35(1), pp 567-572). More generally, about 12% of all human cancers worldwide are caused by oncogenic virus infection (see e.g., Mesri et al., 2014, Cell Host & Microbe 15:266-282; Bouvard, V., et al. (2009). Lancet Oncol. 10, 321–322; Boyle, P., and Levin, B. (2008). World Cancer Report 2008. (Lyon: International Agency for Research on Cancer and World Health Organization Press); de Martel, C., et al., (2012). Lancet Oncol. 13, 607–615).

[0011] The present invention provides an immunotherapeutic treatment of cancers, including virus-induced cancers. These and other advantages are provided in the present disclosure.

BRIEF SUMMARY

[0012] The present disclosure provides compositions and methods for enhancing immunotherapy of cancer, such as virus-induced cancer, through combination of local and systemic immune stimulation. In one embodiment of the present disclosure, a method of treating cancer is provided comprising a) inducing an immune response specific for an immunogen in a subject, comprising administering to the subject a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding the immunogen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence; and b) administering locally to the subject a second composition comprising a pharmaceutically suitable adjuvant, wherein the second composition does not comprise or is substantially devoid of immunogen, wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, and wherein the first composition and second composition are administered concurrently or sequentially. In certain embodiments, administration of the second composition induces expression of one or more cytokines capable of recruiting CD8 T-cells to the local site of administration.

[0012a] In one aspect of the present disclosure, there is provided a composition comprising

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the

polynucleotide is operatively linked to at least one regulatory expression sequence, thereby inducing an immune response against the cancer-associated antigen in the subject; and

b) a second composition comprising a TLR4 agonist; wherein the second composition does not comprise antigen;

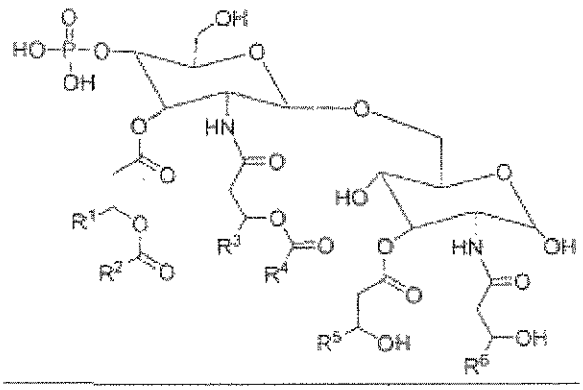
wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, for use in a method of treating a cancer wherein the first composition is to be administered systemically to the subject; and wherein the second composition is to be administered intratumorally, peritumorally, intranodally or intravesically to the subject; and wherein the first composition and second composition are to be administered concurrently or sequentially.

[0012b] In another aspect of the present disclosure, there is provided a kit comprising a first composition and a second composition according to any one of the preceding claims.

[0012c] In a further aspect of the present disclosure, there is provided a composition comprising:

a) a first composition comprising a lentiviral vector particle, the lentiviral vector particle comprising a recombinant expression vector, wherein the recombinant expression vector comprises a polynucleotide encoding an immunogen associated with cancer, wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein; and

b) a second composition comprising a TLR4 agonist, wherein the second composition does not comprise an immunogen; wherein the TLR4 agonist is glucopyranosyl lipid A (GLA) formulated in a stable oil-in-water emulsion, wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient,

for use in a method of treating a cancer in a subject, wherein the first composition is to be administered intramuscularly, intradermally or subcutaneously and induces an immune response specific for the immunogen associated with the cancer in the subject; wherein the second composition is to be administered intratumorally or peritumorally; and wherein the first composition or second composition are to be administered concurrently or sequentially.

[0012d] In another aspect of the present disclosure, there is provided a composition comprising:

a) a first composition comprising

i. antigen-specific T cells specific for a cancer-associated antigen, said antigen-specific T cells having been expanded *ex vivo*,

ii. T cells that have been genetically modified to express a chimeric antigen receptor (CAR) that recognizes the cancer-associated antigen, or

iii. T cells that have been genetically modified to express a specific chimeric T cell receptor (TCR) that recognizes the cancer-associated antigen in the context of MHC; and

b) a second composition comprising a pharmaceutically suitable adjuvant;

wherein the second composition does not comprise an immunogen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient;

for use in a method of treating a cancer in a subject wherein the first composition is to be administered systemically to the subject; and the second composition is to be administered intratumorally and wherein the first composition and second composition are to be administered concurrently or sequentially.

[0012e] In a further aspect of the present disclosure, there is provided a composition comprising:

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, and

b) a second composition comprising a TLR4 agonist wherein the composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, for use in a method of increasing T cells in a tumor microenvironment in a subject, where the first composition is to be administered systemically, thereby inducing an immune response against the cancer-associated antigen in the subject; and where the second composition is to be administered intratumorally or peritumorally to the subject; and where the first and second composition are to be administered concurrently or sequentially; thereby increasing T cells in the tumor microenvironment.

