Claims

1. A pharmaceutical composition for treating human subjects diagnosed with Mucopolysaccharidosis II (MPS II) or Hunter Syndrome, comprising a suspension of replication deficient recombinant adeno-associated virus (rAAV) in a formulation buffer, wherein:

a) the rAAV comprises an AAV9 capsid and a vector genome, wherein the vector genome comprises an AAV 5' inverted terminal repeat (ITR), an expression cassette, and an AAV 3' ITR, wherein the expression cassette comprises a heterologous nucleic acid encoding human iduronate-2-sulfatase (hIDS) operably linked to expression control sequences which direct expression thereof, wherein the expression control sequences comprise a CB7 promoter, a chicken beta actin intron, and a rabbit beta-globin polyadenylation (polyA) signal sequence, wherein the hIDS coding sequence has the nucleic acid of SEQ ID NO: 1, and wherein said vector genome is packaged in the AAV9 capsid;

b) the formulation buffer is a intrathecal formulation buffer which comprises a physiologically compatible aqueous buffer, and surfactants and excipients; and

c) (i) the composition comprises an rAAV Genome Copy (GC) titer of at least 1.0 x 10^{13} GC/ml (+/-20%); or

(ii) the rAAV is to be administered to the human subject in a dose of at least 4.0 x 10^{8} GC/g brain mass to 4.0 x 10^{11} GC/g brain mass.

2. The pharmaceutical composition according to claim 1, wherein the composition is to be administered intrathecally.

3. The pharmaceutical composition according to claim 1 or claim 2, wherein the suspension is an aqueous liquid suspension having a pH in the range of 6 to 9 which comprises an aqueous suspension base and wherein the rAAV comprises a vector genome which comprises nucleotide 2 to nucleotide 3967 of SEQ ID NO: 3.

4. A suspension of replication deficient recombinant adeno-associated virus (rAAV) in a formulation buffer at a dose between 4.0 x 10^{8} GC/g brain mass to 4.0 x 10^{11} GC/g brain mass for use in treating a human subject diagnosed with Mucopolysaccharidosis II (MPS II), wherein the suspension is to be administered to the human subject in need thereof by intrathecal injection, wherein:

a) the rAAV comprises an AAV9 capsid and a vector genome, wherein the vector genome comprises an AAV 5' inverted terminal repeat (ITR), an expression cassette, and an AAV 3' ITR, wherein the expression cassette comprises a heterologous nucleic acid encoding human iduronate-2-sulfatase (hIDS) operably linked to expression control sequences which direct expression thereof, wherein the expression control sequences comprise a CB7 promoter, a chicken beta actin intron, and a rabbit beta-

globin polyadenylation (polyA) signal sequence, wherein the hIDS coding sequence has the nucleic acid of SEQ ID NO: 1, and wherein said vector genome is packaged in the AAV9 capsid;

b) an intrathecal formulation buffer which comprises a physiologically compatible aqueous buffer, a surfactant and excipients; and

c) (i) the suspension comprises an rAAV Genome Copy (GC) titer of at least 1.0 x 10^{13} GC/ml (+/-20%); or

(ii) the rAAV is to be administered to the human subject in a dose of at least 4.0 x 10^{8} GC/g brain mass to 4.0 x 10^{11} GC/g brain mass.

5. The suspension of claim 4, wherein the human subject is age 2 years or older and diagnosed with severe Hunter syndrome, or wherein the human subject has neurocognitive deficits or is at risk of developing neurocognitive deficits.

6. The suspension of claim 4 or claim 5 wherein administration results in an increase in neurocognitive developmental quotient (DQ) in said subject, as assessed using Bayley Scales of Infant Development, or wherein the administration results in a decline of DQ of no more than 15 points in said subject relative to untreated/natural history control data in patients with Hunter Syndrome, or wherein the administration results in an increase in functional hIDS levels, as measured in a serum sample from said patient.

7. The suspension of any one of claims 4 to 6, wherein administration results in a decrease in GAG levels, as measured in a sample of the patient's serum, urine and/or cerebrospinal fluid (CSF).

8. The suspension of any one of claims 4 to 7, further wherein an AAV.hIDS is to be administered to said patient by liver-directed injection,

wherein the liver-directed AAV.hIDS has a capsid selected from AAV8, AAVrh64R1, AAVrh64R2, rh8, rh10, AAV3B, or AAVdj.

9. The pharmaceutical composition of claim 1, wherein potency is measured by an in vitro assay, optionally wherein the in vitro assay comprises transducing HEK293 cells with a known multiplicity of the rAAV GC titer per cell and assaying the supernatant for hIDS activity 72 hours post-transduction using the 4MU-iduronide-2-sulfate enzymatic assay.

10. The pharmaceutical composition of any one of claims 1, 2, or 9, wherein the AAV ITRs are heterologous to AAV9, or wherein the ITRs are from AAV2.

11. The pharmaceutical composition of any one of claims 1, 2, 9 to 10, wherein the suspension has a pH of 6 to 9, optionally wherein the suspension has a pH of 6.8 to 7.8.

12. The pharmaceutical composition of any one of claims 1, 2, 9 to 11, wherein the suspension is formulated for delivery via intrathecal injection.

13. The pharmaceutical composition of any one of claims 1, 2, 9 to 12, wherein the suspension is formulated for delivery to newborn patients and comprises 3.8×10^{12} genome copies (GC) to 1.9×10^{14} GC, or to patients that are about 3 months to about 9 months of age and comprises 6×10^{12} GC to 3×10^{14} GC, or to patients that are about 9 months to about 36 months of age and comprises 1×10^{13} GC to 5×10^{14} GC, or to patients that are about 3 years to about 12 years of age and comprises 1.2×10^{13} GC to 6×10^{14} GC, or to patients that are about 12 years of age or older and comprises 1.4×10^{13} GC to 7×10^{14} GC.

14. A recombinant adeno-associated-virus comprising a hIDS gene (rAAV.hIDS) wherein the hIDS gene is operably linked to expression control sequences which direct expression thereof, wherein the expression control sequences comprise a CB7 promoter, a chicken beta actin intron, and a rabbit beta-globin polyadenylation (polyA) signal sequence, wherein the hIDS gene comprises a nucleic acid sequence of SEQ ID NO: 1 or a sequence at least 80% identical to SEQ ID NO:1 which encodes a functional hIDS, for use in a method of treating a human patient having MPS II or Hunter Syndrome, wherein

a) a sufficient amount of hIDS enzyme to induce transgene-specific tolerance is to be dosed to a patient having MPS II or Hunber Syndrome; and

b) an rAAV.hIDS which rAAV.hIDS directs expression of therapeutic levels of hIDS is to be administered to the patient.

15. The rAAV.hIDS of claim 14, wherein the hIDS of (a) is to be dosed as a recombinant protein.

16. The rAAV.hIDS of claim 14, wherein the administering (b) is performed about three days to 14 days post-dosing (a).

17. A kit for intrathecal administration comprising the suspension of any one of claims 4-8, optionally further comprising a dilution buffer useful for diluting the suspension.