Claims

1. A one-vector expression system comprising:
- a first and a second expression cassette,

wherein said first expression cassette comprises
a nucleotide sequence comprising
a first promoter sequence operably linked to
a first nucleotide sequence,
said first nucleotide sequence encoding a GTP-cyclohydrolase 1 (GCH1; EC 3.5.4.16) polypeptide or a biologically active fragment or variant thereof, and

wherein said second expression cassette comprises
a nucleotide sequence comprising
a second promoter sequence operably linked to
a second nucleotide sequence
said second nucleotide sequence encoding a tyrosine hydroxylase (TH; EC 1.14.16.2) polypeptide or a biologically active fragment or variant thereof, and

wherein the vector is an adeno associated vector (AAV) and

wherein said second nucleotide sequence is operably linked to a Woodchuck hepatitis virus post-transcriptional regulatory element (WPRE), and

wherein both said first and said second promoter are Synapsin1 promoters.

2. The vector of claim 1 wherein the GTP-cyclohydrolase 1 (GCH1; EC 3.5.4.16) polypeptide or the biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, and wherein said tyrosine hydroxylase (TH; EC 1.14.16.2) polypeptide or a biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the

group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12, SEQ ID NO. 13 and SEQ ID NO. 14.

3. The vector of claim 1, wherein said vector does not comprise a nucleotide sequence encoding an aromatic amino acid decarboxylase (AADC) polypeptide.

4. The vector of claim 1, wherein said vector has a packaging capacity from 1 to 40 kb, for example from 1 to 30 kb, such as from 1 to 20 kb, for example from 1 to 15 kb, such as from 1 to 10, for example from 1 to 8 kb, such as from 2 to 7 kb, for example from 3 to 6 kb, such as from 4 to 5 kb.

5. The vector of claim 1, wherein said vector has a packaging capacity from 4.5 to 4.8 kb.

6. The vector of claim 1, wherein the adeno associated vector (AAV) is an AAV2 vector.

7. The vector of claim 6 wherein the capsid of the AAV2 vector is packaged in an AAV capsid other than an AAV2 capsid.

8. The vector of claim 7 wherein the AAV2 vector is packaged in an AAV5 capsid.

9. The vector of any of claims 1 to 8, wherein said vector is functional in mammalian cells.

10. The vector of claim 1, wherein said biologically active fragment comprises at least 50 contiguous amino acids, wherein any amino acid specified in the selected sequence is altered to a different amino acid, provided that no more than 15 of the amino acid residues in the sequence are so altered.

11. The vector of claim 10, wherein said biologically active fragment is the catalytic domain of tyrosine hydroxylase.

12. The vector of claim 1, wherein said biologically active variant is a mutated tyrosine hydroxylase polypeptide, wherein one or more of the residues S19, S31, S40 or S404 have been altered to another amino acid residue.

13. The vector of any of claims 1 to 12, wherein said GTP-cyclohydrolase 1 (GCH1) polypeptide is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 75% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 80% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 85% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 90% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 95% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 96% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 97% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 98% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 99% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably 100% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6.

14. The vector of any of claims 1 to 13, wherein said tyrosine hydroxylase (TH) polypeptide is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ

ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 75% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 80% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 85% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 90% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9. SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 95% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 96% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 97% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 98% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9. SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 99% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably 100% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14.

15. The vector of claim 1, wherein said first expression cassette or said second expression cassette further comprises a polyadenylation sequence.

16. The vector of claim 1, wherein said first expression cassette and said second expression cassette further comprises polyadenylation sequences.

17. The vector of any of claims 15 or 16, wherein said polyadenylation sequence is a SV40 polyadenylation sequence.

18. The vector of any of claims 15 to 17, wherein the 5' of said polyadenylation sequence is operably linked to the 3' of said first and/or said second nucleotide sequence.

19. The vector of claim 1, wherein said first nucleotide sequence encoding a GTP-cyclohydrolase 1 (GCH1) polypeptide or a biologically active fragment or variant thereof comprises the sequence of SEQ ID NO. 18.

20. The vector of claim 1, wherein said second nucleotide sequence encoding a tyrosine hydroxylase (TH) polypeptide or a biologically active fragment or variant thereof comprises the sequence of SEQ ID NO. 21.

