CLAIMS

WHAT IS CLAIMED IS:

1. A recombinant adeno-associated viral (rAAV) vector comprising at least a first polynucleotide that comprises a photoreceptor-specific human rhodopsin kinase (hGRK1) promoter~~, or a ubiquitous truncated chimeric CMV-chicken β-actin promoter,~~ operably linked to at least a first nucleic acid segment that encodes at least a first ~~mammalian~~ biologically-active, retinal-specific human guanylate cyclase protein, peptide, or polypeptide that comprises a first contiguous amino acid sequence region that is at least about 90% identical to an at least 80-contiguous-amino-acid sequence from SEQ ID NO:1.

~~2. The rAAV vector according to claim 1, wherein the at least a first nucleic acid segment encodes at least a first mammalian guanylate cyclase protein, peptide, or polypeptide that comprises at least a first contiguous amino acid sequence region that is at least about 90% identical to at least a first sequence region of at least 80 contiguous amino acids of a sequence as set forth in SEQ ID NO:1 , 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, or SEQ ID NO:11.~~

~~3. The rAAV vector according to claim 1 or claim 2, wherein the at least a first nucleic acid segment encodes at least a first mammalian guanylate cyclase protein, peptide, or polypeptide that comprises at least a first contiguous amino acid sequence region that is at least about 92% identical to at least a first sequence region of at least 100 contiguous amino acids of a sequence as set forth in SEQ ID NO: 1, 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, or SEQ ID NO: 11.~~

~~4.~~ 2. The rAAV vector according to ~~any preceding claim~~ claim 1, wherein the at least a first ~~mammalian~~ biologically-active, retinal-specific human guanylate cyclase protein, peptide, or polypeptide that comprises at least a first contiguous amino acid sequence region that is at least about 95% identical to at least a first sequence region of at least

120 contiguous amino acids of a sequence as set forth in SEQ ID NO:1~~, 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, or SEQ ID NO: 11~~.

~~5. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide comprises at least a first contiguous amino acid sequence region that is at least about 98% identical to at least a first sequence region of at least 140 contiguous amino acids of a sequence as set forth in SEQ ID NO:1, 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, or SEQ ID NO:11.~~

~~6. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide comprises at least an amino acid sequence that is at least about 98% identical to the amino acid sequence of any one of SEQ ID NO: 1, 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, or SEQ ID NO:11.~~

~~7. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide comprises the amino acid sequence of SEQID NO:1, SEQID NO:2, SEQ ID NO:3, SEQID NO:4, SEQID NO:5, SEQID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO: 10, or SEQ ID NO: 11.~~

~~8. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide consists essentially of an amino acid sequence that is at least 98% identical to the sequence of SEQ ID NO: 1, SEQ ID NO:2, SEQID NO:3, SEQ ID NO:4, SEQID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO: 10, or SEQ ID NO: 11.~~

~~9. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide is biologically-active~~

~~and comprises an at least 50 amino acid contiguous sequence from SEQ ID NO: 1, SEQ ID NO:2, SEQID NO:3, SEQID 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, or SEQ ID NO:11.~~

~~10. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide is biologically-active and comprises an at least 75 amino acid contiguous sequence from SEQID NO:1, SEQ ID NO:2, SEQID 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, or SEQ ID NO:11.~~

~~11. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide is biologically-active and comprises an at least 100 amino acid contiguous sequence from SEQ ID NO:1, 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, or SEQ ID NO:11.~~

~~12. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide is biologically-active and comprises an at least 125 amino acid contiguous sequence from SEQ ID NOT , 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, or SEQ ID NO:11.~~

~~13. The rAAV vector according to any preceding claim, wherein the mammalian guanylate cyclase protein, peptide, or polypeptide is biologically-active and comprises a primary amino acid sequence that is at least 95% identical to the primary amino acid sequence from one or more of SEQ ID NO:1, 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, or SEQ ID NO:11.~~

~~14.~~ 3. The rAAV vector according to ~~any preceding claim~~ claim 1 or claim 2, wherein the ~~mammalian~~ at least a first biologically-active, retinal-specific guanylate cyclase protein, peptide, or polypeptide ~~is biologically-active and~~ comprises a contiguous primary amino acid sequence that is at least 99% identical to the primary amino acid

sequence ~~from one or more~~ of SEQ ID NO:1 ~~, 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, or SEQ ID NO:11~~.

