

obesity, laminopathies such as Hutchinson Gilford's progeria, hernia, sarcopenia and cachexia, arthritis, scoliosis and cancer.

It was recently shown that clearance of senescent cells in a genetic fashion could markedly improve the fitness and decrease parameters of aging in a mouse model for accelerated aging. These mice showed reduced signs of aging measured by kyphosis (excessive bone curvature), muscle strength, fat deposition and cataracts. This provided further evidence that cellular senescence and the SASP is causally linked to age-associated phenotypes and cancer (Baker et al., 2011. Nature 479(7372):232-6). This proof-of-concept evidence was obtained in a genetic fashion that has poor therapeutic applicability.

It is clear in the context of the above that there is a need in the art for compounds that selectively induce apoptosis in senescent cells, as cellular senescence is linked to degenerative (loss-of-function) diseases and cancer (gain of function). There are currently no suitable therapeutically applicable compounds that can selectively induce apoptosis in senescent cells in vivo. It is a goal of the present invention to provide new and existing compounds that therapeutically target senescent cells and that can be used in the treatment of diseases associated with cellular senescence.

20

SUMMARY

The inventors solved this problem by their unexpected finding that a peptide according to the disclosure, and the inhibitors NQDI and R406 for use according to the disclosure, selectively induce apoptosis in senescent cells and, as a consequence, are applicable in the treatment of diseases associated with the presence of senescent cells. The inventors showed *inter alia* that compounds according to the disclosure are effective in the (i) removal of senescent cells *in vitro*, *ex vivo* and *in vivo* (ii) countering symptoms of aging and age-related diseases in a fast aging mouse model, (iii) protection of organ function in an *in vivo* model for chemotherapy-

30

induced toxicity and (iv) sensitization of a resistant cancer to a chemotherapeutic agent it was previously resistant to or acquired resistance to.

Thus, in one aspect, the present disclosure refers to a peptide
5 comprising the amino acid sequence
LTLRKEPASEIAQSILEAYSQNGWANRRSGGKRP, wherein the amino acids in said amino acid sequence are D-amino acid residues.

In another aspect, the present disclosure refers to a peptide having at least 70% amino acid sequence identity to the peptide as disclosed herein, or a
10 fragment of the peptide as disclosed herein, wherein said peptide or fragment exhibits apoptosis-inducing activity in senescent cells or cells having an increased FOXO4 expression as compared to a control cell and expressing Ser15 phosphorylated p53 (pSer15-p53).

In another aspect, the present disclosure refers to a pharmaceutical
15 composition comprising a peptide disclosed herein.

In yet another aspect, the present disclosure refers to a retro-inverso peptide of the Forkhead box protein O4, wherein the amino acid sequence of said Forkhead box protein O4 is indicated in Figs 14 or 15, or a homologue or a fragment thereof, wherein said homologue has an amino acid sequence
20 identity of at least 70% to the retro-inverso amino acid sequence of the Forkhead box protein O4, wherein said homologue exhibits apoptosis-inducing activity in senescent cells or cells having an increased FOXO4 expression as compared to a control cell and expressing pSer15-p53, and wherein said fragment has apoptosis-inducing activity in senescent cells or
25 cells having an increased FOXO4 expression as compared to a control cell and expressing pSer15-p53.

In yet another aspect, the present disclosure refers to a nucleic acid encoding a peptide disclosed herein.

In yet another aspect, the present disclosure refers to a host cell
30 comprising the nucleic acid disclosed herein.

In yet another aspect, the present disclosure refers to the peptide disclosed herein, the pharmaceutical composition disclosed herein, or the nucleic acid disclosed herein, for use as a medicament.

In yet another aspect, the present disclosure refers to use of the peptide disclosed herein, the pharmaceutical composition disclosed herein, or the nucleic acid disclosed herein, in the manufacture of a medicament for
5 the treatment of cancer.