[0012f] In another aspect of the present disclosure, there is provided a use of a composition comprising

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, thereby inducing an immune response against the cancer-associated antigen in the subject; and

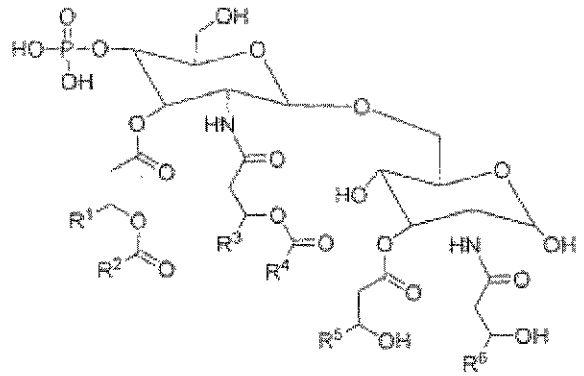
b) a second composition comprising a TLR4 agonist; wherein the second composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, in the manufacture of a medicament for treating a cancer wherein the first composition is to be administered systemically to the subject; and wherein the second composition is to be administered intratumorally, peritumorally, intranodally or intravesically to the subject; and wherein the first composition and second composition are to be administered concurrently or sequentially.

[0012g] In a further aspect of the present disclosure, there is provided a use of a composition comprising:

a) a first composition comprising a lentiviral vector particle, the lentiviral vector particle comprising a recombinant expression vector, wherein the recombinant expression vector comprises a polynucleotide encoding an immunogen associated with cancer, wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein; and

b) a second composition comprising a TLR4 agonist, wherein the second composition does not comprise an immunogen; wherein the TLR4 agonist is glucopyranosyl lipid A (GLA) formulated in a stable oil-in-water emulsion, wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient.

in the manufacture of a medicament for treating a cancer in a subject, wherein the first composition is to be administered intramuscularly, intradermally or subcutaneously and induces an immune response specific for the immunogen associated with the cancer in the subject; wherein the second composition is to be administered intratumorally or peritumorally; and wherein the first composition or second composition are to be administered concurrently or sequentially.

[0012h] In another aspect of the present disclosure, there is provided a use of a composition comprising:

a) a first composition comprising

i. antigen-specific T cells specific for a cancer-associated antigen, said antigen-specific T cells having been expanded *ex vivo*,

ii. T cells that have been genetically modified to express a chimeric antigen receptor (CAR) that recognizes the cancer-associated antigen, or

iii. T cells that have been genetically modified to express a specific chimeric T cell receptor (TCR) that recognizes the cancer-associated antigen in the context of MHC; and

b) a second composition comprising a pharmaceutically suitable adjuvant; wherein the second composition does not comprise an immunogen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient;

in the manufacture of a medicament for treating a cancer in a subject wherein the first composition is to be administered systemically to the subject; and the second composition is to be administered intratumorally and wherein the first composition and second composition are to be administered concurrently or sequentially.

[0012i] In a further aspect of the present disclosure, there is provided a use of a composition comprising:

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, and

b) a second composition comprising a TLR4 agonist wherein the composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, in the manufacture of a medicament for increasing T cells in a tumor microenvironment in a subject, where the first composition is to be administered systemically, thereby inducing an immune response against the cancer-associated antigen in the subject; and where the second composition is to be administered intratumorally or peritumorally to the subject; and where the first and second composition are to be administered concurrently or sequentially; thereby increasing T cells in the tumor microenvironment.

[0013] In another embodiment, an aforementioned method is provided wherein the first composition and the second composition are administered sequentially, and wherein the first composition is administered to the subject at a first site and the second composition is administered to the subject at a second site, wherein the first site and the second site are different. In still another embodiment, an aforementioned method is provided wherein the first composition is administered at the first site via a first route and the second composition is administered at the second site via a second route.

where: R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl; wherein the cytokines are selected from the group consisting of Cxcl9, Cxcl10, CCL2, CCL3, MCP-1, TNFa, IFNc and IP-10; wherein the immunogen associated with a virus-induced cancer is a viral antigen selected from EBNA-1, LMP-1, LMP-2A; E6, E7, E5; HBx; HCV Core, NS3, Ns5A; Tax, HBZ; vFLIP, LANA, vGPCR, vIRF-1; wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, wherein the first composition and second composition are administered concurrently or sequentially, wherein the first composition is administered intramuscularly, intradermally or subcutaneously and the second composition is administered intratumorally or intravesically; and wherein the second composition optionally comprises BCG.

[0026] In another embodiment of the present disclosure, a kit is provided comprising a first composition and a second composition according to any one of the previous methods, above or described herein.

[0027] ~~Another aspect of t~~The present ~~invention~~ disclosure also provides a method of treating an oncogenic virus-induced cancer comprising: a) inducing an immune response specific for an oncogenic virus antigen in a subject, comprising administering to the subject a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding the oncogenic virus antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence; and b) administering to the subject at a local site a second composition comprising a pharmaceutically suitable adjuvant, wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, and wherein the first composition and second composition are administered concurrently or sequentially. In one embodiment, administration of the second composition at the local site induces expression of one or more cytokines capable of recruiting T cells to the local site of administration. In certain embodiments, the T cells are antigen-specific T cells, such as antigen-specific CD4 and/or CD8 T cells, effector memory T cells (T_{EM}), central memory T cells (T_{CM}) and/or tissue-resident memory T cells (T_{RM}). In one embodiment of the method, the local site of local administration is intratumoral, intranodal, intravesical or mucosal. In another embodiment, the expression vector further comprises a polynucleotide encoding a tumor-associated antigen. In a further embodiment, the second composition further comprises the tumor-associated antigen.

[0028] ~~One specific aspect of t~~The present disclosure ~~invention~~ further provides a method for treating an oncogenic virus-induced cancer in a subject comprising: a) inducing an immune response specific for an antigen derived from the oncogenic virus in the subject, comprising administering to the subject a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding the antigen derived from the oncogenic virus, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence; and b) administering to the subject intratumorally a second composition comprising a pharmaceutically suitable adjuvant, wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, and wherein the first composition and the second composition are administered concurrently or sequentially. In one embodiment, administering the second composition intratumorally induces expression of one or more cytokines or chemokines capable of recruiting T cells, in particular antigen-specific T cells (e.g., CD4 and/or CD8 T cells, effector memory T cells (T_{EM}), central memory T cells (T_{CM}) and/or tissue-resident memory T cells (T_{RM})) to the tumor.

