Claims:

1. A gRNA molecule comprising, from 5’ to 3’, [targeting domain]-SEQ ID NO: 7811, wherein said targeting domain comprises SEQ ID NO: 253.

2. The gRNA molecule of claim 1, wherein the targeting domain and said SEQ ID NO: 7811 are disposed on a single nucleic acid molecule, and wherein said single nucleic acid molecule comprises said targeting domain and said SEQ ID NO: 7811 that is disposed immediately 3’ to said targeting domain.

3. The gRNA molecule of claim 1 or 2, wherein said nucleic acid molecule comprises:
(a) three phosphorothioate modifications at the 3’ end of said nucleic acid molecule;
(b) three phosphorothioate modifications at the 5’ end of said nucleic acid molecule;
(c) three 2’-O-methyl modifications at the 3’ end of said nucleic acid molecule;
(d) three 2’-O-methyl modifications at the 5’ end of said nucleic acid molecule; or
(e) any combination thereof.

4. A composition comprising:
(a) the gRNA molecule of any one of claims 1-3 and a Cas9 molecule;
(b) the gRNA molecule of any one of claims 1-3 and a polynucleotide comprising a nucleic acid sequence encoding a Cas9 molecule;
(c) a polynucleotide comprising a nucleic acid sequence encoding the gRNA molecule of any one of claims 1-3 and a Cas9 molecule; or
(d) a polynucleotide comprising a nucleic acid sequence encoding the gRNA molecule of any one of claims 1-3 and a polynucleotide comprising a nucleic acid sequence encoding a Cas9 molecule.

5. A composition comprising the gRNA molecule of any one of claims 1-3, further comprising a Cas9 molecule.

6. The composition of claim 4 or 5, wherein the Cas9 molecule is an active or inactive *S. pyogenes* Cas9.

7. The composition of any one of claims 4-6, wherein the Cas9 molecule comprises SEQ ID NO: 6611.

8. The composition of any one of claims 4-7, wherein the Cas9 molecule comprises:
(a) SEQ ID NO: 7821;
(b) SEQ ID NO: 7822;
(c) SEQ ID NO: 7823;
(d) SEQ ID NO: 7824;
(e) SEQ ID NO: 7825;
(f) SEQ ID NO: 7826;
(g) SEQ ID NO: 7827;
(h) SEQ ID NO: 7828;
(i) SEQ ID NO: 7829;
(j) SEQ ID NO: 7830; or
(k) SEQ ID NO: 7831.

9. The composition of any one of claims 5-8, wherein the gRNA molecule and the Cas9 molecule are present in a ribonuclear protein (RNP) complex.

10. The composition of any one of claims 4-9, formulated in a medium suitable for electroporation.

11. The composition of any one of claims 4-10, wherein said RNP is at a concentration of less than about 10uM.

12. A nucleic acid sequence that encodes the gRNA molecule of any one of claims 1-3.

13. The nucleic acid sequence of claim 12, further comprising a promoter operably linked to the nucleic acid sequence that encodes the gRNA molecule.

14. The nucleic acid sequence of claim 13, wherein the promoter is a promoter recognized by an RNA polymerase II or RNA polymerase III.

15. The nucleic acid sequence of claim 14, wherein the promoter is a U6 promoter or an HI promoter.

16. The nucleic acid sequence of any one of claims 12-15, further comprising a nucleic acid sequence that encodes a Cas9 molecule.

17. The nucleic acid sequence of claim 16, wherein the Cas9 molecule comprises SEQ ID NO: 6611, SEQ ID NO: 7821, SEQ ID NO: 7822, SEQ ID NO: 7823, SEQ ID NO: 7824, SEQ ID NO: 7825, SEQ ID NO: 7826, SEQ ID NO: 7827, SEQ ID NO: 7828, SEQ ID NO: 7829, SEQ ID NO: 7830, or SEQ ID NO: 7831.

18. The nucleic acid sequence of any one of claims 16-17, further comprising a promoter operably linked to the nucleic acid sequence that encodes a Cas9 molecule.

19. The nucleic acid sequence of claim 18, wherein the promoter is an EF-1 promoter, a CMV IE gene promoter, an EF-1α promoter, an ubiquitin C promoter, or a phosphoglycerate kinase (PGK) promoter.

20. A vector comprising the nucleic acid sequence of any one of claims 12-19.

21. The vector of claim 20, wherein the vector is selected from the group consisting of a lentiviral vector, an adenoviral vector, an adeno-associated viral (AAV) vector, a herpes simplex virus (HSV) vector, a plasmid, a minicircle, a nanoplasmid, and an RNA vector.

