CLAIMS

What is claimed is:

1. An antigenic human metapneumovirus (hMPV) prefusion F polypeptide, or a nucleic acid molecule that encodes the same, wherein said prefusion F polypeptide lacks a transmembrane domain and lacks a cytoplasmic tail, and comprises a human rhinovirus 3C (HRV-3C) protease cleavage site.

2. The F polypeptide or nucleic acid molecule of claim 1, wherein:

said prefusion F polypeptide further comprises a F_0 cleavage site mutation comprising amino acid substitutions Q100R and S101R, replacing glutamine at amino acid position 100 of SEQ ID NO: 1 with arginine, and replacing serine at amino acid position 101 of SEQ ID NO: 1 with arginine;

said prefusion F polypeptide comprises a signal peptide;

said prefusion F polypeptide comprises at least one tag sequence that is optionally an 8x His tag and/or a Strep II tag;

said prefusion F polypeptide comprises a foldon domain; and/or

said prefusion F polypeptide comprises an amino acid substitution replacing threonine at amino acid position 160 of SEQ ID NO: 1, and an amino acid substitution replacing asparagine at amino acid position 46 of SEQ ID NO: 1.

3. An antigenic human metapneumovirus (hMPV) prefusion F polypeptide, or a nucleic acid molecule that encodes the same, wherein said prefusion F polypeptide lacks a transmembrane domain and lacks a cytoplasmic tail, and comprises:

an F_0 cleavage site mutation comprising amino acid substitutions Q100R and S101R; replacing glutamine at amino acid position 100 of SEQ ID NO: 1 with arginine, and replacing serine at amino acid position 101 of SEQ ID NO: 1 with arginine;

a human rhinovirus 3C (HRV-3C) protease cleavage site;

a heterologous signal peptide;

an 8x His tag and/or a Strep II tag; and

a foldon domain.

4. An antigenic human prefusion metapneumovirus (hMPV) F polypeptide, or a nucleic acid molecule that encodes the same, wherein said prefusion F polypeptide lacks a transmembrane domain and lacks a cytoplasmic tail, and comprises an amino acid substitution replacing the wild-type amino acid at position 160 of SEQ ID NO: 1, and an amino acid substitution replacing the wild-type amino acid at position 46 of SEQ ID NO: 1.

5. An antigenic human prefusion metapneumovirus (hMPV) F polypeptide, or a nucleic acid molecule that encodes the same, wherein said prefusion F polypeptide lacks a transmembrane domain and lacks a cytoplasmic tail, and comprises an amino acid substitution replacing threonine at amino acid position 160 of SEQ ID NO: 1, and an amino acid substitution replacing asparagine at amino acid position 46 of SEQ ID NO: 1.

6. The hMPV F polypeptide or nucleic acid molecule of claim 4 or 5, wherein:

said prefusion F polypeptide comprises an amino acid substitution replacing the amino acid at position 160 with phenylalanine, tryptophan, tyrosine, valine, alanine, isoleucine or leucine; and/or

said prefusion F polypeptide comprises an amino acid substitution replacing the amino acid at position 46 with valine, alanine, isoleucine, leucine, phenylalanine, tyrosine or proline.

7. The hMPV F polypeptide or nucleic acid molecule of claim 6, wherein:

said prefusion F polypeptide comprises at least 95% sequence identity to SEQ ID NO: 7 or comprises SEQ ID NO: 7; or

said prefusion F polypeptide comprises at least 95% sequence identity to SEQ ID NO: 11 or comprises SEQ ID NO: 11.

8. The hMPV F polypeptide or nucleic acid molecule of any one of claims 4-7, wherein:

said prefusion F polypeptide further comprises an F_0 cleavage site mutation comprising amino acid substitutions Q100R and S101R, replacing glutamine at amino acid position 100 of SEQ ID NO: 1 with arginine, and replacing serine at amino acid position 101 of SEQ ID NO: 1 with arginine;

said prefusion F polypeptide comprises a signal peptide;

said prefusion F polypeptide comprises at least one tag sequence that is optionally an 8x His tag and/or a Strep II tag;

said prefusion F polypeptide comprises a foldon domain;

said F polypeptide comprises a human rhinovirus 3C (HRV-3C) protease cleavage site; and/or

the hMPV is A strain or B strain, optionally wherein the hMPV is A1 subtype, A2 subtype, B1 subtype, or B2 subtype.

9. The F polypeptide or nucleic acid molecule of any of the preceding claims, wherein said prefusion F polypeptide comprises at least 95% sequence identity to SEQ ID NO: 3 or comprises SEQ ID NO: 3.

10. A human metapneumovirus (hMPV) F polypeptide, or a nucleic acid molecule that encodes the same, wherein said F polypeptide comprises at least 95% sequence identity to SEQ ID NO: 7, optionally wherein:

the F polypeptide is a prefusion F polypeptide;

the F polypeptide is antigenic;

the F polypeptide comprises amino acid substitution T160F replacing threonine at amino acid position 160 with phenylalanine, and amino acid substitution N46V replacing asparagine at amino acid position 46 with valine; and/or

the F polypeptide comprises SEQ ID NO: 7.

11. A nucleic acid molecule encoding the polypeptide of claim 10, optionally wherein the nucleic acid molecule comprises at least 95% sequence identity to SEQ ID NO: 8, SEQ ID NO: 18, or SEQ ID NO: 19, or comprising SEQ ID NO: 8, SEQ ID NO: 18, or SEQ ID NO: 19.

