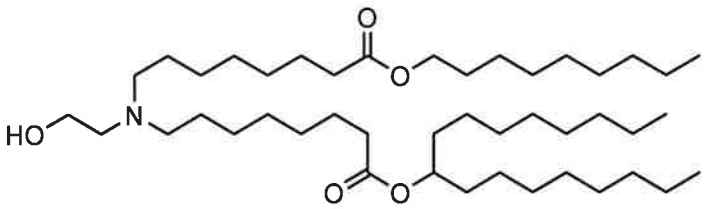
WHAT IS CLAIMED IS:

CLAIMS

1. A composition comprising an RNA polynucleotide having an open reading frame encoding at least one human relaxin protein formulated in an ionizable lipid nanoparticle, wherein the ionizable lipid nanoparticle comprises an ionizable lipid of Compound 18 having the structure:



a salt or a stereoisomers thereof, or any combination thereof.

2. The composition of claim 1, wherein the relaxin protein is a relaxin fusion protein.

3. The composition of claim 2, wherein the relaxin fusion protein includes an immunoglobulin (Ig) fragment.

4. The composition of claim 3, wherein the Ig fragment is a variable chain fragment.

5. The composition of claim 3, wherein the Ig fragment is a constant chain fragment.

6. The composition of claim 3, wherein the Ig fragment is a variable light chain fragment.

7. The composition of claim 6, wherein the relaxin fusion protein includes a variable light chain fragment a VLκ IgG region.

8. The composition of claim 1, wherein the relaxin protein has a nucleotide sequence that is 70%-100% identical to the sequence of SEQ ID NO. 1.

9. The composition of claim 1, wherein the relaxin protein has a nucleotide sequence that is 85%-100% identical to the sequence of SEQ ID NO. 1.

10. The composition of claim 1, wherein the composition is a unit dosage form having a dosage of 25 micrograms to 400 micrograms of the RNA polynucleotide for a dose of up to 2 mg/kg in a human subject.

11. The composition of claim 1, wherein the open reading frame is codon optimized.

12. The composition of claim 1, wherein the RNA comprises at least one chemical modification.

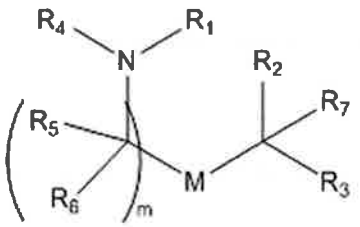
13. The composition of claim 12, wherein the chemical modification is selected from pseudouridine, 1-methylpseudouridine, 1-ethyl pseudouridine, 2-thiouridine, 4'-thiouridine, 5-methylcytosine, 2-thio-1-methyl-1-deaza-pseudouridine, 2-thio-1-methyl-pseudouridine, 2-thio-5-aza-uridine , 2-thio-dihydropseudouridine, 2-thio-dihydrouridine, 2-thio-pseudouridine, 4-methoxy-2-thio-pseudouridine, 4-methoxy-pseudouridine, 4-thio-1-methyl-pseudouridine, 4-thio-pseudouridine, 5-aza-uridine, dihydropseudouridine, 5-methyluridine, 5-methoxyuridine and 2'-O-methyl uridine.

14. The composition of claim 1, wherein the RNA polynucleotide formulated in the ionizable lipid nanoparticle has a therapeutic index of greater than 60% of the therapeutic index of the RNA polynucleotide alone.

15. The composition of claim 1, wherein the RNA polynucleotide formulated in the ionizable lipid nanoparticle has a therapeutic index of greater than 80% of the therapeutic index of the RNA polynucleotide alone.

16. A composition comprising an RNA polynucleotide having an open reading frame encoding at least one human relaxin fusion protein formulated in an ionizable lipid nanoparticle, wherein the ionizable lipid nanoparticle has a molar ratio of about 20-60% ionizable lipid: about 5-25% non-cationic lipid: about 25-55% sterol; and about 0.5-15% PEG-modified lipid.