[0029] ~~Another aspect of t~~The present disclosure ~~invention~~ additionally provides a method of treating a cancer comprising: a) inducing an immune response specific for a cancer-associated antigen in a subject, comprising administering to the subject a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding the cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence; and b) administering intra-tumorally to the subject a second composition comprising a pharmaceutically suitable adjuvant; wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, and wherein the first composition and second composition are administered concurrently or sequentially.

[0030] In certain embodiments of the methods of treating cancer, including oncovirus-induced cancers, the first composition and/or the second composition are administered 2, 3, 4, 5, 6, 7, 8, 9 or 10 times. In other embodiments, the recombinant expression vector is selected from a retroviral vector genome, a lentiviral vector genome, poxvirus vector genome, vaccinia virus vector genome, adenovirus vector genome, adenovirus-associated virus vector genome, herpes virus vector genome, alpha virus vector genome, plasmid DNA and RNA. In

[0034] ~~Another aspect of t~~The present disclosure also provides a method of increasing T cells in the tumor microenvironment comprising, a) administering to a subject having a tumor a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, thereby inducing an immune response against the cancer-associated antigen in the subject; and b) administering intratumorally or peritumorally to the subject a second composition comprising a TLR4 agonist wherein the composition does not comprise antigen; wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, and wherein the first composition and second composition are administered concurrently or sequentially; thereby increasing T cells in the tumor microenvironment. In certain embodiments, the vector particle is a lentiviral vector particle that comprises the lentiviral vector genome; a poxvirus vector particle that comprises the poxvirus vector genome; a vaccinia virus vector particle that comprises the vaccinia virus vector genome; an adenovirus vector particle that comprises the adenovirus vector genome; an adenovirus-associated virus vector particle that comprises the adenovirus-associated virus vector genome; a herpes virus vector particle that comprises the herpes virus vector genome; or an alpha virus vector particle that comprises the alpha virus vector genome. In another embodiment, the vector particle is the lentiviral vector particle and comprises the lentiviral vector genome. In additional embodiments, the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein and in some embodiments, delivers the recombinant expression vector to a dendritic cell. In some embodiments of the method, the TLR4 agonist is a non-toxic lipid A-related adjuvant such as GLA described herein and may be formulated in stable oil-in-water emulsion.

BRIEF DESCRIPTION OF THE DRAWINGS

[0035] **Figure 1**: Prime-Pull Vaccination Strategy. C57BL/6 mice (5 mice per group) were inoculated with B16F10-OVA cells on Day 0. Ten days post-tumor inoculation, tumor-bearing mice were immunized with VP02/OVA or VP02/GFP (control vector) at the tail base.

CLAIMS

We claim the following:

1. A composition comprising

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, thereby inducing an immune response against the cancer-associated antigen in the subject; and

b) a second composition comprising a TLR4 agonist; wherein the second composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, for use in a method of treating a cancer wherein the first composition is to be administered systemically to the subject; and wherein the second composition is to be administered intratumorally, peritumorally, intranodally or intravesically to the subject; and wherein the first composition and second composition are to be administered concurrently or sequentially.

2. The composition of claim 1, wherein the cancer associated antigen is selected from CTA, NY-ESO-1, LAGE-1,MAGE-A1, MAGE-A3, MAGE-A4, MAGE-A10, CT7, CT10, GAGE, PRAME; BAGE; RAGE, SAGE, HAGE, MPHOSPH1, DEPDC1, IMP3, MAGE-A, BK T-antigen, p53, Ras, c-Myc, A-Raf, B-Raf, C-Raf, cyclin-dependent kinases, MAGE-A1, MAGE-A2, MAGE-A3, MAGE-A4, MAGE-A6, MAGE-A10, MAGE-A12, MART-1, BAGE, DAM-6, -10, GAGE-1, -2, -8, GAGE-3, -4, -5, -6, -7B, NA88-A, MART-1, MC1R, Gp100, PSA, PSM, Tyrosinase, TRP-1, TRP-2, ART-4, CAMEL, CEA, Cyp-B, hTERT, hTRT, iCE, MUC1, MUC2, Phosphoinositide 3-kinases (PI3Ks), TRK receptors, PRAME, P15, RU1, RU2, SART-1, SART-3, Wilms' tumor antigen (WT1), AFP, β-catenin/m, Caspase-8/m, CEA, CDK-4/m, ELF2M, GnT-V, G250, HSP70-2M, HST-2, KIAA0205, MUM-1, MUM-2, MUM-3, Myosin/m, RAGE, SART-2, TRP-2/INT2, 707-AP, Annexin II, CDC27/m, TPI/mbcr-abl, BCR-ABL, interferon regulatory factor 4 (IRF4),

