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gctgaaggct gtatgtctgta ataaaggctc gggaaatcac cgttttggcc tctgactgac 180
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tgatacagaa acgaaggctt aggcctgttt ggccctctgac tgacggcctg aaccctttc 480
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SEQ ID NO: 12        moltype = DNA length = 540
FEATURE              Location/Qualifiers
source                1..540
                      mol_type = other DNA
                      organism = synthetic construct

SEQUENCE: 12
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SEQ ID NO: 13        moltype = DNA length = 540
FEATURE              Location/Qualifiers
source                1..540
                      mol_type = other DNA
                      organism = synthetic construct

SEQUENCE: 13
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gctgaaggct gtatgtctgac aatcagatat ggttgcctcg cgttttggcc tctgactgac 180
gccagagcaac tatctgattg tcaggacaca aggcctgtta ctagactca catggaacaa 240
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SEQ ID NO: 14        moltype = DNA length = 6339
FEATURE              Location/Qualifiers
source                1..6339
                      mol_type = other DNA
                      organism = synthetic construct

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gataccgctg	agatctaact	tgtttattgc	agcttataat	ggttacaaat	aaagcaatag	660
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SEQ ID NO: 15 moltype = DNA length = 6339
 FEATURE Location/Qualifiers
 source 1..6339
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 organism = synthetic construct

SEQUENCE: 15

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The invention claimed is:

1. A composition that comprises a recombinant plasmid (RP) a sequence of nucleotides that encode micro-interfering ribonucleic acid (miRNA) that binds to and inactivates and/or degrades messenger ribonucleic acid (mRNA) that encodes for a serotonin receptor, wherein the sequence of nucleotides comprises 95-100% the same nucleotide sequence as SEQ ID NO. 11.

2. The composition of claim 1, wherein the sequence of nucleotides is configured to be delivered to a target cell that has expressed the serotonin receptor.

3. The composition of claim 1, wherein the sequence of nucleotides is encased in a protein coat, a lipid vesicle, or any combination thereof.

4. The composition of claim 1, wherein the sequence of nucleotides is encased in a viral vector.

5. The composition of claim 4, wherein the viral vector is one of a double stranded DNA virus, a single stranded DNA virus, a single stranded RNA virus, or a double stranded RNA virus.

6. The composition of claim 4, wherein the viral vector is an adeno-associated virus.

7. The composition of claim 1 wherein the serotonin receptor is serotonin receptor 5HT4.

8. A composition that comprises a recombinant plasmid (RP) with a sequence of nucleotides for encoding a sequence of micro-interfering ribonucleic acid (miRNA) that binds to and degrades and/or inactivates messenger ribonucleic acid (mRNA) that encodes for a serotonin receptor, wherein the sequence of nucleotides comprises 95-100% of the same nucleotide sequence as SEQ ID NO. 23.

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