21. The vector of claim 1, wherein said Woodchuck hepatitis virus post-transcriptional regulatory element comprises a sequence that has at least 85 percent sequence identity, preferably at least 90 to 95 percent sequence identity, more preferably at least 99 percent sequence identity to SEQ ID NO: 22.

22. The vector of claim 1, wherein said Woodchuck hepatitis virus post-transcriptional regulatory element comprises the sequence of SEQ ID NO. 22.

23. The vector of any of claims 1 to 22 wherein the expression cassettes of said vector comprises a 5' terminal repeat and a 3' terminal repeat.

24. The vector of claim 23, wherein the 5' and 3' terminal repeats are selected from Inverted Terminal Repeats [ITR] and Long Terminal Repeats [LTR].

25. The vector of claim 23, wherein the 5' and 3' terminal repeats are AAV Inverted Terminal Repeats [ITR].

26. The vector of claim 25, wherein said Inverted Terminal Repeats comprises the sequences of SEQ ID NO. 15 and SEQ ID NO. 16.

27. The vector of any of claims 1 to 26 wherein the TH:GCH1 ratio is at least 3:1, such as at least 4:1, for example at least 5:1, such as at least 6:1, for example at least 7:1, such as at least 10:1, for example 15:1, such as 20:1, for example 25:1, such as 30:1, for example 35:1, such as 40:1, for example 45:1, such as 50:1.

28. The vector of any of claims 1 to 27 wherein the TH:GCH1 ratio is 7:1.

29. The vector of any of claims 27 to 28 wherein the ratio is determined by measuring the activity of the expressed TH and GCH1 enzymes.

30. The vector of any of claims 27 to 28 wherein the ratio is determined by measuring the amount of Tetrahydrobiopterin (BH_4).

31. The vector of any of claims 27 to 28 wherein the ratio is determined by the amount of mRNA transcribed.

32. The vector of any of claims 27 to 28 wherein the ratio is determined by the amount of protein expressed.

33. The vector according to any of the proceeding claims wherein said vector is the vector defined in figure 6.

34. The vector of any of claims 1 to 33 wherein said vector is a minimally integrating vector.

35. The vector of any of the claims 1 to 34, for use as a medicament.

36. An isolated host cell comprising the vector of any of claims 1 to 35.

37. The host cell of claim 36, wherein said cell is selected from the group consisting of eukaryotic cells, preferably mammalian cells, more preferably primate cells, more preferably human cells.

38. The host cell of claim 37, wherein the cell is selected from the group consisting of Chinese hamster ovary cells, CHO-K1, baby hamster kidney cells, mouse

fibroblast-3T3 cells, African green monkey cell lines, rat adrenal pheochromocytoma, AT3 cells, rat glial tumor cells, rat neuronal cells and rat hippocampal cells.

39. Use of the vector of any of claims 1 to 35, for the preparation of a medicament for the treatment of a disease associated with catecholamine dysfunction.

40. The use of claim 39, wherein the catecholamine dysfunction is catecholamine deficiency.

41. The use of claim 39 wherein the catecholamine dysfunction is catecholamine excess.

42. The use of claim 40, wherein the catecholamine deficiency is dopamine deficiency.

43. The use of claim 41, wherein the catecholamine excess is dopamine excess.

44. The use of any of claims 39 to 43 wherein said disease associated with catecholamine dysfunction is a disease, disorder or damage of the central and/or peripheral nervous system.

45. The use of claim 44, wherein said disease, disorder or damage of the central and/or peripheral nervous system is a neurodegenerative disorder.

46. The use of any of claims 39 to 45 wherein said disease associated with catecholamine dysfunction is a disease of the basal ganglia.

47. The use of any of claims 39 to 46, wherein said disease is selected from the group consisting of Parkinson's Disease (PD), DOPA responsive dystonia, ADHD, schizophrenia, depression, vascular parkinsonism, essential tremor, chronic stress, genetic dopamine receptor abnormalities, chronic opoid, cocaine, alcohol or marijuana use, adrenal insufficiency, hypertension, noradrenaline deficiency, post-traumatic stress disorder, pathological gambling disorder, dementia, Lewy body dementia.

48. The use of any of claims 39 to 46, wherein said neurodegenerative disorder is Parkinson's Disease (PD).

49. The vector of any of claims 1 to 35, for use in a method of treatment of a disease associated with catecholamine dysfunction.