4. The rAAV vector according to any preceding claim, wherein the at least a first biologically-active retinal-specific human guanylate cyclase protein, peptide, or polypeptide comprises a first contiguous amino acid sequence region that comprises the amino acid sequence of SEQ ID NO:1.

~~15. The rAAV vector according to any preceding claim, wherein the rAAV vector is a recombinant adeno-associated virus serotype 1 (rAAV1) vector, a recombinant adeno-associated virus serotype 2 (rAAV2) vector, recombinant adeno-associated virus serotype 3 (rAAV3) vector, a recombinant adeno-associated virus serotype 4 (rAAV4) vector, a recombinant adeno-associated virus serotype 5 (rAAV5) vector, a recombinant adeno-associated virus serotype 6 (rAAV6) vector, a recombinant adeno-associated virus serotype 7 (rAAV7) vector, a recombinant adeno-associated virus serotype 8 (rAAV8) vector, or a recombinant adeno-associated virus serotype 9 (rAAV) vector.~~

~~16.~~ 5. The rAAV vector according to any preceding claim, wherein the vector is a self-complementary rAAV (scAAV).

~~17. The rAAV vector according to any preceding claim, wherein the photoreceptor-specific human rhodopsin kinase promoter comprises a nucleic acid sequence that consists essentially of an at least 30 contiguous base pair sequence from SEQ ID NO:12, or the truncated chimeric CMV-chicken ß-actin promoter comprises a nucleic acid sequence that consists essentially of an at least 70 contiguous basepair sequence from SEQ ID NO:13.~~

~~18. The rAAV vector according to any preceding claim, wherein the photoreceptor-specific human rhodopsin kinase promoter comprises a nucleic acid sequence that consists essentially of an at least 40 contiguous basepair sequence from SEQ ID NO:12, or the truncated chimeric CMV-chicken ß-actin promoter comprises a nucleic~~

~~acid sequence that consists essentially of an at least 90 contiguous basepair sequence from SEQ ID NO:13.~~

~~19. The rAAV vector according to any preceding claim, wherein the photoreceptor-specific human rhodopsin kinase promoter comprises a nucleic acid sequence that consists essentially of an at least 60 contiguous basepair sequence from SEQ ID NO:12, or the truncated chimeric CMV-chicken ß-actin promoter comprises a nucleic acid sequence that consists essentially of an at least 120 contiguous basepair sequence from SEQ ID NO:13.~~

~~20. The rAAV vector according to any preceding claim, wherein the photoreceptor-specific human rhodopsin kinase promoter comprises the nucleic acid sequence of SEQ ID NO:12, or the truncated chimeric CMV-chicken ß-actin promoter comprises the nucleic acid sequence of SEQ ID NO:13~~.

~~21.~~ 6. The rAAV vector according to any preceding claim, further comprising at least a first enhancer or at least a first mammalian intron sequence operably linked to the at least a first nucleic segment.

~~22. The rAAV vector according to any preceding claim, further comprising at least a first mammalian intron sequence operably linked to the at least a first nucleic segment.~~

~~23. The rAAV vector according to any preceding claim, wherein the at least a first mammalian guanylate cyclase protein, peptide, or polypeptide is of human, non-human primate, murine, feline, canine, porcine, ovine, bovine, equine, epine, caprine, or lupine origin.~~

~~24. The rAAV vector according to any preceding claim, wherein the at least a first nucleic acid segment encodes a biologically-active human guanylate cyclase protein, peptide, or polypeptide ofhuman origin.~~

~~25. The rAAV vector according to any preceding claim, wherein the truncated chimeric CMV-chicken ß-actin promoter comprises a nucleic acid sequence that consists essentially of the sequence of SEQ ID NO:12, or wherein the at least a first nucleic acid segment encodes a mammalian guanylate cyclase protein, peptide, or polypeptide that comprises an at least 100 contiguous amino acid sequence from SEQ ID NO:1.~~

~~26.~~ 7. The rAAV vector according to any preceding claim, comprised within an infectious adeno-associated viral particle, virion, or a plurality of infectious AAV particles.

