

NEUROD1 AND DLX2 VECTOR**CROSS-REFERENCE TO RELATED APPLICATIONS**

[0001] This application claims the benefit of U.S. Provisional Application No. 63/084,945, filed September 29, 2020, and U.S. Provisional Application No. 63/247,442, filed September 23, 2021,
5 both of which are incorporated by reference in their entirety herein.

INCORPORATION OF SEQUENCE LISTING

[0002] A sequence listing contained in the file named "P34838WO00_SL.TXT" which is 28,311 bytes (measured in MS-Windows®) and created on September 27, 2021, is filed electronically herewith and incorporated by reference in its entirety.

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FIELD OF THE INVENTION

[0003] The present disclosure includes methods and compositions using an AAV vector comprising a nucleic acid sequence encoding human NeuroD1 and Dlx2 to convert glial cells to neurons.

BACKGROUND OF THE INVENTION

15 [0004] Neurons are often killed or damaged and unable to regenerate in subjects with a neurological condition or following an injury to the central nervous system (CNS) or peripheral nervous system (PNS).

[0005] Glial cells become reactive following an injury to the CNS or PNS such as a brain injury or neurological condition.

20 [0006] Currently there are no methods available to regenerate functional new neurons in human subjects having a neurological condition using adeno-associated viruses (AAVs).

SUMMARY OF THE INVENTION

[0007] In one aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the
25 nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, where the hNeuroD1 sequence and the hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal
30 ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 sequence and the hDlx2 sequence are operably linked to regulatory elements comprising: (a) glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid

sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0008] In one aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where the hNeuroD1 coding sequence and the hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 coding sequence and the hDlx2 coding sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0009] In an aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a neurogenic differentiation 1 (NeuroD1) nucleic acid coding sequence encoding a NeuroD1 protein and a distal-less homeobox 2 (Dlx2) nucleic acid coding sequence encoding a Dlx2 protein, where the NeuroD1 coding sequence and the Dlx2 coding sequence are

separated by a linker sequence, where the NeuroD1 coding sequence and the Dlx2 coding sequence are operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal sequence.

- 5 **[0010]** In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector for converting glial cells to functional neurons in a human, where the AAV vector comprises a human neurogenic differentiation 1 (hNeuroD1) sequence having a nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence having a nucleic acid sequence of SEQ ID NO: 13, where the hNeuroD1 sequence and the hDlx2
- 10 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 sequence and hDlx2 sequence are operably linked to regulatory elements comprising:
- 15 (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group
- 20 consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the
- 25 nucleic acid sequence of SEQ ID NO: 30.

- [0011]** In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated-virus (AAV) vector for converting glial cells to functional neurons in a human, where the AAV vector comprises a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and
- 30 a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where the hNeuroD1 coding sequence and the hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and
- 35 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence

comprising SEQ ID NO: 3, where the hNeuroD1 coding sequence and the hDlx2 coding sequence are operably linked to regulatory elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0012] In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector for the treatment of a subject in need thereof, where the AAV vector comprises a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, where the NeuroD1 sequence and the Dlx2 sequence are separated by a linker sequence, where the NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal.

[0013] In an aspect, this disclosure provides, and includes, a method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, where said AAV comprises a DNA vector construct comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, where said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic

acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0014] In an aspect, this disclosure provides, and includes, a method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, where said AAV comprises a DNA vector construct comprising a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 coding sequence and hDlx2 coding sequence are operably linked to expression control elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0015] In an aspect, this disclosure provides, and includes, a method of converting glial cells to neurons in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject in need thereof, where said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, where said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, where said

NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising: (a) a glial fibrillary acid protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal sequence, where said vector is capable of converting at least one glial cell to a neuron in said subject in need thereof.

[0016] In an aspect, this disclosure provides, and includes, a method of treating a neurological condition in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject, where said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and distal-less homeobox 2 (Dlx2) sequence, where said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, where said NeuroD1 sequence and said Dlx2 sequence are operably linked to expression control elements comprising: (a) a glial fibrillary acid protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal to said subject in need thereof.

[0017] In an aspect, this disclosure provides, and includes, a composition comprising (i) an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6, and (ii) an adeno-associated virus (AAV) vector comprising a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13; where the hNeuroD1 sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0018] In an aspect, this disclosure provides, and includes, a composition comprising (i) an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6, and (ii) an adeno-associated virus (AAV) vector comprising a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13; where the hNeuroD1 sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c)

a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

- 5 **[0019]** In an aspect, this disclosure provides, and includes, a composition comprising (i) an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10, and (ii) an adeno-associated virus (AAV) vector comprising a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14; where the nucleic acid sequence encoding an hNeuroD1 protein is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

- [0020]** In an aspect, this disclosure provides, and includes, a composition comprising (i) an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10, and (ii) an adeno-associated virus (AAV) vector comprising a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14; where the nucleic acid sequence encoding an hNeuroD1 protein is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

- [0021]** In an aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, where said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence

selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 sequence and said hDlx2 sequence are operably linked to
5 regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the
10 nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0022] In an aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence
15 comprising the nucleic acid sequence of SEQ ID NO: 13, where said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising
20 SEQ ID NO: 3, where said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus
25 posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0023] In an aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1
30 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker
35 comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and

19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) an
 5 enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

10 **[0024]** In an aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid
 15 sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 coding sequence and said hDlx2 coding
 20 sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the
 25 nucleic acid sequence of SEQ ID NO: 29; and (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[0025] In an aspect, this disclosures provides, and includes, an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence
 30 comprising the nucleic acid sequence of SEQ ID NO: 13, where said hNeuroD1 sequence and said hDlx2 sequence are separated by a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18 or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements
 35 comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid

sequence of SEQ ID NO: 26; (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; and (d) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

- 5 [0026] In an aspect, this disclosures provides, and includes, an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18 or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26; (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27; and (d) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

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BRIEF DESCRIPTION OF THE DRAWINGS

- [0027] Figure 1A depicts a map of aCE:Gfa681:NeuroD1:P2A:Dlx2:WPRE:SV40.
 [0028] Figure 1B depicts a map of a EF-1 α : Gfa681:NeuroD1:P2A:Dlx2:WPRE:SV40.
 [0029] Figure 1C depicts a map of a CE:Gfa681:NeuroD1:P2A:Dlx2:WPRE:hGH
 [0030] Figure 1D depicts a map of EF-1 α : Gfa681:NeuroD1:P2A:Dlx2:WPRE:hGH.
 25 [0031] Figure 2A depicts a map of a CE: Gfa681 NeuroD1:GSG-P2A:Dlx2:WPRE:SV40.
 [0032] Figure 2B depicts a map of a EF-1 α : Gfa681 NeuroD1:GSG-P2A:Dlx2:WPRE:SV40.
 [0033] Figure 2C depicts a map of a CE: Gfa681 NeuroD1:GSG-P2A:Dlx2:WPRE:hGH.
 [0034] Figure 2D depicts a map of a EF-1 α : Gfa681 NeuroD1:GSG-P2A:Dlx2:WPRE:hGH.
 [0035] Figure 3A depicts a map of a CE: Gfa681 NeuroD1:T2A:Dlx2:WPRE:SV40
 30 [0036] Figure 3B depicts a map of a EF-1 α : Gfa681 NeuroD1:T2A:Dlx2:WPRE:SV40.
 [0037] Figure 3C depicts a map of a CE: Gfa681 NeuroD1:T2A:Dlx2:WPRE:hGH.
 [0038] Figure 3D depicts a map of a EF-1 α : Gfa681 NeuroD1:T2A:Dlx2:WPRE:hGH.
 [0039] Figure 4A depicts a map of a CE: Gfa681 NeuroD1:GSG-T2A:Dlx2:WPRE:SV40
 [0040] Figure 4B depicts a map of a EF-1 α : Gfa681 NeuroD1:GSG-T2A:Dlx2:WPRE:SV40.
 35 [0041] Figure 4C depicts a map of a CE: Gfa681 NeuroD1:GSG-T2A:Dlx2:WPRE:hGH.

- [0042] Figure 4D depicts a map of a EF-1 α : Gfa681: NeuroD1: GSG- T2A: Dlx2: WPRE: hGH.
- [0043] Figure 5A depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: P2A: Dlx2: SV40.
- [0044] Figure 5B depicts a map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: P2A: Dlx2: SV40.
- [0045] Figure 5C depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: GSG- P2A: Dlx2: SV40.
- 5 [0046] Figure 5D depicts a map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: GSG- P2A: Dlx2: SV40.
- [0047] Figure 6A depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: P2A: Dlx2: hGh.
- [0048] Figure 6B depicts a map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: P2A: Dlx2: hGh.
- [0049] Figure 6C depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: GSG- P2A: Dlx2: hGh.
- 10 [0050] Figure 6D depicts a map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: GSG- P2A: Dlx2: hGh.
- [0051] Figure 7A depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: T2A: Dlx2: SV40.
- [0052] Figure 7B depicts a map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: T2A: Dlx2: SV40.
- [0053] Figure 7C depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: GSG- T2A: Dlx2: SV40.
- [0054] Figure 7D depicts a map of a EF-1 α : Gfa681: NeuroD1: GSG- T2A: Dlx2: SV40.
- 15 [0055] Figure 8A depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: T2A: Dlx2: hGh.
- [0056] Figure 8B depicts a map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: T2A: Dlx2: hGh.
- [0057] Figure 8C depicts a map of a U6: shRNA: CE: Gfa681: NeuroD1: GSG- T2A: Dlx2: hGh.
- [0058] Figure 8D depicts a vector map of a U6: shRNA: EF-1 α : Gfa681: NeuroD1: GSG- T2A: Dlx2: hGh.
- 20 [0059] Figure 9 measures AAV virus production of the P31 plasmid. Titer analysis is performed using gene of interest (GOI) primers, ITR region primers, and reverse packaging primers. Virus titer is calculated as vg/cell.
- [0060] Figure 10 depicts establishment of rat astrocyte primary culture from 3 day post-natal Sprague-Dawley rat brains. Upper left panel presents an image of GFAP stained cells. Upper
- 25 right panel presents an image of SOX9 stained cells. Lower left panel presents an image of DAPI stained cells. Lower right panel presents a merge image of GFAP, SOX9, and DAPI stained cells.
- [0061] Figure 11 depicts comparison of NeuroD1 expression efficiency of plamids. Primary rat astrocyte cells are transfected with either the P14 (CE: GfaABC1D: hNeuroD1- P2A- Dlx2: WPRE: SV40), P31 (EF-1 α : GfaABC1D: NeuroD1- P2A- Dlx2: WPRE: SV40), and P63
- 30 (CE: GfaABC1D: NeuroD1- GSG P2A- Dlx2: WPRE: SV40). Top panels show NeuroD1 staining of cells, middle panels show Dlx2 staining of cells and bottom panels show merged NeuroD1, Dlx2 and DAPI staining of cells.
- [0062] Figure 12 depicts comparison of AAV virus particle transduction at different doses using AAV9-P12 (pGfaABC1D: GFP). Upper left panel shows a dose of 3×10^{10} vg/well. Upper right
- 35 panel shows a dose of 1×10^{10} vg/well. Lower panel shows a dose of 2.5×10^9 vg/well.

[0063] Figure 13A and 13B depict quantitative analysis of AAV particle transduction into primary rate astrocytes. Figure 13A presents the percentage transduction rate of AAV9-P12 (pGfaABC1D:GFP) and AAV5-P7 (pEF-1 α :GFP) at MOIs of 5×10^5 vg/cell, 2×10^5 vg/cell, and 5×10^4 vg/cell. Figure 13B presents the percentage transduction rate of AAV9-P12 (pGfaABC1D:GFP) in cells seeded at a series of densities of 2×10^4 cell/well, 1.5×10^4 cell/well, 1×10^4 cell/well, and 5×10^3 cell/well and infected with virus at a series of amounts of 2 μ l, 1 μ l, 0.5 μ l, 0.25 μ l, 0.125 μ l of 1×10^{13} vg/ml virus in 100 μ l of medium.

[0064] Figure 14 depicts RCAs three weeks post transduction with control plasmid AAV9-P21 (CE-pGFA681-CI-GFP-WPRE-SV40pA) at 2×10^{10} vg/ml. Cells are immunostained with antibodies against neuronal markers NeuN and MAP2, and with DAPI (nuclear stain). GFP fluorescence indicates the presence of cells transduced with the control plasmid.

[0065] Figure 15 depicts RCAs immunostained with an anti-NeuroD1(ND1) antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P134 (CE-pGfa681-CRGI-hND1-oPRE-bGHpA).

[0066] Figure 16 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 6 days post transduction with AAV9-P134 (CE-pGfa681-CRGI-hND1-oPRE-bGHpA) at 2×10^{10} vg/ml.

[0067] Figure 17 depicts RCAs immunostained with anti-NeuN and anti-MAP2 antibodies and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P134 (CE-pGfa681-CRGI-hND1-oPRE-bGHpA) at 2×10^{10} vg/ml. Transduction with the ND1-containing vector generates neurons (NeuN//MAP2+) from the astrocyte culture.

[0068] Figure 18 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P138 (EE-pGfa681-CRGI-hND1-oPRE-bGHpA).

[0069] Figure 19 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 6 days post transduction with AAV9-P138 (EE-pGfa681-CRGI-hND1-oPRE-bGHpA) at 2×10^{10} vg/ml).

[0070] Figure 20 depicts RCAs immunostained with anti-NeuN and anti-MAP2 antibodies and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P138 (EE-pGfa681-CRGI-hND1-oPRE-bGHpA) at 2×10^{10} vg/ml). Transduction with the ND1-containing vector generates neurons (NeuN//MAP2+) from the astrocyte culture.

[0071] Figure 21 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 6 days post transduction with AAV9-P9 (CE-pGfa681-CI-hND1-p2A-GFP-WPRE-SV40pA) at 2×10^{10} vg/ml. GFP fluorescence indicates presence of transduced cells.

[0072] Figure 22 depicts RCAs immunostained with anti-NeuN and anti-MAP2 antibodies and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P9 (CE-pGfa681-CI-hND1-p2A-

GFP-WPRE-SV40pA) at 2×10^{10} vg/ml. Transduction with the ND1-containing vector generates neurons (NeuN//MAP2+) from the astrocyte culture.

[0073] Figure 23 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P22 (CE-pGfa681-CI-hND1-WPRE-SV40pA).

5 [0074] Figure 24 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 6 days post transduction with AAV9-P22 (CE-pGfa681-CI-hND1 WPRE-SV40pA) at 2×10^{10} vg/ml.

[0075] Figure 25 depicts RCAs immunostained with anti-NeuN and anti-MAP2 antibodies and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P22 (CE-pGfa681-CI-hND1-WPRE-SV40pA) at 2×10^{10} vg/ml. Transduction with the ND1-containing vector generates neurons (NeuN//MAP2+) from the astrocyte culture.

[0076] Figure 26 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P35 (EE-pGfa681-CI-hND1-WPRE-SV40pA).

[0077] Figure 27 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 6 days post transduction with AAV9-P35 (EE-pGfa681-CI-hND1 WPRE-SV40pA) at 2×10^{10} vg/ml.

[0078] Figure 28 depicts RCAs immunostained with an anti-NeuN antibody and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P35 (EE-pGfa681-CI-hND1-WPRE-SV40pA) at 2×10^{10} vg/ml. Transduction with the ND1-containing vector generates neurons (NeuN+) from the astrocyte culture.

[0079] Figure 29 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P107 (CE-pGfa681-CI-hND1-bGHpA).

[0080] Figure 30 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P108 (CE-pGfa681-CI-hND1-oPRE-bGHpA).

25 [0081] Figure 31 depicts RCAs immunostained with an anti-ND1 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P109 (CE-pGfa681-CRGI-hND1-bGHpA).

[0082] Figure 32 depicts the brain cortex tissue of mice infected with AAV9-P12 (P12 control group), AAV9-P12 + AAV9-P134 (P134 group), and AAV9-P12 + AAV9-P138 (P138 group) at 10 days post infection (dpi).

30 [0083] Figure 33 depicts the brain cortex tissue of mice infected with AAV9-P12 + AAV9-P134 (P134 group), and AAV9-P12 + AAV9-P138 (P138 group) at 30 days post infection (dpi).

[0084] Figure 34 depicts the brain cortex tissue of mice (bilateral injury model) infected with AAV9-P12 (P12 control group), and AAV9-P12 + AAV9-P134 (P134 group) at 10 dpi.

[0085] Figure 35 is a plot of measurements of AAV virus production of the P134, P130, P138 and P21 plasmids. Titer analysis is performed by qPCR using primers amplifying gene of interest (GOI) and primers specific to the plasmids. Virus yield is calculated as vg/cell.

[0086] Figure 36 depicts the brain cortex tissue of mice infected with AAV9-P12 (P12 control group), AAV9-P12 + AAV9-P134 (P134 group), and AAV9-P12 + AAV9-P138 (P138 group) at 10 days post infection (dpi). Cells are immunostained with antibodies against NeuroD1, GFAP, NeuN, and with DAPI (nuclear stain). GFP fluorescence indicates the presence of cells infected with the control virus.

[0087] Figure 37 depicts the brain cortex tissue of mice infected with AAV9-P12 (P12 control group), AAV9-P12 + AAV9-P134 (P134 group), and AAV9-P12 + AAV9-P138 (P138 group) at 30 days post infection (dpi). Cells are immunostained with antibodies against NeuroD1, GFAP, NeuN, and with DAPI (nuclear stain). GFP fluorescence indicates the presence of cells infected with the control virus.

[0088] Figure 38 depicts the brain cortex tissue of mice (bilateral injury model) infected with AAV9-P12 (P12 control group), and AAV9-P12 + AAV9-P134 (P134 group) at 10 dpi. Cells are immunostained with antibodies against NeuroD1, GFAP, NeuN, and with DAPI (nuclear stain). GFP fluorescence indicates the presence of cells infected with the control virus.

[0089] Figure 39A-B depicts the brain cortex tissue of mice (bilateral injury model) infected with AAV9-P12 (P12 control group), and AAV9-P12 + AAV9-P134 (P134 group) at 30 dpi. Figure 39A depicts the cells that are immunostained with antibodies against NeuroD1, GFAP, NeuN, and with DAPI (nuclear stain). GFP fluorescence indicates the presence of cells infected with the control virus. Figure 39B is a quantification of the glial cell-to-neuron conversion rate at 30 dpi.

[0090] Figure 40 depicts two general maps of ND1 and Dlx2 constructs. Enhancer refers to an efla enhancer or CMV enhancer. pGfa refers to a 2.2 kb or 1.6 kb Gfa promoter. poly(A) signal refers to SV40, bGH or an hGH poly(A) signal.

[0091] Figure 41 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P112 (CE-pGfa681-CI-hDlx2-IRES-hND1-bGHpA).

[0092] Figure 42 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 6 days post transduction with AAV9-P112 (CE-pGfa681-CI-hDlx2-IRES-hND1-bGHpA) at 2×10^{10} vg/ml.

[0093] Figure 43 depicts RCAs immunostained with an anti-NeuN antibody, an anti-MAP2 antibody, and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P112 (CE-pGfa681-CI-hDlx2-IRES-hND1-bGHpA) at 2×10^{10} vg/ml.

- [0094] Figure 44 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P122 (CE-pGfa681-CI-hDlx2-P2A-hND1-bGHpA).
- 5 [0095] Figure 45 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 6 days post transduction with AAV9-P122 (CE-pGfa681-CI-hDlx2-p2A-hND1-bGHpA) at 2×10^{10} vg/ml.
- [0096] Figure 46 depicts RCAs immunostained with an anti-NeuN antibody, an anti-MAP2 antibody, and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P122 (CE-pGfa681-CI-hDlx2-P2A-hND1-bGHpA) at 2×10^{10} vg/ml.
- 10 [0097] Figure 47 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P124 (CE-pGfa681-CI-hND1-P2A-hDlx2-bGHpA).
- [0098] Figure 48 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 6 days post transduction with AAV9-P124 (CE-pGfa681-CI-hND1-p2A-hDlx2-bGHpA) at 2×10^{10} vg/ml.
- 15 [0099] Figure 49 depicts RCAs immunostained with an anti-NeuN antibody, an anti-MAP2 antibody, and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P124 (CE-pGfa681-CI-hND1-P2A-hDlx2-bGHpA) at 2×10^{10} vg/ml.
- [00100] Figure 50 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P20 (CE-pGfa681-CI-hND1-P2A-hDlx2-WPRE-SV40pA).
- 20 [00101] Figure 51 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 6 days post transduction with AAV9-P20 (CE-pGfa681-CI-hDND1-p2A-hDlx2-WPRE-SV40pA) at 2×10^{10} vg/ml.
- 25 [00102] Figure 52 depicts RCAs immunostained with an anti-NeuN antibody, an anti-MAP2 antibody, and DAPI (nuclear stain) 3 weeks post transduction with AAV9-P20 (CE-pGfa681-CI-hND1-P2A-hDlx2-bSV40pA) at 2×10^{10} vg/ml.
- [00103] Figure 53 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P31 (EE-pGfa681-CI-hND1-P2A-hDlx2-WPRE-SV40pA).
- 30 [00104] Figure 54 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P123 (EE-pGfa681-CI-hDlx2-p2A-hND1-bGHpA).

[00105] Figure 55 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P113 (EE-pGfa681-CI-hDlx2-IRES-hND1-bGHpA).

[00106] Figure 56 depicts RCAs immunostained with an anti-ND1 antibody, an anti-Dlx2 antibody, and DAPI (nuclear stain) 24 hours post transfection with NXL-P111 (CE-pGfa681-CI-hDlx2-p2A-hND1-SV40pA).

[00107] Figure 57A-B depicts the brain striatum tissue of mice infected with AAV9-P12 (virus) at 10 dpi and 30 dpi. Figure 57A shows wide infection of mouse striatum by AAV-P12 at 10 dpi as demonstrated by the GFP fluorescence. Figure 57B shows infection of the mouse by AAV9-P12 at 30 dpi. The cells are immunostained with antibodies against GFAP and NeuN.

[00108] Figure 58 depicts the brain striatum tissue of mice co-infected with AAV9-P12 and AAV9-P112 (the P112 group) at 10 dpi. GFP fluorescence identifies the AAV9-P112-infected cells. The cells are immunostained with antibodies against GFAP and NeuN.

[00109] Figure 59 depicts the brain striatum tissue of mice co-infected with AAV9-P12 and AAV9-P112 (the P112 group) at 30 dpi. GFP fluorescence identifies the AAV9-P112-infected cells. The cells are immunostained with antibodies against GFAP and NeuN. The white arrows indicate cells co-infected with AAV9-P12 and AAV9-P112 which became NeuN-positive neurons.

[00110] Figure 60 depicts the brain striatum tissue of mice co-infected with AAV9-P12 and AAV9-P122 (the P122 group) at 10 dpi. GFP fluorescence identifies the AAV9-P112-infected cells. The cells are immunostained with antibodies against GFAP and NeuN.

[00111] Figure 61 depicts the brain striatum tissue of mice co-infected with AAV9-P12 and AAV9-P122 (the P122 group) at 30 dpi. GFP fluorescence identifies the AAV9-P112-infected cells. The cells are immunostained with antibodies against GFAP and NeuN.

[00112] Figure 62 depicts rat cortical astrocytes (RCAs) immunostained with an anti-Dlx2 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P104 (CE-pGfa681-CGRI-Dlx2-bGHpA) or NXL-P105 (CE-pGfa681-CI-Dlx2-oPRE-bGHpA).

[00113] Figure 63 depicts rat cortical astrocytes (RCAs) immunostained with an anti-Dlx2 antibody and DAPI (nuclear stain) 24 hours post transfection with NXL-P133 (EE-pGfa681-CGRI-Dlx2-oPRE-bGHpA), NXL-P137 (EE-pGfa681-CGRI-Dlx2-oPRE-bGHpA), or NXL-P131 (EE-pGfa681-CI-Dlx2-oPRE-bGHpA).

[00114] Figure 64 depicts rat cortical astrocytes (RCAs) immunostained with an anti-Dlx2 antibody and DAPI (nuclear stain) after transduction with AAV9-P133 (CE-pGfa681-CGRI-Dlx2-oPRE-bGHpA).

BRIEF DESCRIPTION OF SEQUENCES

[00115] A listing of nucleic acid sequences and amino acid sequences is provided in Table

1.

5 **Table 1.** Nucleic acid sequences

SEQ ID NO	Sequence Description	Sequence Type	Sequence
1	Upstream AAV2 ITR	Nucleic acid	TGCAGGCAGCTGCGCGCTCGCTCGCTCACTGAGGCCGC CCGGGCGTCGGGCGACCTTTGGTTCGCCCCGGCCTCAGTG AGCGAGCGAGCGCGCAGAGAGGGAGTGGCCAACTCCA TCACTAGGGGTTCTT
2	Efla enhancer	Nucleic acid	TGCAAAGATGGATAAAGTTTTAAACAGAGAGGAATCTT TGCAGCTAATGGACCTTCTAGGTCTTGAAAGGAGTGGG AATTGGCTCCGGTGCCCGTCAGTGGGCAGAGCGCACAT CGCCACAGTCCCCGAGAAGTTGGGGGGAGGGGTCGGC A
3	Internal Ribosomal Entry Sequence (IRES)	Nucleic acid	ACGTTACTGGCCGAAGCCGCTTGAATAAGGCCGGTGT GCGTTTGTCTATATGTTATTTTCCACCATATTGCCGTCTT TTGGCAATGTGAGGGCCCGGAAACCTGGCCCTGTCTTCT TGACGAGCATTCTAGGGGTCTTTCCCTCTCGCCAAAG GAATGCAAGGTCTGTTGAATGTCGTGAAGGAAGCAGTT CCTCTGGAAGCTTCTTGAAGACAAACAACGTCTGTAGC GACCTTTGTCAGGCAGCGGAACCCCCACCTGGCGACA GGTGCCTCTGCGGCCAAAAGCCACGTGTATAAGATACA CCTGCAAAGGCGGCACAACCCACGTGCCACGTTGTGAG TTGGATAGTTGTGGAAAGAGTCAAATGGCTCTCCTCAA GCGTATTCAACAAGGGGCTGAAGGATGCCCAGAAGGTA CCCCATTGTATGGGATCTGATCTGGGGCCTCGGTGCACA TGCTTTACATGTGTTTAGTCGAGGTTAAAAAACGTCTA GGCCCCCGAACCACGGGGACGTGGTTTTCCTTTGAAA AACACGATGATAATA
4	Gfa1.6 promoter	Nucleic acid	CTGCAAGCAGACCTGGCAGCATTGGGCTGGCCGCCCCC CAGGGCCTCCTCTTCATGCCCAGTGAATGACTCACCTTG GCACAGACACAATGTTTCGGGGTGGGCACAGTGCCTGCT TCCCGCCGCACCCCAGCCCCCTCAAATGCCTTCCGAGA AGCCCATGAGTAGGGGGCTTGCAATTGCACCCACGCCT GACAGCCTGGCATCTTGGGATAAAAGCAGCACAGCCCC CTAGGGGCTGCCCTTGCTGTGTGGCGCCACCGGCGGTG GAGAACAAGGCTCTATTCAGCCTGTGCCCAGGAAAGGG GATCAGGGGATGCCCAGGCATGGACAGTGGGTGGCAG GGGGGGAGAGGAGGGCTGTCTGCTTCCAGAAGTCCAA GGACACAAATGGGTGAGGGGACTGGGCAGGGTTCTGAC CCTGTGGGACCAGAGTGGAGGGCGTAGATGGACCTGAA GTCTCCAGGGACAACAGGGGCCAGGTCTCAGGCTCCTA GTTGGGCCAGTGGCTCCAGCGTTTCCAAACCCATCCAT CCCCAGAGGTTCTTCCCATCTCTCCAGGCTGATGTGTGG GAACTCGAGGAAATAAATCTCCAGTGGGAGACGGAGG GGTGGCCAGGGAAACGGGGCGCTGCAGGAATAAAGAC

SEQ ID NO	Sequence Description	Sequence Type	Sequence
			GAGCCAGCACAGCCAGCTCATGCGTAACGGCTTTGTGG AGCTGTCAAGGCCTGGTCTCTGGGAGAGAGGCACAGGG AGGCCAGACAAGGAAGGGGTGACCTGGAGGGACAGAT CCAGGGGCTAAAGTCCTGATAAGGCAAGAGAGTGCCGG CCCCCTCTTGCCCTATCAGGACCTCCACTGCCACATAGA GGCCATGATTGACCCTTAGACAAAGGGCTGGTGTCCAA TCCCAGCCCCCAGCCCCAGAACTCCAGGGAATGAATGG GCAGAGAGCAGGAATGTGGGACATCTGTGTTCAAGGGA AGGACTCCAGGAGTCTGCTGGGAATGAGGCCCTAGTAGG AAATGAGGTGGCCCTTGAGGGTACAGAACAGGTTTCATT CTTCGCCAAATTCCCAGCACCTTGCAGGCACTTACAGCT GAGTGAGATAATGCCTGGGTTATGAAATCAAAAAGTTG GAAAGCAGGTCAGAGGTCATCTGGTACAGCCCTTCCTT CCCTTTTTTTTTTTTTTTTTTTTTTGTGAGACAAGGTCTCTCT CTGTTGCCCAGGCTGGAGTGGCGCAAACACAGCTCACT GCAGCCTCAACCTACTGGGCTCAAGCAATCCTCCAGCC TCAGCCTCCCAAAGTGCTGGGATTACAAGCATGAGCCA CCCCACTCAGCCCTTTCCTTCCTTTTAATTGATGCATAA TAATTGTAAGTATTCATCATGGTCCAACCAACCCTTTCT TGACCCACCTTCCTAGAGAGAGGGTCCTCTTGATTCAGC GGTCAGGGCCCCAGACCCATGGTCTGGCTCCAGGTACC ACCTGCCTCATGCAGGAGTTGGCGTGCCAGGAAGCTC TGCCTCTGGGCACAGTGACCTCAGTGGGGTGAGGGGAG CTCTCCCCATAGCTGGGCTGCGGCCCAACCCACCCCT CAGGCTATGCCAGGGGGTGTGGCCAGGGGCACCCGGGC ATCGCCAGTCTAGCCCACTCCTTCATAAAGCCCTCGCAT CCCAGGAGCGAGCAGAGCCAGAG
5	Chimeric Intron	Nucleic acid	GTAAGTATCAAGGTTACAAGACAGGTTTAAGGAGACCA ATAGAAACTGGGCTTGTCGAGACAGAGAAGACTCTTGC GTTTCTGATAGGCACCTATTGGTCTTACTGACATCCACT TTGCCTTTCTCTCCACAG
6	hND1 (human NeuroD1)	Nucleic acid	ATGACCAAATCGTACAGCGAGAGTGGGCTGATGGGCGA GCCTCAGCCCCAAGGTCCTCCAAGCTGGACAGACGAGT GTCTCAGTTCTCAGGACGAGGAGCACGAGGCAGACAAG AAGGAGGACGACCTCGAAGCCATGAACGCAGAGGAGG ACTCACTGAGGAACGGGGGAGAGGAGGAGGACGAAGA TGAGGACCTGGAAGAGGAGGAAGAAGAGGAAGAGGAG GATGACGATCAAAAAGCCCAAGAGACGCGGCCCAAAA AGAAGAAGATGACTAAGGCTCGCCTGGAGCGTTTTAAA TTGAGACGCATGAAGGCTAACGCCCGGGAGCGGAACCG CATGCACGGAAGTGAACGCGGCGCTAGACAACCTGCGCA AGGTGGTGCCTTGCTATTCTAAGACGCAGAAGCTGTCC AAAATCGAGACTCTGCGCTTGGCCAAGAATACTACATCTG GGCTCTGTCGGAGATCCTGCGCTCAGGCAAAAGCCCAG ACCTGGTCTCCTTCGTTTCAGACGCTTTGCAAGGGCTTAT CCCAACCCACCACCAACCTGGTTGCGGGCTGCCTGCAA CTCAATCCTCGGACTTTTCTGCCTGAGCAGAACCAGGAC ATGCCCCCCCCACCTGCCGACGGCCAGCGCTTCCTTCCCT GTACACCCCTACTCCTACCAGTCGCCTGGGCTGCCAGT CCGCCTTACGGTACCATGGACAGCTCCCATGTCTTCCAC