22. A method of altering a cell at or near a target sequence within said cell, comprising contacting said cell with:
(a) the gRNA molecule of any one of claims 1-3 and a Cas9 molecule;
(b) the gRNA molecule of any one of claims 1-3 and a polynucleotide comprising a nucleic acid sequence encoding a Cas9 molecule;
(c) a nucleic acid molecule encoding the gRNA molecule of any one of claims 1-3 and a Cas9 molecule;
(d) a nucleic acid molecule encoding the gRNA molecule of any one of claims 1-3 and a polynucleotide comprising a nucleic acid encoding a Cas9 molecule;

(e) the composition of any one of claims 4-11; or
(f) the vector of any one of claims 20-21.

23. The method of claim 22, wherein the gRNA molecule or the nucleic acid molecule encoding the gRNA molecule, and the Cas9 molecule or the polynucleotide comprising a nucleic acid sequence encoding the Cas9 molecule, are formulated in a single composition.

24. The method of claim 23, wherein the gRNA molecule or the nucleic acid molecule encoding the gRNA molecule, and the Cas9 molecule or the polynucleotide comprising a nucleic acid sequence encoding the Cas9 molecule, are formulated in more than one composition.

25. The method of claim 24, wherein the more than one composition are delivered simultaneously or sequentially.

26. The method of claim 25, wherein the cell is a hematopoietic stem or progenitor cell (HSPC), a CD34+ cell, a CD34+CD90+ cell or a cell disposed in a composition comprising a population of cells that has been enriched for CD34+ cells.

27. The method of any one of claims 22-26, wherein the cell is a CD34+ cell.

28. The method of any one of claims 22-27, wherein the cell has been isolated from bone marrow, mobilized peripheral blood, or umbilical cord blood.

29. The method of any one of claims 22-28, wherein the altering results in an indel at or near a genomic DNA sequence complementary to the targeting domain of the gRNA molecule.

30. The method of claim 29, wherein the indel is an indel shown in SEQ ID NOs: 2040-2046.

31. The method of claim 29 or 30, wherein the method results in a population of cells, wherein at least about 50% of said population of cells have been altered.

32. The method of any one of claims 22-31, wherein the altering results in a cell that is capable of differentiating into a differentiated cell of an erythroid lineage, and wherein said differentiated cell produces at least about 6 picograms fetal hemoglobin per cell.

33. A cell, altered by the method of any one of claims 22-30.

34. A cell, comprising the gRNA molecule of any one of claims 1-3, the composition of any one of claims 4-11, the nucleic acid of any one of claims 12-19, or the vector of claim 20 or 21.

35. The cell of claim 33 or 34, wherein expression of fetal hemoglobin is increased in said cell or its progeny relative to a cell or its progeny of the same cell type that has not been modified to comprise a gRNA molecule.

36. The cell of any one of claims 33-35, wherein the differentiated cell of an erythroid lineage produces at least about 6 picograms fetal hemoglobin per cell.

37. The cell of any one of claims 33-36, wherein the cell has been contacted with a stem cell expander.

38. The cell of claim 37, wherein the stem cell expander is

(1r,4r)-N^1-(2-benzyl-7-(2-methyl-2H-tetrazol-5-yl)-9H-pyrimido[4,5-b]indol-4-yl)cyclohexane-1,4-diamine,

methyl 4-(3-piperidin-1-ylpropylamino)-9H-pyrimido[4,5-b]indole-7-carboxylate,

4-(2-(2-(benzo[b]thiophen-3-yl)-9-isopropyl-9H-purin-6-ylamino)ethyl)phenol,

(S)-2-(6-(2-(lH-indol-3-yl)ethylamino)-2-(5-fluoropyridin-3-yl)-9H-purin-9-yl)propan-l-ol,

or a combination thereof.

39. The cell of any one of claims 33-38, comprising an indel at or near a genomic DNA sequence complementary to the targeting domain of a gRNA molecule of any one of claims 1-3.

40. The cell of claim 39, wherein the indel is an indel shown in SEQ ID NOs: 2040-2046.

41. The cell of any one of claims 33-40, wherein the cell is an animal cell.

42. The cell of claim 41, wherein the cell is a mammalian, a primate, or a human cell.

43. The cell of any one of claims 33-42, wherein the cell is a hematopoietic stem or progenitor cell (HSPC), a CD34+ cell, a CD34+CD90+ cell, or a cell isolated from bone marrow, mobilized peripheral blood, or umbilical cord blood.

44. The cell of any one of claims 33-43, wherein the cell is a CD34+ cell.

45. A population of cells comprising the cell of any one of claims 33-44.

46. The population of cells of claim 45, wherein at least about 50% of said population of cells are a cell according to any one of claims 33-44.

47. The population of cells of claim 45 or 46, wherein the population of cells is capable of differentiating into a population of differentiated cells of an erythroid lineage, and wherein said population of differentiated cells has an increased fraction of F cells relative to a population of unmodified cells of the same type.

