

also been tested as vaccine carriers for the delivery of a variety of immunogens derived from a variety of infectious agents. Studies conducted in experimental animals (e. g. rodents, canines and nonhuman primates) indicate that recombinant replication-defective human adenoviral vectors carrying transgenes encoding immunogens as well as other antigens elicit both humoral and cell-mediated immune responses against the transgene product. Generally speaking, investigators have reported success using human adenoviral vectors as vaccine carriers in non human experimental systems by either using immunization protocols that utilizes high doses of recombinant adenoviral vectors that are predicted to elicit immune responses; or by using immunization protocols which employ the sequential administration of adenoviral vectors that are derived from different serotypes but which carry the same transgene product as boosting immunizations (Mastrangeli, et. al., Human Gene Therapy, 7: 79-87 (1996)).

Vectors derived from species C adenoviruses (e.g. Ad5, Ad6 and ChAd63) are the most immunogenic (Colloca et al., Sci. Transl. Med. 4 (115), 2012). In particular, viral vectors based on human adenovirus type 5 (Ad5) have been developed for gene therapy and vaccine applications. Although Ad5-based vectors are extremely efficient in animal models, the presence of a pre-existing immunity in humans against Ad5 wild type virus has in clinical trials been demonstrated to reduce the efficiency of gene transduction (Moore JP et al. Science. 2008 May 9; 320(5877):753-5). Thus, immunity in the general population limits the broad application of Ad vectored-vaccines based on Ad5. On the other hand, rare human adenoviruses are less immunogenic than Ad5 (Colloca et al., Sci. Transl. Med. 4 (115), 2012). Vectors based on non human adenoviruses do not have a pre-existing immunity in the general human population (Farina et al., J. Virol. 75 (23), 11603-11613, 2001).

Thus, there is a need for adenovirus vectors with high immunogenicity and a low or absent pre-existing immunity in humans. Preferably, these adenovirus vectors have a high productivity in terms of their replication.

SUMMARY

In one aspect, the present disclosure provides an isolated polynucleotide encoding an adenovirus hexon protein comprising:

- (i) a HVR1 comprising an amino acid sequence according to SEQ ID NO: 11
- (ii) a HVR2 comprising an amino acid sequence according to SEQ ID NO: 12,
- (iii) a HVR3 comprising an amino acid sequence according to SEQ ID NO: 13,

- (iv) a HVR4 comprising an amino acid sequence according to SEQ ID NO: 14
- (v) a HVR5 comprising an amino acid sequence according to SEQ ID NO: 15,
- (vi) a HVR6 comprising an amino acid sequence according to SEQ ID NO: 16, and
- (vii) a HVR7 comprising an amino acid sequence according to SEQ ID NO: 17.

5 In another aspect, the present disclosure provides an isolated polynucleotide encoding an adenovirus comprising the polynucleotide as described herein. In another aspect, the present disclosure provides an isolated adenovirus comprising at least one isolated adenoviral hexon protein as described herein. In another aspect, the present disclosure provides a virus-like particle encoded by an isolated polynucleotide as described herein, wherein the virus-like
10 particle comprises at least one adenoviral hexon protein as described herein. In another aspect, the present disclosure provides a vector comprising an isolated polynucleotide as described herein.

In a further aspect, the present disclosure provides a composition comprising (i) an adjuvant, (ii) an isolated polynucleotide as described herein, at least one isolated adenoviral
15 hexon protein as described herein, an adenovirus as described herein, a virus-like particle as described herein, or a vector as described herein, and optionally (iii) a pharmaceutically acceptable excipient. In a further aspect, the present disclosure provides a cell comprising a polynucleotide as described herein, at least one isolated adenoviral hexon protein as described herein, an adenovirus as described herein, a virus-like particle as described herein, or a vector
20 as described herein.

In yet another aspect, the present disclosure provides use of the polynucleotide as described herein, the at least one isolated adenoviral hexon protein as described herein, the adenovirus as described herein, the virus-like particle as described herein, or the vector as described herein and/or the composition as described herein in the manufacture of a
25 medicament for treating or preventing a disease, In yet another aspect, the present disclosure provides an *in vitro* method for producing an adenovirus or an adenovirus-like particle, comprising the steps of

- (i) expressing an isolated polynucleotide as described herein in a cell such that an adenovirus or an adenovirus-like particle is assembled in the cell,
- 30 isolating the adenovirus or the adenovirus-like particle from the cell or the medium surrounding the cell.