SEQ ID NO	Sequence Description	Sequence Type	Sequence
			GTTAAGCCTCCGCCGCACGCCTACAGCGCAGCGCTGGA GCCCTTCTTTGAAAGCCCTCTGACTGATTGCACCAGCCC TTCCTTTGATGGACCCCTCAGCCCGCCGCTCAGCATCAA TGGCAACTTCTCTTTCAAACACGAACCGTCCGCCGAGTT TGAGAAAAATTATGCCTTTACCATGCACTATCCTGCAGC GACACTGGCAGGGGCCAAAGCCACGGATCAATCTTCT CAGGCACCGCTGCCCCTCGCTGCGAGATCCCCATAGAC AATATTATGTCTTCGATAGCCATTACATCATGAGCGA GTCATGAGTGCCAGCTCAATGCCATATTTTCATGAT
7	WPRE (Woodchuck Hepatitis Virus Posttranscrip- tional Regulatory Element)	Nucleic acid	AATCAACCTCTGGATTACAAAATTTGTGAAAGATTGAC TGGTATTCTTAACTATGTTGCTCCTTTTACGCTATGTGG ATACGCTGCTTTAATGCCTTTGTATCATGCTATTGCTTCC CGTATGGCTTTTCATTTTCTCCTCCTTGATAAATCCTGGT TGCTGTCTCTTTATGAGGAGTTGTGGCCCGTTGTCAGGC AACGTGGCGTGGTGTGCACTGTGTTTGTGACGCAACC CCCACTGGTTGGGGCATTGCCACCACCTGTCAGCTCCTT TCCGGGACTTTCGCTTTCCCCCTCCCTATTGCCACGGCG GAACTCATCGCCGCCTGCCTTGCCCGCTGCTGGACAGG GGCTCGGCTGTTGGGCACTGACAATTCCGTGGTGTGTGTC GGGGAAATCATCGTCCTTTCTTGCTGCTCGCCTGTGT TGCCACCTGGATTCTGCGCGGGACGTCTTCTGCTACGT CCCTTCGGCCCTCAATCCAGCGGACCTTCCTTCCCCGCGG CCTGCTGCCGGCTCTGCGGCCTCTTCCGCGTCTTCGCCT TCGCCCTCAGACGAGTCGGATCTCCCTTTGGGCCGCCCTC CCCGC
8	SV40 poly(A) signal	Nucleic acid	CGATCCACCGGATCTAGATAACTGATCATAATCAGCCA TACCACATTTGTAGAGGTTTTACTTGCTTTAAAAAACCT CCCACACCTCCCCCTGAACCTGAAACATAAAATGAATG CAATTGTTGTTGTTAACTTGTTTATTGCAGCTTATAATG GTTACAAATAAAGCAATAGCATCACAAATTCACAAAT AAAGCATTTTTTCTACTGCATTCTAGTTGTGGTTGTCC AAATCATCAATGTATCTTA
9	Downstream AAV2 ITR	Nucleic acid	AGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCTCTGC GCGCTCGCTCGCTCACTGAGGCCGGGCGACCAAAGGTC GCCCAGCGCCCGGGCTTTGCCCGGGCGGCCTCAGTGAG CGAGCGAGCGCGCAGCTGCCTGCA
10	hND1 (human NeuroD1)	Amino Acid	MTKSYSESGLMGEPQPQGPSSWTDECLSSQDEEHEADKK EDDLEAMNAEEDSLRNGGEEDEDEDELEEEEEEEEEEDDD QKPKRRGPKKKKMTKARLERFKLRRMKANARERNRMHG LNAALDNLKVVPCYSKTQKLSKIETLRLAKNYIWALSEI LRSGKSPDLVSFVQTLCKGLSQPTTNLVAGCLQLNPRTFLP EQNQDMPPHLPTASASFPVHPYSYQSPGLSPPYGTMDSS HVFHVKPPPHAYSAALEPFFESPLTDCTSPSFDGPLSPPLSI NGNFSFKHEPSAEFEKNYAFTMHYPAATLAGAQSHGSIFS GTAAPRCEIPIDNIMSFDSHSHHERVMSAQLNAIFHD
11	CMV enhancer ("CE")	Nucleic Acid	GACATTGATTATTGACTAGTTATTAATAGTAATCAATTA CGGGGTCATTAGTTCATAGCCCATATATGGAGTTCCGCG TTACATAACTTACGGTAAATGGCCCGCCTGGCTGACCG CCCAACGACCCCCGCCCATGACGTCAATAATGACGTA

SEQ ID NO	Sequence Description	Sequence Type	Sequence
			TGTTCCCATAGTAACGCCAATAGGGACTTTCCATTGACG TCAATGGGTGGACTATTTACGGTAAACTGCCCACTTGGC AGTACATCAAGTGTATCATATGCCAAGTACGCCCCCTAT TGACGTCAATGACGGTAAATGGCCCGCCTGGCATTATG CCCAGTACATGACCTTATGGGACTTTCCTACTTGGCAGT ACATCTACGTATTAGTCATCGCTATTACCATG
12	hGFA2.2 promoter	Nucleic Acid	CGCGTCCCACCTCCCTCTCTGTGCTGGGACTCACAGAGG GAGACCTCAGGAGGCAGTCTGTCCATCACATGTCCAAA TGCAGAGCATAACCCTGGGCTGGGCGCAGTGGCGCACAA CTGTAATTCCAGCACTTTGGGAGGCTGATGTGGAAGGA TCACTTGAGCCCAGAAGTTCTAGACCAGCCTGGGCAAC ATGGCAAGACCTATCTCTACAAAAAAGTTAAAAAAT CAGCCACGTGTGGTGACACACACCTGTAGTCCCAGCTA TTCAGGAGGCTGAGGTGAGGGGATCACTTAAGGCTGGG AGGTTGAGGCTGCAGTGAGTCGTGGTTGCGCCACTGCA CTCCAGCCTGGGCAACAGTGAGACCTGTCTCAAAAGA CAAAAAAAAAAAAAAAAAAAAAAAAAAAGAACATATCCTGG TGTGGAGTAGGGGACGCTGCTCTGACAGAGGCTCGGGG GCCTGAGCTGGCTCTGTGAGCTGGGGAGGAGGCAGACA GCCAGGCCTTGTCTGCAAGCAGACCTGGCAGCATTGGG CTGGCCGCCCCCAGGGCCTCCTTTCATGCCCAGTGAA TGACTCACCTTGGCACAGACACAATGTTCCGGGTGGGC ACAGTGCCTGCTTCCCGCCGCACCCCAGCCCCCTCAA TGCCTTCCGAGAAGCCCATTGAGCAGGGGGCTTGCAAT GCACCCAGCCTGACAGCCTGGCATCTTGGGATAAAAG CAGCACAGCCCCCTAGGGGCTGCCCTTGCTGTGTGGCG CCACCGGCGGTGGAGAACAAGGCTCTATTCAGCCTGTG CCCAGGAAAGGGGATCAGGGGATGCCCAGGCATGGAC AGTGGGTGGCAGGGGGGAGAGGAGGGCTGTCTGCTTC CCAGAAGTCCAAGGACACAAATGGGTGAGGGGACTGG GCAGGGTTCTGACCCTGTGGGACCAGAGTGGAGGGCGT AGATGGACCTGAAGTCTCCAGGGACAACAGGGGCCAGG TCTCAGGCTCCTAGTTGGGCCAGTGGCTCCAGCGTTTC CAAACCCATCCATCCCCAGAGGTTCTTCCCATCTCTCCA GGCTGATGTGTGGGAACTCGAGGAAATAAATCTCCAGT GGGAGACGGAGGGGTGGCCAGGGAACGGGGCGCTGC AGGAATAAAGACGAGCCAGCACAGCCAGCTCATGTGTA ACGGCTTTGTGGAGCTGTCAAGGCCTGGTCTCTGGGAG AGAGGCACAGGGAGGCCAGACAAGGAAGGGGTGACCT GGAGGGACAGATCCAGGGGCTAAAGTCCTGATAAGGC AAGAGAGTGCCGGCCCCCTCTTGCCCTATCAGGACCTC CACTGCCACATAGAGGCCATGATTGACCCTTAGACAAA GGGCTGGTGTCCAATCCCAGCCCCCAGCCCCAGAACTC

SEQ ID NO	Sequence Description	Sequence Type	Sequence
			CAGGGAATGAATGGGCAGAGAGCAGGAATGTGGGACA TCTGTGTTCAAGGGAAGGACTCCAGGAGTCTGCTGGGA ATGAGGCCTAGTAGGAAATGAGGTGGCCCTTGAGGGTA CAGAACAGGTTTCATTCTTCGCCAAATTCCCAGCACCTTG CAGGCACTTACAGCTGAGTGAGATAATGCCTGGGTTAT GAAATCAAAAAGTTGGAAAGCAGGTCAGAGGTCATCTG GTACAGCCCTTCCTTCCCTTTTTTTTTTTTTTTTTTGTA GACAAGGTCTCTCTCTGTTGCCCAGGCTGGAGTGGCGC AAACACAGCTCACTGCAGCCTCAACCTACTGGGCTCAA GCAATCCTCCAGCCTCAGCCTCCCAAAGTGCTGGGATT ACAAGCATGAGCCACCCACTCAGCCCTTTCCTTCCTTT TTAATTGATGCATAATAATTGTAAGTATTCATCATGGTC CAACCAACCCTTTCTTGACCCACCTTCCTAGAGAGAGG GTCCTCTTGCTTCAGCGGTCAGGGCCCCAGACCCATGGT CTGGCTCCAGGTACCACCTGCCTCATGCAGGAGTTGGC GTGCCCAGGAAGCTCTGCCTCTGGGCACAGTGACCTCA GTGGGGTGAGGGGAGCTCTCCCCATAGCTGGGCTGCGG CCCAACCCCAACCCCTCAGGCTATGCCAGGGGGTGTTG CCAGGGGCACCCGGGCATCGCCAGTCTAGCCCACTCCT TCATAAAGCCCTCGCATCCAGGAGCGAGCAGAGCCAG AGCAGGTTGGAGAGGAGACGCATCACCTCCGCTGCTCG CCGGG
13	Human Dlx2	Nucleic Acid	ATGACTGGAGTCTTTGACAGTCTAGTGGCTGATATGCAC TCGACCCAGATCGCCGCTCCAGCACGTACCACCAGCA CCAGCAGCCCCCGAGCGGCGGCGGCCCGCCCGGGTG GCAACAGCAGCAGCAGCAGCAGCCTCCACAAGCCCCAG GAGTCGCCACCCCTTCCGGTGTCACCCGCCACCGACAG CAGCTACTACACCAACCAGCAGCACC CGCGGGCGGCG GCGGCGGCGGGGGCTCGCCCTACGCGCACATGGGTTC TACCAGTACCAAGCCAGCGGCCTCAACAACGTCCCTTA CTCCGCCAAGAGCAGCTATGACCTGGGCTACACCGCCG CCTACACCTCCTACGCTCCCTATGGAACCAAGTTCGTC CAGCCAACAACGAGCCTGAGAAGGAGGACCTTGAGCCT GAAATTCGGATAGTGAACGGGAAGCCAAAGAAAGTCC GGAAACCCCGCACCATCTACTCCAGTTTCCAGCTGGCG GCTCTTCAGCGGCGTTTCCAAAAGACTCAATACTTGCC TTGCCGGAGCGAGCCGAGCTGGCGGCCTCTCTGGGCCT CACCAGACTCAGGTCAAAATCTGGTTCCAGAACCGCC GGTCCAAGTTCAAGAAGATGTGGAAAAGTGGTGAGATC CCCTCGGAGCAGCACCCTGGGGCCAGCGCTTCTCCACC TTGTGCTTCGCCGCCAGTCTCAGCGCCGGCCTCCTGGGA CTTTGGTGTGCCGCAGCGGATGGCGGGCGGCGGTGGTC CGGGCAGTGGCGGCAGCGGCGCCGGCAGCTCGGGCTCC

SEQ ID NO	Sequence Description	Sequence Type	Sequence
			AGCCCGAGCAGCGCGGCCTCGGCTTTTCTGGGCAACTA CCCCTGGTACCACCAGACCTCGGGATCCGCCTCACACCT GCAGGCCACGGCGCCGCTGCTGCACCCCACTCAGACCC CGCAGCCGCATCACCACCACCACCATCACGGCGGCGGG GGCGCCCCGGTGAGCGCGGGGACGATTTTCTGA
14	Human Dlx2	Amino Acid	MTGVFDSL VADMHSTQIAASSTYHQHQPPSGGGAGPGG NSSSSSLHKPQESPTLPVSTATDSSYYTNQOHPAGGGGG GGSPYAHMGSYQYQASGLNNVPYSAKSSYDLGYTAAYTS YAPYGTSSSPANNEPEKEDLEPEIRIVNGKPKKVRKPRTIYS SFQLAALQRRFQKTQYLALPERAELAASLGLTQTQVKIWF QNRRSKFKKMWKSGEIPSEQHPGASASPPCASPPVSAPAS WDFGVPQRMAGGGGPGSGGSGAGSSGSSPSAASAFLGN YPWYHQTSGSASHLQATAPLLHPTQTPQPHHHHHHGGG GAPVSAGTIF
15	P2A	Nucleic Acid	GCTACTAAGTTCAGCCTGCTGAAGCAGGCTGGAGACGT GGAGGAGAACCCTGGACCT
16	T2A	Nucleic Acid	GAGGGGAGAGGAAGTCTTCTGACCTGCGGAGACGTCGA AGAGAATCCTGGACCC
17	hGH poly(A) signal	Nucleic Acid	GGGTGGCATCCCTGTGACCCCTCCCCAGTGCCTCTCCTG GCCCTGGAAGTTGCCACTCCAGTGCCACCAGCCTTGTC CTAATAAAATTAAGTTGCATCATTTTGTCTGACTAGGTG TCCTTCTATAATATTATGGGGTGGAGGGGGGTGGTATG GAGCAAGGGGCAAGTTGGGAAGACAACCTGTAGGGCC TGCGGGGTCTATTGGGAACCAAGCTGGAGTGCAGTGGC ACAATCTTGGCTCACTGCAATCTCCGCCTCCTGGGTTCA AGCGATTCTCCTGCCTCAGCCTCCCGAGTTGTTGGGATT CCAGGCATGCATGACCAGGCTCAGCTAATTTTTGTTTTT TTGGTAGAGACGGGGTTTCACCATATTGGCCAGGCTGG TCTCCAACCTCCTAATCTCAGGTGATCTACCCACCTTGGC CTCCCAAATTGCTGGGATTACAGGCGTGAACCACTGCT CCCTTCCCTGTCCTT
18	GSG-P2A	Nucleic Acid	GGAAGCGGAGCTACTAAGTTCAGCCTGCTGAAGCAGGC TGGAGACGTGGAGGAGAACCCTGGACCT
19	GSG-T2A	Nucleic Acid	GGAAGCGGAGCTACTAAGTTCAGCCTGCTGAAGCAGGC TGGAGACGTGGAGGAGAACCCTGGACCT
20	GSG-P2A	Amino Acid	GSGATNFSLLKQAGDVEENPGP
21	GSG-T2A	Amino Acid	GSGEGRGSLLTCGDVEENPGP
22	U6 promoter	Nucleic Acid	GTCCTTTCCACAAGATATATAAACCAAGAAATCGAAA TACTTTCAAGTTACGGTAAGCATATGATAGTCCATTTTA AAACATAATTTTAAACTGCAAACTACCCAAGAAATTA TTACTTTCTACGTCACGTATTTTGTACTAATATCTTTGTG TTTACAGTCAAATTAATTCTAATTATCTCTCTAACAGCC TTGTATCGTATATGCAATATGAAGGAATCATGGGAAA TAGGCCCTC

SEQ ID NO	Sequence Description	Sequence Type	Sequence
23	Htt shRNA 1	Nucleic Acid	CCGGTGGTTCAGTTACGGGTAAATTCTCGAGAATTAACC CGTAACTGAACCATTTTTG
24	Htt shRNA 2	Nucleic Acid	CCGGCAGTTACGGGTAAATTAATACCTGACCCATATTAA TTAACCCGTAACCTGCTTTTTG
25	Htt shRNA 3	Nucleic Acid	CCGGTGTGCGGCAGCATCACTAATCTCGAGATTAGTG ATGCTGCGGCAACATTTTTG
26	pGfa681 promoter (also called “GfaABC1D” promoter)		AACATATCCTGGTGTGGAGTAGGGGACGCTGCTCTGAC AGAGGCTCGGGGGCCTGAGCTGGCTCTGTGAGCTGGGG AGGAGGCAGACAGCCAGGCCTTGTCTGCAAGCAGACCT GGCAGCATTGGGCTGGCCGCCCCCAGGGCCTCCTCTTC ATGCCCAGTGAATGACTCACCTTGGCACAGACACAATG TTCGGGGTGGGCACAGTGCCTGCTTCCCGCCGCACCCC AGCCCCCTCAAATGCCTTCCGAGAAGCCCATTTGAGCA GGGGGCTTGCATTGCACCCAGCCTGACAGCCTGGCAT CTTGGGATAAAAGCAGCACAGCCCCCTAGGGGCTGCCC TTGCTGTGTGGCGCCACCGGCGGTGGAGAACAAGGCTC TATTCAGCCTGTGCCAGGAAAGGGGATCAGGGGATGC CCAGGCATGGACAGTGGGTGGCAGGGGGGGAGAGGAG GGCTGTCTGCTTCCAGAAAGTCCAAGGACACAAATGGG TGAGGGGAGAGCTCTCCCCATAGCTGGGCTGCGGCCCA ACCCACCCCCCTCAGGCTATGCCAGGGGGTGTGTCAG GGGCACCCGGGCATCGCCAGTCTAGCCCACTCCTTCAT AAAGCCCTCGCATCCCAGGAGCGAGCAGAGCCAGAGC AGGTTGGAGAGGAGACGCATCACCTCCGCTGCTCGC
27	CRGI Chimeric Intron		GGAGTCGCTGCGTTGCCTTCGCCCCGTGCCCCGCTCCGC GCCGCCTCGCGCCGCCCCGCCCCGGCTCTGACTGACCGC GTTACTCCCACAGGTGAGCGGGCGGGACGGCCCTTCTC CCTCCGGGCTGTAATTAGCGCTTGGTTTAATGACGGCTC GTTTCTTTTCTGTGGCTGCGTGAAAGCCTTAAAGGGCTC CGGGAGGGCCTTTGTGCGGGGGGGAGCGGCTCGGGGG GTGCGTGCGTGTGTGTGTGCGTGGGGAGCGCCGCGTGC GGCCCGCGCTGCCCGGCGGCTGTGAGCGCTGCGGGGCG GGCGCGGGGCTTTGTGCGCTCCGCGTGTGCGCAGGGG AGCGCGGGCCGGGGCGGTGCCCGCGGTGCGGGGGG GCTGCGAGGGGAACAAAGGCTGCGTGTGCGGGGTGTGTGC GTGGGGGGGTGAGCAGGGGGTGTGGGCGCGGCGGTGCG GGCTGTAACCCCCCTGGCACCCCCCTCCCCAGTTGC TGAGCACGGCCCGGCTTCGGGTGCGGGGCTCCGTGCGG GGCGTGGCGCGGGGCTCGCCGTGCCGGGCGGGGGGTGG CGGCAGGTGGGGGTGCCGGGCGGGGCGGGGCCGCTC GGGCCGGGGAGGGCTCGGGGGAGGGGCGCGGCGGGCC CGGAGCGCCGGCGGCTGTGAGGCGCGGCGAGCCGCA GCCATTGCCTTTTATGGTAATCGTGCGAGAGGGCGCAG GGACTTCCTTTGTCCCAAATCTGGCGGAGCCGAAATCTG GGAGGCGCCGCCGACCCCCCTAGCGGGCGCGGGCGA AGCGGTGCGGCGCCGGCAGGAAGGAAATGGGCGGGGA GGGCCTTCGTGCGTGCCTGCGCGCCGCGCTCCCTTCTCCA TCTCCAGCCTCGGGGCTGCCGAGGGGGACGGCTGCCT TCGGGGGGGACGGGGCAGGGCGGGGTTCGGCTTCTGGC GTGTGACCGGCGGCTTTAGAGCCTCTGCTAACCATGTTT

SEQ ID NO	Sequence Description	Sequence Type	Sequence
			ATGCCTTCTTCTTTTTCTACAGCTCCTGGGCAACGTGC TGGTTGTTGTGCTGTCTCATCATTTTGGCAAAGAT
28	GFAP first intron (GI)		GGCCACTGTGAGGCAGAAAGTGAGGAGGGGATGGGGAA GGGGGGCCTTGTGAGCAGAAGGGGCTGAATCCCCAAGA AGGAGTGCCCGAGAAGTCTCAGGAGGGGCGAACCTC CCTGCTCCCTGGGCCTCCCTACCTCTTGATGGGGCACTA TCCTTGCCCCCAACATGATGGGAGGGACCAGAAACAG GCCAGGGCCCCGGGGATCTGATGCCCGCATGCCTTCT GCCAGGAGTCCAGGGTCCCCTCAGCACCTCCCTACTGG GGAAAGCAGTGCAGGAGCAGCGGGGCCCTGTGTTTCA TTCATGGCTGGGCTTTGTGACTGTGGGCAGCGAGCTCAC CTATTCTGAGCCTGTGTCCATATAAAGGAGGAGTTGGA AGCGGAGAAGGTTGATGTCCATGAGGGAGATTGGATTC TGGGGTGAAGAAAGTGAGGGAAAGAGCAGGCAGGTCT GGGCGCAAAGCACAGGTGACTGCCTGCCACCAGCTTGT GACCCCCATCAAGTTACTTTGACTTGACACAGCTGTGAAG CGGTGGTCATAATAAAATTCATTTCAAAGGTGGTTAC CTGGGATCAGAGGAATCCCCAGGGGCATGGCGCTTCAC TGAGCTGACAGGACATGCATGTGTGCCTTCAAGTGCAG GAGGACATGTGCGTGTGTGTGTGTGTGTGTGCAACAGT GAGTGTATGCTTGTGGATGCGCCTGTGTGAGCAGAAGC AGGTGCACCAACCCTGATAAGGCACCTTAGTAATGAGT TAAGGCAAAAGCCCACATCTGCTCATCCTCCAGACAAG TCCTCTGTCTAAGGCCCCCCAACCTTAATCCTCCTGCT GCTCTACTGGTCTTGGGTGGGGTGGTCTCTGTGACAGC TGCCTCAAGGGGAGACTGAGGCAGGTATTCAAGTGTCT CAGAAGAGCCTGGACCCAGGAATGTGTCCCCCACTCC AGGCTCCAGGATGAAACCAACCTGA
29	Optimized version of WPRE (oPRE),		GAGCATCTTACCGCCATTTATACCCATATTTGTTCTGTTT TTCTTGATTTGGGTATACATTTAAATGTTAATAAAACAA AATGGTGGGGCAATCATTTACATTTTGGGATATGTAA TTACTAGTTCAGGTGATTGCCACAAGACAACATGTTA AGAAACTTTCCCGTTATTTACGCTCTGTTCTGTTAATC AACCTCTGGATTACAAAATTTGTGAAAGATTGACTGAT ATTCTTAACTATGTTGCTCCTTTTACGCTGTGTGGATAT GCTGCTTTATAGCCTCTGTATCTAGCTATTGCTTCCCGT ACGGCTTTCGTTTTCTCCTCCTTGTATAAATCCTGGTTGC TGTCTCTTTTAGAGGAGTTGTGGCCCGTTGTCCGTCAAC GTGGCGTGGTGTGCTCTGTGTTTGTGACGCAACCCCCA CTGGCTGGGGCATTGCCACCACCTGTCAACTCCTTTCTG GGACTTTCGCTTTCCCCCTCCCGATCGCCACGGCAGAAC TCATCGCCGCTGCTTGGCCGCTGCTGGACAGGGGCTA GGTTGCTGGGCACTGATAATTCCGTGGTGTGTGTC
30	bGH poly (A) signal (bGHpA)		CTGTGCCTTCTAGTTGCCAGCCATCTGTTGTTTGCCCCCTC CCCCGTGCCTTCCTTGACCCTGGAAGGTGCCACTCCCAC TGTCCTTTTCTAATAAAATGAGGAAATTGCATCGCATTG TCTGAGTAGGTGTCATTCTATTCTGGGGGGTGGGGTGG GGCAGGACAGCAAGGGGGGAGGATTGGGAAGAGAATAG CAGGCATGCTGGGGA

DETAILED DESCRIPTION

[00116] Unless defined otherwise, all technical and scientific terms used have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. Where a term is provided in the singular, the inventors also contemplate aspects of the disclosure described by the plural of that term. Where there are discrepancies in terms and definitions used in references that are incorporated by reference, the terms used in this application shall have the definitions given herein. Other technical terms used have their ordinary meaning in the art in which they are used, as exemplified by various art-specific dictionaries, for example, "The American Heritage® Science Dictionary" (Editors of the American Heritage Dictionaries, 2011, Houghton Mifflin Harcourt, Boston and New York), the "McGraw-Hill Dictionary of Scientific and Technical Terms" (6th edition, 2002, McGraw-Hill, New York), or the "Oxford Dictionary of Biology" (6th edition, 2008, Oxford University Press, Oxford and New York).

[00117] Any references cited herein, including, *e.g.*, all patents, published patent applications, and non-patent publications, are incorporated herein by reference in their entirety.

[00118] When a grouping of alternatives is presented, any and all combinations of the members that make up that grouping of alternatives is specifically envisioned. For example, if an item is selected from a group consisting of A, B, C, and D, the inventors specifically envision each alternative individually (*e.g.*, A alone, B alone, etc.), as well as combinations such as A, B, and D; A and C; B and C; etc. The term "and/or" when used in a list of two or more items means any one of the listed items by itself or in combination with any one or more of the other listed items. For example, the expression "A and/or B" is intended to mean either or both of A and B – *i.e.*, A alone, B alone, or A and B in combination. The expression "A, B and/or C" is intended to mean A alone, B alone, C alone, A and B in combination, A and C in combination, B and C in combination, or A, B, and C in combination.

[00119] When a range of numbers is provided herein, the range is understood to be inclusive of the edges of the range as well as any number between the defined edges of the range. For example, "between 1 and 10" includes any number between 1 and 10, as well as the number 1 and the number 10.

[00120] When the term "about" is used in reference to a number, it is understood to mean plus or minus 10%. For example, "about 100" would include from 90 to 110.

[00121] As used herein "hND1" refers to a human neuronal differentiation (NeuroD1) gene or protein.

[00122] As used herein "CE" refers to a cytomegalovirus (CMV) promoter enhancer sequence.

- [00123] As used herein “EE” refers to an Ef1 alpha promoter enhancer sequence.
- [00124] As used herein “pGfa681” refers to a human glial fibrillary acid protein (GFAP) promoter truncated sequence of 681 bp size. As used herein “pGfa681,” “Gfa681,” “GfaABC1D,” and “pGfaABC1D” are used interchangeably.
- 5 [00125] As used herein “CI” refers to a chimeric intron composed of the 5’-donor site from the first intron of the human β -globin gene and the branch and 3’-acceptor site from the intron of an immunoglobulin gene heavy chain variable region.
- [00126] As used herein “CRGI” refers to a chimeric intron of rabbit beta-globin and chicken beta actin similar in CAG promoter.
- 10 [00127] As used herein “GI” refers to a human glial fibrillary acid protein (GFAP) first intron.
- [00128] As used herein “WPRE” refers to a Woodchuck Hepatitis Virus (WHV) Posttranscriptional Regulatory Element.
- [00129] As used herein “oPRE” refers to an optimized version of WPRE.
- 15 [00130] As used herein “SV40pA” refers to a poly A signal of SV40 virus.
- [00131] As used herein “bGHpA” refers to a poly A signal of bovine growth hormone.
- [00132] As used herein “vg” refers to a viral genome.
- [00133] As used herein “hDlx2” refers to a human distal-less homeobox 2 gene or protein.
- 20 [00134] Any composition or vector provided herein is specifically envisioned for use with any method provided herein.
- [00135] In an aspect, methods and compositions provided herein comprise a vector. As used herein, the term “vector” refers to a circular, double-stranded DNA molecule that is physically separate from chromosomal DNA. It should be noted that the term “vector” can be
- 25 used interchangeably with the term “plasmid.”
- [00136] In an aspect, a vector provided herein is a recombinant vector. As used herein, the term “recombinant vector” refers to a vector that comprises a recombinant nucleic acid. As used herein, a “recombinant nucleic acid” refers to a nucleic acid molecule formed by laboratory methods of genetic recombination, such as, without being limiting, molecular cloning. A
- 30 recombinant vector can be formed by laboratory methods of genetic recombination, such as, without being limiting, molecular cloning. Also, without being limiting, one skilled in the art can create a recombinant vector *de novo* via synthesizing a plasmid by individual nucleotides, or by splicing together nucleic acid molecules from different pre-existing vectors.
- [00137] Adeno-associated viruses (AAVs) are replication-defective, non-enveloped
- 35 *Dependoparvovirus* viruses that infect humans and additional primate species. AAVs are not

known to cause disease in any species, although they can cause mild immune responses. AAVs can infect dividing and quiescent cells. AAVs are stably integrate into the human genome at a specific site in chromosome 19 termed the AAVS1 locus (nucleotides 7774-11429 of GenBank Accession No. AC010327.8), although random integrations at other loci in the human genome are possible.

[00138] AAVs comprise a linear genome with a single-stranded DNA of about 4700 nucleotides in length. The genome of AAVs also includes a 145 nucleotide-long inverted terminal repeat (ITR) at each end of the genome. The ITRs flank two viral genes *rep* (for replication, encoding non-structural proteins) and *cap* (for capsid, encoding structural proteins). The ITRs contain all of the *cis*-acting elements need for genome rescue, replication, and packaging of the AAV.

[00139] When used in gene therapy approaches, the *rep* and *cap* genes of the AAV genome sequence are removed and replaced with DNA of interest positioned between two AAV ITRs. As used herein, an “AAV vector construct” refers to a DNA molecule comprising a desired sequence inserted between two AAV ITR sequences. As used herein, an “AAV vector” refers to an AAV packaged with a DNA vector construct.

[00140] As used herein, the term “AAV vector serotype” mainly refers to a variation within the *capsid* proteins of an AAV vector.

[00141] In an aspect, an AAV vector is selected from the group consisting of AAV vector serotype 1, AAV vector serotype 2, AAV vector serotype 3, AAV vector serotype 4, AAV vector serotype 5, AAV vector serotype 6, AAV vector serotype 7, AAV vector serotype 8, AAV vector serotype 9, AAV vector serotype 10, AAV vector serotype 11, and AAV vector serotype 12. In one aspect, an AAV vector is selected from the group consisting AAV serotype 2, AAV serotype 5, and AAV serotype 9. In one aspect, an AAV vector is AAV serotype 1. In one aspect, an AAV vector is AAV serotype 2. In one aspect, an AAV vector is AAV serotype 3. In one aspect, an AAV vector is AAV serotype 4. In one aspect, an AAV vector is AAV serotype 5. In one aspect, an AAV vector is AAV serotype 6. In one aspect, an AAV vector is AAV serotype 7. In one aspect, an AAV vector is AAV serotype 8. In one aspect, an AAV vector is AAV serotype 9. In one aspect, an AAV vector is AAV serotype 10. In one aspect, an AAV vector is AAV serotype 11. In one aspect, an AAV vector is AAV serotype 12.

[00142] In an aspect, an AAV vector ITR is selected from the group consisting of an AAV serotype 1 ITR, an AAV serotype 2 ITR, an AAV serotype 3 ITR, an AAV serotype 4 ITR, an AAV serotype 5 ITR, an AAV serotype 6 ITR, an AAV serotype 7 ITR, an AAV serotype 8 ITR, an AAV serotype 9 ITR, an AAV serotype 10 ITR, an AAV serotype 11 ITR, and an AAV serotype 12 ITR. In one aspect, an AAV vector ITR is an AAV serotype 1 ITR. In one aspect,

an AAV vector ITR is an AAV serotype 2 ITR. In one aspect, an AAV vector ITR is an AAV serotype 3 ITR. In one aspect, an AAV vector ITR is an AAV serotype 4 ITR. In one aspect, an AAV vector ITR is an AAV serotype 5 ITR. In one aspect, an AAV vector ITR is an AAV serotype 6 ITR. In one aspect, an AAV vector ITR is an AAV serotype 7 ITR. In one aspect, an AAV vector ITR is an AAV serotype 8 ITR. In one aspect, an AAV vector ITR is an AAV serotype 9 ITR. In one aspect, an AAV vector ITR is an AAV serotype 10 ITR. In one aspect, an AAV vector ITR is an AAV serotype 11 ITR. In one aspect, an AAV vector ITR is an AAV serotype 12 ITR.

[00143] In an aspect, at least one AAV vector ITR nucleic acid sequence is selected from the group consisting of SEQ ID NO: 1 and 9. In one aspect, at least one AAV vector ITR nucleic acid sequence is SEQ ID NO 1. In one aspect, at least one AAV vector ITR nucleic acid sequence is SEQ ID NO 9.

[00144] In an aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid

sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 1, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 1, or the complement thereof.

5 **[00145]** In an aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 9, or the complement thereof. In one aspect, an AAV ITR nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 9, or the complement thereof.

[00146] The terms “percent identity” or “percent identical” as used herein in reference to two or more nucleotide or amino acid sequences is calculated by (i) comparing two optimally aligned sequences (nucleotide or amino acid) over a window of comparison (the “alignable” region or regions), (ii) determining the number of positions at which the identical nucleic acid

base (for nucleotide sequences) or amino acid residue (for proteins and polypeptides) occurs in both sequences to yield the number of matched positions, (iii) dividing the number of matched positions by the total number of positions in the window of comparison, and then (iv) multiplying this quotient by 100% to yield the percent identity. If the “percent identity” is being calculated in relation to a reference sequence without a particular comparison window being specified, then the percent identity is determined by dividing the number of matched positions over the region of alignment by the total length of the reference sequence. Accordingly, for purposes of the present application, when two sequences (query and subject) are optimally aligned (with allowance for gaps in their alignment), the “percent identity” for the query sequence is equal to the number of identical positions between the two sequences divided by the total number of positions in the query sequence over its length (or a comparison window), which is then multiplied by 100%.

[00147] When percentage of sequence identity is used in reference to amino acids it is recognized that residue positions which are not identical often differ by conservative amino acid substitutions, where amino acid residues are substituted for other amino acid residues with similar chemical properties (*e.g.*, charge or hydrophobicity) and therefore do not change the functional properties of the molecule. When sequences differ in conservative substitutions, the percent sequence identity can be adjusted upwards to correct for the conservative nature of the substitution. Sequences that differ by such conservative substitutions are said to have “sequence similarity” or “similarity.”

[00148] For optimal alignment of sequences to calculate their percent identity, various pair-wise or multiple sequence alignment algorithms and programs are known in the art, such as ClustalW or Basic Local Alignment Search Tool® (BLAST™), etc., that can be used to compare the sequence identity or similarity between two or more nucleotide or amino acid sequences. Although other alignment and comparison methods are known in the art, the alignment and percent identity between two sequences (including the percent identity ranges described above) can be as determined by the ClustalW algorithm, *see, e.g.*, Chenna *et al.*, “Multiple sequence alignment with the Clustal series of programs,” *Nucleic Acids Research* 31: 3497-3500 (2003); Thompson *et al.*, “Clustal W: Improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice,” *Nucleic Acids Research* 22: 4673-4680 (1994); Larkin MA *et al.*, “Clustal W and Clustal X version 2.0,” *Bioinformatics* 23: 2947-48 (2007); and Altschul *et al.* “Basic local alignment search tool.” *J. Mol. Biol.* 215:403-410 (1990), the entire contents and disclosures of which are incorporated herein by reference.

