

SYNTHETIC ONCOLYTIC LNP-REPLICON RNA AND USES FOR CANCER IMMUNOTHERAPY

RELATED APPLICATION

5 This application claims priority under 35 U.S.C. § 119(e) to United States Provisional Application Serial Number 62/815,611, filed March 8, 2019, the entire contents of which are incorporated by reference herein.

BACKGROUND

10 The present disclosure relates to synthetic oncolytic viruses comprising a lipid nanoparticle comprising one or more types of lipid and a self-amplifying replicon RNA comprising a sequence that encodes an immunomodulatory molecule.

SUMMARY

15 The present disclosure is based, at least in part, on the unexpected discovery that upon injection at a tumor site, a synthetic oncolytic virus successfully triggers anti-cancer immune response of local tumors and enables systemic immunity against distal tumors.

 In one aspect, the present disclosure refers to a synthetic oncolytic virus, comprising:
20 (i) a lipid nanoparticle comprising N1,N3,N5-tris(3-(didodecylamino)propyl)benzene-1,3,5-tricarboxamide (TT3)~~one or more types of lipid~~; and (ii) a self-amplifying replicon RNA comprising a sequence that encodes an interleukin (IL)-12 molecule; ~~wherein the lipid nanoparticle is capable of triggering immunogenic cell death~~, and wherein the IL-12 molecule is expressed by the self-amplifying replicon RNA.

25 In another aspect, the present disclosure refers to a pharmaceutical composition, comprising: the synthetic oncolytic virus as disclosed herein and a pharmaceutically acceptable carrier.

 In another aspect, the present disclosure refers to a use of the synthetic oncolytic virus as disclosed herein or the pharmaceutical composition as disclosed herein in the manufacture of a medicament for treating cancer in a subject in need thereof.

30 Accordingly, one example of the present disclosure provides a synthetic oncolytic virus comprising a lipid nanoparticle comprising one or more types of lipid and a self-amplifying replicon RNA comprising a sequence that encodes an interleukin (IL)-12 molecule. The lipid nanoparticle is capable of triggering immunogenic cell death. The IL-12 molecule is expressed by the self-amplifying replicon RNA.

35 The synthetic oncolytic virus comprises a lipid nanoparticle. The lipid nanoparticle comprises one or more types of lipids. In some examples, the one or more types of lipids comprises a cationic lipid. In some examples, the cationic lipid is N1,N3,N5-tris(3-(didodecylamino)propyl)benzene-1,3,5-tricarboxamide (TT3). In some examples, the lipid nanoparticle comprises TT3, 1,2-Dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE), cholesterol, and C14-PEG2000.

40 Further, the synthetic oncolytic virus comprises a self-amplifying replicon RNA. In some examples, the self-amplicon RNA is derived from an alphavirus or other Group IV viruses (positive single strand RNA viruses, such as a hepatitis C virus (HCV)). In some examples, the alphavirus can be Venezuela Equine Encephalitis virus, Semliki Forest virus, or Sindbis virus.

What is claimed is:

CLAIMS

1. A synthetic oncolytic virus, comprising:
 - (i) a lipid nanoparticle comprising N1,N3,N5-tris(3-(didodecylamino)propyl)benzene-1,3,5-tricarboxamide (TT3)~~one or more types of lipid~~; and
 - (ii) a self-amplifying replicon RNA comprising a sequence that encodes an interleukin (IL)-12 molecule;
~~wherein the lipid nanoparticle is capable of triggering immunogenic cell death~~, and
wherein the IL-12 molecule is expressed by the self-amplifying replicon RNA .
2. The synthetic oncolytic virus of claim 1, wherein the lipid nanoparticle further comprises one or more types of lipid, where the one or more types of lipid comprises a cationic lipid.
3. ~~The synthetic oncolytic virus of claim 2, wherein the cationic lipid is N1,N3,N5-tris(3-(didodecylamino)propyl)benzene-1,3,5-tricarboxamide (TT3).~~
4. The synthetic oncolytic virus of claim 2, wherein the lipid nanoparticle further comprises ~~TT3~~, 1,2-Dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE), ~~cholesterol~~, and ~~C14-PEG2000~~.
4. The synthetic oncolytic virus of claim 2 or 3, wherein the lipid nanoparticle further comprises cholesterol.
5. The synthetic oncolytic virus of any one of claims 1-4, wherein the self-amplifying replicon RNA is derived from an alphavirus, or a hepatitis C virus.
6. The synthetic oncolytic virus of ~~any one of claims~~claim 5, wherein the alphavirus is Venezuela Equine Encephalitis virus, Semliki Forest virus, or Sindbis virus.
7. The synthetic oncolytic virus of any one of claims 1-6, wherein the sequence that encodes the IL-12 molecule is located in a subgenomic region of the self-amplifying replicon RNA.

