In yet another aspect, the present disclosure refers to an isolated polynucleotide encoding the α chain of a T-cell receptor (TCR) of the present disclosure, and/or the β chain of a TCR of the present disclosure.

In yet another aspect, the present disclosure refers to a vector comprising a polynucleotide as disclosed herein.

In a further aspect, the present disclosure refers to a cell comprising a TCR as disclosed herein, a polynucleotide as disclosed herein, or a vector as disclosed herein, optionally wherein the cell further comprises a vector which encodes one or more CD3 chains, CD8, a suicide gene and/or a selectable marker.

In still another aspect, the present disclosure refers to a method of preparing a cell, which comprises the step of introducing a vector as disclosed herein into a cell *in vitro* or *ex vivo*, e.g. by transfection or transduction.

In yet another aspect, the present disclosure refers to a chimeric molecule comprising a TCR of the present disclosure, conjugated to a non-cellular substrate, a toxin and/or an antibody, wherein the non-cellular substrate is selected from the group consisting of nanoparticles and exosomes.

In yet another aspect, the present disclosure refers to the use of the TCR as disclosed herein, an isolated polynucleotide as disclosed herein, a vector as disclosed herein, a cell as disclosed herein, a cell prepared by the method as disclosed herein, or a chimeric molecule according to claim in the manufacture of a medicament for treating a hematological malignancy or a solid tumor, preferably wherein the hematological malignancy is selected from the group consisting of acute myeloid leukemia (AML), chronic myeloid leukemia (CML), lymphoblastic leukemia, myelodisplastic syndromes, lymphoma, multiple myeloma, non Hodgkin lymphoma, and Hodgkin lymphoma; or preferably wherein the solid tumor is selected from the group consisting of lung cancer, breast cancer, oesophageal cancer, gastric cancer, colon cancer, cholangiocarcinoma, pancreatic cancer, ovarian cancer, head and neck cancers, synovial sarcoma, angiosarcoma, osteosarcoma, thyroid cancer, endometrial cancer, neuroblastoma, rabdomyosarcoma, liver cancer, melanoma, prostate cancer, renal cancer, soft tissue sarcoma, urothelial cancer, biliary cancer, glioblastoma, mesothelioma, cervical cancer, and colorectal cancer. In one aspect, the present disclosure refers to the use of a cell as disclosed herein in the manufacture of a medicament for treating a hematological malignancy or a solid tumor, preferably wherein the hematological malignancy is selected from the group consisting of acute myeloid leukemia (AML), chronic myeloid leukemia (CML), lymphoblastic leukemia, myelodisplastic syndromes, lymphoma, multiple myeloma, non Hodgkin lymphoma, and Hodgkin lymphoma; or preferably wherein

the solid tumor is selected from the group consisting of lung cancer, breast cancer, oesophageal cancer, gastric cancer, colon cancer, cholangiocarcinoma, pancreatic cancer, ovarian cancer, head and neck cancers, synovial sarcoma, angiosarcoma, osteosarcoma, thyroid cancer, endometrial cancer, neuroblastoma, rabdomyosarcoma, liver cancer, melanoma, prostate cancer, renal cancer, soft tissue sarcoma, urothelial cancer, biliary cancer, glioblastoma, mesothelioma, cervical cancer, and colorectal cancer.

In another example, the present disclosure refers to an isolated immunogenic WT1 peptide comprising an amino acid sequence selected from the group consisting of EPASQHTLRSG (SEQ ID NO: 123) YESDNHTTPIL (SEQ ID NO: 126), NHTTPILCGAQYRIH (SEQ ID NO: 127), QCLSAFTVHFSGQFT (SEQ ID NO: 118), EDPMGQQGSLGEQQY (SEQ ID NO: 119), SQLECMTWNQMNLGA (SEQ ID NO: 120), DPGGIWAKLGAAEAS (SEQ ID NO: 251), NHTTPILCGAQYRIH (SEQ ID NO: 252), KRHQRRHTGVKPFQC (SEQ ID NO: 253), PSCQKKFARSDELVR (SEQ ID NO: 254) and variants thereof each having up to three amino acid substitutions, additions or deletions.

In yet another example, the present disclosure refers to the use of a TCR as disclosed herein, an isolated polynucleotide as disclosed herein, a vector as disclosed herein, a cell as disclosed herein, a cell prepared by the method as disclosed herein, or a chimeric molecule as disclosed herein in the manufacture of a medicament for treating and/or preventing a disease associated with expression of WT1 in a subject in need thereof, wherein the use comprises that the medicament is to be administered to the subject.