~~27.~~ 8. A virion or an infectious viral particle comprising the rAAV vector in accordance with any one of claims 1 to ~~26~~ 7.

~~28.~~ 9. A plurality of infectious viral particles prepared from the rAAV vector in accordance with any one of claims 1 to 26 7.

~~29.~~ 10. An isolated mammalian host cell comprising:

(a) the rAAV vector in accordance with any one of claims 1 to ~~26~~ 7;

(b) the virion or infectious viral particle in accordance with claim ~~27~~ 8; or

(c) the plurality of infectious viral particles in accordance with claim ~~28~~ 9.

~~30.~~ 11. The isolated mammalian host cell according to claim ~~27~~ 10, wherein the cell is a human host cell.

~~31.~~ 12. A composition comprising:

(1) (a) the rAAV vector in accordance with any one of claims 1 to ~~26~~ 7;

(b) the virion or infectious viral particle m accordance with claim ~~27~~ 8;

(c) the plurality of infectious viral particles in accordance with claim ~~28~~ 9; or

(d) an isolated mammalian host cell comprising the vector, virion, or infectious particles; and

(2) a pharmaceutically-acceptable buffer, carrier, vehicle, or diluent.

~~32.~~ 13. The composition according to claim ~~31~~ 12, further comprising a lipid, a liposome, a lipid complex, an ethosome, a niosome, a nanoparticle, a microparticle, a liposphere, a nanocapsule, or any combination thereof.

~~33.~~ 14. The composition according to claim ~~31~~ 12 or claim ~~32~~ 13, formulated for administration to the human eye.

~~34.~~ 15. Use of the ~~The~~ composition according to any one of claims ~~31 to 33~~ 12 to 14 ~~, for use~~ in therapy or prophylaxis.

~~35.~~ 16. Use of the ~~The~~ composition according to any one of claims ~~31 to 34~~ 12 to 14~~, for use~~ in the therapy or prophylaxis of a human retinal dystrophy, disease, or disorder.

~~36.~~ 17. Use of the ~~The~~ composition according to any one of claims ~~31 to 54~~ 12 to 14~~, for use~~ in the therapy or prophylaxis of Leber congenital amaurosis-1 (LCA1).

~~37.~~ 18. A therapeutic kit comprising:

(1) a component selected from the group consisting of:

(a) the rAAV vector in accordance with any one of claims 1 to ~~26~~ 7;

(b) the virion or infectious viral particle in accordance with claim ~~27~~ 8;

(c) the plurality of infectious viral particles in accordance with claim ~~28~~ 9;

(d) the isolated mammalian host cell in accordance with claim ~~29~~ 10 or claim ~~30~~ 11; or

(e) the composition in accordance with any one of claims ~~31~~ 12 to ~~36~~ 15; and

(2) instructions for using the component in the diagnosis, prevention, treatment, or amelioration of one or more symptoms of ~~a retinal dystrophy, disease, disorder, or abnormal condition~~ Leber congenital amaurosis-1 (LCA-1) in a human.

~~38. The kit according to claim 35, wherein the retinal dystrophy is diagnosed as Leber congenital amaurosis-1 (LCA-1).~~

~~39. Use of a composition in accordance with claim 31, in the manufacture of a medicament for diagnosing, preventing, treating or ameliorating a disease, disorder, dysfunction, or abnormal condition of a mammalian eye, or one or more symptoms thereof.~~

~~40.~~ 19. Use of a composition according to ~~claim 39~~ claim 12 in the manufacture of a medicament for treating Leber congenital amaurosis-1 (LCA-1) in a human or ameliorating one or more symptoms thereof ~~of Leber congenital amaurosis-1 (LCA-1) in a human~~.