In a first example, the disclosure provides an isolated polynucleotide encoding an adenovirus hexon protein comprising:

- 5 A) (i) a first hypervariable region HVR1 comprising an amino acid sequence according to
SEQ ID NO: 11, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 11 and with no A and preferably with a V at position 27,
- (ii) a second hypervariable region HVR2 comprising an amino acid sequence according to
10 SEQ ID NO: 12, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 12 and with no L and preferably with an I at position 1,
- (iii) a third hypervariable region HVR3 comprising an amino acid sequence according to
SEQ ID NO: 13, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 13 and with no V and preferably with an A at position 7,
- 15 (iv) a fourth hypervariable region HVR4 comprising an amino acid sequence according to
SEQ ID NO: 14, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 14,
- (v) a fifth hypervariable region HVR5 comprising an amino acid sequence according to
SEQ ID NO: 15, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 15,
- (vi) a sixth hypervariable region HVR6 comprising an amino acid sequence according to
SEQ ID NO: 16, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 16, and
- 20 (vii) a seventh hypervariable region HVR7 comprising an amino acid sequence according
to SEQ ID NO: 17, or a variant thereof having at least 85% sequence identity to SEQ
ID NO: 17 with no I and preferably with a V at position 1; or
- B) (i) a first hypervariable region HVR1 comprising an amino acid sequence according to
SEQ ID NO: 18, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 18 with no V and preferably with an E at position 8, with no D and preferably
25 with an E at position 12, with no E and preferably with a D at position 13, and/or with
no L and preferably with a V at position 14,
- (ii) a second hypervariable region HVR2 comprising an amino acid sequence according to
SEQ ID NO: 19, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 19 with no D and preferably with an E at position 10,
- 30 (iii) a third hypervariable region HVR3 comprising an amino acid sequence according to
SEQ ID NO: 20, or a variant thereof having at least 85% sequence identity to SEQ ID
NO: 20 with no T and preferably with an A at position 6,

- (iv) a fourth hypervariable region HVR4 comprising an amino acid sequence according to SEQ ID NO: 21, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 21 with no L and preferably with a M at position 9,
 - (v) a fifth hypervariable region HVR5 comprising an amino acid sequence according to SEQ ID NO: 22, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 22 with no T and preferably with an S at position 3,
 - (vi) a sixth hypervariable region HVR6 comprising an amino acid sequence according to SEQ ID NO: 23, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 23 with no I and preferably with a V at position 9, and
 - (vii) a seventh hypervariable region HVR7 comprising an amino acid sequence according to SEQ ID NO: 24, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 24 with no I and preferably with a V at position 8; or
- C) (i) a first hypervariable region HVR1 comprising an amino acid sequence according to SEQ ID NO: 25, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 25,
- (ii) a second hypervariable region HVR2 comprising an amino acid sequence according to SEQ ID NO: 26, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 26,
 - (iii) a third hypervariable region HVR3 comprising an amino acid sequence according to SEQ ID NO: 27, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 27 with no V and preferably with an A at position 7,
 - (iv) a fourth hypervariable region HVR4 comprising an amino acid sequence according to SEQ ID NO: 28, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 28 with no E and preferably with a Q at position 10,
 - (v) a fifth hypervariable region HVR5 comprising an amino acid sequence according to SEQ ID NO: 29, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 29 with no T and preferably with an S at position 3,
 - (vi) a sixth hypervariable region HVR6 comprising an amino acid sequence according to SEQ ID NO: 30, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 30 with no I and preferably with a V at position 9, and
 - (vii) a seventh hypervariable region HVR7 comprising an amino acid sequence according to SEQ ID NO: 31, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 31 with no I and preferably with a V at position 8 and/or with no T and preferably with an S at position 11; or