In yet another example, the present disclosure refers to an isolated immunogenic WT1 peptide comprising an amino acid sequence selected from the group consisting of EPASQHTLRSG (SEQ ID NO: 123) YESDNHTTPIL (SEQ ID NO: 126), NHTTPILCGAQYRIH (SEQ ID NO: 127), QCLSAFTVHFSGQFT (SEQ ID NO: 118), EDPMGQQGSLGEQQY (SEQ ID NO: 119), SQLECMTWNQMNLGA (SEQ ID NO: 120), APVLDFAPPGA (SEQ ID NO: 117), NQMNLGATLKG (SEQ ID NO: 250), DPGGIWAKLGAAEAS (SEQ ID NO: 251), NHTTPILCGAQYRIH (SEQ ID NO: 252), KRHQRRHTGVKPFQC (SEQ ID NO: 253), PSCQKKFARSDELVR (SEQ ID NO: 254) and variants thereof each having up to three amino acid substitutions, additions or deletions.

In one example, the present disclosure provides a TCR of the present disclosure comprising the following CDR sequences:

(i) CDR1α - KALYS (SEQ ID NO: 1),
CDR2α - LLKGGEQ (SEQ ID NO: 2),
CDR3α - CGTAWINDYKLSF (SEQ ID NO: 3),
CDR1β - SGHDY (SEQ ID NO: 6),
CDR2β - FNNNVP (SEQ ID NO: 7), and
CDR3β - CASRKTGGYSNQPQHF (SEQ ID NO: 8),
or variants thereof each having up to three amino acid substitutions, additions or deletions;

CLAIMS

1. A T-cell receptor (TCR), which binds to a Wilms tumour 1 protein (WT1) peptide when presented by a major histocompatibility complex (MHC), wherein the TCR comprises the following CDR sequences:

CDR1α - KALYS (SEQ ID NO: 1),
CDR2α - LLKGGEQ (SEQ ID NO: 2),
CDR3α - CGTAWINDYKLSF (SEQ ID NO: 3),
CDR1β - SGHDY (SEQ ID NO: 6),
CDR2β - FNNNVP (SEQ ID NO: 7), and
CDR3β - CASRKTGGYSNQPQHF (SEQ ID NO: 8).

2. A TCR according to claim 1 comprising an α chain variable domain comprising the amino acid sequence of SEQ ID NO: 4 or a variant thereof having at least 75% sequence identity thereto; and a β chain variable domain comprising the amino acid sequence of SEQ ID NO: 9 or a variant thereof having at least 75% sequence identity thereto.

3. A TCR according to claim 1 or 2 comprising an α chain comprising the amino acid sequence of SEQ ID NO: 5 or a variant thereof having at least 75% sequence identity thereto; and a β chain comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 10, SEQ ID NO: 11 and variants of SEQ ID NOs: 10 and 11 having at least 75% sequence identity thereto.

4. A TCR according to any preceding claim which binds to an MHC I peptide complex.

5. A TCR according to any preceding claim, which is restricted to a human leukocyte antigen (HLA) allele, preferably wherein the TCR is restricted to a HLA-A allele.

6. A TCR according to any one of claims 1-3, which is restricted to HLA-A*0201.

7. A TCR according to any preceding claim comprising one or more mutations at the α chain/β chain interface, such that when the α chain and the β chain are expressed in a T-cell, the frequency of mispairing between said chains and endogenous TCR α and β chains is reduced.

8. A TCR according to claim 7, wherein the one or more mutations introduce a cysteine residue into the constant region domain of each of the α chain and the β chain, wherein the cysteine residues are capable of forming a disulphide bond between the α chain and the β chain.

9. A TCR according to any preceding claim, which comprises a murinized constant region.

10. A TCR according to any preceding claim, wherein the TCR is a soluble TCR.

11. An isolated polynucleotide encoding the α chain of a T-cell receptor (TCR) according to any preceding claim, and/or the β chain of a TCR according to any preceding claim.

12. An isolated polynucleotide according to claim 11, wherein the polynucleotide encodes the α chain linked to the β chain.

13. An isolated polynucleotide according to claim 11 or 12, which encodes one or more short interfering RNA (siRNA) or other agents capable of reducing or preventing expression of one or more endogenous TCR gene.

14. A vector comprising a polynucleotide according to any one of claims 11-13.

15. A vector according to claim 14 comprising a polynucleotide, which encodes one or more CD3 chains, CD8, a suicide gene, and/or a selectable marker.

16. A cell comprising a TCR according to any one of claims 1-10, a polynucleotide according to any one of claims 11-13 or a vector according to claim 14 or 15,

optionally wherein the cell further comprises a vector which encodes one or more CD3 chains, CD8, a suicide gene and/or a selectable marker.

17. A cell according to claim 16, wherein the cell is a T-cell, a lymphocyte, or a stem cell, optionally wherein the T-cell, the lymphocyte, or the stem cell is selected from the group consisting of CD4 cells, CD8 cells, naive T-cells, memory stem T-cells, central memory T-cells, double negative T-cells, effector memory T-cells, effector T-cells, Th0 cells, Tc0 cells, Th1 cells, Tc1 cells, Th2 cells, Tc2 cells, Th17 cells, Th22 cells, gamma/delta T-cells, natural killer (NK) cells, natural killer T (NKT) cells, hematopoietic stem cells and pluripotent stem cells.