[00149] The terms “percent complementarity” or “percent complementary” as used herein in reference to two nucleotide sequences is similar to the concept of percent identity but refers to

the percentage of nucleotides of a query sequence that optimally base-pair or hybridize to nucleotides a subject sequence when the query and subject sequences are linearly arranged and optimally base paired without secondary folding structures, such as loops, stems or hairpins. Such a percent complementarity can be between two DNA strands, two RNA strands, or a DNA strand and a RNA strand. The “percent complementarity” can be calculated by (i) optimally base-pairing or hybridizing the two nucleotide sequences in a linear and fully extended arrangement (*i.e.*, without folding or secondary structures) over a window of comparison, (ii) determining the number of positions that base-pair between the two sequences over the window of comparison to yield the number of complementary positions, (iii) dividing the number of complementary positions by the total number of positions in the window of comparison, and (iv) multiplying this quotient by 100% to yield the percent complementarity of the two sequences. Optimal base pairing of two sequences can be determined based on the known pairings of nucleotide bases, such as G-C, A-T, and A-U, through hydrogen binding. If the “percent complementarity” is being calculated in relation to a reference sequence without specifying a particular comparison window, then the percent identity is determined by dividing the number of complementary positions between the two linear sequences by the total length of the reference sequence. Thus, for purposes of the present application, when two sequences (query and subject) are optimally base-paired (with allowance for mismatches or non-base-paired nucleotides), the “percent complementarity” for the query sequence is equal to the number of base-paired positions between the two sequences divided by the total number of positions in the query sequence over its length, which is then multiplied by 100%.

[00150] The use of the term “polynucleotide,” “nucleic acid sequence,” or “nucleic acid molecule” is not intended to limit the present disclosure to polynucleotides comprising deoxyribonucleic acid (DNA). For example, ribonucleic acid (RNA) molecules are also envisioned. Those of ordinary skill in the art will recognize that polynucleotides and nucleic acid molecules can comprise ribonucleotides and combinations of ribonucleotides and deoxyribonucleotides. Such deoxyribonucleotides and ribonucleotides include both naturally occurring molecules and synthetic analogues. The polynucleotides of the present disclosure also encompass all forms of sequences including, but not limited to, single-stranded forms, double-stranded forms, hairpins, stem-and-loop structures, and the like. In an aspect, a nucleic acid molecule provided herein is a DNA molecule. In one aspect, a nucleic acid molecule provided herein is an RNA molecule. In one aspect, a nucleic acid molecule provided herein is single-stranded. In one aspect, a nucleic acid molecule provided herein is double-stranded. A nucleic acid molecule can encode a polypeptide or a small RNA.

[00151] As used herein, the term “polypeptide” refers to a chain of at least two covalently linked amino acids. Polypeptides can be encoded by polynucleotides provided herein. Proteins provided herein can be encoded by nucleic acid molecules provided herein. Proteins can comprise polypeptides provided herein. As used herein, a “protein” refers to a chain of amino acid residues that is capable of providing structure or enzymatic activity to a cell. As used herein, a “coding sequence” refers to a nucleic acid sequence that encodes a protein.

[00152] As used herein, the term “CpG site” or “CG site” refers to a region of DNA sequence where a cytosine and guanine is separated by only one phosphate group.

[00153] As used herein, the term “CpG island” or “CG island” refers to CpG sites that occur with a high frequency.

[00154] As used herein, the term “codon” refers to a sequence of three nucleotides.

[00155] As used herein, the term “codon optimized” refers to a code that is modified for enhanced expression in a host cell of interest by replacing at least one codon of a sequence with codons that are more frequently or most frequently used in the genes of the host cell while maintaining the original amino acid sequence.

[00156] As used herein, the term “enhancer” refers to a region of DNA sequence that operates to initiate, assist, affect, cause, and/or promote the transcription and expression of the associated transcribable DNA sequence or coding sequence, at least in certain tissue(s), developmental stage(s) and/or condition(s). In an aspect, an enhancer is a *cis* enhancer. In one aspect, an enhancer is a *trans* enhancer.

[00157] Enhancer sequences can be identified by utilizing genomic techniques well known in the art. Non-limiting examples include use of a reporter gene and next-generation sequencing methods such as chromatin immunoprecipitation sequencing (ChIP-seq), DNase I hypersensitivity sequencing (DNase-seq), micrococcal nuclease sequencing (MNase-seq), formaldehyde-assisted isolation of regulatory elements sequencing (FAIRE-seq), and assay for transposase accessible chromatin sequencing (ATAC-seq).

[00158] As used herein, the term “operably linked” refers to a functional linkage between a promoter or other regulatory element and an associated transcribable DNA sequence or coding sequence of a gene (or transgene), such that the promoter, etc., operates to initiate, assist, affect, cause, and/or promote the transcription and expression of the associated transcribable DNA sequence or coding sequence, at least in certain tissue(s), developmental stage(s) and/or condition(s). As used herein, “regulatory elements” refer to any sequence elements that regulate, positively or negatively, the expression of an operably linked sequence. “Regulatory elements” include, without being limiting, a promoter, an enhancer, a leader, a transcription start site (TSS), a linker, 5' and 3' untranslated regions (UTRs), an intron, a polyadenylation signal, and a

termination region or sequence, etc., that are suitable, necessary or preferred for regulating or allowing expression of the gene or transcribable DNA sequence in a cell. Such additional regulatory element(s) can be optional and used to enhance or optimize expression of the gene or transcribable DNA sequence.

5 **[00159]** As used herein, the term “promoter” refers to a DNA sequence that contains an RNA polymerase binding site, a transcription start site, and/or a TATA box and assists or promotes the transcription and expression of an associated transcribable polynucleotide sequence and/or gene (or transgene). A promoter can be synthetically produced, varied, or derived from a known or naturally occurring promoter sequence or other promoter sequence. A promoter can also
10 include a chimeric promoter comprising a combination of two or more heterologous sequences. A promoter of the present application can thus include variants of promoter sequences that are similar in composition, but not identical to, other promoter sequence(s) known or provided herein.

[00160] As used herein, an “intron” refers to a nucleotide sequence that is removed by RNA splicing as a messenger RNA (mRNA) matures from a mRNA precursor.

15 **[00161]** As used herein, “mRNA” or “messenger RNA” refers to a single stranded RNA that corresponds to the genetic sequence of a gene.

[00162] Expression of mRNA can be measured using any suitable method known in the art. Non-limiting examples of measuring mRNA expression include quantitative reverse transcriptase polymerase chain reaction (qRT-PCR), RNA blot (e.g., a Northern blot), and RNA sequencing.
20 Differences in expression can be described as an absolute quantification or a relative quantification. See, for example, Livak and Schmittgen, *Methods*, 25:402-408 (2001).

[00163] As used herein, “genome editing” or “gene editing” refers to targeted mutagenesis, insertion, deletion, inversion, substitution, or translocation of a nucleotide sequence of interest in a genome using a targeted editing technique. A nucleotide sequence of interest can be of any
25 length, for example, at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 15, at least 20, at least 25, at least 30, at least 35, at least 40, at least 45, at least 50, at least 75, at least 100, at least 250, at least 500, at least 1000, at least 2500, at least 5000, at least 10,000, or at least 25,000 nucleotides. Non-limiting examples of gene editing techniques are small interference RNA (siRNA) technology, small hairpin RNA (shRNA)
30 technology, microRNA (miRNA) technology, antisense oligonucleotides (ASO) technology, or CRISPR/CAS technology.

[00164] As used herein, a “ASO” or “antisense oligonucleotide” is a small, single stranded nucleic acid that bind to their target RNA sequence inside the cells and silence genes.

[00165] As used herein, a “coding region,” a “gene region,” or a “gene” refers to a
35 polynucleotide that can produce a functional unit. Non-limiting examples include a protein, or a

non-coding RNA molecule. A “coding region,” “gene,” or “gene region” can comprise a promoter, an enhancer sequence, a leader sequence, a transcriptional start site, a transcriptional stop site, a polyadenylation site, one or more exons, one or more introns, a 5’-UTR, a 3’-UTR, or any combination thereof.

- 5 **[00166]** In an aspect, gene editing targets mutant Huntingtin (Htt) aggregates. In one aspect gene editing is by non-coding RNA molecules. Non-limiting examples of a non-coding RNA molecule include a microRNA (miRNA), a miRNA precursor (pre-miRNA), a small interfering RNA (siRNA), a small RNA (18-26 nucleotides in length) and precursor encoding same, a heterochromatic siRNA (hc-siRNA), a Piwi-interacting RNA (piRNA), a hairpin double strand
- 10 RNA (hairpin dsRNA), a *trans*-acting siRNA (ta-siRNA), a naturally occurring antisense siRNA (nat-siRNA), a CRISPR RNA (crRNA), a tracer RNA (tracrRNA), a guide RNA (gRNA), and a single-guide RNA (sgRNA). In one aspect, a shRNA targets a Htt gene. In one aspect, a siRNA targets a Htt gene. In one aspect, an ASO targets a Htt gene. In one aspect, miRNA targets a Htt gene. In one aspect, a gRNA targets a Htt gene. In one aspect, a pre-miRNA targets a Htt gene.
- 15 In one aspect, a small RNA targets a Htt gene. In one aspect, a hc-siRNA targets a Htt gene. In one aspect, a piRNA targets a Htt gene. In one aspect, a hairpin dsRNA targets a Htt gene. In one aspect, a ta-siRNA targets a Htt gene. In one aspect, a nat-siRNA targets a Htt gene. In one aspect, a crRNA targets a Htt gene. In one aspect, a tracrRNA targets a Htt gene. In one aspect, a sgRNA targets a Htt gene. In one aspect, a shRNA comprises a nucleic acid sequence selected
- 20 from the group consisting of SEQ ID NOs: 23 to 25. In one aspect, a shRNA comprises a nucleic acid sequence SEQ ID NO: 23. In one aspect, a shRNA comprises a nucleic acid sequence SEQ ID NO: 24. In one aspect, a shRNA comprises a nucleic acid sequence SEQ ID NO: 25.

- [00167]** As used herein a “donor molecule” or “donor sequence” is defined as a nucleic acid sequence that has been selected for site directed, targeted insertion into a genome. In an
- 25 aspect, a donor molecule comprises a “donor sequence.” In one aspect, a targeted editing technique provided herein comprises the use of one or more, two or more, three or more, four or more, or five or more donor molecules or donor sequences. A donor molecule or donor sequence provided herein can be of any length. For example, a donor molecule or donor sequence provided herein is
- 30 between 2 and 50,000, between 2 and 10,000, between 2 and 5000, between 2 and 1000, between 2 and 500, between 2 and 250, between 2 and 100, between 2 and 50, between 2 and 30, between 15 and 50, between 15 and 100, between 15 and 500, between 15 and 1000, between 15 and 5000, between 18 and 30, between 18 and 26, between 20 and 26, between 20 and 50, between 20 and 100, between 20 and 250, between 20 and 500, between 20 and 1000, between 20 and 5000 or between 20 and 10,000 nucleotides in length.

[00168] As used here “HTT” refers to an Htt specific guide RNA (gRNA) and/or a donor sequence.

[00169] In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector where the AAV vector comprises a Cas9 nuclease gene, an Htt specific gRNA, and a donor sequence. In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector where the AAV vector comprises a Cas9 nuclease gene, an Htt specific gRNA, a donor sequence, and a Dlx2 gene sequence. In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector where the AAV vector comprises a Cas9 nuclease gene, an Htt specific shRNA, and a donor sequence. In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector where the AAV vector comprises a Cas9 nuclease gene, an Htt specific shRNA, a donor sequence, and a Dlx2 gene sequence.

[00170] Site-specific nucleases provided herein can be used as part of a targeted editing technique. Non-limiting examples of site-specific nucleases include meganucleases, zinc finger nucleases (ZFNs), transcription activator-like effector nucleases (TALENs), RNA-guided nucleases (*e.g.*, Cas9 and Cpf1), a recombinase (without being limiting, for example, a serine recombinase attached to a DNA recognition motif, a tyrosine recombinase attached to a DNA recognition motif), a transposase (without being limiting, for example, a DNA transposase attached to a DNA binding domain), or any combination thereof.

[00171] Site-specific nucleases, such as meganucleases, ZFNs, TALENs, Argonaute proteins (non-limiting examples of Argonaute proteins include *Thermus thermophilus* Argonaute (TtAgo), *Pyrococcus furiosus* Argonaute (PfAgo), *Natronobacterium gregoryi* Argonaute (NgAgo), homologs thereof, or modified versions thereof), Cas9 nucleases (non-limiting examples of RNA-guided nucleases include Cas1, Cas1B, Cas2, Cas3, Cas4, Cas5, Cas6, Cas7, Cas8, Cas9 (also known as Csn1 and Csx12), Cas10, Csy1, Csy2, Csy3, Cse1, Cse2, Csc1, Csc2, Csa5, Csn2, Csm2, Csm3, Csm4, Csm5, Csm6, Cmr1, Cmr3, Cmr4, Cmr5, Cmr6, Csb1, Csb2, Csb3, Csx17, Csx14, Csx10, Csx16, CsaX, Csx3, Csx1, Csx15, Csf1, Csf2, Csf3, Csf4, Cpf1, CasX, CasY, homologs thereof, or modified versions thereof), induce a double-strand DNA break at the target site of a genomic sequence that is then repaired by the natural processes. Sequence modifications then occur at the cleaved sites, which can include inversions, deletions, or insertions that result in gene disruption or integration of nucleic acid sequences. In an aspect, an RNA-guided nuclease provided herein is selected from the group consisting of Cas1, Cas1B, Cas2, Cas3, Cas4, Cas5, Cas6, Cas7, Cas8, Cas9 (also known as Csn1 and Csx12), Cas10, Csy1, Csy2, Csy3, Cse1, Cse2, Csc1, Csc2, Csa5, Csn2, Csm2, Csm3, Csm4, Csm5, Csm6, Cmr1, Cmr3, Cmr4,

Cmr5, Cmr6, Csb1, Csb2, Csb3, Csx17, Csx14, Csx10, Csx16, CsaX, Csx3, Csx1, Csx15, Csf1, Csf2, Csf3, Csf4, Cpf1, CasX, CasY, homologs thereof, or modified versions thereof

[00172] In an aspect, a targeted editing technique described herein comprises the use of a RNA-guided nuclease.

5 **[00173]** While not being limited by any particular scientific theory, CRISPR/CAS nucleases are part of the adaptive immune system of bacteria and archaea, protecting them against invading nucleic acids such as viruses by cleaving target DNA in a sequence-dependent manner. The immunity is acquired by the integration of short fragments of the invading DNA, known as spacers, between ~20 nucleotide long CRISPR repeats at the proximal end of a CRISPR locus (a
10 CRISPR array). A well described Cas protein is the Cas9 nuclease (also known as Csn1), which is part of the Class 2, type II CRISPR/Cas system in *Streptococcus pyogenes*. See Makarova *et al. Nature Reviews Microbiology* (2015) doi: 10.1038/nrmicro3569. Cas9 comprises an RuvC-like nuclease domain at its amino terminus and an HNH-like nuclease domain positioned in the middle of the protein. Cas9 proteins also contain a PAM-interacting (PI) domain, a recognition lobe
15 (REC), and a BH domain. The Cpf1 nuclease, another type II system, acts in a similar manner to Cas9, but Cpf1 does not require a tracrRNA. See Cong *et al. Science* (2013) 339: 819-823; Zetsche *et al., Cell* (2015) doi: 10.1016/j.cell.2015.09.038; U. S. Patent Publication No. 2014/0068797; U. S. Patent Publication No. 2014/0273235; U. S. Patent Publication No. 2015/0067922; U. S. Patent No. 8,697,359; U. S. Patent No. 8,771,945; U. S. Patent No. 8,795,965; U. S. Patent No.
20 8,865,406; U. S. Patent No. 8,871,445; U. S. Patent No. 8,889,356; U. S. Patent No. 8,889,418; U. S. Patent No. 8,895,308; and U. S. Patent No. 8,906,616, each of which is herein incorporated by reference in its entirety.

[00174] As used herein, the term “glial” or “glial cell” refers to a non-neuronal cell in the CNS or the PNS. In an aspect, at least one glial cell is selected from the group consisting of at
25 least one oligodendrocyte, at least one astrocyte, at least one NG2 cell, at least one ependymal cell, and at least one microglia. In one aspect, at least one glial cell is at least one oligodendrocyte. In one aspect, at least one glial cell is at least one NG2 cell. In one aspect, at least one glial cell is at least one ependymal cell. In one aspect, at least one glial cell is at least one microglia. In one aspect, at least one glial cell is at least one reactive astrocyte. In one aspect, at least one
30 astrocyte is at least one reactive astrocyte.

[00175] As used herein, the term “astrocyte” refers to a glial cell that is an important component of the brain. An astrocyte is involved in supporting neuronal functions such as synapse formation and plasticity, potassium buffering, nutrient supply, the secretion and absorption of neural or glial transmitters, and maintenance of the blood–brain barrier. As used herein, the term
35 “reactive astrocytes” refers to an abnormal status of astrocytes after injury or disease.

[00176] As used herein, the term “NG2 cell” or “polydendrocyte” refers to a glial cell that expresses chondroitin sulfate proteoglycan (CSPG4) and the alpha receptor for platelet-derived growth factor (PDGFRA).

[00177] As used herein, the term “neuron” or “neuronal cell” refers to an electrically excitable cell that communicates with other neurons via synapses. In an aspect, a neuron is selected from the group consisting of an unipolar neuron, a bipolar neuron, a pseudounipolar neuron, and a multipolar neuron. In one aspect, a neuron is an unipolar neuron. In one aspect, a neuron is a bipolar neuron. In one aspect, a neuron is a pseudounipolar neuron. In one aspect, a neuron is a bipolar neuron. In one aspect, a neuron is selected from the group consisting of a sensory neuron, a motor neuron, and an interneuron. In one aspect, a neuron is a sensory neuron. In one aspect, a neuron is a motor neuron. In one aspect, a neuron is an interneuron.

[00178] As used herein, the term “functional neuron” refers to a neuron that can perform biological process. Without being limiting, examples of biological processes include processing and transmission of information and communication via chemical and electrical synapses.

[00179] As used herein, the term “glutamatergic neurons” refers to a subclass of neurons that produce glutamate and establish excitatory synapses. As used herein, the term “excitatory synapse” refers to a synapse in which an action potential in a presynaptic neuron increases the probability of an action potential occurring in a postsynaptic cell. As used herein, the term “action potential” or “nerve impulse” refers to an electrical impulse across the membrane of an axon. As used herein, the term “axon” or “nerve fiber” refers to a neuron that conducts action potentials. As used herein, the term “GABAergic neurons” refers to a subset of neurons that produce GABA and establish inhibitory synapses. As used herein, the term “GABA” or “*gamma*-Aminobutyric acid” refers to a compound that opens ion channels to allow the flow of negatively charged chloride ions into the cell or positively charged potassium ions out of the cell. As used herein, the term “inhibitory synapse” refers to a synapse that moves the membrane potential of a postsynaptic neuron away from the threshold for generating action potentials. As used herein, the term “dopaminergic neuron” refers to a subset of neurons that produce dopamine. As used herein, the term “dopamine” refers to a neurotransmitter. As used herein, the term “neurotransmitter” refers to endogenous chemicals that activate neurotransmissions. As used herein, the term “neurotransmission” refers to a process where neurotransmitters are released by the axon terminal of a neuron. As used herein, the term “acetyl cholinergic neuron” or “cholinergic neuron” refers to a subset of neurons that secrete acetylcholine. As used herein, the term “acetylcholine” refers to neurotransmitter. As used herein, the term “serotonergic neuron” refers to a subset of neurons that synthesizes serotonin. As used herein, the term “serotonin” refers to a neurotransmitter. As used herein, a “epinephrinergic neuron” refers to a neuron that releases epinephrine as

the neurotransmitter. As used herein, the term “motor neuron” refers to a subset of neurons where the cell body is located in the motor cortex, brainstem, or the spinal cord and the axon projects to the spinal cord or outside the spinal cord and directly or indirectly controls muscles and glands. As used herein, the term peptidergic neuron refers to a subset of neurons that utilize small peptide molecules as their neurotransmitter.

[00180] In an aspect, a neuron is a functional neuron. In one aspect, a functional neuron is selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons. In one aspect, a functional neuron is a glutamatergic neuron.

In one aspect, a functional neuron is a GABAergic neuron. In one aspect, a functional neuron is a dopaminergic neuron. In one aspect, a functional neuron is a cholinergic neuron. In one aspect, a functional neuron is a serotonergic neuron. In one aspect, a functional neuron is an epinephrinergic neuron. In one aspect, a functional neuron is a motor neuron. In one aspect, a functional neuron is a peptidergic neuron.

[00181] As used herein, the term “converting” or “converted” refers to a cell type changing its physical morphology and/or biological function into a different physical morphology and/or different biological function. In an aspect, this disclosure provides the conversion of at least one glial cell into at least one neuron. In one aspect, conversion of at least one glial cell to at least one neuron occurs in the CNS or PNS. In one aspect, conversion of at least one glial cell to at least one neuron occurs in the CNS. In one aspect, conversion of at least one glial cell to at least one neuron occurs in the PNS.

[00182] In one aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, where the hNeuroD1 sequence and the hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18 (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, (iii) or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 sequence and the hDlx2 sequence are operably linked to regulatory elements comprising: (a) glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) chimeric intron comprising the nucleic acid sequence selected from the group

consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[00183] In one aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where the hNeuroD1 coding sequence and the hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 coding sequence and the hDlx2 coding sequence is operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 28; (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[00184] In an aspect, this disclosure provides, and includes, an adeno-associated virus (AAV) vector comprising a neurogenic differentiation 1 (NeuroD1) nucleic acid coding sequence encoding a NeuroD1 protein and a distal-less homeobox 2 (Dlx2) nucleic acid coding sequence encoding a Dlx2 protein, where the NeuroD1 coding sequence and the Dlx2 coding sequence are separated by a linker sequence, where the NeuroD1 coding sequence and the Dlx2 coding sequence are operably linked to regulatory elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal sequence.

[00185] In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector for converting glial cells to functional neurons in a human, where the AAV vector comprises a human neurogenic differentiation 1 (hNeuroD1) sequence having a nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence having a nucleic acid sequence of SEQ ID NO: 13, where the hNeuroD1 sequence and the hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, (iii) or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 sequence and hDlx2 sequence are operably linked to regulatory elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[00186] In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated-virus (AAV) vector for converting glial cells to functional neurons in a human, where the AAV vector comprises a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, where the hNeuroD1 coding sequence and the hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, (iii) or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, where the hNeuroD1 coding sequence and the hDlx2 coding sequence are operably linked to regulatory elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha)

promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[00187] In an aspect, this disclosure provides, and includes, a composition comprising an adeno-associated virus (AAV) vector for the treatment of a subject in need thereof, where the AAV vector comprises a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, where the NeuroD1 sequence and the Dlx2 sequence are separated by a linker sequence, where the NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising: (a) a glial fibrillary acidic protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal.

[00188] In an aspect, an AAV vector comprises a nucleic acid sequence encoding an AAV protein. In one aspect, an AAV vector comprises a nucleic acid sequence encoding a viral protein. Non-limiting examples of AAV proteins and viral proteins include *rep* and *cap* proteins.

[00189] Neurogenic differentiation 1 (NeuroD1; also referred to as $\beta 2$) is a basic helix-loop-helix (bHLH) transcription factor that forms heterodimers with other bHLH proteins to activate transcription of genes that contain a DNA sequence known as an E-box.

[00190] In an aspect, a NeuroD1 sequence is a human NeuroD1 (hNeuroD1) sequence. In one aspect, a NeuroD1 sequence is selected from the group consisting of a chimpanzee NeuroD1 sequence, a bonobo NeuroD1 sequence, an orangutan NeuroD1 sequence, a gorilla NeuroD1 sequence, a macaque NeuroD1 sequence, a marmoset NeuroD1 sequence, a capuchin NeuroD1 sequence, a baboon NeuroD1 sequence, a gibbon NeuroD1 sequence, and a lemur NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a chimpanzee NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a bonobo NeuroD1 sequence. In one aspect, a NeuroD1 sequence is an orangutan NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a gorilla NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a macaque NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a marmoset NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a capuchin NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a baboon NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a gibbon NeuroD1 sequence. In one aspect, a NeuroD1 sequence is a lemur NeuroD1 sequence.

[00191] In an aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 6, or the complement thereof. In one aspect, a NeuroD1 nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 6, or the complement thereof.

[00192] In an aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 70% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 75% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 80% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 85% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino

acid sequence at least 90% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 91% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 92% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 93% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 94% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 95% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 96% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 97% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 98% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 99% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 99.5% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 99.8% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence at least 99.9% identical or similar to SEQ ID NO: 10. In one aspect, a nucleic acid coding sequence encodes a NeuroD1 protein comprising an amino acid sequence 100% identical or similar to SEQ ID NO: 10.

[00193] Distal-less homeobox 2 (Dlx2; also referred to as TES1) is a member of the Dlx gene family and is a homeobox containing gene that plays a role in forebrain and craniofacial development.

[00194] In an aspect, a Dlx2 sequence is a human Dlx2 (hDlx2) sequence. In one aspect, a Dlx2 sequence is selected from the group consisting of a chimpanzee Dlx2 sequence, a bonobo Dlx2 sequence, an orangutan Dlx2 sequence, a gorilla Dlx2 sequence, a macaque Dlx2 sequence, a marmoset Dlx2 sequence, a capuchin Dlx2 sequence, a baboon Dlx2 sequence, a gibbon Dlx2 sequence, and a lemur Dlx2 sequence. In one aspect, a Dlx2 sequence is a chimpanzee Dlx2 sequence. In one aspect, a Dlx2 sequence is a bonobo Dlx2 sequence. In one aspect, a Dlx2 sequence is an orangutan Dlx2 sequence. In one aspect, a Dlx2 sequence is a gorilla Dlx2 sequence. In one aspect, a Dlx2 sequence is a macaque Dlx2 sequence. In one aspect, a Dlx2 sequence is a marmoset Dlx2 sequence. In one aspect, a Dlx2 sequence is a capuchin Dlx2

sequence. In one aspect, a Dlx2 sequence is a baboon Dlx2 sequence. In one aspect, a Dlx2 sequence is a gibbon Dlx2 sequence. In one aspect, a Dlx2 sequence is a lemur Dlx2 sequence.

[00195] In an aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 913% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 13, or the complement thereof. In one aspect, a Dlx2 nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 13, or the complement thereof.

[00196] In an aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 70% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 75% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 80% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 85% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid

coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 90% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 91% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 92% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 93% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 94% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 95% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 96% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 97% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 98% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 99% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 99.5% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 99.8% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence at least 99.9% identical or similar to SEQ ID NO: 14. In one aspect, a nucleic acid coding sequence encodes a Dlx2 protein comprising an amino acid sequence 100% identical or similar to SEQ ID NO: 14.

[00197] In an aspect, an AAV vector comprises a NeuroD1 sequence and a Dlx2 sequence. In one aspect, an AAV vector comprises a NeuroD1 sequence. In one aspect, an AAV comprises a Dlx2 sequence.

[00198] As used herein, “linkers” or “spacers” are short sequences that separate multiple protein and coding domains. Linkers can be cleavable or non-cleavable and facilitate multigene co-expression in single vectors.

[00199] As used herein, “2A self-cleaving peptides” or “2A peptides” are a class of linkers that can induce the cleaving of recombinant protein in a cell.

[00200] As used herein, “P2A linker,” “p2A,” or “P2A” refer to the porcine teschovirus-1 (P2A) linker, which is a member of the 2A self-cleaving peptides.

[00201] In one aspect a P2A linker has a nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18. In one aspect, a P2A linker has a nucleic acid sequence of

SEQ ID NO: 15. In one aspect, a P2A linker has a nucleic acid sequence of SEQ ID NO: 18. In one aspect, a P2A is SEQ ID NO: 15. In one aspect, a GSG-P2A is SEQ ID NO: 18. In one aspect, a P2A linker has a nucleic acid sequence of SEQ ID NO: 18. In one aspect, a P2A linker protein has a nucleic acid coding sequence of SEQ ID NO: 20.

5 **[00202]** As used herein, “T2A linker” refers to thosea asigna virus 2A (T2A) linker, which is a member of the 2A self-cleaving peptides.

[00203] In one aspect, a T2A linker has a nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19. In one aspect, a T2A linker has a nucleic acid sequence of SEQ ID NO: 16. In one aspect, a T2A linker has a nucleic acid sequence of SEQ ID NO: 19. In
10 one aspect, a T2A linker protein has a nucleic acid coding sequence of SEQ ID NO: 21

[00204] As used herein, “E2A linker” refers to equine rhinitis A virus (E2A) linker, which is a member of the 2A self-cleaving peptides.

[00205] As used herein, “F2A linker” refers to foot and mouse disease virus (F2A) linker, which is a member of the 2A self-cleaving peptides.

15 **[00206]** In an aspect, a linker is selected from the group consisting of a P2A linker, a T2A linker, a E2A linker, and a F2A linker. In one aspect, a linker is a P2A linker. In one aspect, a linker is a T2A linker. In one aspect, a linker is a E2A linker. In one aspect, a linker is a F2A linker.

[00207] In an aspect, a linker sequence comprises a P2A linker. In one aspect, a P2A linker
20 nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker
25 nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker
30 nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 15, or the complement thereof. In one aspect, a P2A linker
35 nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 15, or the

[00208] In an aspect, a linker sequence comprises a P2A linker. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at

least 99.8% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 18, or the complement thereof. In one aspect, a P2A linker nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 18, or the complement thereof.

- 5 **[00209]** In an aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 70% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 75% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 80% identical or similar to SEQ ID NO:
- 10 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 85% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 90% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 91% identical or similar to SEQ ID NO: 20. In one
- 15 aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 92% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 93% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 94% identical or similar to SEQ ID NO: 20. In one aspect, a
- 20 nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 95% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 96% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 97% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid
- 25 coding sequence encodes a P2A protein comprising an amino acid sequence at least 98% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 99% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 99.5% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding
- 30 sequence encodes a P2A protein comprising an amino acid sequence at least 99.8% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence at least 99.9% identical or similar to SEQ ID NO: 20. In one aspect, a nucleic acid coding sequence encodes a P2A protein comprising an amino acid sequence 100% identical or similar to SEQ ID NO: 20.

[00211] In an aspect, a linker sequence comprises a T2A linker. In one aspect, a T2A linker
30 nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 19, or the
complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at
least 75% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker
nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 19, or the
complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at
35 least 85% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker

nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 19, or the complement thereof. In one aspect, a T2A linker nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 19, or the complement thereof.

[00212] In an aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 70% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 75% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 80% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 85% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 90% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 91% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 92% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 93% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising

an amino acid sequence at least 94% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 95% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 96% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 97% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 98% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 99% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 99.5% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 99.8% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence at least 99.9% identical or similar to SEQ ID NO: 21. In one aspect, a nucleic acid coding sequence encodes a T2A protein comprising an amino acid sequence 100% identical or similar to SEQ ID NO: 21.

[00213] As used herein “IRES” refers to an internal ribosomal entry site of the encephalomyocarditis virus (EMCV).

[00214] In one aspect, an AAV or vector of the present disclosure comprises an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence. In one aspect, the IRES sequence comprises SEQ ID NO: 3. In one aspect, the IRES sequence comprises a sequence at least 70% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 75% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 80% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 85% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 90% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 91% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 92% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 93% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 94% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 95% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 96% identical to SEQ ID NO: 3, or the complement thereof. In one

aspect, the IRES sequence comprises a sequence at least 97% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 98% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 99% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 3, or the complement thereof. In one aspect, the IRES sequence comprises a sequence 100% identical to SEQ ID NO: 3, or the complement thereof.

[00215] Glial fibrillary acid protein (GFAP); also referred to as glial fibrillary acidic protein is a member of the type III intermediate filament family of proteins that is expressed in the central nervous system and plays a role in cell communication and the functioning of the blood-brain barrier.

[00216] In an aspect, the promoter is selected from the group consisting of GFAP promoter, Sox9 promoter, S100b promoter, Aldh1l1 promoter, Lipocalin 2 (Lcn2) promoter, glutamine synthetase promoter, Aquaporin-4 (AQP4) promoter, oligodendrocyte transcription factor (Olig2) promoter, and synapsin promoter, NG2 promoter, ionized calcium binding adaptor molecule 1 (Iba1) promoter, cluster of differentiation 86 (CD86) promoter, platelet-derived growth factor receptor alpha (PDGFRA) promoter, platelet-derived growth factor receptor beta (PDGFRB) promoter, elongation factor 1-alpha (EF1a) promoter, CAG promoter, cytomegalovirus (CMV) promoter, ubiquitin promoter. In one aspect, the promoter is GFAP promoter. In one aspect, the promoter is a truncated GFAP promoter. In one aspect, the promoter is Sox9 promoter. In one aspect, the promoter is S100b promoter. In one aspect, the promoter is Aldh1l1 promoter. In one aspect, the promoter is Lcn2 promoter. In one aspect, the promoter is glutamine synthetase promoter. In one aspect, the promoter is AQP4 promoter. In one aspect, the promoter is Olig2 promoter. In one aspect, the promoter is synapsin promoter. In one aspect, the promoter is Iba1 promoter. In one aspect, the promoter is CD86 promoter. In one aspect, the promoter is PDGFRA promoter. In one aspect, the promoter is PDGFRB promoter. In one aspect, the promoter is EF1a promoter. In one aspect, the promoter is CAG promoter. In one aspect, the promoter is CMV promoter. In one aspect, the promoter is ubiquitin promoter.

[00217] In an aspect, an ubiquitin promoter is selected from the group consisting of U6, H1, 7SK, and U1. In one aspect, an ubiquitin promoter is U6. In one aspect, an ubiquitin promoter is H1. In one aspect, an ubiquitin promoter is H1. In one aspect, an ubiquitin promoter is 7SK.

In one aspect, an ubiquitin promoter is U1. In one aspect, U6 comprises the nucleic acid sequence of SEQ ID NO: 22.

