What is Claimed is:

1. A method for producing transfected cells that produce recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest, said method comprising:

(a) providing one or more plasmids comprising nucleic acids encoding AAV packaging proteins and nucleic acids encoding helper proteins;

(b) providing a plasmid comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest;

(c) providing a solution comprising polyethylenimine (PEI);

(d) mixing the plasmids of steps (a) and (b) with the PEI solution of step (c), wherein said plasmids are in a molar ratio range of 1:0.01 to 1:100, to produce a plasmid/PEI mixture, and optionally incubating said plasmid/PEI mixture for a period in the range of 10 seconds to 4 hours;

(e) contacting cells with said plasmid/PEI mixture of step (d), to produce a plasmid/PEI cell culture;

(f) adding Free PEI to said plasmid/PEI cell culture produced in step (e) to produce a Free PEI/plasmid/PEI cell culture; and

(g) incubating said Free PEI/plasmid/PEI cell culture of step (f) for at least 4 hours, thereby producing transfected cells that produce recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

2. The method according to Claim 1, further comprising the step of harvesting said transfected cells produced in steps (f) or (g) and/or culture medium from said transfected cells produced in steps (f) or (g) to produce a cell and/or culture medium harvest.

3. The method according to Claim 2, further comprising isolating and/or purifying recombinant AAV vector from said cell and/or culture medium harvest of step (g) thereby producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

4. A method for producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest, said method comprising the steps:

(a) providing one or more plasmids comprising nucleic acids encoding AAV packaging proteins and nucleic acids encoding helper proteins;

(b) providing a plasmid comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest;

(c) providing a solution comprising polyethylenimine (PEI);

(d) mixing said plasmids of steps (a) and (b) with said PEI solution of step (c), wherein said plasmids are in a molar ratio range of 1:0.01 to 1:100, to produce a plasmid/PEI mixture, and optionally incubating the plasmid/PEI mixture for a period of time in the range of 10 seconds to 4 hours;

(e) contacting cells with said plasmid/PEI mixture produced in step (d), to produce a plasmid/PEI cell culture;

(f) adding Free PEI to said plasmid/PEI cell culture produced in step (e) to produce a Free PEI/plasmid/PEI cell culture;

(g) incubating said plasmid/PEI cell culture of step (e) or said Free PEI/plasmid/PEI cell culture of step (f) for at least 4 hours to produce transfected cells;

(h) harvesting said transfected cells produced in step (g) and/or culture medium from the transfected cells produced in step (g) to produce a cell and/or culture medium harvest; and

(i) isolating and/or purifying recombinant AAV vector from said cell and/or culture medium harvest produced in step (h) thereby producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

5. A method for producing transfected cells that produce recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest, said method comprising the steps:

(a) providing a mixture of components (i), (ii) and (iii):

(i) one or more plasmids comprising nucleic acids encoding AAV packaging proteins and nucleic acids encoding helper proteins;

(ii) a plasmid comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest; and

(iii) a polyethylenimine (PEI) solution,

(b) mixing said plasmids (i) and (ii) with said PEI solution (iii) so that said plasmids are in a molar ratio range of 1:0.01 to 1:100, to produce a plasmid/PEI mixture, and

optionally incubating said plasmid/PEI mixture for a period of time in the range of 10 seconds to 4 hours;

(c) contacting cells with said plasmid/PEI mixture produced in step (b) to produce a plasmid/PEI cell culture;

(d) adding Free PEI to said plasmid/PEI cell culture produced in step (c) to produce a Free PEI/plasmid/PEI cell culture;

(e) incubating said plasmid/PEI cell culture of step (c) or said Free PEI/plasmid/PEI cell culture of step (d) for at least 4 hours to produce transfected cells that produce recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

6. The method according to Claim 4 or 5, further comprising the step of harvesting the transfected cells produced in step (e) and/or culture medium from the transfected cells produced in step (e) to produce a cell and/or culture medium harvest; and/or isolating and/or purifying recombinant AAV vector from the cell and/or culture medium harvest of step (e), thereby producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