ETV6/AML, LDLR/FUT, Pml/RARα, Tumor-associated calcium signal transducer 1 (TACSTD1) TACSTD2, receptor tyrosine kinases, Epidermal Growth Factor receptor (EGFR), EGFRvIII, platelet derived growth factor receptor (PDGFR), vascular endothelial growth factor receptor (VEGFR), cytoplasmic tyrosine kinases, src-family, syk-ZAP70, integrin-linked kinase (ILK), signal transducers and activators of transcription STAT3, STAT5, and STAT6, hypoxia inducible factors, HIF-1α and HIF-2α, Nuclear Factor-Kappa B (NF-κB), Notch receptors, Notch1-4, c-Met, mammalian targets of rapamycin (mTOR), WNT, extracellular signal-regulated kinases (ERKs), PMSA, PR-3, MDM2, Mesothelin, renal cell carcinoma – 5T4, SM22-alpha, carbonic anhydrases I (CAI) and IX (CAIX), STEAD, TEL/AML1, GD2, proteinase3, hTERT, sarcoma translocation breakpoints, EphA2, ML-IAP, EpCAM, ERG (TMPRSS2 ETS fusion gene), NA17, PAX3, ALK, androgen receptor, cyclin B1, polysialic acid, MYCN, RhoC, GD3, fucosyl GM1, mesothelian, PSCA, sLe, PLAC1, GM3, BORIS, Tn, GLoboH, NY-BR-1, RGs5, SART3, STn, PAX5, OY-TES1, sperm protein 17, LCK, HMWMAA, AKAP-4, SSX2, XAGE 1, B7H3, legumain, TIE2, Page4, MAD-CT-1, FAP, MAD-CT-2, and fos related antigen 1.

3. The composition of any one of claims 1 - 2, wherein the first composition is to be administered systemically via a first route selected from the group consisting of intramuscular, intradermal or subcutaneous.

4. The composition of any one of claims 1 - 3 wherein the TLR4 agonist is a non-toxic lipid A-related adjuvant.

5. The composition of claim 4, wherein the non-toxic lipid A-related adjuvant is glucopyranosyl lipid A (GLA).

6. The composition of claim 5, wherein GLA is formulated in a stable oil-in-water emulsion.

7. The composition of any one of claims 4 - 5 wherein the GLA has the formula:

(I)

L7, L8, L9 and L10 are the same or different, and at any occurrence may be either absent or –C(=O)-; Y1 is an acid functional group; Y2 and Y3 are the same or different and are each independently selected from –OH, -SH, and an acid functional group; Y4 is –OH or –SH; R1, R3, R5 and R6 are the same or different and are each independently selected from the group of C8-C13 alkyl; and R2 and R4 are the same or different and are each independently selected from the group of C6-C11 alkyl.

10. The composition of any one of claims 1 - 9, wherein the first composition is to be administered prior to administration of the second composition.

11. The composition of any one of claims 1 - 9 wherein the first composition and/or the second composition are to be administered 2, 3, 4, 5, 6, 7, 8, 9 or 10 times.

12. The composition of any one of claims 1 - 9, wherein the recombinant expression vector is selected from a retroviral vector genome, a lentiviral vector genome, poxvirus vector genome, vaccinia virus vector genome, adenovirus vector genome, adenovirus-associated virus vector genome, herpes virus vector genome, alpha virus vector genome, plasmid DNA and RNA.

13. The composition of claim 12, wherein the vector particle is a lentiviral vector particle that comprises the lentiviral vector genome; a poxvirus vector particle that comprises the poxvirus vector genome; a vaccinia virus vector particle that comprises the vaccinia virus vector genome; an adenovirus vector particle that comprises the adenovirus vector genome; an adenovirus-associated virus vector particle that comprises the adenovirus-associated virus vector genome; a herpes virus vector particle that comprises the herpes virus vector genome; or an alpha virus vector particle that comprises the alpha virus vector genome.

14. The composition of claim 13, wherein the vector particle is the lentiviral vector particle and comprises the lentiviral vector genome.

15. The composition of claim 13 or 14, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein.

16. The composition of claim 15, wherein the lentiviral vector particle delivers the recombinant expression vector to a dendritic cell.

17. The composition of any one of the preceding claims, wherein the cancer is an oncogenic virus-induced cancer.

18. The composition of claim 17, wherein the cancer-associated antigen is an antigen derived from the oncogenic virus.

19. The composition of any one of claims 1 - 18, wherein the second composition is to be administered in combination with a composition comprising a therapeutic agent selected from the group consisting of a checkpoint inhibitor, a cytokine, chloroquine, an antibody that increase ADCC, an antibody that promotes a co-stimulatory signal, an anti-CD40 antibody.

20. The composition of any one of the preceding claims, wherein the first composition further comprises an adjuvant.

21. The composition of any one of the preceding claims, wherein ~~further comprising administering~~ a boosting composition comprising an adjuvant in combination with a polypeptide is to be further administered, wherein the polypeptide comprises the cancer-associated antigen, or an immunogenic fragment thereof.

22. The composition of any one of the preceding claims, wherein the induced immune response comprises a cytotoxic T lymphocyte immune response.

23. The composition of any one of the preceding claims, wherein administering the second composition induces an increase in CD8+ T cells at the site of administration.

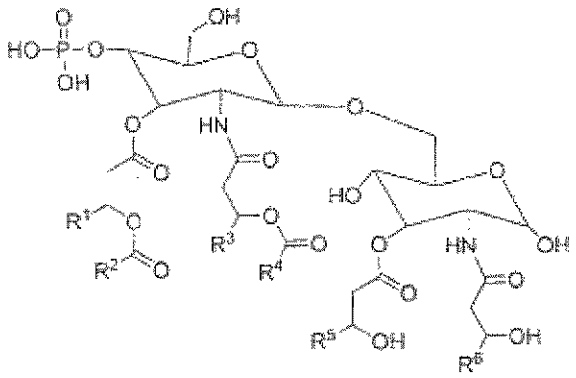
24. A kit comprising a first composition and a second composition according to any one of the preceding claims.