50. The vector of claim 49, wherein the catecholamine dysfunction is catecholamine deficiency.

51. The vector of claim 49 wherein the catecholamine dysfunction is catecholamine excess.

52. The vector of claim 50, wherein the catecholamine deficiency is dopamine deficiency.

53. The vector of claim 51, wherein the catecholamine excess is dopamine excess.

54. The vector of any of claims 49 to 53 wherein said disease associated with catecholamine dysfunction is a disease, disorder or damage of the central and/or peripheral nervous system.

55. The vector of claim 54, wherein said disease, disorder or damage of the central and/or peripheral nervous system is a neurodegenerative disorder.

56. The vector of any of claims 49 to 55 wherein said disease associated with catecholamine dysfunction is a disease of the basal ganglia.

57. The vector of any of claims 49 to 55, wherein said disease is selected from the group consisting of Parkinson's Disease (PD), DOPA responsive dystonia, ADHD, schizophrenia, depression, vascular parkinsonism, essential tremor, chronic stress, genetic dopamine receptor abnormalities, chronic opoid, cocaine, alcohol or marijuana use, adrenal insufficiency, hypertension, noradrenaline deficiency, post-traumatic stress disorder, pathological gambling disorder, dementia, Lewy body dementia.

58. The vector of any of claims 49 to 56, wherein said neurodegenerative disorder is Parkinson's Disease (PD).

59. A method of treating a disease associated with catecholamine dysfunction, in a patient in need thereof, said method comprising administering to said individual the vector of any of claims 1 to 35.

60. The method of claim 59, wherein the catecholamine dysfunction is catecholamine deficiency.

61. The method of claim 59 wherein the catecholamine dysfunction is catecholamine excess.

62. The method of claim 60, wherein the catecholamine deficiency is dopamine deficiency.

63. The method of claim 61, wherein the catecholamine excess is dopamine excess.

64. The method of any of claims 59 to 63 wherein said disease associated with catecholamine dysfunction is a disease, disorder or damage of the central and/or peripheral nervous system.

65. The method of claim 64, wherein said disease, disorder or damage of the central and/or peripheral nervous system is a neurodegenerative disorder.

66. The method of any of claims 59 to 65 wherein said disease associated with catecholamine dysfunction is a disease of the basal ganglia.

67. The method of any of claims 59 to 66, wherein said disease is selected from the group consisting of Parkinson's Disease (PD), DOPA responsive dystonia, ADHD, schizophrenia, depression, vascular parkinsonism, essential tremor, chronic stress, genetic dopamine receptor abnormalities, chronic opoid, cocaine, alcohol or marijuana use, adrenal insufficiency, hypertension, noradrenaline deficiency, post-traumatic stress disorder, pathological gambling disorder, dementia, Lewy body dementia.

68. The method of any of claims 59 to 66, wherein said neurocegenerative disorder is Parkinson's Disease (PD).

69. A pharmaceutical composition for use in a method of treatment of Parkinson's Disease, said composition comprising a one-vector expression system and a formulation for delivering said vector to the basal ganglia, wherein said one-vector expression system comprises:

- a first and a second expression cassette,

wherein said first expression cassette comprises
a nucleotide sequence comprising
a first promoter sequence operably linked to
a first nucleotide sequence,
said first nucleotide sequence encoding a GTP-cyclohydrolase 1 (GCH1; EC 3.5.4.16) polypeptide or a biologically active fragment or variant thereof, and

wherein said second expression cassette comprises
a nucleotide sequence comprising
a second promoter sequence operably linked to
a second nucleotide sequence
said second nucleotide sequence encoding a tyrosine hydroxylase (TH; EC 1.14.16.2) polypeptide or a biologically active fragment or variant thereof, and

wherein said second nucleotide sequence is operably linked to a Woodchuck hepatitis virus post-transcriptional regulatory element (WPRE), and

wherein the vector is an adeno associated vector (AAV) and

wherein both said first and said second promoter are Synapsin1 promoters.

70. The composition of claim 69 wherein the GTP-cyclohydrolase 1 (GCH1) polypeptide or the biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, and wherein said tyrosine hydroxylase (TH) polypeptide or a biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12, SEQ ID NO. 13 and SEQ ID NO. 14.

