

CLAIMS

What is claimed is:

1. A method for automated production of a viral vector, comprising:
 - (a) introducing an engineered viral producer cell into a fully enclosed cell engineering system;
 - (b) transducing the engineered viral producer cell with a vector encoding a gene of interest to produce a transduced viral producer cell;
 - (c) expanding the transduced viral producer cell and producing the viral vector within the transduced viral producer cell;
 - (d) transferring the expanded producer cell to a downstream processing module; and
 - (e) isolating the viral vector; and
 - (f) purifying the viral vector,wherein (a) through (e) are performed in a closed and automated process.
2. The method of claim 1, wherein the engineered viral producer cell is a mammalian cell.
3. The method of claim 2, wherein the mammalian cell is a mammalian cell culture.
4. The method of claim 3, wherein the mammalian cell culture is a suspension culture.
5. The method of any one of claims 1-4, wherein the viral vector is an adeno-associated virus (AAV) vector.
6. The method of any one of claims 1-4, wherein the viral vector is a lentivirus vector.
7. The method of any one of claims 1-4, wherein the viral vector is a retrovirus vector.
8. The method of any one of claims 1-4, wherein the viral vector is a baculovirus vector.
9. The method of any one of claims 2-8, wherein the mammalian cell is a Chinese hamster ovary (CHO) cell.
10. The method of any one of claims 2-8, wherein the mammalian cell is a human cell.

11. The method of claim 10, wherein the human cell is a human embryonic kidney (HEK) cell.
12. The method of claim 10, wherein the human cell is a HEK293T cell.
13. The method of claim 1, wherein the engineered producer cell is an insect cell.
14. The method of claim 1, wherein the engineered producer cell is an Sf9 cell.
15. The method of claim 1, wherein an amount of viral vectors produced is at least about 10^{10} viral vectors.
16. The method of any one of claims 1-15, wherein the closed and automated process comprises:
 - (a) monitoring with one or more of a temperature sensor, a pH sensor, a glucose sensor, a lactose sensor, an oxygen sensor, a carbon dioxide sensor, and an optical density sensor, and
 - (b) automatically adjusting one or more of a temperature, a pH level, a glucose level, a lactose level, an oxygen level, a carbon dioxide level, and an optical density.
17. The method of any one of claims 1-16, wherein the transducing comprises viral infection, electroporation, liposome transfection, or membrane disruption.
18. The method of any one of claims 1-17, wherein the isolating comprises passing the expanded producer cells through an elution column.
19. The method of any one of claims 1-18, wherein the purifying comprises membrane polishing.
20. The method of any one of claims 1-19, further comprising formulating the viral vector.
21. A method for automated production of a viral vector, comprising:
 - (a) introducing a packaging cell into a fully enclosed cell engineering system;

- (b) transducing the packaging cell with one or more vectors encoding a viral helper gene, a viral packing gene, and a gene of interest to produce a transduced cell;
 - (c) expanding the transduced cell and producing the viral vector within the transduced cell;
 - (d) transferring the expanded cell to a downstream processing module; and
 - (e) isolating the viral vector; and
 - (f) purifying the viral vector,
- wherein (a) through (e) are performed in a closed and automated process.
22. The method of claim 21, wherein the packaging cell is a mammalian cell.
23. The method of claim 22, wherein the mammalian cell is a mammalian cell culture.
24. The method of claim 23, wherein the mammalian cell culture is a suspension culture.
25. The method of any one of claims 21-24, wherein the viral vector is an adeno-associated virus (AAV) vector.
26. The method of any one of claims 21-24, wherein the viral vector is a lentivirus vector.
27. The method of any one of claims 21-24, wherein the viral vector is a retrovirus vector.
28. The method of any one of claims 21-24, wherein the viral vector is a baculovirus vector.
29. The method of any one of claims 22-28, wherein the mammalian cell is a Chinese hamster ovary (CHO) cell.
30. The method of any one of claims 22-28, wherein the mammalian cell is a human cell.
31. The method of claim 30, wherein the human cell is a human embryonic kidney (HEK) cell.
32. The method of claim 30, wherein the human cell is a HEK293T cell.
33. The method of claim 21, wherein the packaging cell is an insect cell.

34. The method of claim 21, wherein the packaging cell is an Sf9 cell.
35. The method of claim 21, wherein an amount of viral vectors produced is at least about 10^{10} viral vectors.
36. The method of any one of claims 21-35, wherein the closed and automated process comprises:
- (c) monitoring with one or more of a temperature sensor, a pH sensor, a glucose sensor, a lactose sensor, an oxygen sensor, a carbon dioxide sensor, and an optical density sensor, and
 - (d) automatically adjusting one or more of a temperature, a pH level, a glucose level, a lactose level, an oxygen level, a carbon dioxide level, and an optical density.
37. The method of any one of claims 21-36, wherein the transducing comprises viral infection, electroporation, liposome transfection, or membrane disruption.
38. The method of any one of claims 21-37, wherein the isolating comprises passing the expanded producer cells through an elution column.
39. The method of any one of claims 21-38, wherein the purifying comprises membrane polishing.
40. The method of any one of claims 21-39, further comprising formulating the viral vector.