~~41. A method for preventing, treating or ameliorating a disease, dysfunction, disorder, deficiency, or abnormal condition in a mammal, or one or more symptoms thereof, the method comprising administering to the mammal a composition comprising the rAAV vector in accordance with any one of claims 1 to 26, in an amount and for a time sufficient to prevent, treat or ameliorate the disease, dysfunction, disorder, deficiency, or abnormal condition in the mamma1, or one or more symptoms thereof.~~

~~42. The method according to claim 41, wherein the mammal has, is suspected of having, is at risk for developing, or has been diagnosed with at least a first retinal disorder, disease, or dystrophy.~~

~~43. The method according to claim 41 or claim 42, wherein the mammal is at risk for developing, or has been diagnosed with a deficiency in biologically-active retinal guanylate cyclase protein, peptide, or polypeptide.~~

~~44. The method according to any one of claims 41 to 43, wherein the mammal is a human neonate, newborn, infant, or juvenile that is at risk for developing or has been diagnosed with a congenital retinal dystrophy such as Leber congenital amaurosis-1 (LCA-1).~~

~~45. A method for providing a mammal with a therapeutically-effective amount of a biologically-active mammalian guanylate cyclase peptide, polypeptide, or protein to a mammal in need thereof, comprising introducing into suitable cells of a mammal in need thereof, an effective amount of the rAAV vector in accordance with any one of claims 1 to 26, for a time sufficient to produce a biologically-active guanylate cyclase peptide, polypeptide or protein therefrom in at least a first population of cells or at least a first tissue of the mammal into which the rAAV vector has been introduced.~~

~~46. The method according to claim 45, wherein the mammal in need thereof has a defect, deficiency, or substantial absence of biologically-active retGCI protein in at least a first tissue of the mammal, when compared to the level of biologically-active retGCI protein in a normal mammal.~~

~~47. The method according to claim 45 or claim 46, wherein a plurality of cells from the mammal are provided with the rAAV vector ex-vivo or in vitro, and the method further comprises introducing the plurality of provided cells into at least a first tissue site within or about the body of the mammal.~~

~~48. The method according to any one of claims 45 to 47, wherein the plurality of obtained cells is introduced into at least a first site within one or both eyes of the mammal.~~

~~49. The method according to any one of claims 45 to 48, wherein the plurality of obtained cells is introduced into at least a first site within one or both eyes of the mammal by direct injection into the retina or the surrounding tissue.~~

~~50. The method according to any one of claims 45 to 49, wherein introduction of the rAAV vector preserved cone photoreceptors, and restored cone-mediated function and visual behavior in the eye of the mammal.~~

~~51. The method according to any one of claims 45 to 50, wherein a single introduction of the rAAV vector into the eye of the mammal preserved cone photoreceptors, and restored cone-mediated function and visual behavior in the mammal for a period of at least three months.~~

~~52. The method according to any one of claims 45 to 51, wherein a single introduction of the rAAV vector into the eye of the mammal preserved cone photoreceptors, and restored cone-mediated function and visual behavior in the mammal for a period of at least six months.~~

~~53. The method according to any one of claims 45 to 52, wherein a single introduction of the rAAV vector into the eye of the mammal preserved cone photoreceptors, and restored cone-mediated function and visual behavior in the mammal for a period of at least ten months.~~

~~54. A method for increasing the level of biologically-active retGCI protein in one or more retinal cells of a mammal that has, is suspected of having, is diagnosed with, or is at risk for developing, LCAI, comprising introducing the rAAV vector in accordance with any one of claims 1 to 26, into at least a first population of retinal cells of the mammal, in an amount and for a time effective to increase the level of biologically-active retGCI protein in one or more retinal cells of the mammal.~~

~~55. A method for treating or ameliorating one or more symptoms of retinal dystrophy in a mammal, the method comprising directly administering to the retina or sub-retinal space of the mammal an rAAV vector in accordance with any one of claims 1 to 26, in an amount and for a time sufficient to treat or ameliorate the one or more symptoms of retinal dystrophy in the mammal.~~

~~56. The method according to claim 55, wherein the mammal has been diagnosed with Leber congenital amaurosis-1 (LCA-1).~~

CLAIMS

WHAT IS CLAIMED IS:

1. A recombinant adeno-associated viral (rAAV) vector comprising at least a first polynucleotide that comprises a photoreceptor-specific human rhodopsin kinase (hGRK1) promoter operably linked to at least a first nucleic acid segment that encodes at least a first biologically-active, retinal-specific human guanylate cyclase protein, peptide, or polypeptide that comprises a first contiguous amino acid sequence region that is at least about 90% identical to an at least 80-contiguous-amino-acid sequence from SEQ ID NO:1.

