

CLAIMS

1. An artificial antigen presenting cell (aAPC) comprising a polymeric nanoparticle having attached polypeptide ligands for immune cell activation, the nanoparticle comprising:
a PLGA or PLA polymer core, and
5 a hydrophilic shell formed of PEG brushes, wherein a portion of the PEG polymers have a terminal attachment of a polypeptide ligand.
2. The aAPC of claim 1, wherein the hydrophilic shell further comprises a mixture of PEG brushes having the same or different molecular weights.
- 10 3. The aAPC of claim 1 or 2, wherein the polymer core is based on 1:0 to 1:1 Lactic Acid:Glycolic Acid ratio.
4. The aAPC of claim 3, wherein the polymer core is PLGA having about 1:1 Lactic
15 Acid:Glycolic Acid.
5. The aAPC of claim 3, wherein the polymer core is PLA.
6. The aAPC of any one of claims 1 to 5, wherein the core polymer has a molecular
20 weight of from about 10K to about 50K.
7. The aAPC of any one of claims 1 to 6, wherein the polymeric nanoparticle comprises a PLGA or PLA polymer core, and PLGA-PEG and/or PLA-PEG block co-polymers, the PEG portion forming the hydrophilic shell.
- 25 8. The aAPC of claim 7, wherein the polypeptide ligands are attached through functional groups at the PEG terminus.

9. The aAPC of claim 8, wherein the polypeptide ligands are attached to PEG through primary amines on the polypeptides.

10. The aAPC of claim 8, wherein the polypeptide ligands are attached through an
5 unpaired cysteine side chain.

11. The aAPC of any one of claims 1 to 10, wherein the PLGA-PEG co-polymer contains a PLGA portion having a molecular weight of from about 10K to about 50K, and a PEG portion having molecular weight(s) of from about 2K to about 10K.
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12. The aAPC of claim 11, wherein the PLGA polymer has a molecular weight of about 20K, and the PEG polymer has a molecular weight of about 3K and/or about 5K.

13. The aAPC of claim 11 or 12, wherein the particle comprises PLGA-PEG-MeOH co-
15 polymer, which contains a PLGA portion having a molecular weight of from about 10K to about 30K, and a PEG-MeOH portion having a molecular weight of from about 2K to about 10K.

14. The aAPC of claim 13, wherein the PLGA portion of the PLGA-PEG has a molecular
20 weight of about 20K, and the PEG-MeOH portion has a molecular weight of from about 3K to 5K.

15. The aAPC of claim 13 or 14, wherein the PLGA-PEG and PLGA-PEG-MeOH are present at a ratio of from about 1:15 to about 15:1.
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16. The aAPC of claim 15, wherein the PLGA-PEG and PLGA-PEG-MeOH are present at from about 1:10 to about 2:1.

17. The aAPC of any one of claims 13 to 16, wherein the polymers are present at about 50% PLGA, and at least about 25% PLGA-PEG-MeOH.
18. The aAPC of any one of claims 1 to 10, wherein the PLA-PEG co-polymer contains a
5 PLA portion having a molecular weight of from about 10K to about 50K, and a PEG portion having molecular weight(s) of from about 2K to about 10K.
19. The aAPC of claim 18, wherein the PLA portion of the PLA-PEG has a molecular weight of about 20K, and the PEG portion has a molecular weight of about 3K and/or about
10 5K.
20. The aAPC of claim 18 or 19, wherein the particle comprises PLA-PEG-MeOH co-polymer, which contains a PLA portion having a molecular weight of from about 10K to about 30K, and a PEG-MeOH portion having a molecular weight of from about 2K to about
15 10K.
21. The aAPC of claim 20, wherein the PLA portion of the PLA-PEG polymer has a molecular weight of about 20K, and the PEG-MeOH portion has a molecular weight of from 3K to 5K.
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22. The aAPC of claim 20 or 21, wherein the PLA-PEG and PLA-PEG-MeOH are present at a ratio of from about 1:15 to about 15:1.
23. The aAPC of claim 22, wherein the PLA-PEG and PLA-PEG-MeOH are present at
25 from about 1:10 to about 2:1.
24. The aAPC of any one of claims 21 to 23, wherein the polymers are present at about 50% PLA, and at least about 25% PLA-PEG-MeOH.

25. The aAPC of any one of claims 1 to 24, wherein the nanoparticles have a size in the range of about 20 to 200 nm.

26. The aAPC of any one of claims 1 to 25, wherein the aAPC has a surface charge of about 0 to -20 mV.

27. The aAPC of claim 26, wherein the aAPC has a surface charge of from about -5 to about -10 mV.

28. The aAPC of any one of claims 1 to 27, having less than about 200 ligands per particle.

29. The aAPC of any one of claims 1 to 28, wherein the polypeptide ligands comprise peptide-HLA ligands, and one or more of anti-CD28 or anti-4-1BB ligands.

30. The aAPC of claim 29, comprising an anti-CD28 antibody ligand having a human IGHV4-59 germline framework optionally having from 5 to 15 murine framework residues, and a IGKV4-01 germline framework optionally having from 3 to 15 murine framework residues.