18. A cell according to any one of claims 16-17, wherein an endogenous gene encoding a TCR α chain and/or an endogenous gene encoding a TCR β chain is disrupted, preferably such that the endogenous gene encoding a TCR α chain and/or the endogenous gene encoding a TCR β chain is not expressed,

optionally wherein the endogenous gene encoding a TCR α chain and/or the endogenous gene encoding a TCR β chain is disrupted by insertion of an expression cassette comprising a polynucleotide sequence encoding a TCR of any one of claims 1-10,

further optionally wherein one or more endogenous genes encoding an MHC is disrupted,

further optionally wherein an endogenous gene involved in persistence, expansion, activity, resistance to exhaustion/senescence/inhibitory signals, homing capacity, or other T-cell functions is disrupted, preferably wherein the endogenous gene involved in persistence, expansion, activity, resistance to exhaustion/senescence/inhibitory signals, homing capacity, or other T-cell functions is selected from the group consisting of *PD1, TIM3, LAG3, 2B4, KLRG1, TGFbR, CD160* and *CTLA4*.

19. A cell according to any one of claims 16-18, wherein the cell is a T-cell.

20. A method of preparing a cell, which comprises the step of introducing a vector according to claim 14 and/or 15 into a cell *in vitro* or *ex vivo*, e.g. by transfection or transduction.

21. A method of preparing a cell according to claim 20, which comprises the step of T-cell editing, which comprises disrupting an endogenous gene encoding a TCR α chain and/or an endogenous gene encoding a TCR β chain with an artificial nuclease, preferably wherein the artificial nuclease is selected from the group consisting of zinc finger nucleases (ZFN), transcription activator-like effector nucleases (TALEN) and CRISPR/Cas systems.

22. A method of preparing a cell according to claim 21, which comprises the step of targeted integration of an expression cassette into the endogenous gene encoding the TCR α chain and/or the endogenous gene encoding the TCR β chain disrupted by the artificial nuclease, wherein the expression cassette comprises a polynucleotide sequence encoding a TCR of any one of claims 1-10.

23. A method of preparing a cell according to any one of claims 20-22, which comprises the step of disrupting one or more endogenous genes encoding an MHC, preferably wherein the cell prepared by the method is a non-alloreactive universal T-cell.

24. A method of preparing a cell according to any one of claims 20-23, which comprises the step of disrupting one or more endogenous genes to modify the persistence, expansion, activity, resistance to exhaustion/senescence/inhibitory signals, homing capacity, or other T-cell functions, preferably wherein the method comprises the step of targeted integration of an expression cassette into an endogenous gene involved in persistence, expansion, activity, resistance to exhaustion/senescence/inhibitory signals, homing capacity, or other T-cell functions disrupted by an artificial nuclease, wherein the expression cassette comprises a polynucleotide sequence encoding a TCR of any one of claims 1-10, preferably wherein the

endogenous gene is selected from the group consisting of *PD1, TIM3, LAG3, 2B4, KLRG1, TGFbR, CD160* and *CTLA4*.

25. A cell according to claim 19 for use in adoptive cell transfer, optionally wherein the adoptive cell transfer is allogenic adoptive T-cell transfer, autologous adoptive T-cell transfer, or universal non-alloreactive adoptive T-cell transfer.

26. A chimeric molecule comprising a TCR of any one of claims 1-10 conjugated to a non-cellular substrate, a toxin and/or an antibody, wherein the non-cellular substrate is selected from the group consisting of nanoparticles and exosomes.

27. A cell according to claim 19 for use in therapy.

28. Use of the cell according to claim 19 in the manufacture of a medicament for treating a hematological malignancy or a solid tumor, preferably wherein the hematological malignancy is selected from the group consisting of acute myeloid leukemia (AML), chronic myeloid leukemia (CML), lymphoblastic leukemia, myelodisplastic syndromes, lymphoma, multiple myeloma, non Hodgkin lymphoma, and Hodgkin lymphoma; or preferably wherein the solid tumor is selected from the group consisting of lung cancer, breast cancer, oesophageal cancer, gastric cancer, colon cancer, cholangiocarcinoma, pancreatic cancer, ovarian cancer, head and neck cancers, synovial sarcoma, angiosarcoma, osteosarcoma, thyroid cancer, endometrial cancer, neuroblastoma, rabdomyosarcoma, liver cancer, melanoma, prostate cancer, renal cancer, soft tissue sarcoma, urothelial cancer, biliary cancer, glioblastoma, mesothelioma, cervical cancer, and colorectal cancer.