12. A pharmaceutical composition comprising the F polypeptide, or a nucleic acid molecule that encodes the same, of claim 10, or the nucleic acid molecule of claim 11, optionally wherein the pharmaceutical composition comprises a vaccine.

13. A messenger RNA (mRNA) comprising an open reading frame (ORF) encoding the F polypeptide of any one of claims 1-10.

14. A messenger RNA (mRNA) comprising an open reading frame (ORF) encoding a human metapneumovirus (hMPV) F polypeptide antigen, wherein the hMPV F polypeptide antigen comprises

an amino acid sequence with at least 95% identity to SEQ ID NO: 11 or consists of an amino acid sequence of SEQ ID NO: 11.

15. The mRNA of claim 14, wherein:

the ORF is codon optimized;

the mRNA comprises at least one 5' untranslated region (5' UTR), at least one 3' untranslated region (3' UTR), and at least one polyadenylation (poly(A)) sequence;

the mRNA comprises at least one chemical modification;

at least 20%, at least 30%, at least 40%, at least 50%, at least 60%, at least 70%, at least 80%, at least 85%, at least 90%, at least 95%, or 100% of the uracil nucleotides in the mRNA are chemically modified; and/or

at least 20%, at least 30%, at least 40%, at least 50%, at least 60%, at least 70%, at least 80%, at least 85%, at least 90%, at least 95%, or 100% of the uracil nucleotides in the ORF are chemically modified, optionally wherein the chemical modification is selected from the group consisting of pseudouridine, N1-methylpseudouridine, 2-thiouridine, 4'-thiouridine, 5-methylcytosine, 2-thio-l-methyl-1-deaza-pseudouridine, 2-thio-l-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-l-methyl-pseudouridine, 4-thio-pseudouridine, 5-aza-uridine, dihydropseudouridine, 5-methyluridine, 5-methoxyuridine, and 2'-O-methyl uridine.

16. The mRNA of any one of claims 13-15, wherein the mRNA is formulated in a lipid nanoparticle (LNP), optionally wherein the LNP comprises at least one cationic lipid, optionally wherein the cationic lipid is biodegradable or not biodegradable, or cleavable or not cleavable, optionally wherein the cationic lipid is selected from the group consisting of OF-02, cKK-E10, GL-HEPES-E3-E10-DS-3-E18-1, GL-HEPES-E3-E12-DS-4-E10, and GL-HEPES-E3-E12-DS-3-E14.

17. The mRNA of claim 16, wherein the LNP further comprises a polyethylene glycol (PEG) conjugated (PEGylated) lipid, a cholesterol-based lipid, and a helper lipid.

18. The mRNA of claim 16 or 17, wherein the LNP comprises:

a cationic lipid at a molar ratio of 35% to 55%, optionally 40%,

a polyethylene glycol (PEG) conjugated (PEGylated) lipid at a molar ratio of 0.25% to 2.75%, optionally 1.5%,

a cholesterol-based lipid at a molar ratio of 20% to 45%, optionally 28.5 %, and

a helper lipid at a molar ratio of 5% to 35%, optionally 30%,

wherein all of the molar ratios are relative to the total lipid content of the LNP.

19. The mRNA of claim 17 or 18, wherein:

the PEGylated lipid is dimyristoyl-PEG2000 (DMG-PEG2000) or 2-[(polyethylene glycol)-2000]-N,N-ditetradecylacetamide (ALC-0159);

the cholesterol-based lipid is cholesterol; and/or

the helper lipid is 1,2-dioleoyl-SN-glycero-3-phosphoethanolamine (DOPE) or 1,2-distearoyl-*sn*-glycero-3-phosphocholine (DSPC).

20. The mRNA of any one of claims 16-19, wherein the LNP comprises:

GL-HEPES-E3-E12-DS-4-E10 or cKK-E10 at a molar ratio of 40%,

DMG-PEG2000 at a molar ratio of 1.5%,

cholesterol at a molar ratio of 28.5%, and

DOPE at a molar ratio of 30%.

21. A pharmaceutical composition comprising the mRNA of any one of claims 13-20.

22. A vaccine for use in eliciting an immune response to hMPV or protecting a subject against hMPV infection, comprising the pharmaceutical composition of claim 12 or 21 or mRNA of any one of claims 13-20 to be administered to a subject, optionally wherein:

the subject has a comparable serum concentration of neutralizing antibodies against hMPV after administration of the vaccine, relative to a subject that is administered a protein hMPV vaccine;

the protein hMPV vaccine is to be co-administered with an adjuvant; and/or

the vaccine increases the serum concentration of neutralizing antibodies in a subject with pre-existing hMPV immunity.

23. The F polypeptide or nucleic acid molecule of any one of claims 1-10, a prophylactically effective amount of the mRNA of any one of claims 13-20, or the pharmaceutical composition of claim 12 or 21, for use in treating a subject in need thereof.

24. A vaccine comprising a human metapneumovirus (hMPV) F polypeptide antigen or a nucleic acid molecule that encodes the same, wherein the F polypeptide comprises an amino acid sequence having at least 95% identity to SEQ ID NO: 7 or consisting of an amino acid sequence of SEQ ID NO: 7.