in the vector. However, promoters are often spread over a long range of DNA and poorly characterized, and can thus not be easily reconstituted in their entirety in a vector construct. Furthermore, expression levels from tissue-specific promoters reconstituted in gene-transfer vectors are often not sufficient to achieve phenotypic correction, most likely because of imperfect reconstitution and/or detrimental influence of the chromatin at the semi-random vector integration site. Thus, additional strategies to regulate a transgene are direly needed.

Summary

In one aspect, the present disclosure refers to a use of a gene vector comprising at least one miRNA target sequence corresponding to a miRNA selected from the group consisting of mir-130a and mir-126 operably linked to a nucleotide sequence for the manufacture of a medicament for preventing or reducing expression of the nucleotide sequence in a hematopoietic stem cell or a hematopoietic progenitor cell.

In another aspect, the present disclosure refers to a use of a cell for the manufacture of a medicament for preventing or reducing expression of the nucleotide sequence in a hematopoietic stem cell or a hematopoietic progenitor cell, wherein the cell is infected or transduced with the gene vector as disclosed herein.

In another aspect, the present disclosure refers to a use of the gene vector as disclosed herein, or the cell as disclosed herien for the manufacture of a medicament for treating a disease selected from Globoid Cell Leukodystrophy, Chronic Granulomatous Disease Severe Combined Immunodeficiency (SCID) and solid tumours.

In another aspect, the present disclosure refers to a use of the gene vector as disclosed herein, or the cell as disclosed herein for the manufacture of a medicament for increasing the chances of survival of a hematopoietic stem cell or a hematopoietic progenitor cell in relation to gene therapy.

In another aspect, the present disclosure refers to a use of the gene vector as disclosed herein, or the cell as disclosed herein for the manufacture of a medicament for preventing apoptosis of a hematopoietic stem cell or a hematopoietic progenitor cell, whereby the apoptosis is caused by expression of the nucleotide sequence.

In another aspect, the present disclosure refers to a use of the gene vector as disclosed herein, or the cell as disclosed herein for the manufacture of a medicament for monitoring a stage of differentiation of a hematopoietic stem cell or a hematopoietic progenitor cell.

In another aspect, the present disclosure refers to a method of determining the differentiation stage of a hematopoietic stem cell or a hematopoietic progenitor cell, comprising determining the expression level of a transgene in said hematopoietic stem cell or said hematopoietic progenitor cell, wherein the transgene is operably linked to a miRNA target sequence, whereby the corresponding miRNA prevents or reduces expression of the transgene in a hematopoietic progenitor cell (HSPC) or a hematopoietic stem cell (HSC) but not in a differentiated cell, wherein the miRNA is selected from the group comprising mir-130a and mir-126.

Statements of the Invention

According to one aspect of the present invention there is provided a gene vector for use in gene therapy comprising at least one miRNA sequence target operably linked to a nucleotide sequence having a corresponding miRNA in a hematopoietic progenitor cell (HSPC) which prevents or reduces expression of the nucleotide sequence in a HSPC but not in a differentiated cell.

According to another aspect of the present invention there is provided a gene vector for use in gene therapy comprising at least one miRNA sequence target operably linked to a nucleotide sequence having a corresponding miRNA in a hematopoietic stem cell (HSC) which prevents or reduces expression of the nucleotide sequence in a HSC but not in a differentiated cell.

In other words, the present invention provides a gene vector suitable for use in hematopoietic gene therapy comprising at least one miRNA sequence target for a miRNA which is present in an effective amount in a hematopoietic progenitor cell or hematopoietic stem cell and optionally a transgene. By effective amount we mean that the concentration of the endogenous miRNA is sufficient to reduce or prevent expression of a transgene which is operably linked to the corresponding miRNA target sequence. Thus the present invention employs the use of miRNA which is strongly expressed in cells, such as HSPC and HSC but not in differentiated progeny of e.g. the myeloid and lymphoid lineage, preventing or reducing expression of a potentially toxic transgene in sensitive stem cell populations, whilst maintain expression and therapeutic efficacy in the diseased progeny.

CLAIMS

1. Use of a gene vector comprising at least one miRNA target sequence corresponding to a miRNA selected from the group consisting of mir-130a and mir-126 operably linked to a nucleotide sequence for the manufacture of a medicament for preventing or reducing expression of the nucleotide sequence in a hematopoietic stem cell or a hematopoietic progenitor cell.

2. The use of claim 1, wherein the gene vector comprises at least one miRNA target sequence corresponding to mir-130a and at least one miRNA target sequence corresponding to mir-126.