25. A composition comprising:

a) a first composition comprising a lentiviral vector particle, the lentiviral vector particle comprising a recombinant expression vector, wherein the recombinant expression vector comprises a polynucleotide encoding an immunogen associated with cancer, wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, wherein the lentiviral vector particle further comprises an envelope comprising a

Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein; and

b) a second composition comprising a TLR4 agonist, wherein the second composition does not comprise an immunogen; wherein the TLR4 agonist is glucopyranosyl lipid A (GLA) formulated in a stable oil-in-water emulsion, wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient,

for use in a method of treating a cancer in a subject, wherein the first composition is to be administered intramuscularly, intradermally or subcutaneously and induces an immune response specific for the immunogen associated with the cancer in the subject; wherein the second composition is to be administered intratumorally or peritumorally; and wherein the first composition or second composition are to be administered concurrently or sequentially.

26. A composition comprising:

a) a first composition comprising

i. antigen-specific T cells specific for a cancer-associated antigen, said antigen-specific T cells having been expanded *ex vivo*,

ii. T cells that have been genetically modified to express a chimeric antigen receptor (CAR) that recognizes the cancer-associated antigen, or

iii. T cells that have been genetically modified to express a specific chimeric T cell receptor (TCR) that recognizes the cancer-associated antigen in the context of MHC; and

b) a second composition comprising a pharmaceutically suitable adjuvant; wherein the second composition does not comprise an immunogen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient;

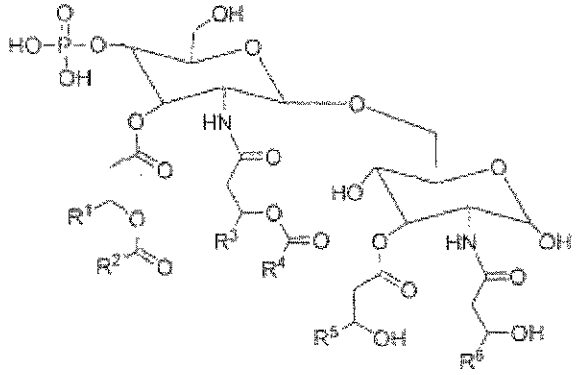
for use in a method of treating a cancer in a subject wherein the first composition is to be administered systemically to the subject; and the second composition is to be administered intratumorally and wherein the first composition and second composition are to be administered concurrently or sequentially.

27. The composition of claim 26, wherein the adjuvant is a non-toxic lipid A-related adjuvant.

28. The composition of claim 26, wherein the adjuvant is glucopyranosyl lipid A (GLA).

29. The composition of claim 28, wherein GLA is formulated in a stable oil-in-water emulsion.

30. The composition of claim 28 or claim 29 wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 are C_{11}-C_{20} alkyl; and R^2 and R^4 are C_{12}-C_{20} alkyl.

31. The composition of claim 30 wherein R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl.

32. A composition comprising:

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, and

b) a second composition comprising a TLR4 agonist wherein the composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, for use in a method of increasing T cells in a tumor microenvironment in a subject, where the first composition is to be administered systemically, thereby inducing an immune response against the cancer-associated antigen in the subject; and where the second composition is to be administered intratumorally or peritumorally to the subject; and where the first and second composition are to be administered concurrently or sequentially; thereby increasing T cells in the tumor microenvironment.

33. The composition of claim 32~~[00373]~~, wherein the vector particle is a lentiviral vector particle that comprises the lentiviral vector genome; a poxvirus vector particle that comprises the poxvirus vector genome; a vaccinia virus vector particle that comprises the vaccinia virus vector genome; an adenovirus vector particle that comprises the adenovirus vector genome; an adenovirus-associated virus vector particle that comprises the adenovirus-associated virus vector genome; a herpes virus vector particle that comprises the herpes virus vector genome; or an alpha virus vector particle that comprises the alpha virus vector genome.

34. The composition of claim 33, wherein the vector particle is the lentiviral vector particle and comprises the lentiviral vector genome.

35. The composition of claim 33 or 34, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an

amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein.

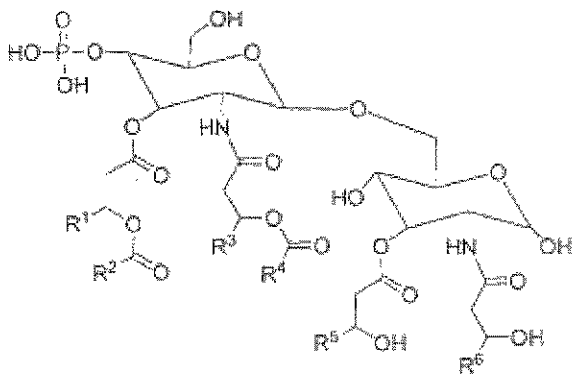
36. The composition of claim 35, wherein the lentiviral vector particle delivers the recombinant expression vector to a dendritic cell.

37. The composition of claim 32~~[00373]~~, wherein the TLR4 agonist is a non-toxic lipid A-related adjuvant.

38. The composition of claim 32~~[00373]~~, wherein the TLR4 agonist is glucopyranosyl lipid A (GLA).

39. The composition of claim 38, wherein the GLA is formulated in a stable oil-in-water emulsion.

40. The composition of claim 38 or claim 39 wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 are C_{11}-C_{20} alkyl; and R^2 and R^4 are C_{12}-C_{20} alkyl.