48. The population of cells of claim 47, wherein the F cells of the population of differentiated cells produce an average of at least about 6 picograms fetal hemoglobin per cell.

49. The population of cells of any one of claims 45-48, comprising:
(a) at least 1e6 CD34+ cells per kg body weight of a patient to whom the cells are to be administered;
(b) at least 2e6 CD34+ cells per kg body weight of a patient to whom the cells are to be administered;

(c) at least 3e6 CD34+ cells per kg body weight of a patient to whom the cells are to be administered;
(d) at least 4e6 CD34+ cells per kg body weight of a patient to whom the cells are to be administered; or
(e) from 2e6 to 10e6 CD34+ cells per kg body weight of a patient to whom the cells are to be administered,
wherein the CD34+ cells are a cell according to any one of claims 33-44.

50. The population of cells of any one of claims 45-49, wherein at least about 40% of said population of cells are CD34+ cells.

51. The population of cells of claim 50, wherein at least about 10% of said population of cells of the population are CD34+CD90+ cells.

52. The population of cells of any one of claims 45-51, wherein the population of cells is isolated from bone marrow, mobilized peripheral blood, or umbilical cord blood.

53. The population of cells of any one of claims 45-52, wherein the population of cells are isolated from a patient that has a hemoglobinopathy.

54. A composition comprising the cell of any one of claims 33-44, or the population of cells of any one of claims 45-53.

55. The composition of claim 54, comprising a pharmaceutically acceptable medium suitable for cryopreservation.

56. A method of preparing a population of cells comprising:
(a) providing a population of HSPCs;
(b) culturing said population of HSPCs ex vivo in a cell culture medium comprising a stem cell expander; and
(c) introducing into said population of HSPCs the gRNA molecule of any one of claims 1-3, a nucleic acid molecule encoding the gRNA molecule of any one of claims 1-3, the composition

of any one of claims 4-11, the nucleic acid of any one of claims 12-19, or the vector of claim 20 or 21.

57. The method of claim 56, wherein the stem cell expander is:

(1r,4r)-N^1-(2-benzyl-7-(2-methyl-2H-tetrazol-5-yl)-9H-pyrimido[4,5-b]indol-4-yl)cyclohexane-1,4-diamine,

methyl 4-(3-piperidin-1-ylpropylamino)-9H-pyrimido[4,5-b]indole-7-carboxylate,

4-(2-(2-(benzo[b]thiophen-3-yl)-9-isopropyl-9H-purin-6-ylamino)ethyl)phenol,

(S)-2-(6-(2-(lH-indol-3-yl)ethylamino)-2-(5-fluoropyridin-3-yl)-9H-purin-9-yl)propan-l-ol, or

a combination thereof.

58. The method of claim 56, wherein the cell culture medium comprises the stem cell expander at a concentration ranging from about 1 nM to about 1 mM.

59. The method of claim 56, wherein the period of culturing before the introducing step (c) is at least 12 hours.

60. The method of any one of claims 56-59, further comprising a period of culturing step after the introducing step (c).

61. The method of claim 60, wherein the period of culturing step after the introducing step (c) is at least 12 hours.

62. The method of any one of claims 56-61, wherein the population of cells is expanded at least 4-fold, relative to cells which are not cultured according to step (b).

63. The method of any one of claims 56-62, wherein the introducing of step (c) comprises an electroporation.

64. The method of any one of claims 56-63, wherein the population of HSPCs provided in step (a) is a population of human cells.

65. The method of claim 64, wherein the population of HSPCs provided in step (a) is isolated from bone marrow, peripheral blood, umbilical cord blood, or bone marrow of a patient suffering from a hemoglobinopathy.

66. The method of any one of claims 56-65, wherein the population of HSPCs provided in step (a) is enriched for CD34+ cells.

67. The method of any one of claims 56-66, wherein subsequent to the introducing step (c), the population of HSPCs is cryopreserved.

68. The method of any one of claims 56-67, wherein after the introducing step (c), at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, at least about 95%, at least about 96%, at least about 97%, at least about 98% or at least about 99% of the cells of the population of cells comprise an indel at or near a genomic DNA sequence complementary to the targeting domain of the gRNA molecule.

69. The method of claim 68, wherein the indel in each of said cells of the population of cells is an indel shown in SEQ ID NOs: 2040-2046.

70. A cell or a population of cells, prepared by the method of any one of claims 56-69.

71. Use of the gRNA molecule of any one of claims 1-3, the composition of any one of claims 4-11 and 54-55, the nucleic acid of any one of claims 12-19, the vector of claim 20 or 21, the cell of any one of claims 33-44 and 70, or the population of cells of any one of claims 45-53, in the manufacture of a medicament for the treatment of a hemoglobinopathy.

72. The use of claim 71, wherein the hemoglobinopathy is beta-thalassemia or sickle cell disease.