[00218] In an aspect, a GFAP promoter is a promoter directing astrocyte-specific expression of a protein called glial fibrillary acidic protein (GFAP) in cells. In one aspect, a GFAP promoter sequence is a human GFAP (hGFAP) promoter sequence. In one aspect, a GFAP promoter is selected from the group consisting of GfaABC1D (also called “pGfa681”), Gfa1.6, and hGFA2.2. In one aspect, a GFAP promoter is GfaABC1D (also called “pGfa681”). In one aspect, a GFAP promoter is Gfa1.6. In one aspect, a GFAP promoter is hGFA2.2. In one aspect, pGfa681 is SEQ ID NO: 26. In one aspect, GFAP Gfa1.6 is SEQ ID NO: 4. In one aspect, hGfa2.2 is SEQ ID NO: 12. In one aspect, a GFAP promoter is selected from the group consisting of SEQ ID NOs: 4, 12, and 26. In one aspect, a GFAP promoter is SEQ ID NO: 4. In one aspect, a GFAP promoter is SEQ ID NO: 12. In one aspect, a GFAP promoter is SEQ ID NO: 26.

[00219] In one aspect, a GFAP promoter sequence is selected from the group consisting of a chimpanzee GFAP promoter sequence, a bonobo GFAP promoter sequence, an orangutan GFAP promoter sequence, a gorilla GFAP promoter sequence, a macaque GFAP promoter sequence, a marmoset GFAP promoter sequence, a capuchin GFAP promoter sequence, a baboon GFAP promoter sequence, a gibbon GFAP promoter sequence, and a lemur GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a chimpanzee GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a bonobo GFAP promoter sequence. In one aspect, a GFAP promoter sequence is an orangutan GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a gorilla GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a macaque GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a marmoset GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a capuchin GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a baboon GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a gibbon GFAP promoter sequence. In one aspect, a GFAP promoter sequence is a lemur GFAP promoter sequence.

[00220] In an aspect, a GFAP promoter sequence comprises at least 100 nucleotides. In one aspect, a GFAP promoter comprises at least 500 nucleotides. In a further aspect, a GFAP promoter comprises at least 1000 nucleotides. In still another aspect, a GFAP promoter comprises at least 1400 nucleotides.

[00221] It is appreciated in the art that a fragment of a promoter sequence can function to drive transcription of an operably linked nucleic acid molecule. For example, without being limiting, if a 1000 nucleotides promoter is truncated to 500 nucleotides, and the 500 nucleotides fragment is capable of driving transcription, the 500 nucleotides fragment is referred to as a “functional fragment.”

[00222] In an aspect, a promoter comprises at least 10 nucleotides. In one aspect, a promoter comprises at least 50 nucleotides. In one aspect, a promoter comprises at least 100 nucleotides. In one aspect, an intron comprises at least 140 nucleotides. In one aspect, a promoter comprises at least 200 nucleotides. In one aspect, a promoter comprises at least 250 nucleotides.

5 In one aspect, a promoter comprises at least 300 nucleotides. In one aspect, a promoter comprises at least 350 nucleotides. In one aspect, a promoter comprises at least 400 nucleotides. In one aspect, a promoter comprises at least 450 nucleotides. In one aspect, a promoter comprises at least 500 nucleotides. In one aspect, a promoter comprises between 50 nucleotides and 7500 nucleotides. In one aspect, a promoter comprises between 50 nucleotides and 5000 nucleotides.

10 In one aspect, a promoter comprises between 50 nucleotides and 2500 nucleotides. In one aspect, a promoter comprises between 50 nucleotides and 1000 nucleotides. In one aspect, a promoter comprises between 50 nucleotides and 500 nucleotides. In one aspect, a promoter comprises between 10 nucleotides and 7500 nucleotides. In one aspect, a promoter comprises between 10 nucleotides and 5000 nucleotides. In one aspect, a promoter comprises between 10 nucleotides and 1000 nucleotides. In one aspect, a promoter comprises between 10 nucleotides and 500 nucleotides

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[00223] In an aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 70% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 75% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 80% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 85% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 90% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 91% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 92% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 93% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 94% identical to a sequence selected from the group consisting of

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SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 95% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 96% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 97% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 98% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 99% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 99.5% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 99.8% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence at least 99.9% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof. In one aspect, a GFAP promoter nucleic acid sequence comprises a sequence 100% identical to a sequence selected from the group consisting of SEQ ID NO: 4, 12, 26, and functional fragment thereof.

- [00224] In an aspect, a nucleic acid sequence as provided herein is codon optimized.
- [00225] In an aspect, a nucleic acid sequence as provided herein is CpG site depleted.
- [00226] As used herein, the term “brain” refers to an organ that functions as the center of the nervous system. In an aspect, a brain comprises a cerebrum, a cerebral cortex, a cerebellum, and/or a brain stem.
- [00227] As used herein, the term “cerebral cortex” refers to the outer layer of neural tissue of the cerebrum.
- [00228] As used herein, the term “striatum” or “corpus striatum” refers to a cluster of neurons in the subcortical basal ganglia of the forebrain and comprises the ventral striatum and dorsal striatum.
- [00229] As used herein, the term “substantia nigra” refers to a cluster of neurons in the subcortical basal ganglia of the midbrain and comprises the pars compacta and the pars reticulata.
- [00230] As used herein, the term “forebrain” refers to the forward-most portion of the brain.

[00231] As used herein, the term “putamen” refers to a round structure at the base of the forebrain and is a component of the dorsal striatum.

[00232] As used herein, the term “caudate nucleus” refers to a structure at the base of the forebrain and is a component of the dorsal striatum.

5 **[00233]** As used herein, the term “subcortical basal ganglia” refers to a cluster of neurons in the deep cerebral hemispheres of the brain.

[00234] As used herein, the term “spinal cord” refers to a structure that functions in the transmission of nerve signals from the motor cortex to the body.

[00235] As used herein, the term “motor cortex” refers to a region in the frontal lobe of the cerebral cortex that is involved in the planning, control, and execution of voluntary movements.

[00236] In an aspect, a method provided herein converts reactive astrocytes to functional neurons in the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a cerebral cortex of the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a striatum of the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a dorsal striatum of the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a spinal cord of the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a putamen of the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a caudate nucleus of the brain. In one aspect, a method provided herein converts reactive astrocytes to functional neurons in a substantia nigra of the brain.

[00237] Elongation factor-1 alpha (EF-1 alpha; also referred to as eEF1a1) is an isoform of the alpha subunit of the elongation factor 1 complex. The complex is involved in the enzymatic delivery of aminoacyl tRNAs to the ribosome. The EF-1 alpha isoform is expressed in the brain, placenta, lung, liver, kidney, and pancreas.

[00238] In an aspect, an enhancer sequence from the EF-1 alpha promoter is a human enhancer sequence from the EF-1 alpha promoter. In one aspect, an enhancer sequence from the EF-1 alpha promoter is selected from the group consisting of a chimpanzee enhancer sequence from the EF-1 alpha promoter, a bonobo enhancer sequence from the EF-1 alpha promoter, an orangutan enhancer sequence from the EF-1 alpha promoter, a gorilla enhancer sequence from the EF-1 alpha promoter, a macaque enhancer sequence from the EF-1 alpha promoter, a marmoset enhancer sequence from the EF-1 alpha promoter, a capuchin enhancer sequence from the EF-1 alpha promoter, a baboon enhancer sequence from the EF-1 alpha promoter, a gibbon enhancer sequence from the EF-1 alpha promoter, and a lemur enhancer sequence from the EF-1 alpha promoter. In one aspect, an enhancer sequence from the EF-1 alpha promoter is a chimpanzee an

enhancer sequence from the EF-1 alpha promoter. In one aspect, an enhancer sequence from the EF-1 alpha promoter is a bonobo enhancer sequence from the EF-1 alpha promoter. In one aspect, an enhancer sequence from the EF-1 alpha promoter is an orangutan enhancer sequence from the EF-1 alpha promoter. In one aspect, an enhancer sequence from the EF-1 alpha promoter is a gorilla enhancer sequence from the EF-1 alpha promoter. In one aspect, an enhancer sequence from the EF-1 alpha promoter is a macaque enhancer sequence from the EF-1 alpha promoter. In one aspect, enhancer sequence from the EF-1 alpha promoter is a marmoset enhancer sequence from the EF-1 alpha promoter. In one aspect, enhancer sequence from the EF-1 alpha promoter is a capuchin enhancer sequence from the EF-1 alpha promoter. In one aspect, enhancer sequence from the EF-1 alpha promoter is a baboon enhancer sequence from the EF-1 alpha promoter. In one aspect, enhancer sequence from the EF-1 alpha promoter is a gibbon enhancer sequence from the EF-1 alpha promoter. In one aspect, enhancer sequence from the EF-1 alpha promoter is a lemur enhancer sequence from the EF-1 alpha promoter.

[00239] In an aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic

acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 2, or the complement thereof. In one aspect, an enhancer from the EF-1 alpha promoter nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 2, or the complement thereof.

[00240] Cytomegalovirus (CMV) is a genus of viruses in the order *Herpesvirale*.

[00241] In an aspect, an enhancer sequence from the CMV is a human enhancer sequence from the CMV. In one aspect, an enhancer sequence from the CMV is selected from the group consisting of a chimpanzee enhancer sequence from the CMV, a bonobo enhancer sequence from the CMV, an orangutan enhancer sequence from the CMV, a gorilla enhancer sequence from the CMV, a macaque enhancer sequence from the CMV, a marmoset enhancer sequence from the CMV, a capuchin enhancer sequence from the CMV, a baboon enhancer sequence from the CMV, a gibbon enhancer sequence from the CMV, and a lemur enhancer sequence from the CMV. In one aspect, an enhancer sequence from the CMV is a chimpanzee an enhancer sequence from the CMV. In one aspect, an enhancer sequence from the CMV is a bonobo enhancer sequence from the CMV. In one aspect, an enhancer sequence from the CMV is an orangutan enhancer sequence from the CMV. In one aspect, an enhancer sequence from the CMV is a gorilla enhancer sequence from the CMV. In one aspect, an enhancer sequence from the CMV is a macaque enhancer sequence from the CMV. In one aspect, enhancer sequence from the CMV is a marmoset enhancer sequence from the CMV. In one aspect, enhancer sequence from the CMV is a capuchin enhancer sequence from the CMV. In one aspect, enhancer sequence from the CMV is a baboon enhancer sequence from the CMV. In one aspect, enhancer sequence from the CMV is a gibbon enhancer sequence from the CMV. In one aspect, enhancer sequence from the CMV is a lemur enhancer sequence from the CMV.

[00242] In an aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence

- at least 85% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 911% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 11, or the complement thereof. In one aspect, an enhancer from the CMV nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 11, or the complement thereof.
- 25 **[00243]** In an aspect, an enhancer is selected from the group consisting of an enhancer from EF1- α promoter and CMV enhancer. In one aspect, an enhancer is from EF1- α promoter. In one aspect, an enhancer is an CMV enhancer.
- [00244]** In an aspect, a vector of the present disclosures comprises a chimeric intron. In an aspect the chimeric intron is composed of the 5'-donor site from the first intron of the human β -globin gene and the branch and 3'-acceptor site from the intron of an immunoglobulin gene heavy chain variable region. In an aspect, the chimeric intron is a chimeric intron of a rabbit beta-globin and a chicken beta actin similar in CAG promoter. In an aspect, a vector of the present disclosure comprises a glial fibrillary acid protein (GFAP) intron. In an aspect, a vector of the present disclosure comprises a glial fibrillary acid protein (GFAP) first intron.

[00245] Introns can be grouped into at least five classes, including: spliceosomal introns; transfer RNA introns; group I introns; group II introns; and group III introns. An intron can be synthetically produced, varied, or derived from a known or naturally occurring intron sequence or other intron sequence. An intron can also include a chimeric intron comprising a combination of two or more heterologous sequences. An intron of the present application can thus include variants of intron sequences that are similar in composition, but not identical to, other intron sequence(s) known or provided herein. In an aspect, an intron comprises at least 10 nucleotides. In one aspect, an intron comprises at least 50 nucleotides. In one aspect, an intron comprises at least 100 nucleotides. In one aspect, an intron comprises at least 140 nucleotides. In one aspect, an intron comprises at least 200 nucleotides. In one aspect, an intron comprises at least 250 nucleotides. In one aspect, an intron comprises at least 300 nucleotides. In one aspect, an intron comprises at least 350 nucleotides. In one aspect, an intron comprises at least 400 nucleotides. In one aspect, an intron comprises at least 450 nucleotides. In one aspect, an intron comprises at least 500 nucleotides. In one aspect, an intron comprises between 50 nucleotides and 7500 nucleotides. In one aspect, an intron comprises between 50 nucleotides and 5000 nucleotides. In one aspect, an intron comprises between 50 nucleotides and 2500 nucleotides. In one aspect, an intron comprises between 50 nucleotides and 1000 nucleotides. In one aspect, an intron comprises between 50 nucleotides and 500 nucleotides. In one aspect, an intron comprises between 10 nucleotides and 7500 nucleotides. In one aspect, an intron comprises between 10 nucleotides and 5000 nucleotides. In one aspect, an intron comprises between 10 nucleotides and 2500 nucleotides. In one aspect, an intron comprises between 10 nucleotides and 1000 nucleotides. In one aspect, an intron comprises between 10 nucleotides and 500 nucleotides.

[00246] In an aspect, a chimeric intron nucleic acid sequence is selected from the group consisting of SEQ ID NOs: 5 and 27. In one aspect, a chimeric intron nucleic acid sequence is SEQ ID NO: 5. In one aspect, a chimeric intron nucleic acid sequence is SEQ ID NO: 27.

[00247] In an aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 5, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 5, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 5, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 5, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 5, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 5, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence

[00248] In an aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a

chimeric intron nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 98% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 99% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 99.5% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 99.8% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 27, or the complement thereof. In one aspect, a chimeric intron nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 27, or the complement thereof.

[00249] The woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) is a DNA sequence that creates a tertiary structure enhancing expression of genes that are delivered in viral vectors.

[00250] In an aspect, a WPRE nucleic acid sequence is an optimized version of WPRE.

[00251] In an aspect, a WPRE nucleic acid sequence is selected from the group consisting of SEQ ID NOs: 7 and 29. In one aspect, a WPRE nucleic acid sequence is SEQ ID NO: 7. In one aspect, a WPRE nucleic acid sequence is SEQ ID NO: 29.

[00252] In an aspect, a WPRE nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 96% identical to SEQ ID NO: 7, or the complement thereof. In one aspect, a WPRE nucleic acid sequence comprises a sequence at least 97% identical to SEQ ID

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acid sequence comprises a sequence 100% identical to SEQ ID NO: 29, or the complement thereof.

[00254] SV40 polyadenylation signal sequence (also refer as SV40 PolyA; Simian virus 40 PolyA; and PolyA) is a DNA sequence that can terminate transcription and add a PolyA tail to the 3' end of a messenger RNA (mRNA).

[00255] hGH polyadenylation signal sequence (also refer as hGH PolyA) is a DNA sequence that can terminate transcription and add a PolyA tail to the 3' end of a messenger RNA (mRNA).

[00256] bGH polyadenylation signal sequence (also refer as bGH PolyA or bGHpA) refers to a PolyA signal or PolyA tail of a bovine growth hormone.

[00257] As used herein, a "PolyA tail" refers to a stretch of RNA that only contains the nucleobase adenine. In an aspect, an RNA molecule transcribed from an AAV vector construct provided herein comprises a PolyA tail. In one aspect, a PolyA tail comprises at least two adenines. In one aspect, a PolyA tail comprises at least ten adenines. In one aspect, a PolyA tail comprises at least 50 adenines. In one aspect, a PolyA tail comprises at least 100 adenines. In one aspect, a PolyA tail comprises at least 140 adenines. In one aspect, a PolyA tail comprises at least 200 adenines. In one aspect, a PolyA tail comprises at least 250 adenines. In one aspect, a PolyA tail comprises between 50 adenines and 300 adenines.

[00258] In an aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 70% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 75% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 80% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 85% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 90% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 91% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 92% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 93% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 94% identical to SEQ ID NO: 8, or the complement thereof. In one aspect, a SV40 polyadenylation signal nucleic acid sequence comprises a sequence at least 95% identical to SEQ ID NO: 8, or the complement thereof. In one

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[illegible]

at least 99.17% identical to SEQ ID NO: 30, or the complement thereof. In one aspect, a bGH polyadenylation signal nucleic acid sequence comprises a sequence at least 99.9% identical to SEQ ID NO: 30, or the complement thereof. In one aspect, a bGH polyadenylation signal nucleic acid sequence comprises a sequence 100% identical to SEQ ID NO: 30, or the complement thereof

5 [00261] As used herein, the term “central nervous system” or “CNS” refers to the brain and spinal cord of a bilaterally symmetric animal. The CNS also includes the retina, the optic nerve, olfactory nerves, and olfactory epithelium.

[00262] As used herein, the term “peripheral nervous system” or “PNS” refers to nerves and ganglia outside of the brain and spinal cord, excluding the retina, the optic nerve, olfactory
10 nerves, and olfactory epithelium. In an aspect, the peripheral nervous system is divided into the somatic nervous system and the autonomic nervous system.

[00263] As used herein, the term “somatic nervous system” refers to the parts of the PNS that are associated with voluntary control of body movements.

[00264] As used herein, the term “autonomic nervous system” refers to the parts of the PNS
15 that regulate the function of internal organs

[00265] As used herein, the term “GFAP positive” refers to a cell having detectable protein accumulation of human glial fibrillary acid protein (GFAP) or detectable accumulation of GFAP mRNA expression using techniques standard in the art. In one aspect, a glial cell is GFAP positive.

20 [00266] As used herein, the term “detectable” refers to protein or mRNA accumulation that is identifiable.

[00267] Protein accumulation can be identified using antibodies. Non limiting examples of measuring protein accumulation include Western blots, enzyme linked immunosorbent assays (ELISAs), immunoprecipitations and immunofluorescence. An antibody provided herein can be
25 a polyclonal antibody or a monoclonal antibody. An antibody having specific binding affinity for a protein provided herein can be generated using methods well known in the art. An antibody provided herein can be attached to a solid support such as a microtiter plate using methods known in the art.

[00268] As used herein, the term “multiplicity of infection” and “MOI” refers to a the
30 number of virions that are added per cell during infection.

[00269] As used herein, the term “virion” refers to the infective form of a virus outside a host cell.

[00270] As used herein, the term “neurological condition” refers to a disorder, illness, sickness, injury, or disease, in the central nervous system or the peripheral nervous system. Non-
35 limiting examples of neurological conditions can be found in Neurological Disorders: course and

treatment, 2nd Edition (2002) (Academic Press Inc.) and Christopher Goetz, Textbook of Clinical Neurology, 3rd Edition (2007) (Saunders).

[00271] As used herein, the term “injury” refers to damage to the central nervous system or peripheral nervous system.

5 **[00272]** In one aspect, a neurological condition is selected from the group consisting of Alzheimer’s Disease, Parkinson’s Disease, amyotrophic lateral sclerosis (ALS), Huntington’s Disease, epilepsy, physical injury, stroke, cerebral aneurysm, traumatic brain injury, concussion, a tumor, inflammation, infection, ataxia, brain atrophy, spinal cord atrophy, multiple sclerosis, traumatic spinal cord injury, ischemic or hemorrhagic myelopathy (myelopathy), global ischemia,
 10 hypoxic ischemic encephalopathy, embolism, fibrocartilage embolism myelopathy, thrombosis, nephropathy, chronic inflammatory disease, meningitis, and cerebral venous sinus thrombosis. In one aspect, a neurological condition is Alzheimer’s Disease. In one aspect, a neurological condition is Parkinson’s Disease. In one aspect, a neurological condition is ALS. In one aspect, a neurological condition is Huntington’s Disease. In one aspect, a neurological condition is
 15 epilepsy. In one aspect, a neurological condition is a physical injury. In one aspect, a neurological condition is stroke. In one aspect, a neurological condition is ischemic stroke. In one aspect, a neurological condition is hemorrhagic stroke. In one aspect, a neurological condition is cerebral aneurysm. In one aspect, a neurological condition is traumatic brain injury. In one aspect, a neurological condition is concussion. In one aspect, a neurological condition is a tumor. In one
 20 aspect, a neurological condition is inflammation. In one aspect, a neurological condition is infection. In one aspect, a neurological condition is ataxia. In, one aspect, a neurological condition is brain atrophy. In, one aspect, a neurological condition is spinal cord atrophy. In one aspect, a neurological condition is multiple sclerosis. In one aspect, a neurological condition is traumatic spinal cord injury. In one aspect, a neurological condition is ischemic or hemorrhagic
 25 myelopathy (myelopathy). In one aspect, a neurological condition is global ischemia. In one aspect, a neurological condition is hypoxic ischemic encephalopathy. In one aspect, a neurological condition is embolism. In one aspect, a neurological condition is fibrocartilage embolism myelopathy. In one aspect, a neurological condition is thrombosis. In one aspect, a neurological condition is nephropathy. In one aspect, a neurological condition is chronic
 30 inflammatory disease. In one aspect, a neurological condition is meningitis. In one aspect, a neurological condition is cerebral venous sinus thrombosis.

[00273] In an aspect, a neurological condition comprises an injury to the CNS or to the PNS. In one aspect, a neurological condition comprises an injury to the CNS. In one aspect, a neurological condition comprises an injury to the PNS.

[00274] In an aspect, this disclosure provides, and includes, a method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[00275] In an aspect, this disclosure provides, and includes, a method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, (iii) or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and hDlx2 coding sequence are operably linked to expression control elements comprising: (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a

nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26; (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11; (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

[00276] In an aspect, this disclosure provides, and includes, a method of converting glial cells to neurons in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising: (a) a glial fibrillary acid protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal sequence, wherein said AAV vector is capable of converting at least one glial cell to a neuron in said subject in need thereof.

[00277] In an aspect, this disclosure provides, and includes, a method of treating a neurological condition in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject, wherein said AAV comprises a DNA vector comprising a neurogenic differentiation 1 (NeuroD1) sequence and distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and said Dlx2 sequence are operably linked to expression control elements comprising: (a) a glial fibrillary acid protein (GFAP) promoter; (b) an enhancer; (c) a chimeric intron; (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and (e) a polyadenylation signal to said subject in need thereof.

[00278] In an aspect, a method as provided herein, is capable of converting at least one glial cell to a neuron. In one aspect, a method as provided herein converts at least one glial cell to a neuron.

[00279] Achaete-scute family BHLH transcription factor 1 (Ascl1; also referred to as ASH1, HASH1, MASH-1, and bHLHa46) encodes a member of the basic helix-loop-helix family of transcription factors and is a gene that plays a role in neuronal commitment and differentiation..

[00280] Insulin gene enhancer protein (ISL1; also known as ISL LIM homeobox-1 and ISLET1) is a gene that encodes a transcription factor containing two N-terminal LIM domains and one C-terminal homeodomain. The encoded protein plays a role in the embryogenesis of pancreatic islets of Langerhans.

5 **[00281]** LIM-homeobox 3 (LHX3; also known as LIM3 and CPHD3) gene encodes for a protein from a family of proteins with a unique cysteine-rich zinc-binding domain (LIM domain).

[00282] Huntingtin (Htt; also known as Huntington Disease gene) gene encodes for the huntingtin protein. The wild type contains 6-35 glutamine residues and the mutated Htt contains more than 36 glutamine residue.

10 **[00283]** In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 coding sequence and a Dlx2 coding sequence in accordance with the present disclosure. In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 coding sequence in combination with a second AAV vector comprising a Dlx2 coding sequence. In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 or a Dlx2 coding
15 sequence in combination with a second AAV vector comprising a third transcription factor coding sequence. In one aspect, a third transcription factor is selected from the group consisting of Ascl1, ISL1, and LHX3. In one aspect, a third transcription factor is Ascl1. In one aspect, a second transcription factor is ISL1. In one aspect, a third transcription factor is LHX3. In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 coding sequence, a Dlx2
20 coding sequence, a second NeuroD1 coding sequence, and a second Dlx2 coding sequence. In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with a second AAV vector comprising a NeuroD1 and a Dlx2 coding sequence.

[00284] In one aspect, a method as provided herein uses an AAV vector comprising a
25 NeuroD1 and a Dlx2 coding sequence in combination with an AAV vector comprising a shRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an AAV vector comprising a shRNA sequence targeting Htt and a second shRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding
30 sequence in combination with an AAV vector comprising a shRNA sequence targeting Htt, a second shRNA sequence targeting Htt, and a third shRNA targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence and a shRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence and a shRNA sequence targeting Htt and a
35 second shRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV

vector comprising a NeuroD1 and a Dlx2 coding sequence and a shRNA sequence targeting Htt, a second shRNA sequence targeting Htt and a third shRNA targeting Htt.

[00285] In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an ASO sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an ASO sequence targeting Htt and a second ASO sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an ASO sequence targeting Htt, a second ASO sequence targeting Htt, and a third ASO targeting Htt.

[00286] In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an siRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an siRNA sequence targeting Htt and a second siRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an siRNA sequence targeting Htt, a second siRNA sequence targeting Htt, and a third siRNA targeting Htt.

[00287] In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an AAV vector comprising a miRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an AAV vector comprising a miRNA sequence targeting Htt and a second miRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an AAV vector comprising a miRNA sequence targeting Htt, a second miRNA sequence targeting Htt, and a third miRNA targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence and a miRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence and a miRNA sequence targeting Htt and a second miRNA sequence targeting Htt. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence and a miRNA sequence targeting Htt, a second miRNA sequence targeting Htt and a third MIRNA targeting Htt.

[00288] In one aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence in combination with an AAV vector comprising a gRNA sequence targeting Htt and a CAS nuclease. In an aspect, a method as provided herein uses an AAV vector comprising a NeuroD1 and a Dlx2 coding sequence and a gRNA sequence targeting Htt and a CAS nuclease.

- [00289] In an aspect, an AAV vector as provided herein, is measured for functionality by assessing transcription levels and protein levels of NeuN, doublecortin (DCX), β 3-tubulin, (neurofilament 200) NF-200, (microtubule-associated protein 2) MAP2, ionized calcium binding adaptor molecule (Iba1).
- 5 [00290] As used herein, the term “NeuN” or “Fox-3” or “Rbfox2” or “Hexaribonucleotide Binding Protein-3” refers to a protein which is a homologue to the protein product of a sex-determining gene in *Caenorhabditis elegans* and is a neuronal nuclear antigen.
- [00291] As used herein, the term “DCX” or “doubling” or “lissencephalin-X” refers to a microtubule-associated protein expressed by neuronal precursor cells and immature neurons in
10 embryonic and adult cortical structures.
- [00292] As used herein, the term “ β 3-tubulin” or “Class III β -tubulin” or “ β -tubulin III” refers to a microtubule element of the tubulin family found in neurons.
- [00293] As used herein, the term “NF-200” refers to a class of protein that is a type IV intermediate filaments found in the cytoplasm of neurons.
- 15 [00294] As used herein, the term “MAP2” refers to a protein that belongs to the microtubule-associated protein family and play a role in determining and stabilizing neuronal morphology during neuron development.
- [00295] As used herein, the term “Iba1” refers to a microglia macrophage-specific calcium binding protein.
- 20 [00296] In an aspect a method provided herein converts glial cells to neurons in combination with gene editing techniques. In one aspect, a gene editing technique targets the mutant Htt. In one aspect, a gene editing technique is selected from the group consisting of siRNA, miRNA, ASO, and CRISPR/CAS. In one aspect, a gene editing technique is siRNA. In one aspect, a gene editing technique is miRNA. In one aspect, a gene editing technique is ASO.
- 25 In one aspect, a gene editing technique is CRISPR/CAS.
- [00297] In an aspect, a composition as provided herein, is capable of converting at least one glial cell to a neuron. In one aspect, a composition as provided herein converts at least one glial cell to a neuron
- [00298] As used herein, the term “mammal” refers to any species classified in the class
30 Mammalia.
- [00299] As used herein, the term “human” refers to a *Homo sapiens*. In an aspect, a human has a neurological disorder.
- [00300] As used herein, the term “living human” refers to a human that has heart, respiration and brain activity.

[00301] As used herein, the term “non-human primate” refers to any species or subspecies classified in the order Primates that are not *Homo sapiens*. Non-limiting examples of non-human primates include chimpanzee, bonobo, orangutan, gorilla, macaque, marmoset, capuchin, baboon, gibbon, and lemur.

5 [00302] As used herein, the term “delivering” or “delivery” refers to treating a mammal with an AAV vector or composition as provided herein. In an aspect, an AAV vector or composition as provided herein is delivered to a subject in need thereof. In one aspect, an AAV vector or composition as provided herein is formulated to be delivered to a subject in need thereof. In one aspect, delivering comprises local delivery. In one aspect, an AAV vector or composition as provided herein is formulated for local delivery. In one aspect, delivering comprises systemic delivery. In one aspect, an AAV vector or composition as provided herein is formulated for systemic delivery. In one aspect, delivery comprises injecting an AAV vector or composition as provided herein into a subject in need thereof. In one aspect, delivering is selected from the group consisting of intraperitoneal, intramuscular, intravenous, intrathecal, intracerebral, intracranial, 10 intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal, intranasal, or oral administration. In one aspect, delivery comprises intraperitoneal delivery. In one aspect, delivery comprises intramuscular delivery. In one aspect, delivery comprises intravenous delivery. In one aspect, delivery comprises intrathecal delivery. In one aspect, delivery comprises intracerebral delivery. In one aspect, delivery comprises intracranial delivery. In one aspect, delivery comprises intra lateral ventricle of the brain delivery. In one aspect, delivery comprises intra cisterna magna delivery. In one aspect, delivery comprises intra vitreous delivery. In one aspect, delivery comprises intra-subretina delivery. In one aspect, delivery comprises intraparenchymal delivery. In one aspect, delivery comprises intranasal delivery. In one aspect, delivery comprises oral administration.

25 [00303] As used herein, the term “injecting” refers to delivering an AAV vector or composition as provided herein under pressure and with force. As a non-limiting example, injecting can comprise the use of a syringe and needle.

[00304] In an aspect, an AAV vector or composition as provided herein is injected into a brain of a subject. In one aspect, an AAV vector or composition is injected into a cerebral cortex of a subject. In one aspect, an AAV vector or composition as provided herein is injected in the striatum of a subject. In one aspect, an AAV vector or composition as provided herein is injected in to a spinal cord or a subject. In one aspect, an AAV vector or composition is injected in the striatum of a subject. In one aspect, an AAV vector or composition is injected in the dorsal striatum of a subject. In one aspect, an AAV vector or composition is injected in the putamen of a subject. In one aspect, an AAV vector or composition is injected in the caudate nucleus of a 35

subject. In one aspect, an AAV vector or composition is injected in the substantia nigra of a subject

[00305] In an aspect, an AAV vector or composition as provided herein has spread in the brain between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the brain between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00306] In an aspect, an AAV vector or composition as provided herein has spread in the cerebral cortex between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the cerebral cortex between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00307] In an aspect, an AAV vector or composition as provided herein has spread in the spinal cord between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the spinal cord between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about

50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00308] In an aspect, an AAV vector or composition as provided herein has spread in the striatum between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the striatum between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00309] In an aspect, an AAV vector or composition as provided herein has spread in the dorsal striatum between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the dorsal striatum between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00310] In an aspect, an AAV vector or composition as provided herein has spread in the putamen between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the putamen between about 1% and about 10%, between 1% and

about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00311] In an aspect, an AAV vector or composition as provided herein has spread in the caudate nucleus between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the caudate nucleus between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00312] In an aspect, an AAV vector or composition as provided herein has a spread at from injection site between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has a spread from injection site between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about

90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00313] In an aspect, an AAV vector or composition as provided herein has spread in the substantia nigra between about 1% and about 100%. In one aspect, an AAV vector or composition as provided herein has spread in the putamen between about 1% and about 10%, between 1% and about 20%, between 1% and about 30%, between 10% and about 20%, between 10% and about 30%, between about 10% and about 40%, between about 20% and about 30%, between about 20% and about 40%, between about 20% and about 50%, between about 30% and about 40%, between about 30% and about 50%, between about 30% and about 60%, between about 40% and about 50%, between about 40% and about 60%, between about 40% and about 70%, between about 50% and about 60%, between about 50% and about 70%, between about 50% and about 80%, between about 60% and about 70%, between about 60% and about 80%, between about 60% and about 90%, between about 70% and about 80%, between about 70% and about 90%, between about 70% and about 100%, between about 80% and about 90%, between about 80% and about 100%, or between about 90% and about 100%.

[00314] As used herein, the term “AAV particle” refers to packaged capsid forms of the AAV virus that transmits its nucleic acid genome to cells.

[00315] In an aspect, a composition comprising an AAV particle encoded by an AAV vector as provided herein is injected at a concentration between 10^{10} AAV particles/mL and 10^{14} AAV particles/mL. In one aspect, a composition comprising an AAV particle encoded by an AAV vector as provided herein is injected at a concentration between 10^{10} AAV particles/mL and 10^{11} AAV particles/mL, between 10^{10} AAV particles/mL and 10^{12} AAV particles/mL, between 10^{10} AAV particles/mL and 10^{13} AAV particles/mL, between 10^{11} AAV particles/mL and 10^{12} AAV particles/mL, between 10^{11} AAV particles/mL and 10^{13} AAV particles/mL, between 10^{11} AAV particles/mL and 10^{14} AAV particles/mL, between 10^{12} AAV particles/mL and 10^{13} AAV particles/mL, between 10^{12} AAV particles/mL and 10^{14} AAV particles/mL, or between 10^{13} AAV particles/mL and 10^{14} AAV particles/mL.

[00316] In an aspect, a composition comprising an AAV particle encoded by an AAV vector as provided herein is injected at volume between 10 μ L and 1000 μ L. In one aspect, a composition comprising an AAV particle encoded by an AAV vector as provided herein is injected at volume between 10 μ L and 100 μ L, between 10 μ L and 200 μ L, between 10 μ L and 300 μ L, between 100 μ L and 200 μ L, between 100 μ L and 300 μ L, between 100 μ L and 400 μ L, between 200 μ L and 300 μ L, between 200 μ L and 400 μ L, between 200 μ L and 500 μ L, between 300 μ L and 400 μ L, between 300 μ L and 500 μ L, between 300 μ L and 600 μ L, between 400 μ L and 500 μ L, between 400 μ L and 600 μ L, between 400 μ L and 700 μ L, between 500 μ L and 600

μL, between 500 μL and 700 μL, between 500 μL and 800 μL, between 600 μL and 700 μL, between 600 μL and 800 μL, between 600 μL and 900 μL, between 700 μL and 800 μL, between 700 μL and 900 μL, between 700 μL and 1000 μL, between 800 μL and 900 μL, between 800 μL and 1000 μL, or between 900 μL and 1000 μL.