41. The composition of claim 40 wherein R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl.

42. Use of a composition comprising

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, thereby inducing an immune response against the cancer-associated antigen in the subject; and

b) a second composition comprising a TLR4 agonist; wherein the second composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, in the manufacture of a medicament for treating a cancer wherein the first composition is to be administered systemically to the subject; and wherein the second composition is to be administered intratumorally, peritumorally, intranodally or intravesically to the subject; and wherein the first composition and second composition are to be administered concurrently or sequentially.

43. The use of claim 42, wherein the cancer associated antigen is selected from CTA, NY-ESO-1, LAGE-1,MAGE-A1, MAGE-A3, MAGE-A4, MAGE-A10, CT7, CT10, GAGE, PRAME; BAGE; RAGE, SAGE, HAGE, MPHOSPH1, DEPDC1, IMP3, MAGE-A, BK T-antigen, p53, Ras, c-Myc, A-Raf, B-Raf, C-Raf, cyclin-dependent kinases, MAGE-A1, MAGE-A2, MAGE-A3, MAGE-A4, MAGE-A6, MAGE-A10, MAGE-A12, MART-1, BAGE, DAM-6, -10, GAGE-1, -2, -8, GAGE-3, -4, -5, -6, -7B, NA88-A, MART-1, MC1R, Gp100, PSA, PSM, Tyrosinase, TRP-1, TRP-2, ART-4, CAMEL, CEA, Cyp-B, hTERT, hTRT, iCE, MUC1, MUC2, Phosphoinositide 3-kinases (PI3Ks), TRK receptors, PRAME, P15, RU1, RU2, SART-1, SART-3, Wilms' tumor antigen (WT1), AFP, β-catenin/m, Caspase-8/m, CEA, CDK-4/m, ELF2M, GnT-V, G250, HSP70-2M, HST-2, KIAA0205, MUM-1, MUM-2, MUM-3, Myosin/m, RAGE, SART-2, TRP-2/INT2, 707-AP, Annexin II, CDC27/m, TPI/mbcr-abl, BCR-ABL, interferon regulatory factor 4 (IRF4), ETV6/AML, LDLR/FUT, Pml/RARα, Tumor-associated calcium signal transducer 1 (TACSTD1) TACSTD2, receptor tyrosine kinases, Epidermal Growth Factor receptor (EGFR), EGFRvIII, platelet derived growth factor receptor (PDGFR), vascular endothelial growth factor receptor (VEGFR), cytoplasmic tyrosine kinases, src-family, syk-ZAP70, integrin-linked kinase (ILK), signal transducers and activators of transcription STAT3, STAT5, and STAT6,

hypoxia inducible factors, HIF-1α and HIF-2α, Nuclear Factor-Kappa B (NF-κB), Notch receptors, Notch1-4, c-Met, mammalian targets of rapamycin (mTOR), WNT, extracellular signal-regulated kinases (ERKs), PMSA, PR-3, MDM2, Mesothelin, renal cell carcinoma – 5T4, SM22-alpha, carbonic anhydrases I (CAI) and IX (CAIX), STEAD, TEL/AML1, GD2, proteinase3, hTERT, sarcoma translocation breakpoints, EphA2, ML-IAP, EpCAM, ERG (TMPRSS2 ETS fusion gene), NA17, PAX3, ALK, androgen receptor, cyclin B1, polysialic acid, MYCN, RhoC, GD3, fucosyl GM1, mesothelian, PSCA, sLe, PLAC1, GM3, BORIS, Tn, GLoboH, NY-BR-1, RGs5, SART3, STn, PAX5, OY-TES1, sperm protein 17, LCK, HMWMAA, AKAP-4, SSX2, XAGE 1, B7H3, legumain, TIE2, Page4, MAD-CT-1, FAP, MAD-CT-2, and fos related antigen 1.

44. The use of any one of claims 42-43, wherein the first composition is to be administered systemically via a first route selected from the group consisting of intramuscular, intradermal or subcutaneous.

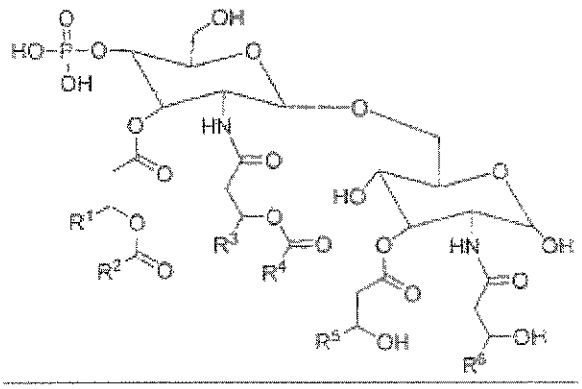
45. The use of any one of claims 42-44 wherein the TLR4 agonist is a non-toxic lipid A-related adjuvant.