17. The composition of claim 16, wherein the ionizable lipid is a lipid of Formula (I):



(I),

or a salt or isomer thereof, wherein:

R_1 is selected from the group consisting of C_{5-30} alkyl, C_{5-20} alkenyl, -R*YR", -YR", and -R"M'R';

R_2 and R_3 are independently selected from the group consisting of H, C_{1-14} alkyl, C_{2-14} alkenyl, -R*YR", -YR", and -R*OR", or R_2 and R_3, together with the atom to which they are attached, form a heterocycle or carbocycle;

R_4 is selected from the group consisting of a C_{3-6} carbocycle, $-(CH_2)_nQ$, $-(CH_2)_nCHQR$,

-CHQR, $-CQ(R)_2$, and unsubstituted C_{1-6} alkyl, where Q is selected from a carbocycle, heterocycle, -OR, $-O(CH_2)_nN(R)_2$, -C(O)OR, -OC(O)R, $-CX_3$, $-CX_2H$, $-CXH_2$, -CN, $-N(R)_2$, $-C(O)N(R)_2$, -N(R)C(O)R, $-N(R)S(O)_2R$, $-N(R)C(O)N(R)_2$, $-N(R)C(S)N(R)_2$, $-N(R)R_8$, $-O(CH_2)_nOR$, $-N(R)C(=NR_9)N(R)_2$, $-N(R)C(=CHR_9)N(R)_2$, $-OC(O)N(R)_2$, -N(R)C(O)OR, -N(OR)C(O)R, $-N(OR)S(O)_2R$, -N(OR)C(O)OR, $-N(OR)C(O)N(R)_2$, $-N(OR)C(S)N(R)_2$, $-N(OR)C(=NR_9)N(R)_2$, $-N(OR)C(=CHR_9)N(R)_2$, $-C(=NR_9)N(R)_2$, $-C(=NR_9)R$, -C(O)N(R)OR, and $-C(R)N(R)_2C(O)OR$, and each n is independently selected from 1, 2, 3, 4, and 5;

each R_5 is independently selected from the group consisting of C_{1-3} alkyl, C_{2-3} alkenyl, and H;

each R_6 is independently selected from the group consisting of C_{1-3} alkyl, C_{2-3} alkenyl, and H;

M and M' are independently selected from -C(O)O-, -OC(O)-, -C(O)N(R')-,

-N(R')C(O)-, -C(O)-, -C(S)-, -C(S)S-, -SC(S)-, -CH(OH)-, -P(O)(OR')O-, $-S(O)_2-$, -S-S-, an aryl group, and a heteroaryl group;

R_7 is selected from the group consisting of C_{1-3} alkyl, C_{2-3} alkenyl, and H;

R_8 is selected from the group consisting of C_{3-6} carbocycle and heterocycle;

R_9 is selected from the group consisting of H, CN, NO_2, C_{1-6} alkyl, -OR, $-S(O)_2R$, $-S(O)_2N(R)_2$, C_{2-6} alkenyl, C_{3-6} carbocycle and heterocycle;

each R is independently selected from the group consisting of C_{1-3} alkyl, C_{2-3} alkenyl, and H;

each R' is independently selected from the group consisting of C_{1-18} alkyl, C_{2-18} alkenyl, -R*YR", -YR", and H;

each R" is independently selected from the group consisting of C_{3-14} alkyl and

C_{3-14} alkenyl;

each R* is independently selected from the group consisting of C_{1-12} alkyl and

C_{2-12} alkenyl;

each Y is independently a C_{3-6} carbocycle;

each X is independently selected from the group consisting of F, Cl, Br, and I; and

m is selected from 5, 6, 7, 8, 9, 10, 11, 12, and 13.

18. The composition claim 16, wherein the nanoparticle has a polydispersity value of less than 0.4.

19. The composition claim 16, wherein the nanoparticle has a net neutral charge at a neutral pH.

20. The composition of claim 16, wherein the RNA comprises at least one chemical modification and wherein the chemical modification is N1-methylpseudouridine.

21. The composition claim 16, wherein the relaxin fusion protein includes an immunoglobulin (Ig) fragment.

22. The composition claim 21, wherein the Ig fragment is a variable chain fragment.

23. The composition claim 21, wherein the Ig fragment is a constant chain fragment.

24. The composition claim 21, wherein the Ig fragment is a variable light chain fragment.

25. The composition claim 21, wherein the variable light chain fragment a VLκ IgG region.

26. The composition of claim 16, wherein the relaxin protein has a nucleotide sequence that is 90%-100% identical to the sequence of SEQ ID NO. 1.

27. The nucleic acid of claim 26, wherein the Ig fragment is a variable chain fragment.

28. The nucleic acid of claim 26, wherein the Ig fragment is a constant chain fragment.

29. The nucleic acid of claim 26, wherein the Ig fragment is a variable light chain fragment.