71. The pharmaceutical composition of any of claims 69 to 70, wherein the formulation comprises manitol, heparin or gadolinium based MRI contrast agents.

72. The pharmaceutical composition of any of claims 69 to 71 further comprising trophic factors or reversible proteasome inhibitors.

73. The pharmaceutical composition of any of claims 69 to 72, and a pharmaceutically acceptable carrier or diluent.

74. The pharmaceutical composition of any of claims 69 to 73, formulated for administration by injection, sublingual tablet or spray, cutaneous administration or inhalation.

75. The pharmaceutical composition of claim 74, wherein the injection is intracranial, intracerebral, intravenous, intravitreous, intranasal, intramuscular, intraspinal, intraperitoneal, subcutaneous, a bolus or a continuous administration.

76. The pharmaceutical composition according to any of claims 69 to 75, wherein the pH of the composition is between pH 4 and pH 10.

77. The pharmaceutical composition according to any of claims 69 to 76, wherein the dosage of the vector is between 1.5E+10 and 2.2E+12 vector genome copies per milliliter putaminal grey matter.

78. The pharmaceutical composition according to any of claims 69 to 76, wherein administration occurs at intervals of 30 minutes to 24 hours.

79. The pharmaceutical composition according to any of claims 69 to 76, wherein administration occurs at intervals of 1 to 6 hours.

80. The pharmaceutical composition according to any of claims 69 to 76, wherein the duration of the treatment is from 6 to 72 hours.

81. The pharmaceutical composition according to any of claims 69 to 76, wherein the duration of the treatment is life long.

82. A method for determining the expression ratio of GTP cyclohydroxylase1 polypeptide (SEQ ID NOs. 1, 2, 3, 4, 5 and 6) and tyrosine hydroxylase polypeptide (SEQ ID NOs. 7, 8, 9, 10, 11, 12, 13 and 14), respectively, or variants thereof, said polypeptide encoded by the vector as defined in any of claims 1 to 34, comprising measuring the:
    a. enzymatic activity of the polypeptides, or
    b. amount of BH_4 produced, or
    c. amount expressed of said polypeptides, or
    d. level of transcribed mRNA corresponding to the polypeptides of GTP cyclohydroxylase1 polypeptide and tyrosine hydroxylase, respectively.

83. The method of claim 82, wherein the ratio is determined by a combination of the activity of said polypeptides and the amount present of said polypeptides.

84. A kit in parts, said kit comprising the pharmaceutical composition of any of claims 69 to 83, and instructions for administration.

CLAIMS

1. A one-vector expression system comprising a first and a second expression cassette, in which the first first expression cassette comprises a nucleotide sequence comprising a first promoter sequence operably linked to a first nucleotide sequence, the first nucleotide sequence encoding a GTP-cyclohydrolase 1 (GCH1; EC 3.5.4.16) polypeptide or a biologically active fragment or variant thereof, and in which the second expression cassette comprises a nucleotide sequence comprising a second promoter sequence operably linked to a second nucleotide sequence the second nucleotide sequence encoding a tyrosine hydroxylase (TH; EC 1.14.16.2) polypeptide or a biologically active fragment or variant thereof, and in which the vector is an adeno associated vector (AAV) and in which the vector does not comprise a nucleotide sequence encoding an aromatic amino acid decarboxylase (AADC) polypeptide, and in which the TH:GCH1 ratio expressed from the vector is between 3:1 and 10:1.

2. A vector according to Claim 1, in which the first promoter is a Synapsin 1 promoter.

3. A vector according to Claim 1 or 2, in which the second promoter is a constitutively active promoter selected from the group consisting of CAG, CMV, human UbiC, RSV, EF-1alpha, SV40 and Mt1 promoters.

4. A vector according to Claim 1 or 2, in which the second promoter is a CMV promoter.

5. A vector according to any preceding claim, in which the vector further comprises one or more enhancers or regulatory elements.

6. A vector according to Claim 6, in which the enhancer or regulatory element is an intron.

7. A vector according to Claim 1, in which the GTP-cyclohydrolase 1 (GCH1; EC 3.5.4.16) polypeptide or the biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, and in which the tyrosine hydroxylase (TH; EC 1.14.16.2) polypeptide or a biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12, SEQ ID NO. 13 and SEQ ID NO. 14.