- 5 **[00317]** As used herein, the term “subject” refers to any animal subject. Non-limiting examples of animal subjects include humans, laboratory animals (*e.g.*, primates, rats, mice), livestock (*e.g.*, cows, sheep, goats, pigs, turkeys, chickens), and household pets (*e.g.*, dogs, cats, rodents, etc.).

- [00318]** As used herein, “a subject in need thereof” refers to a subject with a neurological condition. In an aspect, a subject in need thereof has a neurological condition selected from the group consisting of Alzheimer’s Disease, Parkinson’s Disease, amyotrophic lateral sclerosis (ALS), Huntington’s Disease, epilepsy, physical injury, stroke, cerebral aneurysm, traumatic brain injury, concussion, a tumor, inflammation, infection, ataxia, brain atrophy, spinal cord atrophy, multiple sclerosis, traumatic spinal cord injury, ischemic or hemorrhagic myelopathy (myelopathy), global ischemia, hypoxic ischemic encephalopathy, embolism, fibrocartilage embolism myelopathy, thrombosis, nephropathy, chronic inflammatory disease, meningitis, and cerebral venous sinus thrombosis. In one aspect, a subject in need thereof has Alzheimer’s Disease. In one aspect, a subject in need thereof has Parkinson’s Disease. In one aspect, a subject in need thereof has ALS. In one aspect, a subject in need thereof has Huntington’s Disease. In one aspect, a subject in need thereof has epilepsy. In one aspect, a subject in need thereof has a physical injury. In one aspect, a subject in need thereof has a stroke. In one aspect, a subject in need thereof has ischemic stroke. In one aspect, a subject in need thereof has hemorrhagic stroke. In one aspect, a subject in need thereof has a cerebral aneurysm. In one aspect, a subject in need thereof has traumatic brain injury. In one aspect, a subject in need thereof has concussion. In one aspect, a subject in need thereof has a tumor. In one aspect, a subject in need thereof has inflammation. In one aspect, a subject in need thereof has an infection. In, one aspect, a subject in need thereof has ataxia. In, one aspect, a subject in need thereof has brain atrophy. In one aspect, a subject in need thereof has spinal cord atrophy. In one aspect, a subject in need thereof has multiple sclerosis. In one aspect, a subject in need thereof has a traumatic spinal cord injury. In one aspect, a subject in need thereof has ischemic or hemorrhagic myelopathy (myelopathy). In one aspect, a subject in need thereof has global ischemia. In one aspect, a subject in need thereof has hypoxic ischemic encephalopathy. In one aspect, a subject in need thereof has an embolism. In one aspect, a subject in need thereof has fibrocartilage embolism myelopathy. In one aspect, a subject in need thereof has thrombosis. In one aspect, a subject in need thereof has nephropathy. In one aspect, a subject in need thereof has chronic inflammatory disease. In one

aspect, a subject in need thereof has meningitis. In one aspect, a subject in need thereof has cerebral venous sinus thrombosis.

[00319] In an aspect, a subject in need thereof is a mammal. In one aspect, a subject in need thereof is a human. In one aspect, a subject in need thereof is a non-human primate. In one aspect, a subject in need thereof is selected from the group consisting of chimpanzee, bonobo, orangutan, gorilla, macaque, marmoset, capuchin, baboon, gibbon, and lemur. In one aspect, a subject in need thereof is a chimpanzee. In one aspect, a subject in need thereof is a bonobo. In one aspect, a subject in need thereof is orangutan. In one aspect, a subject in need thereof is gorilla. In one aspect, a subject in need thereof is a macaque. In one aspect, a subject in need thereof is marmoset. In one aspect, a subject in need thereof is a capuchin. In one aspect, a subject in need thereof is a baboon. In one aspect, a subject in need thereof is a gibbon. In one aspect, a subject in need thereof is lemur.

[00320] In one aspect, a subject in need thereof is a male. In one aspect, a subject in need thereof is a female. In one aspect, a subject in need thereof is gender neutral. In one aspect, a subject in need thereof is a premature newborn. In one aspect, a premature newborn is born before 36 weeks gestation. In one aspect, a subject in need thereof is a term newborn. In one aspect, a term newborn is below about 2 months old. In one aspect, a subject in need thereof is a neonate. In one aspect, a neonate is below about 1 month old. In one aspect, a subject in need thereof is an infant. In one aspect, an infant is between 2 months and 24 months old. In one aspect, an infant is between 2 months and 3 months, between 2 months and 4 months, between 2 months and 5 months, between 3 months and 4 months, between 3 months and 5 months, between 3 months and 6 months, between 4 months and 5 months, between 4 months and 6 months, between 4 months and 7 months, between 5 months and 6 months, between 5 months and 7 months, between 5 months and 8 months, between 6 months and 7 months, between 6 months and 8 months, between 6 months and 9 months, between 7 months and 8 months, between 7 months and 9 months, between 7 months and 10 months, between 8 months and 9 months, between 8 months and 10 months, between 8 months and 11 months, between 9 months and 10 months, between 9 months and 11 months, between 9 months and 12 months, between 10 months and 11 months, between 10 months and 12 months, between 10 months and 13 months, between 11 months and 12 months, between 11 months and 13 months, between 11 months and 14 months, between 12 months and 13 months, between 12 months and 14 months, between 12 months and 14 months, between 13 months and 14 months, between 13 months and 16 months, between 14 months and 14 months, between 14 months and 16 months, between 14 months and 17 months, between 14 months and 16 months, between 14 months and 17 months, between 14 months and 18 months, between 16 months and 17 months, between 16 months and 18 months,

between 16 months and 19 months, between 17 months and 18 months, between 17 months and 19 months, between 17 months and 20 months, between 18 months and 19 months, between 18 months and 20 months, between 18 months and 21 months, between 19 months and 20 months, between 19 months and 21 months, between 19 months and 22 months, between 20 months and 21 months, between 20 months and 22 months, between 20 months and 23 months, between 21 months and 22 months, between 21 months and 23 months, between 21 months and 24 months, between 22 months and 23 months, between 22 months and 24 months, and between 23 months and 24 months old. In one aspect, a subject in need thereof is a toddler. In one aspect, a toddler is between 1 year and 4 years old. In one aspect, a toddler is between 1 year and 2 years, between 1 year and 3 years, between 1 year and 4 years, between 2 years and 3 years, between 2 years and 4 years, and between 3 years and 4 years old. In one aspect, a subject in need thereof is a young child. In one aspect, a young child is between 2 years and 5 years old. In one aspect, a young child is between 2 years and 3 years, between 2 years and 4 years, between 2 years and 5 years, between 3 years and 4 years, between 3 years and 5 years, and between 4 years and 5 years old. In one aspect, a subject in need thereof is a child. In one aspect, a child is between 6 years and 12 years old. In one aspect, a child is between 6 years and 7 years, between 6 years and 8 years, between 6 years and 9 years, between 7 years and 8 years, between 7 years and 9 years, between 7 years and 10 years, between 8 years and 9 years, between 8 years and 10 years, between 8 years and 11 years, between 9 years and 10 years, between 9 years and 11 years, between 9 years and 12 years, between 10 years and 11 years, between 10 years and 12 years, and between 11 years and 12 years old. In one aspect, a subject in need thereof is an adolescent. In one aspect, an adolescent is between 13 years and 19 years old. In one aspect, an adolescent is between 13 years and 14 years, between 13 years and 14 years, between 13 years and 16 years, between 14 years and 14 years, between 14 years and 16 years, between 14 years and 17 years, between 14 years and 16 years, between 14 years and 17 years, between 14 years and 18 years, between 16 years and 17 years, between 16 years and 18 years, between 16 years and 19 years, between 17 years and 18 years, between 17 years and 19 years, and between 18 years and 19 years old. In one aspect, a subject in need thereof is a pediatric subject. In one aspect, a pediatric subject between 1 day and 18 years old. In one aspect, a pediatric subject is between 1 day and 1 year, between 1 day and 2 years, between 1 day and 3 years, between 1 year and 2 years, between 1 year and 3 years, between 1 year and 4 years, between 2 years and 3 years, between 2 years and 4 years, between 2 years and 5 years, between 3 years and 4 years, between 3 years and 5 years, between 3 years and 6 years, between 4 years and 5 years, between 4 years and 6 years, between 4 years and 7 years, between 5 years and 6 years, between 5 years and 7 years, between 5 years and 8 years, between 6 years and 7 years, between 6 years and 8 years, between 6 years and 9 years,

between 7 years and 8 years, between 7 years and 9 years, between 7 years and 10 years, between 8 years and 9 years, between 8 years and 10 years, between 8 years and 11 years, between 9 years and 10 years, between 9 years and 11 years, between 9 years and 12 years, between 10 years and 11 years, between 10 years and 12 years, between 10 years and 13 years, between 11 years and 12 years, between 11 years and 13 years, between 11 years and 14 years, between 12 years and 13 years, between 12 years and 14 years, between 12 years and 14 years, between 13 years and 14 years, between 13 years and 14 years, between 13 years and 16 years, between 14 years and 14 years, between 14 years and 16 years, between 14 years and 17 years, between 14 years and 16 years, between 14 years and 17 years, between 14 years and 18 years, between 16 years and 17 years, between 16 years and 18 years, and between 17 years and 18 years old. In one aspect, a subject in need thereof is a geriatric subject. In one aspect, a geriatric subject is between 65 years and 95 or more years old. In one aspect, a geriatric subject is between 65 years and 70 years, between 65 years and 75 years, between 65 years and 80 years, between 70 years and 75 years, between 70 years and 80 years, between 70 years and 85 years, between 75 years and 80 years, between 75 years and 85 years, between 75 years and 90 years, between 80 years and 85 years, between 80 years and 90 years, between 80 years and 95 years, between 85 years and 90 years, and between 85 years and 95 years old. In one aspect, a subject in need thereof is an adult. In one aspect, an adult subject is between 20 years and 95 or more years old. In one aspect, an adult subject is between 20 years and 25 years, between 20 years and 30 years, between 20 years and 35 years, between 25 years and 30 years, between 25 years and 35 years, between 25 years and 40 years, between 30 years and 35 years, between 30 years and 40 years, between 30 years and 45 years, between 35 years and 40 years, between 35 years and 45 years, between 35 years and 50 years, between 40 years and 45 years, between 40 years and 50 years, between 40 years and 55 years, between 45 years and 50 years, between 45 years and 55 years, between 45 years and 60 years, between 50 years and 55 years, between 50 years and 60 years, between 50 years and 65 years, between 55 years and 60 years, between 55 years and 65 years, between 55 years and 70 years, between 60 years and 65 years, between 60 years and 70 years, between 60 years and 75 years, between 65 years and 70 years, between 65 years and 75 years, between 65 years and 80 years, between 70 years and 75 years, between 70 years and 80 years, between 70 years and 85 years, between 75 years and 80 years, between 75 years and 85 years, between 75 years and 90 years, between 80 years and 85 years, between 80 years and 90 years, between 80 years and 95 years, between 85 years and 90 years, and between 85 years and 95 years old. In one aspect, a subject in need thereof is between 1 year and 5 years, between 2 years and 10 years, between 3 years and 18 years, between 21 years and 50 years, between 21 years and 40 years, between 21 years and 30 years, between 50 years and 90 years, between 60 years and 90 years, between 70

years and 90 years, between 60 years and 80 years, or between 65 years and 75 years old. In one aspect, a subject in need thereof is a young old subject (65 to 74 years old). In one aspect, a subject in need thereof is a middle old subject (75 to 84 years old). In one aspect, a subject in need thereof is an old subject (>85 years old).

- [00321]** As used herein, the term “flow rate” refers to the rate of delivery of an AAV vector or composition. In an aspect, the flow rate is between 0.1 μL/minute and 5.0 μL/minute. In one aspect, the flow rate is between 0.1 μL/minute and 0.2 μL/minute, between 0.1 μL/minute and 0.3 μL/minute, between 0.1 μL/minute and 0.4 μL/minute, between 0.2 μL/minute and 0.3 μL/minute, between 0.2 μL/minute and 0.4 μL/minute, between 0.2 μL/minute and 0.5 μL/minute, between 0.3 μL/minute and 0.4 μL/minute, between 0.3 μL/minute and 0.5 μL/minute, between 0.3 μL/minute and 0.6 μL/minute, between 0.4 μL/minute and 0.5 μL/minute, between 0.4 μL/minute and 0.6 μL/minute, between 0.4 μL/minute and 0.7 μL/minute, between 0.5 μL/minute and 0.6 μL/minute, between 0.5 μL/minute and 0.7 μL/minute, between 0.5 μL/minute and 0.8 μL/minute, between 0.6 μL/minute and 0.7 μL/minute, between 0.6 μL/minute and 0.8 μL/minute, between 0.6 μL/minute and 0.9 μL/minute, between 0.7 μL/minute and 0.8 μL/minute, between 0.7 μL/minute and 0.9 μL/minute, between 0.7 μL/minute and 1.0 μL/minute, between 0.8 μL/minute and 0.9 μL/minute, between 0.8 μL/minute and 1.0 μL/minute, between 0.8 μL/minute and 1.1 μL/minute, between 0.9 μL/minute and 1.0 μL/minute, between 0.9 μL/minute and 1.1 μL/minute, between 0.9 μL/minute and 1.2 μL/minute, between 1.0 μL/minute and 1.1 μL/minute, between 1.0 μL/minute and 1.2 μL/minute, between 1.0 μL/minute and 1.3 μL/minute, between 1.1 μL/minute and 1.2 μL/minute, between 1.1 μL/minute and 1.3 μL/minute, between 1.1 μL/minute and 1.4 μL/minute, between 1.2 μL/minute and 1.3 μL/minute, between 1.2 μL/minute and 1.4 μL/minute, between 1.2 μL/minute and 1.5 μL/minute, between 1.3 μL/minute and 1.4 μL/minute, between 1.3 μL/minute and 1.5 μL/minute, between 1.3 μL/minute and 1.6 μL/minute, between 1.4 μL/minute and 1.5 μL/minute, between 1.4 μL/minute and 1.6 μL/minute, between 1.4 μL/minute and 1.7 μL/minute, between 1.5 μL/minute and 1.6 μL/minute, between 1.5 μL/minute and 1.7 μL/minute, between 1.5 μL/minute and 1.8 μL/minute, between 1.6 μL/minute and 1.7 μL/minute, between 1.6 μL/minute and 1.8 μL/minute, between 1.6 μL/minute and 1.9 μL/minute, between 1.7 μL/minute and 1.8 μL/minute, between 1.7 μL/minute and 1.9 μL/minute, between 1.7 μL/minute and 2.0 μL/minute, between 1.8 μL/minute and 1.9 μL/minute, between 1.8 μL/minute and 2.0 μL/minute, between 1.8 μL/minute and 2.1 μL/minute, between 1.9 μL/minute and 2.0 μL/minute, between 1.9 μL/minute and 2.1 μL/minute, between 1.9 μL/minute and 2.2 μL/minute, between 2.0 μL/minute and 2.1 μL/minute, between 2.0 μL/minute and 2.2 μL/minute, between 2.0 μL/minute and 2.3 μL/minute, between 2.1 μL/minute and 2.2 μL/minute, between 2.1 μL/minute and 2.3 μL/minute, between 2.1 μL/minute and 2.4 μL/minute, between 2.2

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μL/minute and 5.0 μL/minute, 4.8 μL/minute and 4.9 μL/minute, between 4.8 μL/minute and 5.0 μL/minute, or between 4.9 μL/minute and 5.0 μL/minute.

[00322] As used herein, the term “therapeutically effective dose” or “pharmaceutically active dose” refers to an amount of AAV particles or composition as provided herein which is effective in treating a neurological condition. In an aspect, an AAV particle or composition as provided herein can be provided together with a pharmaceutically acceptable carrier. As used herein, a “pharmaceutically acceptable carrier” refers to a non-toxic solvent, dispersant, excipient, adjuvant, or other material which is mixed with an AAV particles or composition as provided herein.

[00323] Non-limiting examples of a pharmaceutically acceptable carrier include a liquid (e.g., saline), gel, nanoparticles, exosomes, lipid vesicles, or solid form of diluents, adjuvant, excipients or an acid resistant encapsulated ingredient. Non-limiting examples of suitable diluents and excipients include pharmaceutical grades of physiological saline, dextrose, glycerol, mannitol, lactose, starch, magnesium stearate, sodium saccharin, cellulose, magnesium carbonate, and the like, and combinations thereof. In an aspect, a therapeutic effective dose contains auxiliary substances such as wetting or emulsifying agents, stabilizing or pH buffering agents. In one aspect, a therapeutically effective dose of an AAV particle or composition as provided herein is injected to a subject. In one aspect, a therapeutically effective dose of an AAV particle or composition as provided herein is delivered into a subject. In one aspect, a therapeutically effective dose is administered with at least one pharmaceutically acceptable carrier. In one aspect, a therapeutic effective dose contains between about 1% and about 5%, between about 5% and about 10%, between about 10% and about 14%, between about 14% and about 20%, between about 20% and about 25%, between about 25% and about 30%, between about 30% and about 35%, between about 40 and about 45%, between about 50% and about 55%, between about 1% and about 95%, between about 2% and about 95%, between about 5% and about 95%, between about 10% and about 95%, between about 14% and about 95%, between about 20% and about 95%, between about 25% and about 95%, between about 30% and about 95%, between about 35% and about 95%, between about 40% and about 95%, between about 45% and about 95%, between about 50% and about 95%, between about 55% and about 95%, between about 60% and about 95%, between about 65% and about 95%, between about 70% and about 95%, between about 45% and about 95%, between about 80% and about 95%, or between about 85% and about 95% of AAV particle or composition as provided herein.

[00324] In an aspect, a therapeutically effective dose is delivered to subject in need thereof at least once daily or at least once weekly for at least two consecutive days or weeks. In one aspect, a therapeutically effective dose is delivered to subject in need thereof at least once daily

or at least once weekly for at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 14 consecutive days or weeks. In one aspect, a therapeutically effective dose is delivered to subject in need thereof at least once daily or at least once weekly for at least 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 consecutive weeks. In one aspect, a therapeutically effective dose is delivered to subject in need thereof at least once daily or at least once weekly for at most 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 14, 16, 17, 18, 19, or 20 consecutive days or weeks. In one aspect, a therapeutically effective dose is delivered to subject in need thereof at least once daily or at least once weekly for at most 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 consecutive weeks or months. In one aspect, a therapeutically effective dose is delivered to subject in need thereof is administered at least once for at least 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 consecutive months or years, chronically for a subject's entire life span, or an indefinite period of time. In one aspect, a therapeutically effective dose is delivered to subject in need thereof once a year for 2 consecutive years, 3 consecutive years, or 5 consecutive years. In one aspect, a therapeutically effective dose is delivered to subject in need thereof once a year for 2 consecutive years. In one aspect, a therapeutically effective dose is delivered to subject in need thereof once a year for 3 consecutive years. In one aspect, a therapeutically effective dose is delivered to subject in need thereof once a year for 5 consecutive years.

[00325] As used herein, the term "remission", "cure," or "resolution rate" refers to the percentage of subjects in need thereof that are cured or obtain remission or complete resolution of a neurological condition in response to a therapeutically effective dose.

[00326] As used herein, the term "response rate" refers to the percentage of subjects in need thereof that respond positively (*e.g.*, reduced severity or frequency of one or more symptoms) to a therapeutically effective dose.

[00327] In one aspect, a therapeutically effective dose achieves a remission, cure, response rate, or resolution rate of a neurological condition of at least about 50%. In one aspect, a therapeutically effective dose eliminates, reduces, slows, or delays, one or more neurological condition symptoms. Non-limiting examples of neurological condition symptoms include tremor, slowed movement (bradykinesia), rigid muscles, impaired posture and balance, loss of automatic movements, uncoordinated movement, uncontrolled movement, spontaneous jerking movement, speech changes, numbness, writing changes, involuntary movement such as chorea movement, uncontrolled posture, mood change, and sleep disorder. In an aspect, a neurological condition symptom is a movement symptom. Non-limiting examples of movement symptoms include impairment of an involuntary movement or an impairment of a voluntary movement. In one aspect, a neurological condition symptom is a cognitive symptom. Non-limiting examples of cognitive symptoms include fine motor skills, tremors, seizures, chorea, dystonia, dyskinesia, slow or abnormal eye movements, impaired gait, impaired posture, impaired balance, difficulty

with speech, difficulty with swallowing, difficulty organizing, difficulty prioritizing, difficulty focusing on tasks, lack of flexibility, lack of impulse control, outbursts, lack of awareness of one's own behaviors and/or abilities, slowness in processing thoughts, difficulty in learning new information, difficulty in remember things, difficulty in communications, difficulty in following
 5 orders, difficulty in executing tasks.

[00328] In an aspect, neurological condition symptom is a psychiatric symptom. Non-limiting examples of psychiatric symptoms include depression, irritability, sadness or apathy, social withdrawal, insomnia, fatigue, lack of energy, obsessive-compulsive disorder, mania, bipolar disorder, and weight loss. In one aspect, a neurological condition symptom is at least one
 10 damaged blood vessel. In one aspect, a neurological condition symptom, is a damaged blood brain barrier. In one aspect, a neurological condition symptom is damaged blood flow. Non-limiting examples of tests to evaluate the elimination, reduction, slow, or delay, of neurological condition symptoms include the unified Huntington's disease rating scale (UHDRS) score, UHDRS Total Functional Capacity (TFC), UHDRS Functional Assessment, UHDRS Gait score, UHDRS Total
 15 Motor Score (TMS), Hamilton depression scale (HAM-D), Columbia-suicide severity rating scale (C-SSRS), Montreal cognitive assessment (MoCA), modified Rankin Scale (mRS), National Institutes of Health Stroke Scale (NIHSS), and Barthel Index (BI), Timed Up and Go Test (TUG), Chedoke Arm and Hand Activity Inventory (CAHAI), Symbol Digit Modalities Test, Controlled Oral Word Association tasks, magnetic resonance imaging (MRI), functional magnetic resonance
 20 imaging (fMRI), and positron emission tomography (PET) scanning.

[00329] In an aspect, a therapeutically effective dose achieves remission, cure, response rate, or resolution rate of a neurological condition of between about 10% and about 99% or more. In one aspect, a therapeutically effective dose achieves remission, cure, response rate, or resolution rate of a neurological condition between 10% and 100%, such as between 10% and 14
 25 %, between 10% and 20%, between 10% and 25%, between 14% and 20%, between 14% and 25 %, between 14% and 30%, between 20% and 25%, between 20% and 30%, between 20% and 35%, between 25% and 30%, between 25% and 35%, between 25% and 40%, between 30% and 35%, between 30% and 40%, between 35% and 45%, between 35% and 50%, between 40% and 45%, between 40% and 50%, between 40% and 55%, between 45% and 50%, between 45% and
 30 55%, between 45% and 60%, between 50% and 55%, between 50% and 60%, between 50% and 65%, between 55% and 60%, between 55% and 65%, between 55% and 70%, between 60% and 65%, between 60% and 70%, between 60% and 75%, between 65% and 70%, between 65% and 75%, between 65% and 80%, between 70% and 75%, between 70% and 80%, between 70% and 85%, between 75% and 80%, between 75% and 85%, between 75% and 90%, between 80% and
 35 85%, between 80% and 90%, between 80% and 95%, between 85% and 90%, between 85% and

95%, between 85% and 100%, between 90% and 95%, between 90% and 100%, or between 95% and 100%.

[00330] In an aspect, a therapeutically effective dose eliminates, reduces, slows, or delays, one or more neurological condition symptoms between 10% and 100%, such as between 10% to about 14%, between 10% and 20%, between 10% and 25%, between 14% and 20%, between 14% and 25%, between 14% and 30%, between 20% and 25%, between 20% and 30%, between 20% and 35%, between 25% and 30%, between 25% and 35%, between 25% and 40%, between 30% and 35%, between 30% and 40%, between 35% and 45%, between 35% and 50%, between 40% and 45%, between 40% and 50%, between 40% and 55%, between 45% and 50%, between 45% and 55%, between 45% and 60%, between 50% and 55%, between 50% and 60%, between 50% and 65%, between 55% and 60%, between 55% and 65%, between 55% and 70%, between 60% and 65%, between 60% and 70%, between 60% and 75%, between 65% and 70%, between 65% and 75%, between 65% and 80%, between 70% and 75%, between 70% and 80%, between 70% and 85%, between 75% and 80%, between 75% and 85%, between 75% and 90%, between 80% and 85%, between 80% and 90%, between 80% and 95%, between 85% and 90%, between 85% and 95%, between 85% and 100%, between 90% and 95%, between 90% and 100%, or between 95% and 100%.

[00331] In an aspect, a neurological condition symptom is assessed on the day of treatment, 1 day post treatment, 3 months post treatment, 6 months post treatment, 1 year post treatment and every year thereafter post treatment.

[00332] In an aspect, a neurological condition symptom is assessed between 1 day post treatment and 7 days post treatment. In one aspect, symptoms can be assessed between 1 day post treatment and 2 days post treatment, between 1 day post treatment and 3 days post treatment, between 1 day post treatment and 4 days post treatment, between 2 days post treatment and 3 days post treatment, between 2 days post treatment and 4 days post treatment, between 2 days post treatment and 5 days post treatment, between 3 days post treatment and 4 days post treatment, between 3 days post treatment and 5 days post treatment, between 3 days post treatment and 6 days post treatment, between 4 days post treatment and 5 days post treatment, between 4 days post treatment and 6 days post treatment, between 4 days post treatment and 7 days post treatment, between 5 days post treatment and 6 days post treatment, between 5 days post treatment and 7 days post treatment, or between 6 days post treatment and 7 days post treatment. In one aspect, symptoms can be assessed between 1 week post treatment and 4 weeks post treatment. In one aspect, symptoms can be assessed between 1 week post treatment and 2 weeks post treatment, between 1 week post treatment and 3 weeks post treatment, between 1 week post treatment and 4 weeks post treatment, between 2 weeks post treatment and 3 weeks post treatment, between 2 weeks post

treatment and 4 weeks post treatment, or between 3 weeks post treatment and 4 weeks post treatment. In one aspect, symptoms can be assessed between 1 month post treatment and 12 months post treatment. In one aspect, symptoms can be assessed between 1 month post treatment and 2 months post treatment, between 1 month post treatment and 3 months post treatment, between 1 month post treatment and 4 months post treatment, between 2 months post treatment and 3 months post treatment, between 2 months post treatment and 4 months post treatment, between 2 months post treatment and 5 months post treatment, between 3 months post treatment and 4 months post treatment, between 3 months post treatment and 5 months post treatment, between 3 months post treatment and 6 months post treatment, between 4 months post treatment and 5 months post treatment, between 4 months post treatment and 6 months post treatment, between 4 months post treatment and 7 months post treatment, between 5 months post treatment and 6 months post treatment, between 5 months post treatment and 7 months post treatment, between 5 months post treatment and 8 months post treatment, between 6 months post treatment and 7 months post treatment, between 6 months post treatment and 8 months post treatment, between 6 months post treatment and 9 months post treatment, between 7 months post treatment and 8 months post treatment, between 7 months post treatment and 9 months post treatment, between 7 months post treatment and 10 months post treatment, between 8 months post treatment and 9 months post treatment, between 8 months post treatment and 10 months post treatment, between 8 months post treatment and 11 months post treatment, between 9 months post treatment and 10 months post treatment, between 9 months post treatment and 11 months post treatment, between 9 months post treatment and 12 months post treatment, between 10 months post treatment and 11 months post treatment, between 10 months post treatment and 12 months post treatment, or between 11 months post treatment and 12 months post treatment. In one aspect, symptoms can be assessed between 1 year post treatment and about 20 years post treatment. In one aspect symptoms can be assessed between 1 year post treatment and 5 years post treatment, between 1 year post treatment and 10 years post treatment, between 1 year post treatment and 14 years post treatment, between 5 years post treatment and 10 years post treatment, between 5 years post treatment and 14 years post treatment, between 5 years post treatment and 20 years post treatment, between 10 years post treatment and 14 years post treatment, between 10 years post treatment and 20 years post treatment, or between 14 years post treatment and 20 years post treatment.

[00333] As used herein, the term “survival rate” refers to a cohort of subjects in a treatment group still alive after a given period of time after diagnosis of a neurological condition.

[00334] In an aspect, a therapeutically effective dose achieves increase survival rate of between about 10% and 99% or more. In one aspect, a therapeutically effective dose achieves an increase in survival rate of between 10% and 100%, such as between 10% and 14%, between 10%

and 20%, between 10% and 25%, between 14% and 20%, between 14% and 25%, between 14% and 30%, between 20% and 25%, between 20% and 30%, between 20% and 35%, between 25% and 30%, between 25% and 35%, between 25% and 40%, between 30% and 35%, between 30% and 40%, between 35% and 45%, between 35% and 50%, between 40% and 45%, between 40% and 50%, between 40% and 55%, between 45% and 50%, between 45% and 55%, between 45% and 60%, between 50% and 55%, between 50% and 60%, between 50% and 65%, between 55% and 60%, between 55% and 65%, between 55% and 70%, between 60% and 65%, between 60% and 70%, between 60% and 75%, between 65% and 70%, between 65% and 75%, between 65% and 80%, between 70% and 75%, between 70% and 80%, between 70% and 85%, between 75% and 80%, between 75% and 85%, between 75% and 90%, between 80% and 85%, between 80% and 90%, between 80% and 95%, between 85% and 90%, between 85% and 95%, between 85% and 100%, between 90% and 95%, between 90% and 100%, or between 95% and 100%.

[00335] As used herein, the term “life expectancy” refers to a period of time a subject is expected to live.

[00336] In an aspect, a therapeutically effective dose increases life expectancy of between about 10% and 99% or more. In one aspect, a therapeutically effective dose increases life expectancy of between 10% and 100%, such as between 10% and 14%, between 10% and 20%, between 10% and 25%, between 14% and 20%, between 14% and 25%, between 14% and 30%, between 20% and 25%, between 20% and 30%, between 20% and 35%, between 25% and 30%, between 25% and 35%, between 25% and 40%, between 30% and 35%, between 30% and 40%, between 35% and 45%, between 35% and 50%, between 40% and 45%, between 40% and 50%, between 40% and 55%, between 45% and 50%, between 45% and 55%, between 45% and 60%, between 50% and 55%, between 50% and 60%, between 50% and 65%, between 55% and 60%, between 55% and 65%, between 55% and 70%, between 60% and 65%, between 60% and 70%, between 60% and 75%, between 65% and 70%, between 65% and 75%, between 65% and 80%, between 70% and 75%, between 70% and 80%, between 70% and 85%, between 75% and 80%, between 75% and 85%, between 75% and 90%, between 80% and 85%, between 80% and 90%, between 80% and 95%, between 85% and 90%, between 85% and 95%, between 85% and 100%, between 90% and 95%, between 90% and 100%, or between 95% and 100%.

[00337] In an aspect, a therapeutically effective dose reduces the amount of atrophy within the brain of a subject in need thereof between about 10% and 99% or more. In one aspect, a therapeutically effective dose reduces the amount of atrophy within the brain of a subject in need thereof between 10% and 100%, such as between 10% and 14%, between 10% and 20%, between 10% and 25%, between 14% and 20%, between 14% and 25%, between 14% and 30%, between 20% and 25%, between 20% and 30%, between 20% and 35%, between 25% and 30%, between

25% and 35%, between 25% and 40%, between 30% and 35%, between 30% and 40%, between 35% and 45%, between 35% and 50%, between 40% and 45%, between 40% and 50%, between 40% and 55%, between 45% and 50%, between 45% and 55%, between 45% and 60%, between 50% and 55%, between 50% and 60%, between 50% and 65%, between 55% and 60%, between 55% and 65%, between 55% and 70%, between 60% and 65%, between 60% and 70%, between 60% and 75%, between 65% and 70%, between 65% and 75%, between 65% and 80%, between 70% and 75%, between 70% and 80%, between 70% and 85%, between 75% and 80%, between 75% and 85%, between 75% and 90%, between 80% and 85%, between 80% and 90%, between 80% and 95%, between 85% and 90%, between 85% and 95%, between 85% and 100%, between 90% and 95%, between 90% and 100%, or between 95% and 100%.

[00338] In an aspect, the amount of atrophy within the brain of a subject in need thereof is assessed on the day of treatment, 1 day post treatment, 3 months post treatment, 6 months post treatment, 1 year post treatment and every year thereafter post treatment.