- D) (i) a first hypervariable region HVR1 comprising an amino acid sequence according to SEQ ID NO: 32, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 32,
- 5 (ii) a second hypervariable region HVR2 comprising an amino acid sequence according to SEQ ID NO: 33, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 33,
- (iii) a third hypervariable region HVR3 comprising an amino acid sequence according to SEQ ID NO: 34, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 34 with no T and preferably with an A at position 6,
- 10 (iv) a fourth hypervariable region HVR4 comprising an amino acid sequence according to SEQ ID NO: 35, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 35 with no Q and preferably with a K at position 6 and/or with no E and preferably with a Q at position 10,
- (v) a fifth hypervariable region HVR5 comprising an amino acid sequence according to SEQ ID NO: 36, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 36 with no T and preferably with an S at position 3,
- 15 (vi) a sixth hypervariable region HVR6 comprising an amino acid sequence according to SEQ ID NO: 37, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 37 with no K and preferably with a T at position 1 and/or with no I and preferably with a V at position 9, and
- 20 (vii) a seventh hypervariable region HVR7 comprising an amino acid sequence according to SEQ ID NO: 38, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 38 with no I and preferably with a V at position 8; or
- E) (i) a first hypervariable region HVR1 comprising an amino acid sequence according to SEQ ID NO: 39, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 39 with no A and preferably with a V at position 27,
- 25 (ii) a second hypervariable region HVR2 comprising an amino acid sequence according to SEQ ID NO: 40, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 40,
- 30 (iii) a third hypervariable region HVR3 comprising an amino acid sequence according to SEQ ID NO: 41, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 41,

- (iv) a fourth hypervariable region HVR4 comprising an amino acid sequence according to SEQ ID NO: 42, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 42,
- (v) a fifth hypervariable region HVR5 comprising an amino acid sequence according to SEQ ID NO: 43, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 43,
- (vi) a sixth hypervariable region HVR6 comprising an amino acid sequence according to SEQ ID NO: 44, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 44, and
- (vii) a seventh hypervariable region HVR7 comprising an amino acid sequence according to SEQ ID NO: 45, or a variant thereof having at least 85% sequence identity to SEQ ID NO: 45 with no I and preferably with a V at position 1.

In a second example, the disclosure provides an isolated polynucleotide encoding an adenovirus, preferably a replication-incompetent adenovirus comprising the polynucleotide the first example.

In a third example, the disclosure provides at least one isolated adenoviral capsid polypeptide encoded by an isolated polynucleotide of the first example.

In a fourth example, the disclosure provides an adenovirus encoded by an isolated polynucleotide of the first example or an isolated adenovirus, preferably a replication-incompetent adenovirus, comprising an isolated polynucleotide according to the first example and/or at least one isolated adenoviral capsid polypeptide according to the third example.

In a fifth example, the disclosure provides a virus-like particle encoded by an isolated polynucleotide of the first example.

In a sixth example, the disclosure provides a vector comprising an isolated polynucleotide of the first example.

In a seventh example, the disclosure provides a composition comprising (i) an adjuvant, (ii) an isolated polynucleotide of the first or second example, at least one isolated adenoviral capsid polypeptide of the third example, an adenovirus of the fourth example, a virus-like particle of the fifth example, or a vector of the sixth example, and optionally (iii) a pharmaceutically acceptable excipient.

In an eighth example, the disclosure provides a cell comprising an isolated polynucleotide of the first or second example, at least one isolated adenoviral capsid polypeptide of the third example, an adenovirus of the fourth example, a virus-like particle of the fifth example, or a vector of the sixth example.

In a ninth example, the disclosure provides an isolated polynucleotide of the first or second example, at least one isolated adenoviral capsid polypeptide of the third example, an adenovirus of the fourth example, a virus-like particle of the fifth example, or a vector of the sixth example and/or the composition of the seventh example for use in treating or preventing a disease.

In a tenth example, the disclosure relates to an *in vitro* method for producing an adenovirus or an adenovirus-like particle, comprising the steps of

- (i) expressing an isolated polynucleotide of the first or second example in a cell such that an adenovirus or an adenovirus-like particle is assembled in the cell,
- (ii) isolating the adenovirus or the adenovirus-like particle from the cell or the medium surrounding the cell.

DETAILED DESCRIPTION OF THE INVENTION

Before the present invention is described in detail below, it is to be understood that this invention is not limited to the particular methodology, protocols and reagents described herein as these may vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to limit the scope of the present invention which will be limited only by the appended claims. Unless defined otherwise, all technical and scientific terms used herein have the same meanings as commonly understood by one of ordinary skill in the art.

Preferably, the terms used herein are defined as described in "A multilingual glossary of biotechnological terms: (IUPAC Recommendations)", Leuenberger, H.G.W, Nagel, B. and Klbl, H. eds. (1995), Helvetica Chimica Acta, CH-4010 Basel, Switzerland) and as described in "Pharmaceutical Substances: Syntheses, Patents, Applications" by Axel Kleemann and Jurgen Engel, Thieme Medical Publishing, 1999; the "Merck Index: An Encyclopedia of Chemicals, Drugs, and Biologicals", edited by Susan Budavari et al., CRC Press, 1996, and the United States Pharmacopeia-25/National Formulary-20, published by the United States Pharmacopeial Convention, Inc., Rockville Md., 2001.