8. The synthetic oncolytic virus of any one of claims 1-7, wherein the self-amplifying replicon RNA comprises a nucleotide sequence that ~~is~~ has at least 90% ~~identical sequence identity to the sequence set forth in~~ SEQ ID NO: 1.

5 ~~9. The synthetic oncolytic virus of claim 8, wherein the self-amplifying replicon RNA is not identical to SEQ ID NO: 1 and is capable of expressing the IL-12 molecule at a higher level compared to the self-amplifying replicon RNA comprising SEQ ID NO: 1.~~

10 ~~109.~~ The synthetic oncolytic virus of claim ~~98~~, wherein the self-amplifying replicon RNA comprises a point mutations of G3936C and/or A4758G of SEQ ID NO: 1.

~~11. The synthetic oncolytic virus of any one of claims 1-10, wherein the self-amplifying replicon RNA further comprises a serum albumin coding sequence.~~

15 ~~12. The synthetic oncolytic virus of any one of claims 1-11, wherein the self-amplifying replicon RNA further comprises a lumican coding sequence.~~

~~13~~10. The synthetic oncolytic virus of any one of claims 1-~~129~~, wherein the IL-12 molecule is selected from the group consisting of IL-12, an IL-12 subunit, or a mutant IL-12 molecule that retains the immunomodulatory function.

~~14~~11. The synthetic oncolytic virus of claim ~~13~~10, wherein the IL-12 molecule comprises IL-12 α and/or IL-12 β subunits.

25 ~~15~~12. The synthetic oncolytic virus of any one of claims 1-~~14~~11, wherein the lipid nanoparticle has a diameter of ~~about~~ 100-120 nm and/or has a zeta potential of 3-6 mv.

~~16. The synthetic oncolytic virus of any one of claims 1-15, wherein the lipid nanoparticle has a zeta potential of about 3-6 mv.~~

30 ~~17. The synthetic oncolytic virus of any one of claims 1-16, wherein the lipid and the self-amplifying replicon RNA have a mass ratio of about 1:1.~~

13. The synthetic oncolytic virus of any one of claims 1-12, wherein the lipid nanoparticle is capable of triggering immunogenic cell death.

~~18~~14. A pharmaceutical composition, comprising:

5 the synthetic oncolytic virus of any one of claims 1-~~17~~13 and a pharmaceutically acceptable carrier.

15. The pharmaceutical composition of claim 14, further comprising C14-PEG2000.

10 ~~19~~16. The pharmaceutical composition of claim ~~18~~14 or 15, wherein the pharmaceutical composition is formulated for intratumoral injection.

~~20~~17. Use of the synthetic oncolytic virus of any one of claims 1-~~17~~13 or the pharmaceutical composition of any one of claims ~~18 or 19~~14-16 in the manufacture of a
15 medicament for treating cancer in a subject in need thereof.

~~21~~18. The use of claim ~~20~~17, wherein the subject is a human patient having or suspected of having a cancer.

20 ~~22~~19. The use of claim ~~21~~18, wherein the human patient has a cancer selected from the group consisting of melanoma, breast cancer, ~~and~~ colon cancer, squamous cell cancer, small-cell lung cancer, non-small cell lung cancer, adenocarcinoma of the lung, squamous carcinoma of the lung, cancer of the peritoneum, hepatocellular cancer, gastrointestinal cancer, pancreatic cancer, glioblastoma, cervical cancer, ovarian cancer, liver cancer, bladder
25 cancer, hepatoma, colorectal cancer, endometrial or uterine carcinoma, salivary gland carcinoma, kidney cancer, prostate cancer, vulval cancer, thyroid cancer, hepatic carcinoma, gastric cancer, squamous cell head and neck cancer, renal-cell cancer, urothelial carcinoma, and Hodgkin's lymphoma.

30 ~~23~~20. The use of any one of claims ~~19~~17-22~~19~~, wherein the synthetic oncolytic virus or the pharmaceutical composition is to be administered to the subject in a single dose.

2421. The use of any one of claims ~~1917-2320~~, wherein the synthetic oncolytic virus or the pharmaceutical composition is to be administered to the subject by intratumoral injection, intramuscular injection, subcutaneous injection, or intravenous injection.