8. A vector according to Claim 1, in which the vector has a packaging capacity from 1 to 40 kb, for example from 1 to 30 kb, such as from 1 to 20 kb, for example from 1 to 15 kb, such as from 1 to 10, for example from 1 to 8 kb, such as from 2 to 7 kb, for example from 3 to 6 kb, such as from 4 to 5 kb.

9. A vector according to Claim 1, in which the vector has a packaging capacity from 4.5 to 4.8 kb.

10. A vector according to Claim 1, in which the adeno associated vector (AAV) is an AAV2 vector.

11. A vector according to Claim 10, in which the capsid of the AAV2 vector is packaged in an AAV capsid other than an AAV2 capsid.

12. A vector according to Claim 11, in which the AAV2 vector is packaged in an AAV5 capsid.

13. A vector according to any preceding claim, in which the vector is functional in mammalian cells.

14. A vector according to Claim 1, in which the biologically active fragment comprises at least 50 contiguous amino acids, in which any amino acid specified in the selected sequence is altered to a different amino acid, provided that no more than 15 of the amino acid residues in the sequence are so altered.

15. A vector according to Claim 14, in which the biologically active fragment is the catalytic domain of tyrosine hydroxylase.

16. A vector according to Claim 1, in which the biologically active variant is a mutated tyrosine hydroxylase polypeptide, in which one or more of the residues S19, S31, S40 or S404 have been altered to another amino acid residue.

17. A vector according to any preceding claim, in which the GTP-cyclohydrolase 1 (GCH1) polypeptide is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 75% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 80% identical to a polypeptide selected from the

group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 85% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 90% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 95% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 96% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 97% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 98% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably at least 99% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, more preferably 100% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6.

18. A vector according to any preceding claim, in which the tyrosine hydroxylase (TH) polypeptide is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 75% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 80% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 85% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 90% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 95% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID

NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 96% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 97% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 98% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably at least 99% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14, more preferably 100% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12 SEQ ID NO. 13 and SEQ ID NO. 14.

19. A vector according to Claim 1, in which the first expression cassette or the second expression cassette further comprises a polyadenylation sequence.

20. A vector according to Claim 1, in which the first expression cassette and the second expression cassette further comprises polyadenylation sequences.

21. A vector according to Claim 19 or 20, in which the polyadenylation sequence is a SV40 polyadenylation sequence.

22. A vector according to Claim 19 or 20, in which the 5’ of the polyadenylation sequence is operably linked to the 3’ of the first and/or the second nucleotide sequence.

23. A vector according to Claim 1, in which the first nucleotide sequence encoding a GTP-cyclohydrolase 1 (GCH1) polypeptide or a biologically active fragment or variant thereof comprises the sequence of SEQ ID NO. 18.

24. A vector according to Claim 1, in which the second nucleotide sequence encoding a tyrosine hydroxylase (TH) polypeptide or a biologically active fragment or variant thereof comprises the sequence of SEQ ID NO. 21.

25. A vector according to Claim 1, in which the second nucleotide is operably liked to a Woodchuck hepatitis virus post-transcriptional regulatory element (WPRE).

26. A vector according to Claim 25, in which the Woodchuck hepatitis virus post-transcriptional regulatory element comprises a sequence that has at least 85 percent sequence identity, preferably at least 90 to 95 percent sequence identity, more preferably at least 99 percent sequence identity to SEQ ID NO: 22.

27. A vector according to Claim 1, in which the Woodchuck hepatitis virus post-transcriptional regulatory element comprises the sequence of SEQ ID NO. 22.

28. A vector according to any preceding claim, in which the expression cassettes of the vector comprises a 5' terminal repeat and a 3' terminal repeat.

29. A vector according to Claim 28, in which the 5' and 3' terminal repeats are selected from Inverted Terminal Repeats [ITR] and Long Terminal Repeats [LTR].

30. A vector according to Claim 28, in which the 5' and 3' terminal repeats are AAV Inverted Terminal Repeats [ITR].

31. A vector according to Claim 30, in which the Inverted Terminal Repeats comprises the sequences of SEQ ID NO. 15 and SEQ ID NO. 16.