46. The use of claim 45, wherein the non-toxic lipid A-related adjuvant is glucopyranosyl lipid A (GLA).

47. The use of claim 46, wherein GLA is formulated in a stable oil-in-water emulsion.

48. The use of any one of claims 45-46 wherein the GLA has the formula:

(I)

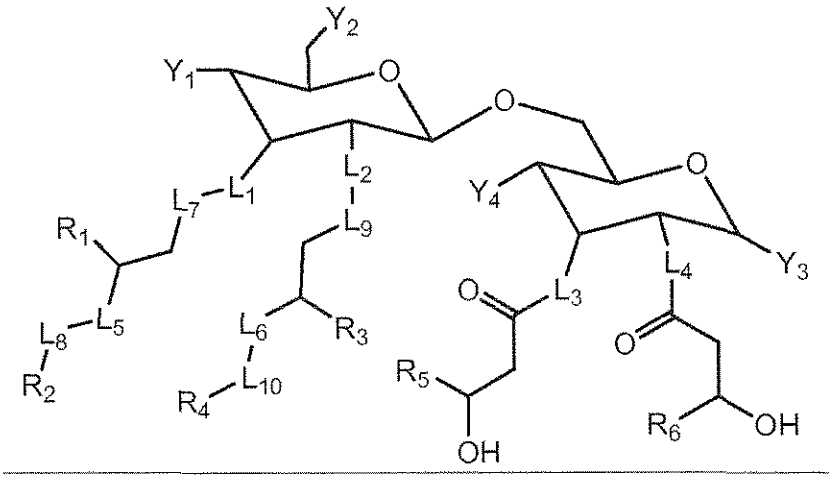


where: R^1, R^3, R^5 and R^6 are C_{11}-C_{20} alkyl; and R^2 and R^4 are C_{12}-C_{20} alkyl.

49. The use of claim 48 wherein R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl.

50. The use of any one of claims 45-46 wherein the GLA has the formula:

(II)



or a pharmaceutically acceptable salt thereof, wherein: L1, L2, L3, L4, L5 and L6 are the same or different and are independently selected from –O-, -NH-, and –(CH2)-; L7, L8, L9 and L10 are the same or different, and at any occurrence may be either absent or –C(=O)-; Y1 is an acid functional group; Y2 and Y3 are the same or different and are each independently selected from –OH, -SH, and an acid functional group; Y4 is –OH or –SH; R1, R3, R5 and R6 are the same or different and are each independently selected from the group of C8-C13 alkyl; and R2 and R4 are the same or different and are each independently selected from the group of C6-C11 alkyl.

51. The use of any one of claims 42-50, wherein the first composition is to be administered prior to administration of the second composition.

52. The use of any one of claims 42-50 wherein the first composition and/or the second composition are to be administered 2, 3, 4, 5, 6, 7, 8, 9 or 10 times.

53. The use of any one of claims 42-50, wherein the recombinant expression vector is selected from a retroviral vector genome, a lentiviral vector genome, poxvirus vector genome, vaccinia virus vector genome, adenovirus vector genome, adenovirus-associated virus vector genome, herpes virus vector genome, alpha virus vector genome, plasmid DNA and RNA.

54. The use of claim 53, wherein the vector particle is a lentiviral vector particle that comprises the lentiviral vector genome; a poxvirus vector particle that comprises the poxvirus vector genome; a vaccinia virus vector particle that comprises the vaccinia virus vector genome; an adenovirus vector particle that comprises the adenovirus vector genome; an adenovirus-associated virus vector particle that comprises the adenovirus-associated virus vector genome; a herpes virus vector particle that comprises the herpes virus vector genome; or an alpha virus vector particle that comprises the alpha virus vector genome.

55. The use of claim 53, wherein the vector particle is the lentiviral vector particle and comprises the lentiviral vector genome.

56. The use of claim 53 or 54, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein.

57. The use of claim 56, wherein the lentiviral vector particle delivers the recombinant expression vector to a dendritic cell.

58. The use of any one of claims 42-57, wherein the cancer is an oncogenic virus-induced cancer.

59. The use of claim 58, wherein the cancer-associated antigen is an antigen derived from the oncogenic virus.

60. The use of any one of claims 42-59, wherein the second composition is to be administered in combination with a composition comprising a therapeutic agent selected from

the group consisting of a checkpoint inhibitor, a cytokine, chloroquine, an antibody that increase ADCC, an antibody that promotes a co-stimulatory signal, an anti-CD40 antibody.

61. The use of any one of claims 42-60, wherein the first composition further comprises an adjuvant.

62. The use of any one of claims 42-61, wherein a boosting composition comprising an adjuvant is to be administered in combination with a polypeptide, wherein the polypeptide comprises the cancer-associated antigen, or an immunogenic fragment thereof.

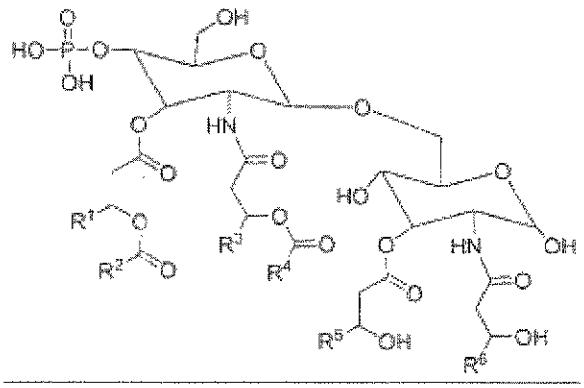
63. The use of any one of claims 42-62, wherein the induced immune response comprises a cytotoxic T lymphocyte immune response.

64. The use of any one of claims 42-63, wherein administering the second composition induces an increase in CD8+ T cells at the site of administration.

65. Use of a composition comprising:

a) a first composition comprising a lentiviral vector particle, the lentiviral vector particle comprising a recombinant expression vector, wherein the recombinant expression vector comprises a polynucleotide encoding an immunogen associated with cancer, wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein; and

b) a second composition comprising a TLR4 agonist, wherein the second composition does not comprise an immunogen; wherein the TLR4 agonist is glucopyranosyl lipid A (GLA) formulated in a stable oil-in-water emulsion, wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient,

in the manufacture of a medicament for treating a cancer in a subject, wherein the first composition is to be administered intramuscularly, intradermally or subcutaneously and induces an immune response specific for the immunogen associated with the cancer in the subject; wherein the second composition is to be administered intratumorally or peritumorally; and wherein the first composition or second composition are to be administered concurrently or sequentially.

