Claims

1. A recombinant adeno-associated virus rh.74 (rAAVrh.74) comprising a polynucleotide which comprises a first AAV inverted terminal repeat (ITR), a promoter, a nucleotide sequence comprising the nucleotide sequence of SEQ ID NO: 2 and a second AAV ITR.

2. The rAAVrh.74 of claim 1 wherein the promoter is a muscle-specific promoter.

3. The rAAVrh.74 of claim 2, wherein the muscle-specific promoter comprises one or more of a human skeletal actin gene element, a cardiac actin gene element, a desmin promoter, a skeletal alpha-actin (ASKA) promoter, a troponin I (TNNI2) promoter, a myocyte-specific enhancer binding factor (MEF) binding element, a muscle creatine kinase (MCK) promoter, a truncated MCK (tMCK) promoter, a myosin heavy chain (MHC) promoter, a hybrid a-myosin heavy chain enhancer-/MCK enhancer-promoter (MHCK7) promoter, a C5-12 promoter, a murine creatine kinase enhancer element, a skeletal fast-twitch troponin C gene element, a slow-twitch cardiac troponin C gene element, a slow-twitch troponin I gene element, hypoxia- inducible nuclear factor (HIF)-response element (HRE), a steroid-inducible element, and a glucocorticoid response element (GRE).

4. The rAAVrh.74 of claim 3, wherein the muscle-specific promoter is an MCK promoter, a tMCK promoter, or an MHCK7 promoter.

5. The rAAVrh.74 of claim 3, wherein the muscle-specific promoter is a truncated MCK promoter comprising the nucleotide sequence of SEQ ID NO: 3.

6. The rAAVrh.74 of any one of claims 1-5, wherein the first and second AAV inverted terminal repeats are AAV2 inverted terminal repeats.

7. A recombinant adeno-associated virus rh.74 (rAAVrh.74) comprising a polynucleotide which comprises a first AAV inverted terminal repeat (ITR), a promoter comprising the nucleotide sequence of SEQ ID NO: 3, a nucleotide sequence comprising the nucleotide sequence of SEQ ID NO: 2 encoding a protein with calpain 3 (CAPN3) activity and a second AAV ITR.

8. A composition comprising the rAAVrh.74 of any one of claims 1-7.

9. A polynucleotide for making an rAAV comprising a sequence which is at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identical to SEQ ID NO: 1.

10. The polynucleotide of claim 9, wherein the polynucleotide comprises a sequence at least 95% identical to SEQ ID NO: 1.

11. The polynucleotide of claim 9 or 10, wherein the polynucleotide comprises the sequence of SEQ ID NO: 1.

12. A composition for use in treating limb girdle muscular dystrophy 2A, wherein the composition comprises a therapeutically effective amount of the rAAVrh.74 of any one of claims 1-7 or the composition of claim 8.

13. The composition of claim 12, wherein the treatment with the composition results in one or more of:

(a) an increased muscle fiber diameter,

(b) a decreased number of small lobulated muscle fibers,

(c) a decreased number of fibers with internal nuclei,

(d) a decreased endomysial connective tissue content,

(e) correction of muscle atrophy, and

(f) an increased muscle force generation.

14. The composition of claim 13, wherein the muscle fiber comprises one or more of slow twitch oxidative (STO) muscle fiber, fast twitch oxidative (FTO) muscle fiber, and fast twitch glycolytic (FTG) fiber.

15. The composition of any one of claims 12-14, wherein the treatment with the composition results in one or more of:

(a) at least a 5%, 10%, 15%, 20%, 25%, 30%, or 35%, or 40% decrease of total muscle fiber number per mm^2 by 4 weeks after administration;

(b) at least a 5%, 10%, 15%, 20%, or 25% increase of muscle fiber diameter by 4 weeks after administration;

(c) at least a 5%, 10%, 15%, 20%, 25%, 30%, 35%, or 42% decrease of STO muscle fiber number per mm^2 by 4 weeks after administration;

(d) at least a 5%, 10%, 15%, 20%, or 25% increase of STO muscle fiber diameter by 4 weeks after administration;

(e) at least a 5%, 10%, 15%, or 20% decrease of FTO muscle fiber number per mm^2 by 4 weeks after administration;

(f) at least a 5%, 10%, 15%, or 20% increase of FTO muscle fiber diameter by 4 weeks after administration;

(g) at least a 5%, 10%, 15%, 20%, 25%, 30%, or 35% decrease of FTG muscle fiber number per mm^2 by 4 weeks after administration; and

(h) at least a 5%, 10%, 15%, 20%, or 25% increase of FTG muscle fiber diameter by 4 weeks after administration.

16. The composition of any one of claims 12-15, wherein the composition is formulated for administration by intramuscular injection or intravenous injection.

17. The composition of any one of claims 12-15, wherein after treatment with the composition, the heart muscle of the subject shows minimum or low calpain 3 protein expressed from the rAAVrh.74.

18. Use of a therapeutically effective amount of the rAAV of any one of claims 1-7 or the composition of claim 8 in the preparation of a medicament for the treatment of limb girdle muscular dystrophy 2A.

19. The use of claim 18, wherein the treatment with the medicament results in one or more of:

(a) an increased muscle fiber diameter,

(b) a decreased number of small lobulated muscle fibers,

(c) a decreased number of fibers with internal nuclei,

(d) a decreased endomysial connective tissue content,

(e) correction of muscle atrophy, and

(f) an increased muscle force generation.

20. The use of claim 19, wherein the muscle fiber comprises one or more of slow twitch oxidative (STO) muscle fiber, fast twitch oxidative (FTO) muscle fiber, and fast twitch glycolytic (FTG) fiber.

21. The use of claim 19, wherein the treatment with the medicament results in one or more of:

(a) at least a 5%, 10%, 15%, 20%, 25%, 30%, or 35%, or 40% decrease of

total muscle fiber number per mm^2 by 4 weeks after administration;

(b) at least a 5%, 10%, 15%, 20%, or 25% increase of muscle fiber diameter by 4 weeks after administration;

(c) at least a 5%, 10%, 15%, 20%, 25%, 30%, 35%, or 42% decrease of STO muscle fiber number per mm^2 by 4 weeks after administration;

(d) at least a 5%, 10%, 15%, 20%, or 25% increase of STO muscle fiber diameter by 4 weeks after administration;

(e) at least a 5%, 10%, 15%, or 20% decrease of FTO muscle fiber number per mm^2 by 4 weeks after administration;

(f) at least a 5%, 10%, 15%, or 20% increase of FTO muscle fiber diameter by 4 weeks after administration;

(g) at least a 5%, 10%, 15%, 20%, 25%, 30%, or 35% decrease of FTG muscle fiber number per mm^2 by 4 weeks after administration; and

(h) at least a 5%, 10%, 15%, 20%, or 25% increase of FTG muscle fiber diameter by 4 weeks after administration.

22. The use of claim 18, wherein the medicament is formulated for administration by intramuscular injection or intravenous injection.

23. The use of claim 18, wherein after treatment with the medicament, the heart muscle of the subject shows minimum or low calpain 3 protein expressed from the rAAV of any one of claims 1-7 or the composition of claim 8.