[00339] In an aspect, the amount of atrophy within the brain of a subject in need thereof is assessed between 1 day post treatment and 7 days post treatment. In one aspect, symptoms can be assessed between 1 day post treatment and 2 days post treatment, between 1 day post treatment and 3 days post treatment, between 1 day post treatment and 4 days post treatment, between 2 days post treatment and 3 days post treatment, between 2 days post treatment and 4 days post treatment, between 2 days post treatment and 5 days post treatment, between 3 days post treatment and 4 days post treatment, between 3 days post treatment and 5 days post treatment, 3 days post treatment and 6 days post treatment, between 4 days post treatment and 5 days post treatment, between 4 days post treatment and 6 days post treatment, between 4 days post treatment and 7 days post treatment, between 5 days post treatment and 6 days post treatment, between 5 days post treatment and 7 days post treatment, or between 6 days post treatment and 7 days post treatment. In one aspect, symptoms can be assessed between 1 week post treatment and 4 weeks post treatment. In one aspect, symptoms can be assessed between 1 week post treatment and 2 weeks post treatment, between 1 week post treatment and 3 weeks post treatment, between 1 week post treatment and 4 weeks post treatment, between 2 weeks post treatment and 3 weeks post treatment, between 2 weeks post treatment and 4 weeks post treatment, or between 3 weeks post treatment and 4 weeks post treatment. In one aspect, symptoms can be assessed between 1 month post treatment and 12 months post treatment. In one aspect, symptoms can be assessed between 1 month post treatment and 2 months post treatment, between 1 month post treatment and 3 months post treatment, between 1 month post treatment and 4 months post treatment, between 2 months post treatment and 3 months post treatment, between 2 months post treatment and 4 months post treatment, between 2 months post treatment and 5 months post treatment, between 3 months post

treatment and 4 months post treatment, between 3 months post treatment and 5 months post treatment, between 3 months post treatment and 6 months post treatment, between 4 months post treatment and 5 months post treatment, between 4 months post treatment and 6 months post treatment, between 4 months post treatment and 7 months post treatment, between 5 months post treatment and 6 months post treatment, between 5 months post treatment and 7 months post treatment, between 5 months post treatment and 8 months post treatment, between 6 months post treatment and 7 months post treatment, between 6 months post treatment and 8 months post treatment, between 6 months post treatment and 9 months post treatment, between 7 months post treatment and 8 months post treatment, between 7 months post treatment and 9 months post treatment, between 7 months post treatment and 10 months post treatment, between 8 months post treatment and 9 months post treatment, between 8 months post treatment and 10 months post treatment, between 8 months post treatment and 11 months post treatment, between 9 months post treatment and 10 months post treatment, between 9 months post treatment and 11 months post treatment, between 9 months post treatment and 12 months post treatment, between 10 months post treatment and 11 months post treatment, between 10 months post treatment and 12 months post treatment, or between 11 months post treatment and 12 months post treatment. In one aspect, symptoms can be assessed between 1 year post treatment and about 20 years post treatment. In one aspect symptoms can be assessed between 1 year post treatment and 5 years post treatment, between 1 year post treatment and 10 years post treatment, between 1 year post treatment and 14 years post treatment, between 5 years post treatment and 10 years post treatment, between 5 years post treatment and 14 years post treatment, between 5 years post treatment and 20 years post treatment, between 10 years post treatment and 14 years post treatment, between 10 years post treatment and 20 years post treatment, or between 14 years post treatment and 20 years post treatment.

25 **[00340]** Non-limiting examples of tests to evaluate the amount of atrophy within the brain of a subject in need thereof include Nissle staining, MRI, functional magnetic resonance fMRI, and PET scanning

30 **[00341]** While the present disclosure has been described with reference to preferred embodiments, it will be understood by those skilled in the art that various changes may be made and equivalents may be substituted for elements thereof to adapt to particular situations without departing from the scope of the present disclosure. Therefore, it is intended that the present disclosure not be limited to the particular embodiments disclosed as the best mode contemplated for carrying out the present disclosure, but that the present disclosure will include all embodiments falling within the scope and spirit of the appended claims.

[00342] The examples set out herein illustrate several embodiments of the present disclosure but should not be construed as limiting the scope of the present disclosure in any manner.

EXAMPLES**Example 1. AAV vector constructs**

[00343] Forty eight AAV vector constructs:

- EF-1 α :GfaABC1D:NeuroD1:P2A:Dlx2:WPRE:SV40 (Figure 1B);
- 5 EF-1 α :Gfa1.6:NeuroD1:P2A:Dlx2:WPRE:SV40;
- EF-1 α :GFA2.2:NeuroD1:P2A:Dlx2:WPRE:SV40;
- EF-1 α :GfaABC1D:NeuroD1:P2A:Dlx2:WPRE:hGH;
- EF-1 α :Gfa1.6:NeuroD1:P2A:Dlx2:WPRE:hGH;
- EF-1 α :GFA2.2:NeuroD1:P2A:Dlx2:WPRE:hGH;
- 10 CE:GfaABC1D:NeuroD1:P2A:Dlx2:WPRE:SV40 (P31) (Figure 1A);
- CE:Gfa1.6:NeuroD1:P2A:Dlx2:WPRE:SV40;
- CE:GFA2.2:NeuroD1:P2A:Dlx2:WPRE:SV40;
- CE:GfaABC1D:NeuroD1:P2A:Dlx2:WPRE:hGH;
- CE:Gfa1.6:NeuroD1:P2A:Dlx2:WPRE:hGH;
- 15 CE:GFA2.2:NeuroD1:P2A:Dlx2:WPRE:hGH;
- EF-1 α :GfaABC1D:NeuroD1:T2A:Dlx2:WPRE:SV40 (Figure 3B);
- EF-1 α :Gfa1.6:NeuroD1:T2A:Dlx2:WPRE:SV40;
- EF-1 α :GFA2.2:NeuroD1:T2A:Dlx2:WPRE:SV40;
- EF-1 α :GfaABC1D:NeuroD1:T2A:Dlx2:WPRE:hGH (Figure 3D);
- 20 EF-1 α :Gfa1.6:NeuroD1:T2A:Dlx2:WPRE:hGH;
- EF-1 α :GFA2.2:NeuroD1:T2A:Dlx2:WPRE:hGH;
- CE:GfaABC1D:NeuroD1:T2A:Dlx2:WPRE:SV40 (Figure 3A);
- CE:Gfa1.6:NeuroD1:T2A:Dlx2:WPRE:SV40;
- CE:GFA2.2:NeuroD1:T2A:Dlx2:WPRE:SV40;
- 25 CE:GfaABC1D:NeuroD1:T2A:Dlx2:WPRE:hGH (Figure 3C);
- CE:Gfa1.6:NeuroD1:T2A:Dlx2:WPRE:hGH;
- CE:GFA2.2:NeuroD1:T2A:Dlx2:WPRE:hGH;
- EF-1 α :GfaABC1D:NeuroD1:GSG-P2A:Dlx2:WPRE:SV40 (Figure 2B);
- EF-1 α :Gfa1.6:NeuroD1:GSG-P2A:Dlx2:WPRE:SV40;
- 30 EF-1 α :GFA2.2:NeuroD1:GSG-P2A:Dlx2:WPRE:SV40;
- EF-1 α :GfaABC1D:NeuroD1:GSG-P2A:Dlx2:WPRE:hGH (Figure 2D);
- EF-1 α :Gfa1.6:NeuroD1:GSG-P2A:Dlx2:WPRE:hGH;

- EF-1 α :GFA2.2:NeuroD1:GSG-P2A:Dlx2:WPRES:hGH; CE:GfaABC1D:NeuroD1:GSG-P2A:Dlx2:WPRES:SV40 (Figure 2A); CE:Gfa1.6:NeuroD1:GSG-P2A:Dlx2:WPRES:SV40;
- CE:GFA2.2:NeuroD1:GSG-P2A:Dlx2:WPRES:SV40; CE:GfaABC1D:NeuroD1:GSG-P2A:Dlx2:WPRES:hGH (Figure 2C);
- CE:Gfa1.6:NeuroD1:GSG-P2A:Dlx2:WPRES:hGH;
- CE:GFA2.2:NeuroD1:GSG-P2A:Dlx2:WPRES:hGH;
- EF-1 α :GfaABC1D:NeuroD1:GSG-T2A:Dlx2:WPRES:SV40 (Figure 4B);
- EF-1 α :Gfa1.6:NeuroD1:GSG-T2A:Dlx2:WPRES:SV40;
- EF-1 α :GFA2.2:NeuroD1:GSG-T2A:Dlx2:WPRES:SV40;
- EF-1 α :GfaABC1D:NeuroD1:GSG-T2A:Dlx2:WPRES:hGH (Figure 4D);
- EF-1 α :Gfa1.6:NeuroD1:GSG-T2A:Dlx2:WPRES:hGH;
- EF-1 α :GFA2.2:NeuroD1:GSG-T2A:Dlx2:WPRES:hGH; CE:GfaABC1D:NeuroD1:GSG-T2A:Dlx2:WPRES:SV40 (Figure 4A); CE:Gfa1.6:NeuroD1:GSG-T2A:Dlx2:WPRES:SV40;
- CE:GFA2.2:NeuroD1:GSG-T2A:Dlx2:WPRES:SV40; CE:GfaABC1D:NeuroD1:GSG-T2A:Dlx2:WPRES:hGH (Figure 4C);
- CE:Gfa1.6:NeuroD1:GSG-T2A:Dlx2:WPRES:hGH; and CE:GFA2.2:NeuroD1:GSG-T2A:Dlx2:WPRES:hGH, are constructed.
- [00344]** All 48 vectors constructs utilize pHSG-299 (Takara, Mountain View, CA), a pUC based vector constructs which contains an origin of replication, a Kanamycin resistance gene and a multiple cloning site (MSC) with lacZ gene as backbone.
- [00345]** The 5' end of the expression cassette is an enhancer from a human elongation factor-1 alpha promoter (EF-1 alpha enhancer; SEQ ID NO: 2) or the cytomegalovirus enhancer (CMV enhancer; SEQ ID NO: 11) placed 5' to either a 758-nucleotide GFAP promoter (GfaABC1D; SEQ ID NO: 26), 1667-nucleotide GFAP promoter (Gfa1.6; SEQ ID NO: 4), or a 2214-nucleotide GFAP promoter (GFA2.2 SEQ ID NO: 12).
- [00346]** Following (*e.g.*, 3' to) the enhancer/GFAP promoter, several additional sequences are introduced into the expression cassette in 5' to 3' direction, including: a chimeric intron (SEQ ID NO: 5); a human NeuroD1 coding sequence (hNeuroD1; SEQ ID NO: 6); a human Dlx2 coding sequence (hDlx2; SEQ ID NO: 13); a linker sequence (P2A; SEQ ID NO: 15), (GSG-P2A; SEQ ID NO: 18), (T2A; SEQ ID NO: 16), or (GSG-T2A; SEQ ID NO: 19); and a woodchuck hepatitis virus posttranscriptional regulatory element (WPRES; SEQ ID NO: 7). These sequences are all operably linked to an SV40 poly(A) signal (SEQ ID NO: 8) or hGH poly (A) signal (SEQ ID NO: 17) or bGH poly (A) signal (SEQ ID NO: 30). The enhancer,

GFAP promoter, chimeric intron, hNeuroD1 coding sequence, hDlx2 coding sequence, linker, WPRE, and SV40 poly(A) signal are flanked by two AAV ITR sequences.

Example 2. AAV virus production

- 5 [00347] Each of the 24 plasmids is co-transfected into 293AAV cells using polyethylenimine along with Rep-Cap plasmid (a plasmid comprising a promoter driving the expression of AAV *rep* and *cap* genes) and Helper plasmid (a plasmid comprising a promoter driving the expression of E2A, E4, and VA RNA (of Adenovirus) to produce recombinant AAV virus particles.
- 10 [00348] Transfected cells are scraped and centrifuged at 72 hours after transfection. Cell pellets are frozen and thawed being placed in a dry ice/ethanol mixture followed by being placed in a 37°C water bath. The freeze/thaw cycle is repeated three additional times. An AAV lysate is purified (*e.g.*, cellular debris is removed) by ultra-centrifugation at 350,000 g for 1 hour in discontinuous iodixanol gradients. The virus-containing layer is collected and then concentrated
- 15 by using Millipore Amicon Ultra Centrifugal Filters. Virus titers are then determined by qPCR using primers amplifying ITR region or gene/expression cassette specific sequences.

Example 3. Astrocytes cell cultures

- [00349] Human cortical astrocytes (HA1800; ScienCell Research Laboratories, Inc., Carlsbad, California) are subcultured when they are over 90% confluent. For subculture, cells are
- 20 trypsinized using TrypLE™ Select (Invitrogen, Carlsbad, California), centrifuged for 5 minutes at $200 \times g$, then resuspended and plated on a medium comprising DMEM/F12 (Gibco); 10% fetal bovine serum (Gibco); penicillin/streptomycin (Gibco); 3.5 mM glucose (Sigma-Aldrich); B27 (Gibco); 10 ng/mL epidermal growth factor (Invitrogen); and 10 ng/mL fibroblast growth factor 2 (Invitrogen). The astrocytes are cultured on poly-D-lysine (Sigma-Aldrich) coated coverslips
- 25 (12 mm) at a density of approximately 20,000 cells per coverslip in 24-well plates (BD Biosciences).

- [00350] Rat primary astrocytes (isolated from Sprague Dawley Rat cortex or striatum) are cultured in media comprising DMEM/F12 (Gibco); 10% fetal bovine serum (Gibco), penicillin/streptomycin (Gibco); 3.5 mM glucose (Gibco).
- 30 [00351] All cells are maintained at 37°C in humidified air with 5% carbon dioxide.

Example 4. Testing AAV vector in astrocyte cell cultures (*in vitro*)

- [00352] Recombinant AAV obtained from the method of Example 2 are used to infect human cortical astrocytes and rat primary astrocytes of Example 3 at a concentration range of 10^{10} particles/mL and 10^{14} particles/mL. Twenty-four hours after infection of the cells, the culture

medium is replaced by differentiation medium comprising DMEM/F12 (Gibco); N2 supplement (Gibco); and 20 ng/mL brain-derived neurotrophic factor (Invitrogen). The differentiation medium is added to the cell cultures every four days. *See Song et al., Nature*, 417:39-44 (2002).

[00353] Empty space in the cell cultures is filled with additional human astrocytes to support the functional development of converted neurons as astrocytes or rat primary astrocytes convert to neurons.

Example 5. Testing of AAV vector potency

[00354] Recombinant AAV obtained from the method of Example 2 are used to infect human cortical astrocytes and rat primary astrocytes from Example 3 (or astrocytes from other brain regions or the spinal cord) at passage number 4 to 7 at a concentration range of 10^{10} particles/mL and 10^{14} particles/mL. qPCR, enzyme-linked immunosorbent (ELISA), and western blot are performed to determine expression of NeuroD1 transcript and protein levels.

[00355] Expression of NeuN, doublecortin (DCX), β 3-tubulin, NF-200, and MAP2, are assessed by qPCR, ELISA, western blot, and immunostaining to determine functional output of recombinant AAV.

Example 6. Testing of AAV vector titration and infection rate

[00356] A purified AAV vector is treated with DNaseI to eliminate remnant plasmid contamination. A series of AAV vector dilutions are performed at 100 times, 500 times, 2500 times, and 12500 times. The AAV plasmid is diluted to generate a standard curve by serial dilutions. The plasmid is diluted to 10^4 , 10^5 , 10^6 , 10^7 , and 10^8 molecules/ul. qPCR is performed on the diluted AAV vectors and the diluted AAV plasmid. The primers used are against the ITR region (Forward ITR primer, 5'-GGAACCCCTAGTGATGGAGTT, reverse ITR primer, 5'-CGGCCTCAGTGAGCGA). The qPCR mix comprises 10 uL Universal SYBR Master Mix 2X, 2 uL of 5 uM forward ITR primer, 2 uL of 5 uM reverse ITR primer, 5 uL of tested sample or diluted standard and 1 uL H₂O. The qPCR program is 95 °C for 10 minutes followed by 40 cycles of 95 °C for 15 seconds, 60 °C for 30 seconds followed by a melt curve. The data is analyzed using the qPCR cyclers software. The physical titer of the AAV sample (viral genomes (vg)/ml) is calculated based on the standard curve.

[00357] The AAV vector infection rate is tested by using the 50% tissue culture infection dose (TCID₅₀) assay performed using a standard protocol from the American Type Culture Collection (ATCC; Manassas, VA).

Example 7. Testing of AAV dose range (in vivo)

[00358] Recombinant AAV obtained from the method of Example 2 is injected into C57/BL6 mice by bilateral intracranial injection into the motor cortex. Each AAV is injected at a dosage of 1×10^{11} , 3×10^{11} , 1×10^{12} , 3×10^{12} , 1×10^{12} , 3×10^{12} , 1×10^{13} viral genomes/mL at 1 uL of volume. Each dosage is assessed at 4 days, 20 days, and 60 days post injection to determine the optimal effective dose (OED), maximum tolerable dose (MTD), and minimum effective dose (MED) at a cell and tissue level. There are three mice per time point. The OED, MTD, and MED are determined by assessment of astrocyte-to-neuron conversion efficiency and potential toxicity via immunostaining of NeuroD1, GFAP, NeuN, and Iba1. If the first dose range is not sufficient to determine the OED, MTD, and MED, a second dosage range is performed at 1×10^{10} viral genomes/mL to 1×10^{14} GC/mL, at 1 uL of volume.

Example 8. Comparison of neuron conversion rate of recombinant AAVs obtained from various AAV vector constructs in human cell cultures (*in vitro*)

AAV vector constructs are design as described in Example 1 to express either NeuroD1 alone or Dlx2 alone. Recombinant AAV is obtained as described in Example 2 for (1) AAV vector constructs expressing NeuroD1 alone; (2) AAV vector constructs expressing Dlx2 alone; (3) a combination of AAV vector constructs (1) and (2); and (4) AAV vector constructs expressing NeuroD1 and a linker with Dlx2. Resulting recombinant AAVs are used to infect human cortical astrocytes and rat primary astrocytes of Example 3. Twenty-four hours after infection of the cells, the culture medium is replaced by differentiation medium comprising DMEM/F12 (Gibco); N2 supplement (Gibco); and 20 ng/mL brain-derived neurotrophic factor (Invitrogen). The differentiation medium is added to the cell cultures every four days. See Song *et al.*, *Nature*, 417:39-44 (2002). Empty space in the cell cultures is filled with additional human astrocytes to support the functional development of converted neurons as astrocytes or microglial cells convert to neurons. Neuron conversion levels of each treatment are measured and compared.

Example 9. Testing AAV vector in human subjects (*in vivo*)

[00359] Recombinant AAV obtained from the method of Example 2 are used to infect human brain or spinal cord astrocytes *in vivo*. Recombinant AAV is injected at a concentration range of 10^{10} particles/mL and 10^{14} particles/mL with a volume ranging from 10 μ L to 1 mL into the brain or spinal cord of a human subject with a neurological condition. The human subject's neurological condition symptoms, brain imaging including MRI, PET scan, or combination of MRI and PET, and behavioral metrics are observed before, during, and post injection. Post injection observations are performed once a week until the first month post injection. After the first month post injection, observations are performed once a month for the next 11 months, and may be extended to 2 years following viral injection.

Example 10. Dose Scale Assay in non-human primates

[00360] The volume of brain tissue expressing NeuroD1 from Example 7 divided by the number of vector genomes ($\text{mm}^3/\text{vector genomes}$) is used to determine the viral infection rate of brain tissue. The volume (mm^3) of specific brain region to be treated in non-human primates is calculated and a dose range of vector genomes is scaled according to the infection rate obtained in Example 7. A dose range study is performed as in Example 7 and the OED, MTD, and MED are determined by assessment of astrocyte-to-neuron conversion efficiency and potential toxicity via immunostaining of NeuroD1, GFAP, NeuN, and Iba1.

Example 11. Treatment of a subject in need thereof with Huntington's Disease (*in vivo*)

10 [00361] A subject with Huntington's Disease is treated with recombinant AAV obtained from the method of Example 2. The subject's neurological symptoms include involuntary movement such as chorea movement, uncontrolled posture, mood change, sleep disorder, speech changes, difficulty with swallowing, and impairment of cognitive functions such as deficits in learning and memory. Recombinant AAV is injected at a concentration range of 10^{10} particles/mL and 10^{14} particles/mL with a volume ranging from 10 μL to 1000 μL into the striatum (putamen and caudate nucleus) of a human subject with a neurological condition. The human subject's neurological condition symptoms, brain imaging including MRI, PET scan, or combination of MRI and PET, and behavioral metric's are observed before, during, and post injection. Post injection observations are performed once a week until the first month post injection. After the first month post injection, observations are performed once a month for the next 11 months, and may be extended to 2 years following viral injection.

Example 12. A combination approach to directly converting glial cells to neurons coupled with shRNA for knockdown of the Htt gene expression

25 [00362] A target sequence is identified that is complementary to the Htt gene. An shRNA is designed to target the Htt gene. NeuroD1, Dlx2, and the target shRNA are packaged in to an AAV vector (hU6::Htt shRNA-hGFAP::hNeuroD1-P2A-hDlx2)(Figure 5A - 8D) and recombinant AAV is produced as described in Example 2. Recombinant AAV is injected into the striatum of mice with mutant Htt gene. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2, and (iii) receiving recombinant AAV (hU6::Htt shRNA-hGFAP::hNeuroD1-P2A-hDlx2)(Figure 5A - 8D).

[00363] Alternatively, the target shRNA is packaged in to an AAV vector (hU6::hHtt shRNA) and another recombinant AAV is produced as described in Example 2. The two recombinant AAVs are injected into the striatum of mice with mutant Htt. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2 alone, and (iii) receiving recombinant AAV (hU6::hHtt shRNA) in combination with recombinant AAV from Example 2.

Example 13. A combination approach to directly converting glial cells to neurons coupled with CRISPR/CAS gene editing of the Htt gene

[00364] A target sequence is identified that is complementary to the Htt gene. A guide RNA (gRNA) sequence is designed to target the Htt gene. A donor sequence is designed to modify the number of CAG repeats of the Htt gene to less than 36. The Cas9 nuclease, an Htt specific gRNA, and donor sequence are packaged into AAV vectors (AAV-Cas9-HTT). Recombinant AAV is produced as described in Example 2.

[00365] Recombinant AAV (AAV-Cas9-HTT) is injected into the striatum of mice with mutant Htt simultaneously with recombinant AAV from Example 2. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2, and (iii) receiving recombinant AAV-Cas9-HTT with recombinant AAV from Example 2 to identify synergistic effects between mutant Htt gene editing and glia-to-neuron conversion. Recombinant AAV-Cas9-HTT and recombinant AAV from Example 2 can be injected simultaneously or at different times.

[00366] Alternatively, NeuroD1, a linker (P2A), Dlx2, a second linker (P2A), Cas9 nuclease, the Htt specific gRNA, and the donor sequence are packaged into AAV vectors (AAV-hNeuroD1-P2A-hDlx2-P2A-Cas9-HTT). Recombinant AAV is produced as described in Example 2. Recombinant AAV (AAV-hNeuroD1-P2A-hDlx2-P2A-Cas9-HTT) is injected into the striatum of mice with mutant Htt simultaneously with recombinant AAV from Example 2. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2, and (iii) receiving recombinant AAV-hNeuroD1-P2A-hDlx2-P2A-Cas9-HTT with recombinant AAV

from Example 2 to identify synergistic effects between mutant Htt gene editing and glia-to-neuron conversion.

Example 14. A combination approach to directly converting glial cells to neurons coupled with antisense oligonucleotide (ASO) to knock down the Htt gene expression

- 5 [00367] A target sequence is identified that is complementary to the Htt gene. An ASO is designed and synthesized to knock down the Htt gene expression. Recombinant AAV from Example 2 is injected together with Htt ASO into the striatum of mice with mutant Htt. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of
- 10 MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2, and (iii) receiving recombinant AAV from Example 2 together with Htt ASO.

Example 15. A combination approach to directly converting glial cells to neurons coupled with siRNA to knock down the Htt gene expression

- 15 [00368] A target sequence is identified that is complementary to the Htt gene. An siRNA is designed and synthesized to knock down the Htt gene expression. Recombinant AAV from example 2 is injected together with Htt siRNA into the striatum of mice with mutant Htt. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of
- 20 MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2, and (iii) receiving recombinant AAV from Example 2 together with Htt siRNA.

Example 16. A combination approach to directly converting glial cells to neurons coupled with miRNA to knock down the Htt gene expression

- 25 [00369] A miRNA is identified that is regulating the Htt gene expression. NeuroD1, Dlx2, and the miRNA are packaged into an AAV vector (CAG::Htt miRNA-hGFAP::hNeuroD1-P2A-hDlx2) and recombinant AAV is produced as described in Example 2. Recombinant AAV is injected into the striatum of mice with mutant Htt. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength
- 30 and brain imaging including MRI, PET scan, or combination of MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2, and (iii) receiving recombinant AAV (CAG::Htt miRNA-hGFAP::hNeuroD1-P2A-hDlx2).

[00370] Alternatively, the target miRNA is packaged in to an AAV vector (CAG::hHtt miRNA) and recombinant AAV is produced as described in Example 2. Recombinant AAV is injected into the striatum of mice with mutant Htt. Mice receiving the treatment are tested for behavioral metrics, such as cat walk, open field test, clasping, mouse weight, and grip strength and brain imaging including MRI, PET scan, or combination of MRI and PET. Behavioral test results and brain imaging are compared among the groups (i) receiving no treatment, (ii) receiving recombinant AAV from Example 2 alone, and (iii) receiving recombinant AAV (CAG::hHtt miRNA) in combination with recombinant AAV from Example 2.

Example 17. AAV virus production of P31

10 [00371] Recombinant AAV is obtained as described in Example 2. The P31 plasmid is co-transfected into AAV293 cells with a Rep-Cap plasmid expressing serotype 5 capsid protein and the Helper plasmid P40Helper (P40H) or pALD-X80 (X80) to produce recombinant AAV virus particles (P31-P40H or P31-X80). Virus titers are determined by qPCR using primers amplifying gene of interest (GOI) primers specific to the P31 plasmid and the ITR region.

15 Reverse packaging primers are used to evaluate nonspecific packaging. Increased viral production is observed with the X80 helper plasmid compared to the P40H helper plasmid (Figure 9).

Example 18. Successful establishment of rat astrocytes primary culture

[00372] Cortical and striatum tissue is isolated from 3 day post-natal Sprague-Dawley rat brains. Tissue is treated with papain to generate single cell suspension and seeded in flasks coated with poly-D-lysine. Cells are immunostained with GFAP antibody and SOX9 antibody. Cells are counter stained with DAPI antibody. More than 95% of cells are astrocytes identified by GFAP and SOX9 staining (Figure 10). Far left panel presents an image of GFAP stained cells. Middle left panel presents an image of SOX9 stained cells. Middle right panel presents an image of DAPI stained cells. Far right panel presents a merge image of GFAP, SOX9, and DAPI stained cells.

Example 19. Comparison of plasmid transfection

[00373] Primary rat astrocytes are seeded and transfected as described in Example 18 with expression vectors P14 (CE:GfaABC1D:hNeuroD1-P2A-Dlx2:WPRE:SV40), P31 (EF-1 α :GfaABC1D:NeuroD1-P2A-Dlx2:WPRE:SV40), and P63 (CE:GfaABC1D:NeuroD1-GSG P2A-Dlx2:WPRE:SV40) to test the expression efficiency of NeuroD1 and Dlx2 in transfected cells. P14 resulted in the NeuroD1 and Dlx2 expression shown by NeuroD1 staining and Dlx2 staining of cells (Figure 11; top panels show NeuroD1 staining of cells,

middle panels show Dlx2 staining of cells, and bottom panels show merged NeuroD1, Dlx2, and DAPI staining of cells).

Example 20. Successful transduction of AAV virus particles into primary rat astrocytes

[00374] Recombinant AAV obtained from the method of Example 2 is transduced into primary rat astrocytes seeded in a 96 well plate with AAV9-P12 (pGfaABC1D:GFP) at various doses of either 3×10^{10} vg /well, 1×10^{10} vg/well, and 2.5×10^9 vg /well. RCAs of passage 5-7 are seeded on glass cover slips coated with poly-D-lysine (PDL) in 96-well plates at 50% confluency 24 hours prior to transduction. Cells are transduced with virus in fresh astrocyte media at the designated titer. Media are refreshed the next day and every 3-4 days. Images acquired six days post transduction of GFP positives cells show that the transduction rate is higher when virus titer is higher (Figure 12).

Example 21. Quantitative analysis of transduction of AAV virus particles into primary rat astrocytes.

[00375] Recombinant AAV obtained from the method of Example 2 is transduced into primary rat astrocytes seeded in 24-well plates or 96-well plates with viral particles AAV9-P12 (pGfaABC1D:GFP) and AAV5-P7 (pEF-1 α :GFP). Cells are harvested seven days post-infection by trypsinization. The cells are fixed, washed, and suspended in PBS. The viral transduction rate is analyzed using flow cytometry to count GFP positive cells compared with all cells (Figure 13A and 13B). Figure 13A shows the % transduction rate at different MOI. Cells seeded in 24-well plates at 1×10^5 cells/well are infected at MOI of either 5×10^5 vg/cell, 2×10^5 vg/cell and 5×10^4 vg/cell. The viral transduction rate decreases as the MOI decreases. Figure 13B shows the transduction rate of AAV viral particles in cells seeded in 96 well plates at a series of densities of 2×10^4 cell/well, 1.5×10^4 cell/well, 1×10^4 cell/well, and 5×10^3 cell/well, and infected with virus at a series of amounts of 2 μ l, 1 μ l, 0.5 μ l, 0.25 μ l, 0.125 μ l of 1×10^{13} vg/ml virus in 100 μ l of medium. This is equivalent to 2×10^{10} vg, 1×10^{10} vg, 5×10^9 vg, 2.5×10^9 vg, and 1.25×10^9 vg each well respectively. The viral transduction rate is unchanged as the number of cells per well decreases.

Example 22. *In vitro* transgene expression and astrocyte-to-neuron conversion induced by NeuroD1 vectors.

Materials and Methods

[00376] Primary Rat Astrocyte Culture: Rat cortical astrocytes (RCA) are isolated from 3-day postnatal Sprague Dawley rat cortical tissue. Cells are maintained in astrocyte media (AM)

composed of DMEM supplemented with 10% FBS, 2.5 mM Glutamine, 3.5 mM Glucose, pen/strep. Cells are sub-cultured at 1:3-1:4 ratio for first two passages at low cell density to promote residual progenitor differentiation. Subsequent sub-cultures are at 1:2 or 1:3 ratio when reaching 90-100% confluent. Cells at passage 5-7 are used for transfection and transduction.

- 5 Immunostaining with a GFAP antibody shows that >90% cells are GFAP positive astrocytes. Culture astrocytes are immunostained with astrocyte markers GFAP and Sox9 at passage 6 (Figure 14).

[00377] Vectors: AAVs are produced with selected vectors and tested *in vitro* using rat astrocytes:

- 10 • NXL-P9 (CE-pGfa681-CI-hND1-p2A-GFP-WPRE-SV40pA)
- NXL-P22 (CE-pGfa681-CI-hND1-WRPE-SV40pA)
- NXL-P35 (EE-pGfa681-CI-hND1-WRPE-SV40pA)
- NXL-P37 (EE-pGfa681-CI-hND1-p2A-GFP-WPRE-SV40pA)
- NXL-P107 (CE-pGfa681-CI-hND1-bGHpA)
- 15 • NXL-P108 (CE-pGfa681-CI-hND1-oPRE-bGHpA)
- NXL-P109 (CE-pGfa681-CRGI-hND1-bGHpA)
- NXL-P130 (CE-pGfa681-GI-hND1-oPRE-bGHpA)
- NXL-P134 (CE-pGfa681-CRGI-hND1-oPRE-bGHpA)
- NXL-P136 (EE-Gfa681-CRGI-hND1-bGHpA)
- 20 • NXL-P138 (EE-Gfa681-CRGI-hND1-oPRE-bGHpA)

[00378] Viral Production: Virus used for *in vitro* studies are produced using adherent AAV293 cells by triple transfections (GOI, helper, and Rep/Cap plasmids) with polyethylenimine (PEI). Virus recovery and purification is achieved via ultra-centrifugation or the use of commercial purification kits.

- 25 [00379] Specifically, AAV293 cells (Cell Biolabs, Cat# AAV-100) are seeded in 15-cm culture dishes 24 hours prior to transfection. Cells at 70-85% confluency are transfected per dish with 10 ug GOI, 10 ug of Rep/Cap, and 14 ug of pALD-X80 (Aldevron) or pHelper (Cell Biolabs) using polyethylenimine (PEI) at a DNA:PEI ratio of 1:4. Multiple dishes are transfected for production based on the scale needed. Culture media is refreshed daily. Seventy-two hours post
- 30 transfection, cells are collected and lysed to harvest the virus using an AAVpro purification kit (Takara, Cat# 6666, 6675, 6235) following the manufacturer's protocol.

[00380] Viral titers are determined by real-time quantitative PCR using a primer pair in the ITR region, primers amplifying a gene of interest (GOI), or vector specific primers. Plasmid DNA is used as a standard. The production yield is $\sim 10^3 - 10^4$ vg/cell level. Figure 35 depicts how each

of the P134, P130, P138 and P21 plasmids co-transfected into AAV293 cells with a Rep-Cap plasmid expressing a serotype 9 capsid protein and the Helper plasmid pALD-X80 (X80) produced recombinant AAV virus particles as measured by qPCR.

[00381] Transfection and Immunofluorescence: Rat cortical astrocytes (RCAs) of passage 5-7 are seeded on glass cover slips coated with poly-D-lysine (PDL) in 24-well plates at 30-50% confluency 24-48 hours prior to transfection. Cells are transfected with 300 ng of vector DNA using Lipofectamine reagent (Thermo Fisher Cat# 15338) following the manufacturer's protocol. At 24-48 hours post transfection, cells are fixed with 4% paraformaldehyde in PBS and subsequently washed and immunostained with anti-NeuroD1 (anti-ND1) antibody (Abcam Cat# ab60704) and followed with secondary antibodies conjugated with fluorescent dyes (Invitrogen, Alexa Fluor). Images are captured under a fluorescent microscope (Zeiss Axiovert A1, Zen Blue). Gene expression levels are assessed by comparing the fluorescence intensity.

[00382] Transduction and Immunofluorescence: RCAs of passage 5-7 are seeded on glass cover slips coated with poly-D-lysine (PDL) in 24-well plates at 30-50% confluency 24-48 hours prior to transduction. Cells are transduced with AAVs at $2-6 \times 10^{10}$ viral genome (vg)/ml in fresh astrocyte media. Media are refreshed the next day and every 3-4 days. Three to six days post transduction, cells are fixed with 4% paraformaldehyde in PBS and subsequently washed and immunostained with anti-ND1 antibody (Abcam Cat# ab60704) and followed with secondary antibodies conjugated with fluorescent dyes (Invitrogen, Alexafluor) for observation and image capturing under a fluorescent microscope (Zeiss Axiovert A1, Zen Blue). Gene expression levels are assessed by comparing the fluorescence intensity.

[00383] Astrocyte-to-neuron conversion assessment RCAs of passage 5-7 are seeded on glass cover slips coated with poly-D-lysine (PDL) in 24-well plates at 30-50% confluency 24-48 hours prior to transduction. Cells are transduced with virus at $2-6 \times 10^{10}$ vg/ml in 500 ul of fresh astrocyte media (DMEM supplemented with 10% FBS, 2.5 mM Glutamine, 3.5 mM Glucose, penn/strep). At 48 hours post transduction, media is replaced with 5% FBS astrocyte media. Subsequently, 100 ul of conversion media (DMEM/F12 + 1% FBS + B27 + N2 and 1 uM Rock inhibitor and 10 ng/ml BDNF) is added daily for 4 days. After the 4 days, the media is completely replaced with conversion media.