In yet another example, the present disclosure refers to the peptide disclosed herein, the pharmaceutical composition disclosed herein, or the nucleic acid disclosed herein for use in countering p21cip1 expression in a human subject, for use in removing cells that express p16INK4a in a
5 subject, and/or for use in countering or in reducing the number of nuclear serine-15-phosphorylated p53 foci in a subject, whereby said subject is suffering from, or expected to suffer from atherosclerosis; chronic inflammatory diseases such as arthritis or arthrosis; cancer; osteoarthritis; glomerulosclerosis, diabetes including diabetes type I and II; diabetic ulcers;
10 kyphosis; scoliosis; hepatic insufficiency; cirrhosis; Hutchinson-Gilford progeria syndrome (HGPS); laminopathies; osteoporosis; dementia; (cardio)vascular diseases; myocardial infarction; obesity; metabolic syndrome; acute myocardial infarction; emphysema; insulin sensitivity; boutonneuse fever; sarcopenia; neurodegenerative diseases such as
15 Alzheimer's, Huntington's or Parkinson's disease; cataracts; anemia; hypertension; fibrosis; age-related macular degeneration; COPD; asthma; renal insufficiency; reducing or preventing graft failure after organ or tissue transplantation; ischemia reperfusion injury; hearing loss such as deafness; vision loss such as blindness.

20 More specifically, the inventors solved this problem by providing a peptide comprising the amino acid sequence
LTLRKEPASEIAQSILEAYSQNGWANRRSGGKRP, wherein the amino acids in said amino acid sequence are D-amino acid residues.

Alternatively, provided is a peptide having at least 70%, preferably at
25 least 90%, amino acid sequence identity to the peptide as disclosed herein, or a fragment of the peptide as disclosed herein, wherein said peptide or fragment exhibits apoptosis-inducing activity in senescent cells, preferably wherein

Claims

1. A peptide comprising the amino acid sequence
LTLRKEPASEIAQSILEAYSQNGWANRRSGGKRP, wherein the amino acids
in said amino acid sequence are D-amino acid residues.
- 5 2. A peptide having at least 70% amino acid sequence identity to the
peptide of claim 1, or a fragment of the peptide of claim 1, wherein said peptide
or fragment exhibits apoptosis-inducing activity in senescent cells or cells
having an increased FOXO4 expression as compared to a control cell and
expressing Ser15 phosphorylated p53 (pSer15-p53).
- 10 3. A peptide according to claim 1 or 2, wherein said peptide has at least
90% amino acid sequence identity to the peptide of claim 1 or a fragment of the
peptide of claim 1.
- 15 4. A peptide according to any of claims 1-3, wherein said fragment has the
amino acid sequence SEIAQSILEAYSQNGW, and wherein at least 90% of the
amino acids in said amino acid sequence or in said fragment are D-amino acid
residues.
- 20 5. A peptide according to any of claims 1-4, further comprising a cell-
penetrating peptide sequence, preferably said cell-penetrating peptide
sequence has the amino acid sequence PPRRRQRRKKRG, preferably wherein
the amino acids in said cell-penetrating peptide sequence are D-amino acid
residues.
- 25 6. Peptide according to claim 5, wherein said cell-penetrating peptide is
fused to the C-terminal part of said peptide.

7. A pharmaceutical composition comprising a peptide according to any one of claims 1-6.
8. The pharmaceutical composition according to claim 7, further comprising
5 a chemotherapeutic agent.
9. A retro-inverso peptide of the Forkhead box protein O4, wherein the amino acid sequence of said Forkhead box protein O4 is indicated in Figs 14 or 15, or a homologue or a fragment thereof, wherein said homologue has an
10 amino acid sequence identity of at least 70% to the retro-inverso amino acid sequence of the Forkhead box protein O4, wherein said homologue exhibits apoptosis-inducing activity in senescent cells or cells having an increased FOXO4 expression as compared to a control cell and expressing pSer15-p53, and wherein said fragment has apoptosis-inducing activity in senescent cells or
15 cells having an increased FOXO4 expression as compared to a control cell and expressing pSer15-p53.
10. A nucleic acid encoding a peptide according to any one of claims 1-6 or 9.
- 20 11. A nucleic acid according to claim 10, wherein said nucleic acid is comprised in an expression vector.
12. A host cell comprising the nucleic acid according to claim 10 or 11.
- 25 13. The peptide of any one of claims 1-6 or 9, the pharmaceutical composition according to claim 7 or 8, or the nucleic acid according to claim 10 or 11, for use as a medicament.

14. Use of the peptide of any one of claims 1-6 or 9, the pharmaceutical composition according to claim 7 or 8, or the nucleic acid according to claim 10 or 11, in the manufacture of a medicament for the treatment of cancer.