

CLAIMS

1. A lipid nanoparticle comprising a polynucleotide encoding a human IL12 polypeptide, wherein the polynucleotide comprises an ORF encoding a human IL12B polypeptide operably linked to a human IL12A polypeptide, wherein the lipid nanoparticle comprises a molar ratio of about 20-60% ionizable amino lipid: 5-25% phospholipid: 25-55% sterol; and 0.5-15% PEG-modified lipid.
2. The lipid nanoparticle of claim 1, wherein the IL12B polypeptide is operably linked to the IL12A polypeptide by a peptide linker.
3. The lipid nanoparticle of claim 1 or 2, wherein the IL12B polypeptide is located at the 5' terminus of the IL12A polypeptide or the peptide linker, or wherein the IL12A polypeptide is located at the 5' terminus of the IL12B polypeptide or the peptide linker.
4. The lipid nanoparticle of any one of claims 1 to 3, wherein the peptide linker comprises a Gly/Ser linker.
5. The lipid nanoparticle of claim 4, wherein the Gly/Ser linker comprises $(G_nS)_m$, wherein n is 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, or 20 and m is 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, or 20..
6. The lipid nanoparticle of claim 5, wherein n is 6 and m is 1.
7. The lipid nanoparticle of any one of claims 1 to 6, wherein the ORF encodes a signal peptide. wherein the signal peptide is an human IL12B signal peptide.
8. The lipid nanoparticle of claim 7, wherein the human IL12B signal peptide comprises an amino acid sequence set forth in amino acids 1 to 22 of SEQ ID NO: 48.

9. The lipid nanoparticle of any one of claims 1 to 8, wherein the human IL12B polypeptide comprises an amino acid sequence set forth in amino acids 23 to 328 of SEQ ID NO: 48.

10. The lipid nanoparticle of any one of claims 1 to 9, wherein the human IL12A polypeptide comprises an amino acid sequence set forth in amino acids 336 to 532 of SEQ ID NO: 48.

11. The lipid nanoparticle of any one of claims 1 to 10, wherein the human IL12 polypeptide comprises an amino acid sequence set forth in SEQ ID NO: 48.

12. The lipid nanoparticle of any one of claims 1 to 11, wherein the polynucleotide comprises a miRNA binding site.

13. The lipid nanoparticle of claim 12, wherein the miRNA binding site is a miR-122 binding site, wherein the miR-122 binding site is a miR-122-3p or miR-122-5p binding site.

14. The lipid nanoparticle of claim 13, wherein the miRNA binding site is a miR-122-5p binding site comprising the sequence set forth in SEQ ID NO: 54.

15. The lipid nanoparticle of any one of claims 1 to 14, wherein the polynucleotide comprises a 3' UTR.

16. The lipid nanoparticle of claim 15, wherein the 3' UTR comprises a sequence set forth in SEQ ID NO:240.

17. The lipid nanoparticle of claim 15 or 16, wherein the miRNA binding site is located in the 3' UTR.

18. The lipid nanoparticle of any one of claims 1 to 17, wherein the polynucleotide comprises a 5' UTR.

19. The lipid nanoparticle of any one of claims 1 to 18, wherein the polynucleotide comprises a 5' terminal cap structure.

20. The lipid nanoparticle of claim 19, wherein the 5' terminal cap structure is a Cap0, Cap1, ARCA, inosine, N1-methyl-guanosine, 2'fluoro-guanosine, 7-deaza-guanosine, 8-oxo-guanosine, 2-amino-guanosine, LNA-guanosine, 2-azidoguanosine, Cap2, Cap4, 5' methylG cap, or an analog thereof.

21. The lipid nanoparticle of any one of claims 1 to 20, wherein the polynucleotide comprises a 3' polyA tail.

22. The lipid nanoparticle of any one of claims 1 to 21, wherein the polynucleotide comprises a nucleotide sequence at least 80%, at least 90%, at least 95%, at least 98%, or at least 99% identical to SEQ ID NO: 236 or 237.

23. The lipid nanoparticle of any one of claims 1 to 21, wherein the polynucleotide comprises the sequence set forth in SEQ ID NO: 236 or 237.

24. The lipid nanoparticle of any one of claims 1 to 23, wherein the polynucleotide comprises an ORF comprising at least one modified nucleoside.

25. The lipid nanoparticle of claim 24, wherein the at least one chemically modified nucleoside is (i) selected from the group consisting of pseudouridine, N1-methylpseudouridine, 5-methylcytosine, 5-methoxyuridine, and a combination thereof, or (ii) N1-methylpseudouridine.

26. The lipid nanoparticle of any of claim 1 to 25, wherein the ionizable amino lipid is Compound 18.

27. Use of a lipid nanoparticle of any one of claims 1 to 26 in the manufacture of a medicament for treating or delaying progression of cancer in an individual, wherein the medicament comprises the lipid nanoparticle, and wherein the treatment comprises the

medicament to be administered in combination with a composition comprising a checkpoint inhibitor polypeptide.

28. A kit comprising a medicament comprising a lipid nanoparticle of any one of claims 1 to 26, and a package insert comprising instructions for administration of the medicament alone or in combination with a composition comprising a checkpoint inhibitor polypeptide for use in treating or delaying progression of cancer in an individual.

29. The kit of claim 28, wherein the kit further comprises a package insert comprising instructions for administration of the first medicament and the second medicament for use in treating or delaying progression of cancer in an individual.

30. A composition comprising the lipid nanoparticle of any one of claims 1 to 26 for use in treating or delaying progression of cancer in an individual, wherein the treatment comprises said lipid nanoparticle to be administered in combination with a second composition, wherein the second composition comprises a checkpoint inhibitor polypeptide.

31. A pharmaceutical composition comprising the lipid nanoparticle of any one of claims 1 to 26 and a pharmaceutically acceptable carrier or excipient.

32. The pharmaceutical composition of claim 31, wherein the lipid nanoparticle is formulated for intratumoral administration.

33. A kit comprising a container comprising the pharmaceutical composition of claim 31 or claim 32 and a package insert comprising instructions for administration of the pharmaceutical composition for use in treating or delaying progression of cancer in an individual.

34. The kit of claim 33, wherein the package insert further comprises instructions for administration of the pharmaceutical composition in combination with a composition comprising

a checkpoint inhibitor polypeptide for use in treating or delaying progression of cancer in an individual.