[00384] Cells are fixed with 4% paraformaldehyde in PBS at various desired time points (three days, one to five weeks post transduction) and subsequently washed and immunostained with antibodies against ND1 (Abcam Cat# ab60704), NeuN (Millipore, Cat# ABN78), Map2 (Invitrogen, Cat# PA5-17646), followed with secondary antibodies conjugated with fluorescent dyes (Invitrogen, Alexafluor) for observation and imaging under a fluorescent microscope (Zeiss Axiovert A1, Zen Blue).

***In Vitro* Studies Results:**

[00385] All tested NeuroD1 (ND1) plasmids are effective in driving the expression of NeuroD1 (Figures 16-31). The expression level of NeuroD1 is affected by the elements in the vector. Among three versions of the GFA promoter, the 681bp promoter shows the highest
 5 NeuroD1 expression level and the 1.6 kb promoter shows the weakest NeuroD1 expression level. Promoter enhancer elements significantly affect the expression level of NeuroD1. The CMV enhancer increases the expression level of NeuroD1 more than the efl α enhancer. Chimeric introns and WPREs also increase the expression level of NeuroD1.

10 [00386] All tested ND1-containing AAVs are effective in driving the expression of ND1 and inducing an astrocyte-to-neuron conversion in cultured rat astrocytes as shown by positive staining of NeuN and/or MAP2 (Figures 17, 30, 22, 25, and 28). The conversion rate is higher when astrocytes are transduced by the vectors driving a higher ND1 expression. Vectors NXL-P134 and NXL-P138, and the viruses generated using these vectors, i.e., AAV9-P134 and AAV9-P138 respectively, are the most effective in driving expression of ND1 and inducing astrocyte-to-neuron conversion, with AAV-P134 being the most effective (Figures 15-20). Plasmid AAV9-P21 (CE-pGFA681-CI-GFP-WPRE-SV40pA), which does not contain an ND1 sequence, is used as a control, and it does not induce an astrocyte-to-neuron conversion, as shown by the lack of positive staining for NeuN and/or Map2 (Figure 14).

20 [00387] NeuN/RBFOX3 (Neuronal nuclear protein) is a neuron differentiation marker, which stains nuclei and perinuclear cytoplasm in neurons. MAP2 (microtubule associated protein 2) is another neuronal marker which stains cytoplasm microtubules including dendrites in neurons.

[00388] One week post transduction by ND1-containing AAVs, small number of NeuN and MAP2 positive cells (neurons) are observed. By two and three weeks, more NeuN/MAP2 positive
 25 cells are observed. Some NeuN/MAP2 positive cells show typical neuronal morphology.

Example 23. *In vivo* transgene expression and astrocyte-to-neuron conversion induced by NeuroD1 viral vectors.

[00389] AAV9-P134 and AAV9-P138 viruses are used for the *in vivo* studies. AAV9-P12, which drives the expression of GFP alone (no ND1) under a GFAP promoter, is used for the control and to identify cells expressing GFAP (astrocytes).
 30

[00390] Single strand adenovirus-associated viral (ssAAV, AAV for short) vectors NXL-P12, NXL-P134 and NXL-P138 are packaged into AAV, serotype 9 (AAV9), followed by a subsequent iodixanol gradient ultracentrifuge and concentration. Purified AAV viruses are titered
 35 using a quantitative PCR-based method. All AAV used in this study is prepared in 0.001%

Pluronic F-68 (Poloxamer 188 Solution, PFL01-100ML, Caisson Laboratories, Smithfield, UT, USA) in PBS (pH 7.4)

[00391] Normal C57BL/6 mice older than 8 weeks are injected with AAV9-P134, AAV-P138, and AAV9-P12 viruses as follows:

- 5
 - P12 control group: AAV9-P12 5×10^{11} GC/ml, 1 μ L, 1 injection in cortex (unilateral) (n=6)
 - P134 group: AAV9-P12 2.5×10^{11} GC/ml + AAV9-P134 2.5×10^{11} GC/ml, 1 μ L, 1 injection in cortex (unilateral) (n=6)
 - P138 group: AAV9-P12 2.5×10^{11} GC/ml + AAV9-P138 2.5×10^{11} GC/ml, 1 μ L, 1 injection in cortex (unilateral) (n=6)

- 10 **[00392]** Mice are sacrificed and brain cortex tissue analyzed at 10 days post infection (dpi) and at 30 dpi. The animals are anesthetized with 1.25 % Avertin and then sequentially perfused intracardially first with saline solution (0.9 % NaCl) and then with 4 % paraformaldehyde (PFA). The brains are collected and post-fixed in 4 % PFA overnight and are sequentially placed in 20 % and 30 % sucrose at 4 °C until the tissue sank. The dehydrated
 - 15 brains are embedded in Optimal Cutting Temperature (Tissue-Tek® O.C.T. Compound, Sakura® Finetek, Torrance, CA, USA), and then serially sectioned at the coronal plane on the cryostat (Thermo Scientific, Shanghai, China) at 30 μ m thickness. For immunofluorescence, free floating brain sections are first washed with PBS and blocked for 1 hour at room temperature (RT) in 5 % normal donkey serum, 3 % bovine serum albumin and 0.3 % TritonX-100 prepared
 - 20 in PBS, and then incubated overnight at 4°C with primary antibodies diluted in blocking solution. After additional washing with 0.2 % PBST (0.2 % tween-20 in PBS), the samples are incubated with 0.5 μ g/ μ L 4',6-diamidino-2-phenylindole (DAPI; F. Hoffmann-La Roche, Natley, NJ, USA) and appropriate donkey anti-mouse/rabbit secondary antibodies conjugated to Alexa Fluor 555, goat anti-chicken secondary antibodies conjugated to Alexa Fluor 488 (1:1000,
 - 25 Life technologies, Carlsbad, CA, USA), and goat anti-rat (Life technologies)/guinea pig (Jackson immune research) secondary antibodies conjugated to Alexa Fluor 647(1:500) for 2 hours at room temperature, followed by extensive washing with PBS. Samples are finally mounted with VECTASHIELD® mounting medium (VECTOR Laboratories, Burlingame, CA, USA) and sealed with nail polish. Representative Images are taken with a confocal microscope
 - 30 (LSM880, Zeiss, Jena, Germany). Primary antibodies used are as follow: rat anti-GFAP (a marker for astrocytes, 1:1000, Cat# 13-0300, Invitrogen), guinea pig anti-NeuN (a marker for neurons 1:1000, Cat# ABN90, Millipore), mouse anti-NeuroD1 (1:500, Cat# ab60704, Abcam), and chicken anti-GFP (1:1000, Cat# ab13970, Abcam). Representative images are captured by either Zeiss Axioplan fluorescent microscope (Axio Imager Z2, Zeiss, Göttingen, Germany) or
 - 35 confocal microscope (LSM880, Zeiss, Jena, Germany). Quantitative analysis is performed based

on 4 randomly chosen fields (212 μm x 212 μm , acquired at 400 magnification from LSM880 confocal microscope) from 3 brain slices per mouse (3 mice per group). The data is shown as mean $\pm$ SEM.

[00393] Control virus P12, which expresses GFP reporter alone, is first compared with NeuroD1-expressing viruses P134 and P138 (both added P12 together to trace converted neurons). When the control virus P12 is injected in the uninjured mouse cortex, the infected cells are primarily astrocytes without NeuroD1 expression at 10 dpi (days post injection, Figure 36). In contrast, NeuroD1 expression is detected clearly in both P134 and P138 groups. While most NeuroD1-expressing cells in the P138 group at 10 dpi are still astrocytes, a portion of NeuroD1-expressing cells in P134 group are NeuN⁺ neurons already (Figure 36), suggesting that P134 might have better conversion capability than P138. Additionally, at 10 dpi, analysis of the cortex brain tissue of the mice in the P134 group shows a high level of conversion of astrocytes into neurons, as demonstrated by the morphological changes, such as the presence of long processes, in GFP positive cells (Figure 32). The P138 group shows a lower level of conversion.

[00394] At 30 days after virus injection, the infected cells in the control group (P12) remain as astrocytes, but most GFP positive cells in the P134 group are neurons expressing NeuN (Figure 37). However, the conversion rate of the P138 group is lower than the P134. Most infected cells in the P138 group at this stage are still astrocytes, and the GFP signal in the converted neurons is weak (Figure 37). Additionally, at 30 dpi, analysis of the cortex brain tissue of the mice in the P134 group shows an even higher level of conversion of astrocytes into neurons, as demonstrated by the presence of long processes in GFP positive cells (Figure 33)

[00395] The AAV9-P134 virus is also effective in a bilateral injury mouse model. Ischemic stroke is induced in normal C57BL/6J mice (older than 8 weeks) by injecting 1 μL of Endothelin 1, 1-31 aa (1 $\mu\text{g}/\mu\text{L}$) in each side of the cortex. Mice are anesthetized with 20 mg/kg 1.25 % Avertin (a mixture of 12.5 mg/mL of 2,2,2-Tribromoethanol and 25 $\mu\text{L}/\text{mL}$ 2-Methyl-2-butanol, Sigma, St. Louis, MO, USA) through intraperitoneal injection and then placed in a prone position in the stereotaxic frame. Endothelin-1 (ET-1) and virus is injected through glass pipette into motor cortex at the coordinate +0.2 mm anterior-posterior (AP from Bregma), -1.5 mm medial-lateral (ML from Bregma, left side), -0.7 mm dorsal-lateral (DV from dura). The injection speed is 80 nL/min. The pipette is kept in place after injection for about 10 minutes and then slowly withdrawn. Seven days after injection of Endothelin 1, mice are injected with the AAV9-P12 and AAV9-P134 viruses as follow:

- P12 Group: AAV9-P12 5×10^{11} GC/ml, 1 μL , 1 injection in each side of cortex (bilateral)
- P14 Group: AAV9-P12 2.5×10^{11} GC/ml + AAV9-P134 2.5×10^{11} GC/ml, 1 μL , 1 injection in each side of cortex (bilateral)

[00396] Mice are sacrificed at 10 days post injection (dpi) of the viruses and the brain cortex tissue analyzed. When the control virus P12 is injected in the ET-1 lesioned mouse cortex, the infected cells are primarily astrocytes without NeuroD1 expression at 10 dpi (days post injection, Figure 38). In contrast, NeuroD1 expression is detected in the P134 group (Figure 38). Mice are sacrificed at 10 days post injection (dpi) of the viruses and the brain cortex tissue analyzed. At 10 dpi, analysis of the cortex brain tissue of the mice in the P134 group shows a high level of conversion of astrocytes into neurons, as demonstrated by the morphological changes observed in GFP positive cells, such as the presence of long processes (Figure 34).

10 **Example 24. *In vitro* transgene expression and astrocyte-to-neuron conversion induced by expression of NeuroD1 and Dlx2**

Materials and Methods

[00397] Vectors: The vectors are tested via transfection of rat cortical astrocytes (RCAs). Additionally, AAVs are produced with selected vectors and tested *in vitro* using rat astrocytes via transduction:

- NXL-P20 (CE-pGfa681-CI-hND1-p2A-hDLX2-WPRE-SV40pA)
- NXL-P31 (EE-pGfa681-CI-hND1-p2A-hDlx2-WPRE-SV40pA)
- NXL-P111 (CE-pGfa1681-CI-hDlx2-IRES-hND1-SV40pA)
- NXL-P112 (CE-pGfa1681-CI-hDlx2-IRES-hND1-bGHpA)
- NXL-P113 (EE-pGfa1681-CI-hDlx2-IRES-hND1-bGHpA)
- NXL-P122 (CE-pGfa681-CI-hDlx2-P2A-hND1-bGHpA)
- NXL-P123 (EE-pGfa681-CI-hDlx2-P2A-hND1-bGHpA)
- NXL-P124 (CE-pGfa681-CI-hND1-P2A-hDlx2-bGHpA)

25 [00398] Figure 40 depicts two general maps of the constructs.

[00399] Cell Culture: Rat cortical astrocytes (RCAs) of passage 5-7 are seeded on glass cover slips coated with poly-D-lysine (PDL) in 24-well plates at 30-50% confluency 24-48 hours prior to transduction.

[00400] Transduction and Immunofluorescence: Cells are transduced with virus at 2-6X10¹⁰ vg/ml in 500 ul of fresh astrocyte media (DMEM supplemented with 10% FBS, 2.5 mM Glutamine, 3.5 mM Glucose, pen/strep). At 48 hours post transduction, media is replaced with 5% FBS astrocyte media. For the next 4 days, 100 ul of conversion media (DMEM/F12 + 1% FBS + B27 + N2 and 1 uM Rock inhibitor and 10 ng/ml BDNF) is added daily. Then the media is completely replaced with conversion media.

[00401] Cells are fixed with 4% paraformaldehyde in PBS at various desired time points (e.g., three days, one to five weeks post transduction) and subsequently washed and immunostained with antibodies against ND1 (Abcam Cat# ab60704), Dlx2 (Millipore Cat# AB5726), NeuN (Millipore, Cat# ABN78), Map2 (Invitrogen, Cat# PA5-17646) and followed with secondary antibodies conjugated with fluorescent dyes (Invitrogen, Alexafluor). Images are captured under a fluorescent microscope (Zeiss Axiovert A1, Zen Blue). Gene expression levels are assessed by comparing the fluorescence intensity.

***In Vitro* Studies Results:**

10 [00402] The tested ND1-Dlx2 constructs are effective in driving the expression of ND1 and Dlx2 by transfection and/or transduction of the cultured rat astrocytes as demonstrated by the positive staining of ND1 and Dlx2 in these cells (Figures 41-56). Additionally, ND1/Dlx2-containing AAVs (AAV9-P122, AAV-P122, AAV-P124, and AAV-P20) are effective in driving the expression of ND1, Dlx2, and inducing an astrocyte-to-neuron conversion in cultured rat astrocytes as shown by positive staining of neuronal markers NeuN and/or MAP2 (Figures 43, 46, 15 and 49, and 52). Some NeuN/MAP2 positive cells show typical neuronal morphology.

Example 25. *In vivo* transgene expression and astrocyte-to-neuron conversion induced by expression of NeuroD1 and Dlx2

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[00403] AAV9-P112 and AAV9-P122 viruses are used for the *in vivo* studies. AAV9-P12, which drives the expression of GFP alone (no ND1 nor Dlx2) under a GFAP promoter, is used for the control and to identify cells expressing GFAP (astrocytes).

25 [00404] Normal C57BL/6 mice older than 8 weeks are injected with AAV-P12, AAV9-P112, and AAV9-P122 viruses as follows:

- P12 control group: AAV9-P12 2.5×10^{11} GC/ml, 2 μ L, 1 injection in striatum (n=6)
- P112 group: AAV9-P12 2.5×10^{11} GC/ml + AAV9-P112 2.5×10^{11} GC/ml, 2 μ L, 1 injection in striatum (n=6)
- 30 • P122 group: AAV9-P12 2.5×10^{11} GC/ml + AAV9-P122 2.5×10^{11} GC/ml, 2 μ L, 1 injection in striatum (n=6)

[00405] Mice are sacrificed and brain cortex tissue are analyzed at 10 days post infection (dpi) and at 30 dpi. The infection of mouse striatum by AAV9-P12 is confirmed at 10 dpi by the presence of GFP fluorescence. At 30 dpi, AAV9-P12 infected cells are still GFAP-positive astrocytes, suggesting that AAV9-P12 is specific in infecting astrocytes (Figure 57A-B). In the

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P112 group, the majority of cells co-infected with AAV9-P12 and AAV9-P112 are GFAP-positive stained astrocytes at 10 dpi (Figure 58). By 30 days post viral infection, many cells co-infected with AAV9-P12 and AAV9-P112 become NeuN-positive neurons (Figure 59, white arrows). In the P122 group, the majority of cells co-infected with AAV9-P12 and AAV9-P122 are GFAP-positive stained astrocytes at 10 dpi (Figure 60). By 30 days post viral infection, many cells co-infected with AAV9-P12 and AAV9-P122 become NeuN-positive neurons (Figure 61, white arrows).

Example 19. *In vitro* transgene expression of Dlx2

10 [00406] Vectors: The vectors are tested via transfection of rat cortical astrocytes (RACs). Additionally, AAVs are produced with selected vectors and tested *in vitro* via transduction: NXL-P44: EE-pGfa681-CI-Dlx2-WPRE-SV40pA

- NXL-P60: EE-pGfa681-Dlx2-WPRE-SV40pA
 - NXL-P75: CE-pGfa681-CI-Dlx2-WPRE-SV40pA
 - 15 • NXL-P104: CE-pGfa681-CGRI-Dlx2-bGHpA
 - NXL-P105: CE-pGfa681-CI-Dlx2-oPRE-bGHpA
 - NXL-P131: EE-pGfa681-CI-Dlx2-oPRE-bGHpA
 - NXL-P133: CE-pGfa681-CGRI-Dlx2-oPRE-bGHpA
 - NXL-P137: EE-pGfa681-CGRI-Dlx2-oPRE-bGHpA
- 20 [00407] NXL-P104 and NXL-P105 constructs are effective in driving the expression of Dlx2 24 hours post transfection of the cultured RACs, as demonstrated by the positive Dlx2 staining in these cells (Figure 62). NXL-P133, NXL-P137, and NXL-P131 constructs are effective in driving the expression of Dlx2 24 hours post transfection of the cultured RACs, as demonstrated by the positive Dlx2 staining in these cells (Figure 63). AAV9-P133 (the AAV produced with
- 25 NLX-P133) is effective in driving the expression of Dlx2 after transducing cultured RACs with this virus, as demonstrated by the positive Dlx2 staining in these cells (Figure 64).

[00408] A variety of further modifications and improvements in and to the compositions and methods of the present disclosure will be apparent to those skilled in the art based. The following non-limiting embodiments are envisioned:

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1. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of

SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
- (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

2. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, (iii) or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;

- (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
3. An adeno-associated virus (AAV) vector comprising a neurogenic differentiation 1 (NeuroD1) nucleic acid coding sequence encoding a NeuroD1 protein and a distal-less homeobox 2 (Dlx2) nucleic acid coding sequence encoding a Dlx2 protein, wherein said NeuroD1 coding sequence and said Dlx2 coding sequence are separated by a linker sequence, wherein said NeuroD1 coding sequence and said Dlx2 coding sequence are operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter;
- (b) an enhancer;
- (c) a chimeric intron;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
- (e) a polyadenylation signal sequence.
4. A composition comprising an adeno-associated virus (AAV) vector for converting glial cells to functional neurons in a human, wherein said AAV vector comprises a human neurogenic differentiation 1 (hNeuroD1) sequence having a nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence having a nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and hDlx2 sequence are operably linked to regulatory elements comprising:

- (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
 - (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
 - (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
5. A composition comprising an adeno-associated-virus (AAV) vector for converting glial cells to functional neurons in a human, wherein said AAV vector comprises a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are operably linked to regulatory elements comprising:
- (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
 - (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;

- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
6. A composition comprising an adeno-associated virus (AAV) vector for the treatment of a subject in need thereof, wherein said AAV vector comprises a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and said Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter;
- (b) an enhancer;
- (c) a chimeric intron;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
- (e) a polyadenylation signal.
7. The AAV vector of any one of embodiments 1-3, or the composition of any one of embodiments 4-6, wherein said AAV vector is selected from the group consisting of AAV serotype 2, AAV serotype 5, and AAV serotype 9.
8. The AAV vector or composition of embodiment 7, wherein said AAV vector is AAV serotype 2.
9. The AAV vector or composition of embodiment 7, wherein said AAV vector is AAV serotype 5.
10. The AAV vector or composition of embodiment 7, wherein said AAV vector is AAV serotype 9.
11. The composition of embodiment 4 or 5, wherein said glial cells are reactive astrocytes.
12. The composition of embodiment 4 or 5, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic

neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons.

13. The composition of embodiment 4 or 5, wherein said human has a neurological condition.
- 5 14. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said NeuroD1 is a human NeuroD1 (hNeuroD1).
15. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said Dlx2 is a human Dlx2 (hDlx2).
- 10 16. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said NeuroD1 is selected from the group consisting of a chimpanzee NeuroD1, a bonobo NeuroD1, an orangutan NeuroD1, a gorilla NeuroD1, a macaque NeuroD1, a marmoset NeuroD1, a capuchin NeuroD1, a baboon NeuroD1, a gibbon NeuroD1, and a lemur NeuroD1.
- 15 17. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said Dlx2 is selected from the group consisting of a chimpanzee Dlx2, a bonobo Dlx2, an orangutan Dlx2, a gorilla Dlx2, a macaque Dlx2, a marmoset Dlx2, a capuchin Dlx2, a baboon Dlx2, a gibbon Dlx2, and a lemur Dlx2.
- 20 18. The AAV vector or composition of embodiment 14, wherein said hNeuroD1 comprises a nucleic acid sequence encoding an amino acid sequence at least 80% identical or similar to SEQ ID NO: 10.
19. The AAV vector or composition of embodiment 15, wherein said hDlx2 comprises a nucleic acid sequence encoding an amino acid sequence at least 80% identical or similar to SEQ ID NO: 14.
- 25 20. The AAV vector or composition of embodiment 14, wherein said hNeuroD1 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 6, or the complement thereof.
21. The AAV vector or composition of embodiment 15, said hDlx2 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof.

22. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said linker is selected from the group consisting of P2A and T2A.
23. The AAV vector or composition of embodiment 22, wherein said linker is said P2A.
24. The AAV vector or composition of embodiment 22, wherein said linker is said T2A
- 5 25. The AAV vector or composition of embodiment 22, wherein said P2A linker comprises a nucleic acid sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 15 and 18, or the complement thereof.
26. The AAV vector or composition of embodiment 22, wherein said T2A linker comprises a nucleic acid sequence at least 80% identical to the sequence selected from the group
10 consisting of SEQ ID NO: 16 and 19, or the complement thereof.
27. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said GFAP promoter is a human GFAP (hGFAP) promoter.
28. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said GFAP promoter is selected from the group consisting of a chimpanzee GFAP promoter,
15 a bonobo GFAP promoter, an orangutan GFAP promoter, a gorilla GFAP promoter, a macaque GFAP promoter, a marmoset GFAP promoter, a capuchin GFAP promoter, a baboon GFAP promoter, a gibbon GFAP promoter, and a lemur GFAP promoter.
29. The AAV vector or composition of any one of the preceding embodiments, wherein said IRES sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO:
20 3, or the complement thereof.
30. The AAV vector or composition of embodiment 27, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 4 or the complement thereof.
31. The AAV vector or composition of embodiment 27, wherein said hGFAP promoter
25 comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 12 or the complement thereof.
32. The AAV vector or composition of embodiment 27, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 26 or the complement thereof.

33. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said enhancer is selected from the group consisting of an enhancer from human elongation factor-1 alpha (EF1- α) promoter and cytomegalovirus (CMV) enhancer
- 5 34. The AAV vector or composition of embodiment 33, wherein said EF1- α comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 2, or the complement thereof.
35. The AAV vector or composition of embodiment 33, wherein said CMV enhancer comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 11, or the complement thereof.
- 10 36. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said chimeric intron comprises a nucleic acid sequence at least 80% identical to a nucleic acid selected from the group consisting of SEQ ID NOs: 5 and 27, or the complement thereof.
37. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said WPRE comprises a nucleic acid sequence at least 80% identical to a nucleic acid
15 selected from the group consisting of SEQ ID NOs: 7 and 29, or the complement thereof.
38. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said polyadenylated signal is selected from the group consisting of SV40 polyadenylation signal, a hGH polyadenylation signal, and a bGH polyadenylation signal.
39. The AAV vector or composition of embodiment 38, wherein said SV40 polyadenylated
20 signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 8, or the complement thereof.
40. The AAV vector or composition of embodiment 38, wherein said hGH polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 17, or the complement thereof.
- 25 41. The AAV vector or composition of embodiment 38, wherein said bGH polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 30, or the complement thereof.
42. The AAV vector of embodiment 3, or the composition of embodiment 6, wherein said AAV vector further comprises a nucleic acid sequence encoding an AAV protein
30 sequence.

43. The AAV vector of any one of embodiments 1-3, or the composition of any one of
embodiments 4-6, wherein said AAV vector comprises AAV serotype 2 inverted
terminal repeats (ITRs).
44. The AAV vector of any one of embodiments 1-3, or the composition of any one of
embodiments 4-6, wherein said AAV vector comprises AAV serotype 5 inverted
terminal repeats (ITRs).
45. The AAV vector of any one of embodiments 1-3, or the composition of any one of
embodiments 4-6, wherein said AAV vector comprises AAV serotype 9 inverted
terminal repeats (ITRs).
46. The AAV vector of any one of embodiments 1-3, or the composition of any one of
embodiments 4-6, wherein said AAV vector comprises at least one ITR nucleic acid
sequence at least 80% identical to SEQ ID NO: 1.
47. The AAV vector of any one of embodiments 1-3, or the composition of any one of
embodiments 4-6, wherein said AAV vector comprises at least one ITR nucleic acid
sequence at least 80% identical to SEQ ID NO: 9.
48. The composition of embodiment 6, wherein said subject in need thereof is a mammal.
49. The composition of embodiment 48, wherein said mammal is a human.
50. The composition of embodiment 48, wherein said mammal is a non-human primate.
51. The composition of embodiment 6, wherein said subject in need thereof has a
neurological condition.
52. The composition of embodiment 13 or 51, wherein said neurological condition
comprises an injury to the central nervous system (CNS) or peripheral nervous system.
53. The composition of embodiment 13 or 51, wherein said wherein said neurological
condition comprises an injury to the CNS.
54. The composition of embodiment 13 or 51, wherein said neurological condition is
selected from the group consisting of Alzheimer's Disease, Parkinson's Disease,
amyotrophic lateral sclerosis (ALS), Huntington's Disease, epilepsy, physical injury,
stroke, cerebral aneurysm, traumatic brain injury, concussion, a tumor, inflammation,
infection, ataxia, brain atrophy, spinal cord atrophy, multiple sclerosis, traumatic spinal

cord injury, ischemic or hemorrhagic myelopathy (myelopathy), global ischemia, hypoxic ischemic encephalopathy, embolism, fibrocartilage embolism myelopathy, thrombosis, nephropathy, chronic inflammatory disease, meningitis, and cerebral venous sinus thrombosis.

- 5 55. The composition of embodiment 13 or 51, wherein said neurological condition is Alzheimer's Disease.
56. The composition of embodiment 13 or 51, wherein said neurological condition is Parkinson's Disease.
57. The composition of embodiment 13 or 51, wherein said neurological condition is ALS.
- 10 58. The composition of embodiment 13 or 51, wherein said neurological condition is Huntington's Disease.
59. The composition of embodiment 13 or 51, wherein said neurological condition is a stroke.
60. The composition of embodiment 59, wherein said stroke is an ischemic stroke.
- 15 61. The composition of embodiment 59, wherein said stroke is a hemorrhagic stroke.
62. The composition of embodiment 51, wherein said composition is capable of converting at least one glial cell to a neuron.
63. The composition of embodiment 62, wherein said glial cells are selected from the group consisting of astrocytes and NG2 cells.
- 20 64. The composition of embodiment 62, wherein said glial cells are astrocytes.
65. The composition of embodiment 62, wherein said astrocytes are reactive astrocytes.
66. The composition of embodiment 62, wherein said glial cells are GFAP positive.
67. The composition of embodiment 62, wherein said neurons are functional neurons.
- 25 68. The composition of embodiment 62, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons.

69. The composition of embodiment 68, wherein said functional neurons are glutamatergic neurons.
70. The composition of embodiment 6, wherein said composition is formulated to be delivered to a subject in need thereof.
- 5 71. The composition of embodiment 70, wherein said composition is formulated for local delivery.
72. The composition of embodiment 70, wherein said composition is formulated for systemic delivery.
- 10 73. The composition of any one of embodiments 70-72, wherein said composition is formulated for delivery via intraperitoneal, intramuscular, intravenous, intrathecal, intracerebral, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal, intranasal, or oral administration.
74. A method comprising delivering the composition of embodiment 6 to said subject in need thereof.
- 15 75. The method of embodiment 74, wherein said composition is formulated to be delivered to a subject in need thereof.
76. The method of embodiment 74, wherein said delivering comprises local administration.
77. The method of embodiment 74, wherein said delivering comprises systemic administration.
- 20 78. The method of any one of embodiments 74-77, wherein said delivering comprises an intraperitoneal, intramuscular, intravenous, intrathecal, intracerebral, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal, intranasal, or oral administration.
- 25 79. A method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said

hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

- (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
- (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

80. A method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and hDlx2 coding sequence are operably linked to expression control elements comprising:

- (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
 - (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
 - (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
81. A method of converting glial cells to neurons in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising:
- (a) a glial fibrillary acid protein (GFAP) promoter;
 - (b) an enhancer;
 - (c) a chimeric intron;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
 - (e) and a polyadenylation signal sequence,
- wherein said AAV vector is capable of converting at least one glial cell to a neuron in said subject in need thereof.
82. A method of treating a neurological condition in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject, wherein said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and said Dlx2 sequence are operably linked to expression control elements comprising:

- (a) a glial fibrillary acid protein (GFAP) promoter;
(b) an enhancer;
(c) a chimeric intron;
(d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
5 (e) a polyadenylation signal to said subject in need thereof.
83. The method of any one of embodiments 79-82, wherein said AAV is selected from the group consisting of AAV serotype 2, AAV serotype 5, and AAV serotype 9.
84. The method of embodiment 83, wherein said AAV is AAV serotype 2.
85. The method of embodiment 83, wherein said AAV is AAV serotype 5.
- 10 86. The method of embodiment 83, wherein said AAV is AAV serotype 9.
87. The method of embodiments 79 or 80, wherein said functional neurons are glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons.
- 15 88. The method of embodiments 81 or 82, wherein said NeuroD1 is human NeuroD1 (hNeuroD1).
89. The method of embodiments 81 or 82 wherein said Dlx2 is human Dlx2 (hDlx2).
90. The method of embodiments 81 or 82, wherein said NeuroD1 is selected from the group consisting of a chimpanzee NeuroD1, a bonobo NeuroD1, an orangutan NeuroD1, a gorilla NeuroD1, a macaque NeuroD1, a marmoset NeuroD1, a capuchin NeuroD1, a baboon NeuroD1, a gibbon NeuroD1, and a lemur NeuroD1.
- 20 91. The method of embodiments 75 or 76, wherein said Dlx2 is selected from the group consisting of a chimpanzee Dlx2, a bonobo Dlx2, an orangutan Dlx2, a gorilla Dlx2, a macaque Dlx2, a marmoset Dlx2, a capuchin Dlx2, a baboon Dlx2, a gibbon Dlx2, and a lemur Dlx2.
- 25 92. The method of embodiment 88, wherein said hNeuroD1 comprises an amino acid sequence encoding an amino acid coding sequence at least 80% identical or similar to SEQ ID NO: 10.
93. The method of embodiment 89, said hDlx2 comprises a amino acid sequence encoding an amino acid sequence at least 80% identical or similar to SEQ ID NO: 14.

94. The method of embodiment 88, wherein said hNeuroD1 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 6, or the complement thereof.
95. The method of embodiment 89, said hDlx2 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof.
96. The method of embodiments 81 or 82, wherein said GFAP promoter is a human GFAP (hGFAP) promoter.
97. The method of embodiments 81 or 82, wherein said GFAP promoter is selected from the group consisting of a chimpanzee GFAP promoter, a bonobo GFAP promoter, an orangutan GFAP promoter, a gorilla GFAP promoter, a macaque GFAP promoter, a marmoset GFAP promoter, a capuchin GFAP promoter, a baboon GFAP promoter, a gibbon GFAP promoter, and a lemur GFAP promoter.
98. The method of any one of embodiments 79-97, wherein said IRES sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 3, or the complement thereof.
99. The method of embodiment 96, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 4, or the complement thereof.
100. The method of embodiment 96, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 12, or the complement thereof.
101. The method of embodiment 96, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 26, or the complement thereof.
102. The method of embodiments 81 or 82, wherein said linker is selected from the group consisting of P2A and T2A.
103. The method of embodiment 102, wherein said P2A linker comprises a nucleic acid sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 15 and 18, or the complement thereof.

104. The method of embodiment 102, wherein said T2A linker comprises a nucleic acid sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 16 and 19, or the complement thereof.
105. The method of embodiments 81 or 82, wherein said enhancer is selected from the group consisting of an enhancer from human elongation factor-1 alpha (EF1- α) promoter and cytomegalovirus (CMV) enhancer.
106. The method of embodiment 105, wherein said EF1- α comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 2, or the complement thereof.
107. The method of embodiment 105, wherein said CMV enhancer comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 11, or the complement thereof.
108. The method of embodiments 81 or 82, wherein said chimeric intron comprises a nucleic acid sequence at least 80% identical to nucleic acid selected from the group consisting of SEQ ID NOs: 5 and 27, or the complement thereof.
109. The method of embodiments 81 or 82, wherein said WPRE comprises a nucleic acid sequence at least 80% identical to a nucleic acid selected from the group consisting of SEQ ID NOs: 7 and 29, or the complement thereof.
110. The method of embodiments 81 or 82, wherein said polyadenylated signal selected from the group consisting of SV40 polyadenylation signal and a hGH polyadenylation signal
111. The method of embodiment 110, wherein said SV40 polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 8, or the complement thereof.
112. The method of embodiment 110, wherein said hGH polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof.
113. The method of embodiments 81 or 82, wherein said vector further comprises a nucleic acid sequence encoding an AAV protein sequence.