32. A vector according to any preceding claim, in which the TH:GCH1 ratio is determined by measuring the activity of the expressed TH and GCH1 enzymes.

33. A vector according to any preceding claim, in which the TH:GCH1 ratio is determined by measuring the amount of Tetrahydrobiopterin (BH_4).

34. A vector according to any preceding claim, in which the TH:GCH1 ratio is determined by the amount of mRNA transcribed.

35. A vector according to any preceding claim, in which the TH:GCH1 ratio is determined by the amount of protein expressed.

36. A vector according to any preceding claim, in which the vector is the vector defined in Figure 6.

37. A vector according to any preceding claim, in which the vector is a minimally integrating vector.

38. A vector according to any preceding claim for use as a medicament.

39. An isolated host cell comprising a vector according to any preceding claim.

40. A host cell according to Claim 39, in which the cell is selected from the group consisting of eukaryotic cells, preferably mammalian cells, more preferably primate cells, more preferably human cells.

41. A host cell according to Claim 40, in which the cell is selected from the group consisting of Chinese hamster ovary cells, CHO-K1, baby hamster kidney cells, mouse fibroblast-3T3 cells, African green monkey cell lines, rat adrenal pheochromocytoma, AT3 cells, rat glial tumor cells, rat neuronal cells and rat hippocampal cells.

42. Use of a vector according to any of Claims 1 to 38, for the preparation of a medicament for the treatment of a disease associated with catecholamine dysfunction.

43. Use according to Claim 42, in which the catecholamine dysfunction is catecholamine deficiency.

44. Use according to Claim 42, in which the catecholamine dysfunction is catecholamine excess.

45. Use according to Claim 43, in which the catecholamine deficiency is dopamine deficiency.

46. Use according to Claim 44, in which the catecholamine excess is dopamine excess.

47. Use according to any of Claims 42 to 46, in which the disease associated with catecholamine dysfunction is a disease, disorder or damage of the central and/or peripheral nervous system.

48. Use according to Claim 47, in which the disease, disorder or damage of the central and/or peripheral nervous system is a neurodegenerative disorder.

49. Use according to any of Claims 42 to 48, in which the disease associated with catecholamine dysfunction is a disease of the basal ganglia.

50. Use according to any of Claims 42 to 49, in which the disease is selected from the group consisting of Parkinson's Disease (PD), DOPA responsive dystonia, ADHD, schizophrenia, depression, vascular parkinsonism, essential tremor, chronic stress, genetic dopamine receptor abnormalities, chronic opoid, cocaine, alcohol or marijuana use, adrenal insufficiency,

hypertension, noradrenaline deficiency, post-traumatic stress disorder, pathological gambling disorder, dementia, Lewy body dementia.

51. Use according to any of Claims 42 to 49, in which the neurodegenerative disorder is Parkinson's Disease (PD).

52. A vector according to any of Claims 1 to 38, for use in a method of treatment of a disease associated with catecholamine dysfunction.

53. A vector according to Claim 52, in which the catecholamine dysfunction is catecholamine deficiency.

54. A vector according to Claim 52, in which the catecholamine dysfunction is catecholamine excess.

55. A vector according to Claim 53, in which the catecholamine deficiency is dopamine deficiency.

56. A vector according to Claim 54, in which the catecholamine excess is dopamine excess.

57. A vector according to any of Claims 52 to 56, in which the disease associated with catecholamine dysfunction is a disease, disorder or damage of the central and/or peripheral nervous system.

58. A vector according to Claim 57, in which the disease, disorder or damage of the central and/or peripheral nervous system is a neurodegenerative disorder.

59. A vector according to any of Claims 52 to 58, in which the disease associated with catecholamine dysfunction is a disease of the basal ganglia.

60. A vector according to any of Claims 52 to 58, in which the disease is selected from the group consisting of Parkinson's Disease (PD), DOPA responsive dystonia, ADHD, schizophrenia, depression, vascular parkinsonism, essential tremor, chronic stress, genetic dopamine receptor abnormalities, chronic opoid, cocaine, alcohol or marijuana use, adrenal insufficiency, hypertension, noradrenaline deficiency, post-traumatic stress disorder, pathological gambling disorder, dementia, Lewy body dementia.