3. The use of claim 2, wherein the number of copies of the miRNA target sequence corresponding to mir-130a is twice the number of copies of the miRNA target sequence corresponding to mir-126.

4. The use of any one of claims 1 to 3, wherein the nucleotide sequence controls the expression of the vector.

5. The use of any one of claims 1 to 4, wherein the nucleotide sequence is a transgene.

6. The use of any one of claims 1 to 5, wherein the vector is a viral vector.

7. The use of any one of claims 1 to 6, wherein the vector is derivable from a lentivirus.

8. The use of any one of claims 1 to 7, wherein the vector comprises a tissue specific promoter.

9. The use of claim 8, wherein the tissue specific promoter is selected from the group consisting of CD11b, c-Fes and CYBB and TEK.

10. The use of claim 8, wherein the tissue specific promoter is a myeloid specific promoter.

11. The use of claim 5, wherein the transgene codes for an enzyme.

12. The use of any one of claims 1 to 10, wherein the nucleotide sequence is a transgene encoding an enzyme selected from the group lysosomal enzyme galactocerebrosidase, and gp91 phox; or wherein the nucleotide sequence is a transgene encoding interferon-alpha.

13. The use of any one of claims 1-12, wherein the gene vector is in the form of a viral vector particle.

14. Use of a cell for the manufacture of a medicament for preventing or reducing expression of the nucleotide sequence in a hematopoietic stem cell or a hematopoietic progenitor cell, wherein the cell is infected or transduced with the gene vector as defined in any one of claims 1 to 13.

15. The use of claim 14, wherein the cell is a hematopoietic stem cell or a hematopoietic progenitor cell.

16. Use of the gene vector as defined in any one of claims 1-13, or the cell as defined in claim 14 or 15 for the manufacture of a medicament for treating a disease selected from Globoid Cell Leukodystrophy, Chronic Granulomatous Disease Severe Combined Immunodeficiency (SCID) and solid tumours.

17. Use of the gene vector as defined in any one of claims 1-13, or the cell as defined in claim 14 or 15 for the manufacture of a medicament for increasing the chances of survival of a hematopoietic stem cell or a hematopoietic progenitor cell in relation to gene therapy.

18. Use of the gene vector as defined in any one of claims 1-13, or the cell as defined in claims 14 or 15 for the manufacture of a medicament for preventing apoptosis of a hematopoietic stem cell or a hematopoietic progenitor cell, whereby the apoptosis is caused by expression of the nucleotide sequence.

19. Use of the gene vector as defined in any one of claims 1-13, or the cell as defined in claim 14 or 15 for the manufacture of a medicament for monitoring a stage of differentiation of a hematopoietic stem cell or a hematopoietic progenitor cell.

20. The use of any one of claims 1-19, wherein the medicament is for hematopoietic cell therapy.

21. A method of determining the differentiation stage of a hematopoietic stem cell or a hematopoietic progenitor cell, comprising determining the expression level of a transgene in said hematopoietic stem cell or said hematopoietic progenitor cell, wherein the transgene is operably linked to a miRNA target sequence, whereby the corresponding miRNA prevents or reduces expression of the transgene in a hematopoietic progenitor cell (HSPC) or a hematopoietic stem cell (HSC) but not in a differentiated cell, wherein the miRNA is selected from the group comprising mir-130a and mir-126.

22. The gene vector as defined in any one of claims 1-13 or the cell as defined in claim 14 or 15 when used for preventing or reducing expression of the nucleotide sequence in a hematopoietic stem cell or a hematopoietic progenitor cell.

23. The gene vector as defined in any one of claims 1-13 or the cell as defined in claim 14 or 15 when used for treating a disease selected from Globoid Cell Leukodystrophy, Chronic Granulomatous Disease Severe Combined Immunodeficiency (SCID) and solid tumours.

24. The gene vector as defined in any one of claims 1-13 or the cell as defined in

claim 14 or 15 when used for increasing the chances of survival of a hematopoietic stem cell or a hematopoietic progenitor cell in relation to gene therapy.

25. The gene vector as defined in any one of claims 1-13 or the cell as defined in claim 14 or 15 when used for preventing apoptosis of a hematopoietic stem cell or a hematopoietic progenitor cell, whereby the apoptosis is caused by expression of the nucleotide sequence.

26. The gene vector as defined in any one of claims 1-13 or the cell as defined in claim 14 or 15 when used for monitoring a stage of differentiation of a hematopoietic stem cell or a hematopoietic progenitor cell.