114. The method of any one of embodiments 79-82, wherein said vector comprises AAV serotype 2 inverted terminal repeats (ITRs).
115. The method of any one of embodiments 79-82, wherein said vector comprises AAV serotype 5 inverted terminal repeats (ITRs).
- 5 116. The method of any one of embodiments 79-82, wherein said vector comprises AAV serotype 9 inverted terminal repeats (ITRs).
117. The method of any one of embodiments 79-82, wherein said vector comprises at least one ITR nucleic acid sequence at least 80% identical to SEQ ID NO: 1.
118. The method of any one of embodiments 79-82, wherein said vector comprises at
10 least one ITR nucleic acid sequence at least 80% identical to SEQ ID NO: 9.
119. The method of embodiment 81, wherein said converting occurs in the central nervous system (CNS) or peripheral nervous system.
120. The method of embodiment 81, wherein said converting occurs in the CNS.
121. The method of embodiment 81 or 82, wherein said subject in need thereof is a
15 mammal.
122. The method of embodiment 121, wherein said mammal is a human.
123. The method of embodiment 121, wherein said mammal is a non-human primate.
124. The method of embodiment 81 or 82, wherein said delivering comprises a local administration.
- 20 125. The method of embodiment 81 or 82, wherein said delivering comprises systemic administration.
126. The method of embodiment 81 or 82, wherein said delivering comprises an administration selected from the group consisting of an intraperitoneal administration, intramuscular administration, intravenous administration, intrathecal administration,
25 intracerebral administration, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal administration, intranasal administration, and oral administration.

127. The method of embodiment 79 or 80, wherein said injecting comprises an injection selected from the group consisting of an intraperitoneal injection, intramuscular injection, intravenous injection, intrathecal injection, intracerebral injection, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal injection, intranasal injection, and oral injection.
128. The method of embodiments 81 or 82, wherein said delivering comprises injecting.
129. The method of any one of embodiments 79, 80 or 128, wherein said injecting is performed at a concentration of between 10^{10} particles/mL and 10^{14} particles/mL.
130. The method of embodiment 129, wherein said injecting further comprises a flow rate of between 0.1 μ L/minute and 5.0 μ L/minute.
131. The method of embodiment 81, wherein said at least one glial cell is selected from the group consisting of at least one astrocyte and at least one NG2 cell.
132. The method of embodiment 131, wherein said at least one glial cell is at least one astrocyte.
133. The method of embodiment 131 or 132, wherein said at least one astrocyte is a reactive astrocyte.
134. The method of embodiment 81, wherein said neuron is a functional neuron.
135. The method of any one of embodiments 79, 80 and 134, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons..
136. The method of embodiment 81, wherein said subject exhibits an improvement of at least one neurological condition symptom as compared to said subject prior to said delivering.
137. The method of embodiment 136, wherein said improvement is measured within 1 year of said delivering.
138. The method of any one of embodiments 79, 80, or 128, wherein said method comprises directly injecting said AAV into the brain of said subject.

139. The method of any one of embodiments 79 or 80 wherein said wherein said converting is in the striatum of said brain.
140. The method of any one of embodiments 79, 80, or 128, wherein said method comprises directly injecting said AAV into the spinal cord of said subject.
- 5 141. The method of embodiment 82, wherein said neurological condition comprises an injury to the central nervous system (CNS) or peripheral nervous system.
142. The method of embodiment 82, wherein said neurological condition is selected from the group consisting of Alzheimer's Disease, Parkinson's Disease, amyotrophic lateral sclerosis (ALS), Huntington's Disease, epilepsy, physical injury, stroke, cerebral aneurysm, traumatic brain injury, concussion, a tumor, inflammation, infection, ataxia, 10 brain atrophy, spinal cord atrophy, multiple sclerosis, traumatic spinal cord injury, ischemic or hemorrhagic myelopathy (myelopathy), global ischemia, hypoxic ischemic encephalopathy, embolism, fibrocartilage embolism myelopathy, thrombosis, nephropathy, chronic inflammatory disease, meningitis, and cerebral venous sinus 15 thrombosis.
143. The method of embodiment 82, wherein said neurological condition is Alzheimer's Disease.
144. The method of embodiment 82, wherein said neurological condition is Parkinson's Disease.
- 20 145. The method of embodiment 82, wherein said neurological condition is ALS.
146. The method of embodiment 82, wherein said neurological condition is Huntington's Disease.
147. The method of embodiment 82, wherein said neurological condition is a stroke.
148. The method of embodiment 147, wherein said stroke is an ischemic stroke.
- 25 149. The method of embodiment 147, wherein said stroke is a hemorrhagic stroke.
150. The method of embodiment 82, wherein said method is capable of converting at least one glial cell into a neuron.

151. The method of embodiment 150, wherein said glial cells are selected from the group consisting of astrocytes and NG2 cells.
152. The method of embodiment 150, wherein said glial cells are astrocytes.
153. The method of embodiment 152, wherein said astrocytes are reactive astrocytes.
- 5 154. The method of embodiment 150, wherein said glial cells are GFAP positive.
155. The method of embodiment 150, wherein said neurons are functional neurons.
156. The method of embodiment 145, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor
10 neurons, and peptidergic neurons.
157. The method of embodiments 79 or 80, wherein a therapeutically effective dose of said AAV is injected into said subject.
158. The method of embodiments 81 or 82, wherein a therapeutically effective dose of said AAV is delivered to said subject.
- 15 159. The method of embodiment 147 or 148, wherein said therapeutically effective dose is administered with a pharmaceutically acceptable carrier.
160. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6, and (ii) an adeno-associated virus (AAV) vector comprising
20 a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13;
wherein the hNeuroD1 sequence is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- 25 (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE)
30 comprising the nucleic acid sequence of SEQ ID NO: 29; and

(e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

161. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6, and (ii) an adeno-associated virus (AAV) vector comprising a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13;

wherein the hNeuroD1 sequence is operably linked to regulatory elements comprising:

(a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;

(b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;

(c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;

(d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and

(e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

162. The composition of embodiment 160 or 161, wherein (ii) comprises an AAV vector comprising the hDlx2 sequence comprising the nucleic acid sequence of SEQ ID NO: 13 operably linked to regulatory elements comprising:

(a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence of SEQ ID NO: 26;

(b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;

(c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;

(d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and

(e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

163. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10, and (ii) an adeno-associated virus (AAV) vector comprising a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14; wherein the nucleic acid sequence encoding an hNeuroD1 protein is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
 - (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;
 - (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
 - (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
164. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10, and (ii) an adeno-associated virus (AAV) vector comprising a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14; wherein the nucleic acid sequencing encoding an hNeuroD1 protein is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
 - (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and

(e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30

165. The composition of embodiment 163 or 164, wherein (ii) comprises an AAV vector comprising a nucleic acid coding sequence encoding the hDlx2 protein, wherein the nucleic acid is operably linked to regulatory elements comprising:

(a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence of SEQ ID NO: 26;

(b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;

(c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;

(d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and

(e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

166. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

(a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;

(b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;

(c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 5;

- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
- 5 167. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:
- 10 (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5;
- 20 (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
168. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:
- 25 30

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;
- 5 (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
- 10 169. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2
- 15 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence
- 20 and said hDlx2 coding sequence is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- 25 (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
- 30 170. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by a P2A linker comprising the nucleic acid sequence selected from the

group consisting of SEQ ID NO: 15 and 18 or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

- 5 (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5; and
- 10 (d) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

171. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence
15 encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18 or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3,
20 wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- 25 (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5; and
- (d) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

CLAIMS:

1. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:
 - (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
 - (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
 - (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
2. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, (iii)

or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:

- 5 (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
- (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- 10 (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- 15 (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

3. An adeno-associated virus (AAV) vector comprising a neurogenic differentiation 1 (NeuroD1) nucleic acid coding sequence encoding a NeuroD1 protein and a distal-less homeobox 2 (Dlx2) nucleic acid coding sequence encoding a Dlx2 protein, wherein said 20 NeuroD1 coding sequence and said Dlx2 coding sequence are separated by a linker sequence, wherein said NeuroD1 coding sequence and said Dlx2 coding sequence are operably linked to regulatory elements comprising:

- 25 (a) a glial fibrillary acidic protein (GFAP) promoter;
- (b) an enhancer;
- (c) a chimeric intron;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
- (e) a polyadenylation signal sequence.

4. A composition comprising an adeno-associated virus (AAV) vector for converting glial 30 cells to functional neurons in a human, wherein said AAV vector comprises a human neurogenic differentiation 1 (hNeuroD1) sequence having a nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence having a nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected

from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and hDlx2 sequence are operably linked to regulatory elements comprising:

- (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
- (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

5. A composition comprising an adeno-associated-virus (AAV) vector for converting glial cells to functional neurons in a human, wherein said AAV vector comprises a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are operably linked to regulatory elements comprising:
 - (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;

- (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
6. A composition comprising an adeno-associated virus (AAV) vector for the treatment of a subject in need thereof, wherein said AAV vector comprises a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and said Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter;
- (b) an enhancer;
- (c) a chimeric intron;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
- (e) a polyadenylation signal.
7. The AAV vector of any one of claims 1-3, or the composition of any one of claims 4-6, wherein said AAV vector is selected from the group consisting of AAV serotype 2, AAV serotype 5, and AAV serotype 9.
8. The AAV vector or composition of claim 7, wherein said AAV vector is AAV serotype 2.
9. The AAV vector or composition of claim 7, wherein said AAV vector is AAV serotype 5.
10. The AAV vector or composition of claim 7, wherein said AAV vector is AAV serotype 9.

11. The composition of claim 4 or 5, wherein said glial cells are reactive astrocytes.
12. The composition of claim 4 or 5, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons.
13. The composition of claim 4 or 5, wherein said human has a neurological condition.
14. The AAV vector of claim 3, or the composition of claim 6, wherein said NeuroD1 is a human NeuroD1 (hNeuroD1).
15. The AAV vector of claim 3, or the composition of claim 6, wherein said Dlx2 is a human Dlx2 (hDlx2).
16. The AAV vector of claim 3, or the composition of claim 6, wherein said NeuroD1 is selected from the group consisting of a chimpanzee NeuroD1, a bonobo NeuroD1, an orangutan NeuroD1, a gorilla NeuroD1, a macaque NeuroD1, a marmoset NeuroD1, a capuchin NeuroD1, a baboon NeuroD1, a gibbon NeuroD1, and a lemur NeuroD1.
17. The AAV vector of claim 3, or the composition of claim 6, wherein said Dlx2 is selected from the group consisting of a chimpanzee Dlx2, a bonobo Dlx2, an orangutan Dlx2, a gorilla Dlx2, a macaque Dlx2, a marmoset Dlx2, a capuchin Dlx2, a baboon Dlx2, a gibbon Dlx2, and a lemur Dlx2.
18. The AAV vector or composition of claim 14, wherein said hNeuroD1 comprises a nucleic acid sequence encoding an amino acid sequence at least 80% identical or similar to SEQ ID NO: 10.
19. The AAV vector or composition of claim 15, wherein said hDlx2 comprises a nucleic acid sequence encoding an amino acid sequence at least 80% identical or similar to SEQ ID NO: 14.
20. The AAV vector or composition of claim 14, wherein said hNeuroD1 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 6, or the complement thereof.

21. The AAV vector or composition of claim 15, said hDlx2 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof.
22. The AAV vector of claim 3, or the composition of claim 6, wherein said linker is
5 selected from the group consisting of P2A and T2A.
23. The AAV vector or composition of claim 22, wherein said linker is said P2A.
24. The AAV vector or composition of claim 22, wherein said linker is said T2A.
25. The AAV vector or composition of claim 22, wherein said P2A linker comprises a
10 nucleic acid sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 15 and 18, or the complement thereof.
26. The AAV vector or composition of claim 22, wherein said T2A linker comprises a nucleic acid sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 16 and 19, or the complement thereof.
27. The AAV vector of claim 3, or the composition of claim 6, wherein said GFAP promoter
15 is a human GFAP (hGFAP) promoter.
28. The AAV vector of claim 3, or the composition of claim 6, wherein said GFAP promoter is selected from the group consisting of a chimpanzee GFAP promoter, a bonobo GFAP promoter, an orangutan GFAP promoter, a gorilla GFAP promoter, a macaque GFAP promoter, a marmoset GFAP promoter, a capuchin GFAP promoter, a baboon GFAP
20 promoter, a gibbon GFAP promoter, and a lemur GFAP promoter.
29. The AAV vector or composition of any one of the preceding claims, wherein said IRES sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 3, or the complement thereof.
30. The AAV vector or composition of claim 27, wherein said hGFAP promoter comprises a
25 nucleic acid sequence at least 80% identical to SEQ ID NOs: 4 or the complement thereof.
31. The AAV vector or composition of claim 27, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 12 or the complement thereof.

32. The AAV vector or composition of claim 27, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 26 or the complement thereof.
- 5 33. The AAV vector of claim 3, or the composition of claim 6, wherein said enhancer is selected from the group consisting of an enhancer from human elongation factor-1 alpha (EF1- α) promoter and cytomegalovirus (CMV) enhancer
34. The AAV vector or composition of claim 33, wherein said EF1- α comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 2, or the complement thereof.
- 10 35. The AAV vector or composition of claim 33, wherein said CMV enhancer comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 11, or the complement thereof.
36. The AAV vector of claim 3, or the composition of claim 6, wherein said chimeric intron comprises a nucleic acid sequence at least 80% identical to a nucleic acid selected from the group consisting of SEQ ID NOs: 5 and 27, or the complement thereof.
- 15 37. The AAV vector of claim 3, or the composition of claim 6, wherein said WPRE comprises a nucleic acid sequence at least 80% identical to a nucleic acid selected from the group consisting of SEQ ID NOs: 7 and 29, or the complement thereof.
- 20 38. The AAV vector of claim 3, or the composition of claim 6, wherein said polyadenylated signal is selected from the group consisting of SV40 polyadenylation signal, a hGH polyadenylation signal, and a bGH polyadenylation signal.
39. The AAV vector or composition of claim 38, wherein said SV40 polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 8, or the complement thereof.
- 25 40. The AAV vector or composition of claim 38, wherein said hGH polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 17, or the complement thereof.
41. The AAV vector or composition of claim 38, wherein said bGH polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 30, or the complement thereof.

42. The AAV vector of claim 3, or the composition of claim 6, wherein said AAV vector further comprises a nucleic acid sequence encoding an AAV protein sequence.
43. The AAV vector of any one of claims 1-3, or the composition of any one of claims 4-6, wherein said AAV vector comprises AAV serotype 2 inverted terminal repeats (ITRs).
- 5 44. The AAV vector of any one of claims 1-3, or the composition of any one of claims 4-6, wherein said AAV vector comprises AAV serotype 5 inverted terminal repeats (ITRs).
45. The AAV vector of any one of claims 1-3, or the composition of any one of claims 4-6, wherein said AAV vector comprises AAV serotype 9 inverted terminal repeats (ITRs).
- 10 46. The AAV vector of any one of claims 1-3, or the composition of any one of claims 4-6, wherein said AAV vector comprises at least one ITR nucleic acid sequence at least 80% identical to SEQ ID NO: 1.
47. The AAV vector of any one of claims 1-3, or the composition of any one of claims 4-6, wherein said AAV vector comprises at least one ITR nucleic acid sequence at least 80% identical to SEQ ID NO: 9.
- 15 48. The composition of claim 6, wherein said subject in need thereof is a mammal.
49. The composition of claim 48, wherein said mammal is a human.
50. The composition of claim 48, wherein said mammal is a non-human primate.
51. The composition of claim 6, wherein said subject in need thereof has a neurological condition.
- 20 52. The composition of claim 13 or 51, wherein said neurological condition comprises an injury to the central nervous system (CNS) or peripheral nervous system.
53. The composition of claim 13 or 51, wherein said wherein said neurological condition comprises an injury to the CNS.
- 25 54. The composition of claim 13 or 51, wherein said neurological condition is selected from the group consisting of Alzheimer's Disease, Parkinson's Disease, amyotrophic lateral sclerosis (ALS), Huntington's Disease, epilepsy, physical injury, stroke, cerebral aneurysm, traumatic brain injury, concussion, a tumor, inflammation, infection, ataxia, brain atrophy, spinal cord atrophy, multiple sclerosis, traumatic spinal cord injury,

ischemic or hemorrhagic myelopathy (myelopathy), global ischemia, hypoxic ischemic encephalopathy, embolism, fibrocartilage embolism myelopathy, thrombosis, nephropathy, chronic inflammatory disease, meningitis, and cerebral venous sinus thrombosis.

- 5 55. The composition of claim 13 or 51, wherein said neurological condition is Alzheimer's Disease.
56. The composition of claim 13 or 51, wherein said neurological condition is Parkinson's Disease.
57. The composition of claim 13 or 51, wherein said neurological condition is ALS.
- 10 58. The composition of claim 13 or 51, wherein said neurological condition is Huntington's Disease.
59. The composition of claim 13 or 51, wherein said neurological condition is a stroke.
60. The composition of claim 59, wherein said stroke is an ischemic stroke.
61. The composition of claim 59, wherein said stroke is a hemorrhagic stroke.
- 15 62. The composition of claim 51, wherein said composition is capable of converting at least one glial cell to a neuron.
63. The composition of claim 62, wherein said glial cells are selected from the group consisting of astrocytes and NG2 cells.
64. The composition of claim 62, wherein said glial cells are astrocytes.
- 20 65. The composition of claim 62, wherein said astrocytes are reactive astrocytes.
66. The composition of claim 62, wherein said glial cells are GFAP positive.
67. The composition of claim 62, wherein said neurons are functional neurons.
68. The composition of claim 62, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and
- 25 peptidergic neurons.

69. The composition of claim 68, wherein said functional neurons are glutamatergic neurons.
70. The composition of claim 6, wherein said composition is formulated to be delivered to a subject in need thereof.
71. The composition of claim 70, wherein said composition is formulated for local delivery.
- 5 72. The composition of claim 70, wherein said composition is formulated for systemic delivery.
73. The composition of any one of claims 70-72, wherein said composition is formulated for delivery via intraperitoneal, intramuscular, intravenous, intrathecal, intracerebral, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-
10 subretina, intraparenchymal, intranasal, or oral administration.
74. A method comprising delivering the composition of claim 6 to said subject in need thereof.
75. The method of claim 74, wherein said composition is formulated to be delivered to a subject in need thereof.
- 15 76. The method of claim 74, wherein said delivering comprises local administration.
77. The method of claim 74, wherein said delivering comprises systemic administration.
78. The method of any one of claims 74-77, wherein said delivering comprises an intraperitoneal, intramuscular, intravenous, intrathecal, intracerebral, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina,
20 intraparenchymal, intranasal, or oral administration.
79. A method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the
25 nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO:

16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

- 5 (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;
- (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- 10 (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- 15 (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

80. A method of converting reactive astrocytes to functional neurons in a brain of a living human comprising: injecting an adeno-associated virus (AAV) into a subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a nucleic acid coding sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and hDlx2 coding sequence are operably linked to expression control elements comprising:

- (a) a human glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence selected from the group consisting of SEQ ID NOs: 4, 12, and 26;

- (b) an enhancer from the human elongation factor-1 alpha (EF-1 alpha) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

81. A method of converting glial cells to neurons in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject in need thereof, wherein said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and a distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and Dlx2 sequence are operably linked to expression control elements comprising:

- (a) a glial fibrillary acid protein (GFAP) promoter;
- (b) an enhancer;
- (c) a chimeric intron;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
- (e) and a polyadenylation signal sequence,

wherein said AAV vector is capable of converting at least one glial cell to a neuron in said subject in need thereof.

82. A method of treating a neurological condition in a subject in need thereof comprising: delivering an adeno-associated virus (AAV) to said subject, wherein said AAV comprises a DNA vector construct comprising a neurogenic differentiation 1 (NeuroD1) sequence and distal-less homeobox 2 (Dlx2) sequence, wherein said NeuroD1 sequence and Dlx2 sequence are separated by a linker sequence, wherein said NeuroD1 sequence and said Dlx2 sequence are operably linked to expression control elements comprising:

(a) a glial fibrillary acid protein (GFAP) promoter;

(b) an enhancer;

- (c) a chimeric intron;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE); and
- (e) a polyadenylation signal to said subject in need thereof.

83. The method of any one of claims 79-82, wherein said AAV is selected from the group
5 consisting of AAV serotype 2, AAV serotype 5, and AAV serotype 9.
84. The method of claim 83, wherein said AAV is AAV serotype 2.
85. The method of claim 83, wherein said AAV is AAV serotype 5.
86. The method of claim 83, wherein said AAV is AAV serotype 9.
87. The method of claims 79 or 80, wherein said functional neurons are glutamatergic
10 neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic
neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons.
88. The method of claims 81 or 82, wherein said NeuroD1 is human NeuroD1 (hNeuroD1).
89. The method of claims 81 or 82 wherein said Dlx2 is human Dlx2 (hDlx2).
90. The method of claims 81 or 82, wherein said NeuroD1 is selected from the group
15 consisting of a chimpanzee NeuroD1, a bonobo NeuroD1, an orangutan NeuroD1, a
gorilla NeuroD1, a macaque NeuroD1, a marmoset NeuroD1, a capuchin NeuroD1, a
baboon NeuroD1, a gibbon NeuroD1, and a lemur NeuroD1.
91. The method of claims 75 or 76, wherein said Dlx2 is selected from the group consisting
20 of a chimpanzee Dlx2, a bonobo Dlx2, an orangutan Dlx2, a gorilla Dlx2, a macaque
Dlx2, a marmoset Dlx2, a capuchin Dlx2, a baboon Dlx2, a gibbon Dlx2, and a lemur
Dlx2.
92. The method of claim 88, wherein said hNeuroD1 comprises an amino acid sequence
encoding an amino acid coding sequence at least 80% identical or similar to SEQ ID NO:
10.
93. The method of claim 89, said hDlx2 comprises a amino acid sequence encoding an
25 amino acid sequence at least 80% identical or similar to SEQ ID NO: 14.
94. The method of claim 88, wherein said hNeuroD1 coding sequence comprises a nucleic
acid sequence at least 80% identical to SEQ ID NO: 6, or the complement thereof.

95. The method of claim 89, said hDlx2 coding sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof.
96. The method of claims 81 or 82, wherein said GFAP promoter is a human GFAP (hGFAP) promoter.
- 5 97. The method of claims 81 or 82, wherein said GFAP promoter is selected from the group consisting of a chimpanzee GFAP promoter, a bonobo GFAP promoter, an orangutan GFAP promoter, a gorilla GFAP promoter, a macaque GFAP promoter, a marmoset GFAP promoter, a capuchin GFAP promoter, a baboon GFAP promoter, a gibbon GFAP promoter, and a lemur GFAP promoter.
- 10 98. The method of any one of claims 79-97, wherein said IRES sequence comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 3, or the complement thereof.
99. The method of claim 96, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 4, or the complement thereof.
- 15 100. The method of claim 96, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 12, or the complement thereof.
101. The method of claim 96, wherein said hGFAP promoter comprises a nucleic acid sequence at least 80% identical to SEQ ID NOs: 26, or the complement thereof.
102. The method of claims 81 or 82, wherein said linker is selected from the group
20 consisting of P2A and T2A.
103. The method of claim 102, wherein said P2A linker comprises a nucleic acid sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 15 and 18, or the complement thereof.
104. The method of claim 102, wherein said T2A linker comprises a nucleic acid
25 sequence at least 80% identical to the sequence selected from the group consisting of SEQ ID NO: 16 and 19, or the complement thereof.
105. The method of claims 81 or 82, wherein said enhancer is selected from the group consisting of an enhancer from human elongation factor-1 alpha (EF1- α) promoter and cytomegalovirus (CMV) enhancer.

106. The method of claim 105, wherein said EF1- α comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 2, or the complement thereof.
107. The method of claim 105, wherein said CMV enhancer comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 11, or the complement thereof.
- 5 108. The method of claims 81 or 82, wherein said chimeric intron comprises a nucleic acid sequence at least 80% identical to nucleic acid selected from the group consisting of SEQ ID NOs: 5 and 27, or the complement thereof.
109. The method of claims 81 or 82, wherein said WPRE comprises a nucleic acid sequence at least 80% identical to a nucleic acid selected from the group consisting of
10 SEQ ID NOs: 7 and 29, or the complement thereof.
110. The method of claims 81 or 82, wherein said polyadenylated signal selected from the group consisting of SV40 polyadenylation signal and a hGH polyadenylation signal
111. The method of claim 110, wherein said SV40 polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 8, or the complement
15 thereof.
112. The method of claim 110, wherein said hGH polyadenylated signal comprises a nucleic acid sequence at least 80% identical to SEQ ID NO: 13, or the complement thereof.
113. The method of claims 81 or 82, wherein said vector further comprises a nucleic acid sequence encoding an AAV protein sequence.
20
114. The method of any one of claims 79-82, wherein said vector comprises AAV serotype 2 inverted terminal repeats (ITRs).
115. The method of any one of claims 79-82, wherein said vector comprises AAV serotype 5 inverted terminal repeats (ITRs).
- 25 116. The method of any one of claims 79-82, wherein said vector comprises AAV serotype 9 inverted terminal repeats (ITRs).
117. The method of any one of claims 79-82, wherein said vector comprises at least one ITR nucleic acid sequence at least 80% identical to SEQ ID NO: 1.

118. The method of any one of claims 79-82, wherein said vector comprises at least one ITR nucleic acid sequence at least 80% identical to SEQ ID NO: 9.
119. The method of claim 81, wherein said converting occurs in the central nervous system (CNS) or peripheral nervous system.
- 5 120. The method of claim 81, wherein said converting occurs in the CNS.
121. The method of claim 81 or 82, wherein said subject in need thereof is a mammal.
122. The method of claim 121, wherein said mammal is a human.
123. The method of claim 121, wherein said mammal is a non-human primate.
124. The method of claim 81 or 82, wherein said delivering comprises a local
10 administration.
125. The method of claim 81 or 82, wherein said delivering comprises systemic administration.
126. The method of claim 81 or 82, wherein said delivering comprises an
15 administration selected from the group consisting of an intraperitoneal administration, intramuscular administration, intravenous administration, intrathecal administration, intracerebral administration, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal administration, intranasal administration, and oral administration.
127. The method of claim 79 or 80, wherein said injecting comprises an injection
20 selected from the group consisting of an intraperitoneal injection, intramuscular injection, intravenous injection, intrathecal injection, intracerebral injection, intracranial, intra lateral ventricle of the brain, intra cisterna magna, intra vitreous, intra-subretina, intraparenchymal injection, intranasal injection, and oral injection.
128. The method of claims 81 or 82, wherein said delivering comprises injecting.
129. The method of any one of claims 79, 80 or 128, wherein said injecting is
25 performed at a concentration of between 10^{10} particles/mL and 10^{14} particles/mL.
130. The method of claim 129, wherein said injecting further comprises a flow rate of between 0.1 μ L/minute and 5.0 μ L/minute.

131. The method of claim 81, wherein said at least one glial cell is selected from the group consisting of at least one astrocyte and at least one NG2 cell.
132. The method of claim 131, wherein said at least one glial cell is at least one astrocyte.
- 5 133. The method of claim 131 or 132, wherein said at least one astrocyte is a reactive astrocyte.
134. The method of claim 81, wherein said neuron is a functional neuron.
135. The method of any one of claims 79, 80 and 134, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons,
10 dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons..
136. The method of claim 81, wherein said subject exhibits an improvement of at least one neurological condition symptom as compared to said subject prior to said delivering.
137. The method of claim 136, wherein said improvement is measured within 1 year
15 of said delivering.
138. The method of any one of claims 79, 80, or 128, wherein said method comprises directly injecting said AAV into the brain of said subject.
139. The method of any one of claims 79 or 80 wherein said wherein said converting is in the striatum of said brain.
- 20 140. The method of any one of claims 79, 80, or 128, wherein said method comprises directly injecting said AAV into the spinal cord of said subject.
141. The method of claim 82, wherein said neurological condition comprises an injury to the central nervous system (CNS) or peripheral nervous system.
- 25 142. The method of claim 82, wherein said neurological condition is selected from the group consisting of Alzheimer's Disease, Parkinson's Disease, amyotrophic lateral sclerosis (ALS), Huntington's Disease, epilepsy, physical injury, stroke, cerebral aneurysm, traumatic brain injury, concussion, a tumor, inflammation, infection, ataxia, brain atrophy, spinal cord atrophy, multiple sclerosis, traumatic spinal cord injury, ischemic or hemorrhagic myelopathy (myelopathy), global ischemia, hypoxic ischemic

encephalopathy, embolism, fibrocartilage embolism myelopathy, thrombosis, nephropathy, chronic inflammatory disease, meningitis, and cerebral venous sinus thrombosis.

- 5 143. The method of claim 82, wherein said neurological condition is Alzheimer's Disease.
144. The method of claim 82, wherein said neurological condition is Parkinson's Disease.
145. The method of claim 82, wherein said neurological condition is ALS.
- 10 146. The method of claim 82, wherein said neurological condition is Huntington's Disease.
147. The method of claim 82, wherein said neurological condition is a stroke.
148. The method of claim 147, wherein said stroke is an ischemic stroke.
149. The method of claim 147, wherein said stroke is a hemorrhagic stroke.
- 15 150. The method of claim 82, wherein said method is capable of converting at least one glial cell into a neuron.
151. The method of claim 150, wherein said glial cells are selected from the group consisting of astrocytes and NG2 cells.
152. The method of claim 150, wherein said glial cells are astrocytes.
153. The method of claim 152, wherein said astrocytes are reactive astrocytes.
- 20 154. The method of claim 150, wherein said glial cells are GFAP positive.
155. The method of claim 150, wherein said neurons are functional neurons.
- 25 156. The method of claim 145, wherein said functional neurons are selected from the group consisting of glutamatergic neurons, GABAergic neurons, dopaminergic neurons, cholinergic neurons, serotonergic neurons, epinephrinergic neurons, motor neurons, and peptidergic neurons.

157. The method of claims 79 or 80, wherein a therapeutically effective dose of said AAV is injected into said subject.
158. The method of claims 81 or 82, wherein a therapeutically effective dose of said AAV is delivered to said subject.
- 5 159. The method of claim 147 or 148, wherein said therapeutically effective dose is administered with a pharmaceutically acceptable carrier.
160. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6, and (ii) an adeno-associated virus (AAV) vector comprising
10 a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13;
wherein the hNeuroD1 sequence is operably linked to regulatory elements comprising:
(a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
15 (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;
(c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;
(d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE)
20 comprising the nucleic acid sequence of SEQ ID NO: 29; and
(e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
161. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid
25 sequence of SEQ ID NO: 6, and (ii) an adeno-associated virus (AAV) vector comprising a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13;
wherein the hNeuroD1 sequence is operably linked to regulatory elements comprising:
(a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid
30 sequence of SEQ ID NO: 26;
(b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;

- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
 - 5 (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
162. The composition of claim 160 or 161, wherein (ii) comprises an AAV vector comprising the hDlx2 sequence comprising the nucleic acid sequence of SEQ ID NO: 13 operably linked to regulatory elements comprising:
- 10 (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence of SEQ ID NO: 26;
 - (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - 15 (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;
 - (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
 - 20 (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
163. A composition comprising (i) an adeno-associated virus (AAV) vector comprising
- 25 a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10, and (ii) an adeno-associated virus (AAV) vector comprising a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14;
- 30 wherein the nucleic acid sequence encoding an hNeuroD1 protein is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
 - (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising
 - 35 the nucleic acid sequence of SEQ ID NO: 2;

- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- 5 (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
164. A composition comprising (i) an adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10, and (ii) an adeno-associated virus (AAV) vector comprising a nucleic acid coding sequence encoding a
- 10 human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14;
- wherein the nucleic acid sequencing encoding an hNeuroD1 protein is operably linked to regulatory elements comprising:
- 15 (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ
- 20 ID NO: 27;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30
- 25 165. The composition of claim 163 or 164, wherein (ii) comprises an AAV vector comprising a nucleic acid coding sequence encoding the hDlx2 protein, wherein the nucleic acid is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising a nucleic acid sequence of SEQ ID NO: 26;
- 30 (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2 or a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 5 and 27;

- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence selected from the group consisting of SEQ ID NOs: 7 and 29; and
- (e) a SV40 polyadenylation signal sequence comprising the nucleic acid sequence of SEQ ID NO: 8, a hGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 17, or a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
166. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5 or SEQ ID NO: 5;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.
167. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ

ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- 5 (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- 10 (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

168. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence
15 encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO:
20 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- 25 (b) an enhancer from a human elongation factor-1 alpha (EF1- α) promoter comprising the nucleic acid sequence of SEQ ID NO: 2;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5;
- (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- 30 (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

169. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the

amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by (i) a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18, (ii) a T2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 16 and 19, or (iii) an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:

- 10 (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5;
- 15 (d) a woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) comprising the nucleic acid sequence of SEQ ID NO: 29; and
- (e) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

170. An adeno-associated virus (AAV) vector comprising a human neurogenic differentiation 1 (hNeuroD1) sequence comprising the nucleic acid sequence of SEQ ID NO: 6 and a human distal-less homeobox 2 (hDlx2) sequence comprising the nucleic acid sequence of SEQ ID NO: 13, wherein said hNeuroD1 sequence and said hDlx2 sequence are separated by a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18 or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 sequence and said hDlx2 sequence are operably linked to regulatory elements comprising:

- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
- 30 (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
- (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5; and
- (d) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.

171. An adeno-associated virus (AAV) vector comprising a nucleic acid sequence encoding a human neurogenic differentiation 1 (hNeuroD1) protein comprising the amino acid coding sequence of SEQ ID NO: 10 and a nucleic acid coding sequence encoding a human distal-less homeobox 2 (hDlx2) protein comprising the amino acid sequence of SEQ ID NO: 14, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence are separated by a P2A linker comprising the nucleic acid sequence selected from the group consisting of SEQ ID NO: 15 and 18 or an internal ribosomal entry site of the encephalomyocarditis virus (IRES) sequence comprising SEQ ID NO: 3, wherein said hNeuroD1 coding sequence and said hDlx2 coding sequence is operably linked to regulatory elements comprising:
- (a) a glial fibrillary acidic protein (GFAP) promoter comprising the nucleic acid sequence of SEQ ID NO: 26;
 - (b) a cytomegalovirus (CMV) enhancer comprising the nucleic acid sequence of SEQ ID NO: 11;
 - (c) a chimeric intron comprising the nucleic acid sequence of SEQ ID NO: 5; and
 - (d) a bGH polyadenylation sequence comprising the nucleic acid sequence of SEQ ID NO: 30.