Throughout this specification and the claims which follow, unless the context requires otherwise, the word "comprise", and variations such as "comprises" and "comprising", will be understood to imply the inclusion of a stated feature, integer or step or group of features, integers or steps but not the exclusion of any other feature, integer or step or group of integers or steps. In the following passages different aspects of the invention are defined in more

detail. Each aspect so defined may be combined with any other aspect or aspects unless clearly indicated to the contrary. In particular, any feature indicated as being preferred or advantageous

CLAIMS

1. An isolated polynucleotide encoding an adenovirus hexon protein comprising:
 - (i) a HVR1 comprising an amino acid sequence according to SEQ ID NO: 11
 - 5 (ii) a HVR2 comprising an amino acid sequence according to SEQ ID NO: 12,
 - (iii) a HVR3 comprising an amino acid sequence according to SEQ ID NO: 13,
 - (iv) a HVR4 comprising an amino acid sequence according to SEQ ID NO: 14
 - (v) a HVR5 comprising an amino acid sequence according to SEQ ID NO: 15,
 - (vi) a HVR6 comprising an amino acid sequence according to SEQ ID NO: 16, and
 - 10 (vii) a HVR7 comprising an amino acid sequence according to SEQ ID NO: 17.
2. The isolated polynucleotide of claim 1, wherein the hexon protein comprises an amino acid sequence according to SEQ ID NO: 46, or a variant thereof having at least 85% sequence identity thereto.
- 15 3. The isolated polynucleotide of any one of claims 1 to 2, further encoding an adenoviral penton protein comprising an amino acid sequence according to SEQ ID NO: 51 or 52, or a variant thereof having at least 85% sequence identity thereto.
- 20 4. The isolated polynucleotide of any one of claims 1 to 3, further encoding an adenoviral fiber protein comprising an amino acid sequence according to SEQ ID NO: 53 or 54, or a variant thereof having at least 85% sequence identity thereto.
- 25 5. The isolated polynucleotide of any one of claims 1 to 4, further encoding a VA RNA II non-coding RNA comprising a nucleotide sequence according to SEQ ID NO: 57, or a variant thereof having at least 85% sequence identity, and/or a VA RNA I non-coding RNA comprising a nucleotide sequence according to SEQ ID NO: 55 or 56, or a variant thereof having at least 85% sequence identity thereto.
- 30 6. An isolated polynucleotide encoding an adenovirus comprising the polynucleotide of any one of claims 1 to 5.
7. The isolated polynucleotide of claim 6, wherein the adenovirus is a chimeric adenovirus, carries a non-adenoviral gene, protein or fragment thereof, and/or is replication-incompetent.

8. An isolated adenoviral hexon protein encoded by an isolated polynucleotide of any one of claims 1-2.

9. An isolated adenovirus comprising at least one isolated adenoviral hexon protein according to claim 8.

10. A virus-like particle encoded by an isolated polynucleotide of any one of claims 1-5, wherein the virus-like particle comprises at least one adenoviral hexon protein according to claim 8.

11. A vector comprising an isolated polynucleotide of any one of claims 1-7.

12. A composition comprising (i) an adjuvant, (ii) an isolated polynucleotide of any one of claims 1-7, at least one isolated adenoviral hexon protein of claim 8, an adenovirus of claim 9, a virus-like particle of claim 10, or a vector of claim 11, and optionally (iii) a pharmaceutically acceptable excipient.

13. A cell comprising a polynucleotide of any one of claims 1-7, at least one isolated adenoviral hexon protein of claim 8, an adenovirus of claim 9, a virus-like particle of claim 10, or a vector of claim 11.

14. Use of the polynucleotide of any one of claims 1-7, the at least one isolated adenoviral hexon protein of claim 8, the adenovirus of claim 9, the virus-like particle of claim 10, or the vector of claim 11 and/or the composition of claim 12 in the manufacture of a medicament for treating or preventing a disease.

15. An *in vitro* method for producing an adenovirus or an adenovirus-like particle, comprising the steps of

- (i) expressing an isolated polynucleotide of any one of claims 1-7 in a cell such that an adenovirus or an adenovirus-like particle is assembled in the cell,
- (ii) isolating the adenovirus or the adenovirus-like particle from the cell or the medium surrounding the cell.