61. A vector according to any of Claims 52 to 49, in which the neurodegenerative disorder is Parkinson's Disease (PD).

62. A pharmaceutical composition for use in a method of treatment of Parkinson's Disease, the composition comprising a one-vector expression system and a formulation for delivering the vector to the basal ganglia, in which the one-vector expression system comprises: a first and a second expression cassette, in which the first expression cassette comprises a nucleotide sequence comprising a first promoter sequence operably linked to a first nucleotide sequence, the first nucleotide sequence encoding a GTP-cyclohydrolase 1 (GCH1; EC 3.5.4.16) polypeptide or a biologically active fragment or variant thereof, and in which the second expression cassette comprises a nucleotide sequence comprising a second promoter sequence operably linked to a second nucleotide sequence, the second nucleotide sequence encoding a tyrosine hydroxylase (TH; EC 1.14.16.2) polypeptide or a biologically active fragment or variant thereof, and in which the vector is an adeno associated vector (AAV), and in which the vector does not comprise a nucleotide sequence encoding an aromatic amino acid decarboxylase (AADC) polypeptide and in which the TH:GCH1 ratio expressed from the vector is between 3:1 and 10:1.

63. A pharmaceutical composition according to Claim 62, in which the GTP-cyclohydrolase 1 (GCH1) polypeptide or the biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 1, SEQ ID NO. 2, SEQ ID NO. 3, SEQ ID NO. 4, SEQ ID NO. 5 and SEQ ID NO. 6, and in which the tyrosine hydroxylase (TH) polypeptide or a biologically active fragment or variant thereof is at least 70% identical to a polypeptide selected from the group consisting of SEQ ID NO. 7, SEQ ID NO. 8, SEQ ID NO. 9, SEQ ID NO. 10, SEQ ID NO. 11, SEQ ID NO. 12, SEQ ID NO. 13 and SEQ ID NO. 14.

64. A pharmaceutical composition according to any of Claims 62 to 63, in which the formulation comprises manitol, heparin or gadolinium based MRI contrast agents.

65. A pharmaceutical composition according to any of Claims 62 to 64 further comprising trophic factors or reversible proteasome inhibitors.

66. A pharmaceutical composition according to any of Claims 62 to 65, and a pharmaceutically acceptable carrier or diluent.

67. A pharmaceutical composition according to any of Claims 62 to 66, formulated for administration by injection, sublingual tablet or spray, cutaneous administration or inhalation.

68. A pharmaceutical composition according to Claim 67, in which the injection is intracranial, intracerebral, intravenous, intravitreous, intranasal, intramuscular, intraspinal, intraperitoneal, subcutaneous, a bolus or a continuous administration.

69. A pharmaceutical composition according to any of Claims 62 to 64, in which the pH of the composition is between pH 4 and pH 10.

70. A pharmaceutical composition according to any of Claims 62 to 69, in which the dosage of the vector is between 1.5E+10 and 2.2E+12 vector genome copies per milliliter putaminal grey matter.

71. A pharmaceutical composition according to any of Claims 62 to 70, in which administration occurs at intervals of 30 minutes to 24 hours.

72. A pharmaceutical composition according to any of Claims 62 to 71, in which administration occurs at intervals of 1 to 6 hours.

73. A pharmaceutical composition according to any of Claims 62 to 71, in which the duration of the treatment is from 6 to 72 hours.

74. A pharmaceutical composition according to any of Claims 62 to 71, in which the duration of the treatment is life long.

75. A method for determining the expression ratio of GTP cyclohydroxylase1 polypeptide (SEQ ID NOs. 1, 2, 3, 4, 5 and 6) and tyrosine hydroxylase polypeptide (SEQ ID NOs. 7, 8, 9, 10, 11, 12, 13 and 14), respectively, or variants thereof, the polypeptide encoded by the vector as defined in any of claims 1 to 37, comprising measuring the enzymatic activity of the polypeptides, or amount of BH_4 produced, or amount expressed of the polypeptides, or level of transcribed mRNA corresponding to the polypeptides of GTP cyclohydroxylase1 polypeptide and tyrosine hydroxylase, respectively.

76. A method according to Claim 75, in which the ratio is determined by a combination of the activity of the polypeptides and the amount present of the polypeptides.

77. A kit in parts, the kit comprising a pharmaceutical composition according to any of Claims 62 to 74, and instructions for administration.