31. The aAPC of claim 30, wherein the anti-CD28 ligand comprises an antigen-binding antibody fragment or portion.

32. The aAPC of claim 31, wherein the anti-CD28 ligand comprises a scFv.

33. The aAPC of claim 29, wherein the HLA ligand is dimeric.

34. The aAPC of claim 33, wherein the HLA is HLA-A*02:01.

35. The aAPC of claim 33 or 34, wherein the HLA comprises a fusion with immunoglobulin sequences.
- 5 36. The aAPC of claim 35, wherein the anti-CD28 antibody ligand and the HLA immunoglobulin fusion have an IgG4 constant region with mutations at S241 and L248, and an unpaired cysteine at codon 473.
- 10 37. The aAPC of claim 36, wherein the anti-CD28 antibody ligands are conjugated to the particles through the unpaired cysteine at codon 473 of the immunoglobulin heavy chain.
38. The aAPC of claim 36 or 37, wherein the HLA ligands are conjugated to the particles through the unpaired cysteine at codon 473 of the immunoglobulin heavy chain.
- 15 39. The aAPC of claim 35, wherein the HLA ligands comprise an extracellular domain of HLA fused to an immunoglobulin hinge region, without immunoglobulin variable domain sequences.
- 20 40. A method for inducing the formation of antigen-specific cytotoxic T cells, comprising administering a composition comprising the aAPC of any one of claims 1 to 39 to a patient.
41. The method of claim 40, wherein the patient is a cancer patient.
- 25 42. The method of claim 40 or 41, wherein the patient is undergoing or has undergone therapy with one or more checkpoint inhibitors.
43. The method of claim 40 or 41, wherein the patient is undergoing adoptive T cell therapy.

44. The method of any one of claims 40 to 43, wherein the aAPC composition is administered by intravenous administration, intra-arterial administration, subcutaneous administration, intradermal administration, intralymphatic administration, or intra-tumoral administration.
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Claims

1. An artificial antigen presenting cell (aAPC) comprising a polymeric nanoparticle having attached polypeptide ligands suitable for activating or inhibiting antigen-specific T cells, the nanoparticle having a size in the range of 20 to 200 nm, the nanoparticle comprising:

5 a poly(lactic-co-glycolic acid) (PLGA) or polylactic acid (PLA) polymer core (PLGA/PLA), and

a hydrophilic shell formed of polyethylene glycol (PEG),

wherein the polymeric nanoparticle comprises PLGA/PLA-PEG and PLGA/PLA-methoxyPEG (mPEG) block co-polymers with the PLGA/PLA portion having
10 molecular weights of from about 10 kDa to about 30 kDa and the PEG portion having molecular weight(s) of from about 2 kDa to about 10 kDa,

wherein a portion of the PLGA/PLA-PEG polymers have a terminal attachment of a polypeptide ligand, and

wherein the polypeptide ligands comprise HLA ligands presenting one or more peptide
15 antigens.

2. The aAPC of claim 1, wherein the PEG portions of the block co-polymers have a molecular weight of from about 3000 and/or about 5000 Da.

20 3. The aAPC of claim 1, wherein the PLGA/PLA portions of the block co-polymers have a molecular weight of about 20,000 Da.

4. The aAPC of claim 1, wherein the polymer core is based on a 20:1 to 1:20 Lactic Acid:Glycolic Acid ratio.
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5. The aAPC of claim 1, wherein the polymer core comprises PLA.

6. The aAPC of claim 1, wherein the polypeptide ligands are attached to PEG through primary amines on the polypeptides.
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7. The aAPC of claim 1, wherein the polypeptide ligands are attached to PEG maleimide functional groups through an unpaired cysteine side chain.

8. The aAPC of claim 1, wherein the aAPC has a surface charge of about 0 to -20 mV.

9. The aAPC of claim 1, wherein the aAPC has a surface charge of from about -5 to about -10 mV.

10. The aAPC of claim 1, having less than about 500 polypeptide ligands per particle.

11. The aAPC of claim 1, having less than about 300 polypeptide ligands per particle.

12. The aAPC of claim 1, wherein the polypeptide ligands further comprise a co-stimulatory or inhibitory ligand.

13. The aAPC of claim 12, wherein the polypeptide ligands comprise an anti-CD28 agonistic antibody or an antigen-binding portion thereof.

14. The aAPC of claim 12, wherein the polypeptide ligands comprise an antibody that binds to Fas.

15. The aAPC of claim 1, wherein the HLA ligands are dimeric.

16. The aAPC of claim 15, wherein the HLA polypeptide ligands are fused with co-stimulatory or inhibitory ligands.

17. The aAPC of claim 15, wherein the HLA comprises a fusion with immunoglobulin sequences.

18. The aAPC of claim 17, wherein the aAPC comprises an anti-CD28 antibody ligand, and each of the HLA immunoglobulin fusion polypeptide ligands and the anti-CD28 antibody ligands

have an IgG4 constant region with mutations at codons S241 and L248, and an unpaired cysteine at codon 473.

19. The aAPC of claim 17, wherein the HLA ligands comprise an extracellular domain of HLA
5 fused to an immunoglobulin hinge region, without immunoglobulin variable domain sequences.