

[0010a] In one aspect, the present disclosure provides a composition for treating a muscular dystrophy in a human subject comprising a recombinant adeno-associated virus (rAAV), wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, and wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard. In yet another aspect, the present disclosure provides a composition for increasing localization of alpha-sarcoglycan to a cell membrane in a human subject in need thereof, wherein the composition comprises a recombinant adeno-associated virus (rAAV), wherein the rAAV comprises a nucleotide sequence of SEQ ID NO: 19, and wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard. In yet another aspect, the present disclosure provides a composition for increasing sarcoglycan expression in muscle tissue of a human subject, wherein the composition comprises a rAAV comprising a nucleotide sequence encoding a first sarcoglycan, wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard, wherein administration of the composition results in increased expression of at least a second sarcoglycan in the cell membrane of the cell expressing said first sarcoglycan.

[0010b] In a further aspect, the present disclosure provides a composition, comprising an rAAV suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard, wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, a buffer agent, an ionic strength agent, and a surfactant, wherein the surfactant comprises a poloxamer at a concentration of 0.00001% to 1%. In a further aspect, the present disclosure provides a polynucleotide sequence comprising a nucleotide sequence that is at least 90%, 95%, or 99% identical to SEQ ID NO: 24.

[0010c] In a further aspect, the present disclosure provides a method of generating a recombinant AAV, comprising transferring to a cell the polynucleotide sequence as described herein. In a further aspect, the present disclosure provides a cell, comprising the polynucleotide sequence as described herein.

[0010d] In a further aspect, the present disclosure provides an rAAV comprising the nucleotide sequence of SEQ ID NO: 19.

[0010e] In a further aspect, the present disclosure provides use of a recombinant adeno-associated virus (rAAV) for the preparation of a medicament for treating muscular dystrophy in a human subject in need thereof, wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard.

[0011] In one example, described herein is a method of treating muscular dystrophy in a subject in need thereof comprising the step of administering a recombinant adeno-associated virus (rAAV) scAAVrh74.MHCK7.hSGCB to a subject in need thereof, wherein the rAAV is administered using a systemic route of administration and at a dose of about 1.0×10^{12} vg/kg to about 5.0×10^{14} vg/kg based on a supercoiled plasmid as the quantitation standard or 1.0×10^{13} vg/kg to about 1.0×10^{14} vg/kg based on a linearized plasmid as the quantitation standard; wherein the serum creatine kinase (CK) level in the subject is decreased after administration of the rAAV as compared to serum CK level before administration of the rAAV.

[0012] In another example, provided is a method of treating muscular dystrophy in a subject in need thereof comprising the step of administering a recombinant adeno-associated virus (rAAV) scAAVrh74.MHCK7.hSGCB, wherein the level of beta-sarcoglycan gene expression in a cell of the subject is increased after administration of the rAAV as compared to the level of beta-sarcoglycan gene expression before administration of the rAAV; wherein the number of beta-sarcoglycan positive fibers in the muscle tissue of the subject is increased after administration of the rAAV as compared to the number of beta-sarcoglycan positive fibers before administration of the rAAV; or wherein motor function is improved in said subject as compared to the motor function of said subject before administration of the rAAV, and wherein the motor function is determined by a 100 meter timed walk test.

[0013] In another example, this disclosure provides a method of treating a limb-girdle muscular dystrophy in a subject in need, comprising administering to the subject an rAAV intravenous infusion over approximately 1 to 2 hours at a dose of about 5.0×10^{13} vg/kg or

Claims

What is claimed is:

1. A composition for treating a muscular dystrophy in a human subject comprising a recombinant adeno-associated virus (rAAV), wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, and wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard.
2. The composition of claim 1, wherein the muscular dystrophy is limb-girdle muscular dystrophy.
3. The composition of claim 1 or 2, wherein the muscular dystrophy is limb-girdle muscular dystrophy type 2E.
4. A composition for increasing localization of alpha-sarcoglycan to a cell membrane in a human subject in need thereof, wherein the composition comprises a recombinant adeno-associated virus (rAAV), wherein the rAAV comprises a nucleotide sequence of SEQ ID NO: 19, and wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard.
5. A composition for increasing sarcoglycan expression in muscle tissue of a human subject, wherein the composition comprises a rAAV comprising a nucleotide sequence encoding a first sarcoglycan, wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard, wherein administration of the composition results in increased expression of at least a second sarcoglycan in the cell membrane of the cell expressing said first sarcoglycan.
6. The composition of claim 5, wherein said first sarcoglycan is β -sarcoglycan (SGCB), and said second sarcoglycan is α -sarcoglycan (SGCA), γ -sarcoglycan (SGCG), and/or γ -sarcoglycan (SGCD).
7. The composition of any one of claims 1 to 6, wherein the human subject is 4 to 15 years of age.

8. The composition of any one of claims 1 to 6, wherein the human subject is a pediatric subject, an adolescent subject or a young adult subject.
9. The composition of any one of claims 1 to 6, wherein the human subject is 25 to 55 years of age.
10. The composition of any one of claims 1 to 6, wherein the human subject is over 50 years of age.
11. The composition of any one of claims 1-10, wherein the composition is formulated for administration by infusion over approximately 1-2 hours.
12. The composition of any one of claims 1-11, wherein the rAAV is of a serotype selected from AAV-1, AAV-2, AAV-3, AAV-4, AAV-5, AAV-6, AAV-7, AAV-8, AAV-9, AAV-10, AAV-11, AAV-12, AAV-13, or AAV rh.74, or a variant thereof.
13. The composition of claim 12, wherein the rAAV serotype is serotype AAVrh.74.
14. A composition, comprising
an rAAV suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard, wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19,
a buffer agent,
an ionic strength agent, and
a surfactant, wherein the surfactant comprises a poloxamer at a concentration of 0.00001% to 1%.
15. The composition of claim 14, wherein the Poloxamer comprises one or more of Poloxamer 124, Poloxamer 181, Poloxamer 184, Poloxamer 188, Poloxamer 237, Poloxamer 331, Poloxamer 338, and Poloxamer 407.
16. A polynucleotide sequence comprising a nucleotide sequence that is at least 90%, 95%, or 99% identical to SEQ ID NO: 24.
17. The polynucleotide sequence of claim 16, wherein the polynucleotide sequence comprises SEQ ID NO: 24.
18. The polynucleotide sequence of claim 16 or 17, is a plasmid.

19. A method of generating a recombinant AAV, comprising transferring to a cell the polynucleotide sequence of any one of claims 16 to 18.
20. The method of claim 19, further comprising transferring a packaging plasmid and/or a helper virus to the cell.
21. The method of claim 19 or 20, wherein the cell comprises a stably integrated AAV cap gene.
22. The method of any one of claims 19 to 21, wherein the cell comprises a stably integrated AAV rep gene.
23. A cell comprising the polynucleotide sequence of any one of claims 16 to 18.
24. The cell of claim 23, wherein the cell is an insect cell, a silkworm cell, a mosquito cell, a bacterial cell or a mammalian cell.
25. The cell of claim 24, wherein the cell is a HEK293 cell, a COS cell, a HeLa cells, or a KB cell.
26. Use of a recombinant adeno-associated virus (rAAV) for the preparation of a medicament for treating muscular dystrophy in a human subject in need thereof, wherein the rAAV comprises the nucleotide sequence of SEQ ID NO: 19, wherein the rAAV is suitable for administration at a dose of about 7.41×10^{13} vg/kg based on a linearized plasmid as the quantitation standard.
27. An rAAV comprising the nucleotide sequence of SEQ ID NO: 19.