2. The rAAV vector according to claim 1, wherein the at least a first biologically-active, retinal-specific human guanylate cyclase protein, peptide, or polypeptide that comprises at least a first contiguous amino acid sequence region that is at least about 95% identical to at least a first sequence region of at least 120 contiguous amino acids of a sequence as set forth in SEQ ID NO:1.

3. The rAAV vector according to claim 1 or claim 2, wherein the at least a first biologically-active, retinal-specific guanylate cyclase protein, peptide, or polypeptide comprises a contiguous primary amino acid sequence that is at least 99% identical to the primary amino acid sequence of SEQ ID NO:1.

4. The rAAV vector according to any preceding claim, wherein the at least a first biologically-active retinal-specific human guanylate cyclase protein, peptide, or polypeptide comprises a first contiguous amino acid sequence region that comprises the amino acid sequence of SEQ ID NO:1.

5. The rAAV vector according to any preceding claim, wherein the vector is a self-complementary rAAV (scAAV).

6. The rAAV vector according to any preceding claim, further comprising at least a first enhancer or at least a first mammalian intron sequence operably linked to the at least a first nucleic segment.

7. The rAAV vector according to any preceding claim, comprised within an infectious adeno-associated viral particle, virion, or a plurality of infectious AAV particles.

8. A virion or an infectious viral particle comprising the rAAV vector in accordance with any one of claims 1 to 7.

9. A plurality of infectious viral particles prepared from the rAAV vector in accordance with any one of claims 1 to 7.

10. An isolated mammalian host cell comprising:

(a) the rAAV vector in accordance with any one of claims 1 to 7;

(b) the virion or infectious viral particle in accordance with claim 8; or

(c) the plurality of infectious viral particles in accordance with claim 9.

11. The isolated mammalian host cell according to claim 10, wherein the cell is a human host cell.

12. A composition comprising:

(1) (a) the rAAV vector in accordance with any one of claims 1 to 7;

(b) the virion or infectious viral particle in accordance with claim 8;

(c) the plurality of infectious viral particles in accordance with claim 9; or

(d) an isolated mammalian host cell comprising the vector, virion, or infectious particles; and

(2) a pharmaceutically-acceptable buffer, carrier, vehicle, or diluent.

13. The composition according to claim 12, further comprising a lipid, a liposome, a lipid complex, an ethosome, a niosome, a nanoparticle, a microparticle, a liposphere, a nanocapsule, or any combination thereof.

14. The composition according to claim 12 or claim 13, formulated for administration to the human eye.

15. Use of the composition according to any one of claims 12 to 14 in therapy or prophylaxis.

16. Use of the composition according to any one of claims 12 to 14 in the therapy or prophylaxis of a human retinal dystrophy, disease, or disorder.

17. Use of the composition according to any one of claims 12 to 14 in the therapy or prophylaxis of Leber congenital amaurosis-1 (LCA1).

18. A therapeutic kit comprising:

(1) a component selected from the group consisting of:

(a) the rAAV vector in accordance with any one of claims 1 to 7;

(b) the virion or infectious viral particle in accordance with claim 8;

(c) the plurality of infectious viral particles in accordance with claim 9;

(d) the isolated mammalian host cell in accordance with claim 10 or claim ~~30~~ 11; or

(e) the composition in accordance with any one of claims 12 to 15; and

(2) instructions for using the component in the diagnosis, prevention, treatment, or amelioration of one or more symptoms of Leber congenital amaurosis-1 (LCA-1) in a human.

19. Use of a composition according to claim 12 in the manufacture of a medicament for treating Leber congenital amaurosis-1 (LCA-1) in a human or ameliorating one or more symptoms thereof.