66. Use of a composition comprising:

a) a first composition comprising

i. antigen-specific T cells specific for a cancer-associated antigen, said antigen-specific T cells having been expanded *ex vivo*,

ii. T cells that have been genetically modified to express a chimeric antigen receptor (CAR) that recognizes the cancer-associated antigen, or

iii. T cells that have been genetically modified to express a specific chimeric T cell receptor (TCR) that recognizes the cancer-associated antigen in the context of MHC; and

b) a second composition comprising a pharmaceutically suitable adjuvant; wherein the second composition does not comprise an immunogen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient;

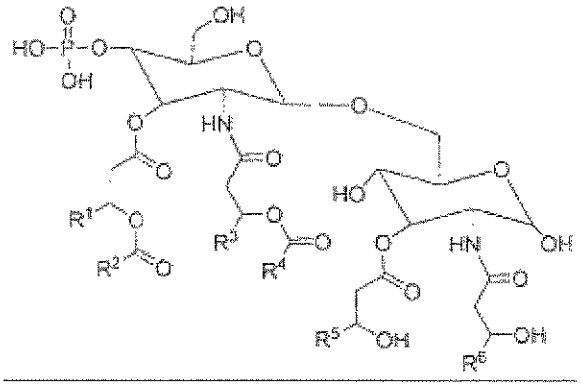
in the manufacture of a medicament for treating a cancer in a subject wherein the first composition is to be administered systemically to the subject; and the second composition is to be administered intratumorally and wherein the first composition and second composition are to be administered concurrently or sequentially.

67. The use of claim 66, wherein the adjuvant is a non-toxic lipid A-related adjuvant.

68. The use of claim 66, wherein the adjuvant is glucopyranosyl lipid A (GLA).

69. The use of claim 68, wherein GLA is formulated in a stable oil-in-water emulsion.

70. The use of claim 68 or claim 69 wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 are C_{11}-C_{20} alkyl; and R^2 and R^4 are C_{12}-C_{20} alkyl.

71. The use of claim 70 wherein R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl.

72. Use of a composition comprising:

a) a first composition comprising a vector particle, the vector particle comprising a recombinant expression vector wherein the recombinant expression vector comprises a polynucleotide encoding a cancer-associated antigen, and wherein the polynucleotide is operatively linked to at least one regulatory expression sequence, and

b) a second composition comprising a TLR4 agonist wherein the composition does not comprise antigen;

wherein the first composition and the second composition each further comprise a pharmaceutically suitable excipient, in the manufacture of a medicament for increasing T cells in a tumor microenvironment in a subject, where the first composition is to be administered systemically, thereby inducing an immune response against the cancer-associated antigen in the subject; and where the second composition is to be administered intratumorally or peritumorally to the subject; and where the first and second composition are to be administered concurrently or sequentially; thereby increasing T cells in the tumor microenvironment.

73. The use of claim 72, wherein the vector particle is a lentiviral vector particle that comprises the lentiviral vector genome; a poxvirus vector particle that comprises the poxvirus vector genome; a vaccinia virus vector particle that comprises the vaccinia virus vector genome; an adenovirus vector particle that comprises the adenovirus vector genome; an adenovirus-associated virus vector particle that comprises the adenovirus-associated virus vector genome; a herpes virus vector particle that comprises the herpes virus vector genome; or an alpha virus vector particle that comprises the alpha virus vector genome.

74. The use of claim 73, wherein the vector particle is the lentiviral vector particle and comprises the lentiviral vector genome.

75. The use of claim 73 or 74, wherein the lentiviral vector particle further comprises an envelope comprising a Sindbis virus E2 glycoprotein having at least one amino acid change compared to SEQ ID NO:1, wherein residue 160 is either absent or an amino acid other than glutamic acid, and wherein E2 glycoprotein is not part of a fusion protein with Sindbis virus E3 protein.

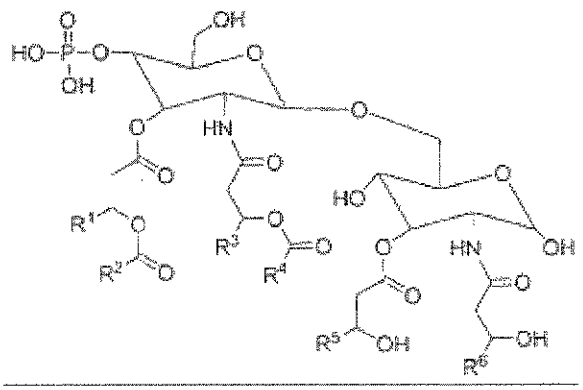
76. The use of claim 75, wherein the lentiviral vector particle delivers the recombinant expression vector to a dendritic cell.

77. The use of claim 72, wherein the TLR4 agonist is a non-toxic lipid A-related adjuvant.

78. The use of claim 72, wherein the TLR4 agonist is glucopyranosyl lipid A (GLA).

79. The use of claim 78, wherein the GLA is formulated in a stable oil-in-water emulsion.

80. The use of claim 78 or claim 79 wherein the GLA has the formula:



where: R^1, R^3, R^5 and R^6 are C_{11}-C_{20} alkyl; and R^2 and R^4 are C_{12}-C_{20} alkyl.

81. The use of claim 80 wherein R^1, R^3, R^5 and R^6 equal to undecyl, and R^2 and R^4 equal to tridecyl.