7. A method for producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest, comprising the step:

(a) providing a mixture of components (i), (ii) and (iii):

(i) one or more plasmids comprising nucleic acids encoding AAV packaging proteins and nucleic acids encoding helper proteins;

(ii) a plasmid comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest; and

(iii) a polyethylenimine (PEI) solution,

(b) mixing said plasmids (i) and (ii) with said PEI solution (iii) so that said plasmids are in a molar ratio range of 1:0.01 to 1:100, to produce a plasmid/PEI mixture, and optionally incubating said plasmid/PEI mixture for a period of time from 10 seconds to 4 hours;

(c) contacting cells with said plasmid/PEI mixture produced in step (b) to produce a plasmid/PEI cell culture;

(d) adding Free PEI to said plasmid/PEI cell culture produced in step (c) to produce a Free PEI/plasmid/PEI cell culture;

(e) incubating said plasmid/PEI cell culture of step (c) or said Free PEI/plasmid/PEI cell culture of step (d) for at least 4 hours to produce transfected cells;

(f) harvesting said transfected cells produced in step (e) and/or culture medium from the transfected cells produced in step (e) to produce a cell and/or culture medium harvest; and

(g) isolating and/or purifying recombinant AAV vector from the cell and/or culture medium harvest produced in step (f), thereby producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

8. A method for producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest, comprising the steps:

(a) providing a mixture of components (i), (ii) and (iii):

(i) one or more plasmids comprising nucleic acids encoding AAV packaging proteins and nucleic acids encoding helper proteins;

(ii) a plasmid comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest; and

(iii) a polyethylenimine (PEI) solution,

wherein said plasmids (i) and (ii) are in a molar ratio range of 1:0.01 to 1:100, and wherein the mixture of components (i), (ii) and (iii) has optionally been incubated for a period of time from 10 seconds to 4 hours.

(b) contacting cells with the mixture produced in step (a) to produce a plasmid/PEI cell culture;

(c) adding Free PEI to said plasmid/PEI cell culture produced in step (b) to produce a Free PEI/plasmid/PEI cell culture;

(d) incubating said plasmid/PEI cell culture of step (b) or said Free PEI/plasmid/PEI cell culture of step (c) for at least 4 hours to produce transfected cells;

(e) harvesting said transfected cells produced in step (d) and/or culture medium from the transfected cells produced in step (d) to produce a cell and/or culture medium harvest; and

(f) isolating and/or purifying recombinant AAV vector from the cell and/or culture medium harvest produced in step (e), thereby producing recombinant AAV vector comprising a nucleic acid that encodes a protein or is transcribed into a transcript of interest.

9. The method according to any of Claims 1 to 8, wherein said plasmid/PEI mixture has a PEI:plasmid weight ratio in the range of 0.1:1 to 5:1, or wherein said Free PEI/plasmid/PEI cell culture has a PEI:plasmid weight ratio in the range of 0.1:1 to 5:1.

10. The method according to any of claims 1 to 9, wherein the PEI of said plasmid/PEI mixture and/or the Free PEI comprises a hydrolyzed linear polyethylenimine.

11. The method according to any of claims 1 to 10, wherein the PEI of said plasmid/PEI mixture and/or said Free PEI comprises a hydrolyzed linear polyethylenimine with a molecular weight in the range of 4,000 to 160,000 and/or in the range of 2,500 to 250,000 molecular weight in free base form.

12. The method according to Claims 1 to 11, wherein the molar ratio of nitrogen (N) in Total PEI to phosphate (P) in plasmid is in the range of 1:1 to 50:1 (N:P) in said Free PEI/plasmid/PEI cell culture.

13. The method according to any of Claims 1 to 12, wherein said plasmid/PEI mixture is incubated for a period of time in the range of 30 seconds to 4 hours.

14. The method according to any of Claims 1 to 13, wherein the amount of Free PEI is in the range of 10% to 90% of Total PEI.

15. The method according to any of Claims 1 to 14, wherein said Free PEI is added to said cells before, at the same time as, or after said plasmid/PEI mixture is contacted with said cells.

16. The method according to any of Claims 1 to 15, wherein said cells are in suspension culture.