Claims

1. A recombinant pre-fusion respiratory syncitial virus (RSV) Fusion (F) polypeptide, comprising at least one epitope that is specific to the pre-fusion conformation F protein, wherein the at least one epitope is recognized by a pre-fusion specific monoclonal antibody, comprising a heavy chain CDR1 region of SEQ ID NO: 54, a heavy chain CDR2 region of SEQ ID NO: 55, a heavy chain CDR3 region of SEQ ID NO: 56 and a light chain CDR1 region of SEQ ID NO: 62, a light chain CDR2 region of SEQ ID NO: 63, and a light chain CDR3 region of SEQ ID NO: 64 and/or a pre-fusion specific monoclonal antibody, comprising a heavy chain CDR1 region of SEQ ID NO: 58, a heavy chain CDR2 region of SEQ ID NO: 59, a heavy chain CDR3 region of SEQ ID NO: 60 and a light chain CDR1 region of SEQ ID NO: 66, a light chain CDR2 region of SEQ ID NO: 67, and a light chain CDR3 region of SEQ ID NO: 68, wherein the polypeptide comprises a mutation of the amino acid residue N/T on position 67 into I; and/or a mutation of the amino acid residue S on position 215 into P and wherein the amino acid positions are given in reference to the sequence of RSV F protein from the A2 strain (SEQ ID NO: 1).

2. Pre-fusion RSV F polypeptide according to claim 1, wherein the polypeptide is trimeric.

3. Pre-fusion RSV F polypeptide according to claim 1 or 2, wherein the polypeptide comprises a F1 domain and a F2 domain, and a linking sequence comprising from 1 to 10 amino acid residues, linking said F1 domain to said F2 domain.

4. Pre-fusion RSV F polypeptide according to claim 1, 2 or 3, comprising a truncated F1 domain and a F2 domain, and a linking sequence comprising 1 to 10 amino acid residues, linking said F1 to said F2 domain, wherein polypeptide comprises at least one stabilizing mutation in the F1 and/or F2 domain as compared to the RSV F1 and/or F2 domain in the wild-type RSV F protein, wherein the at least one stabilizing mutation is selected from the group consisting of:

   (a) a mutation of amino acid residue N/T on position 67 into I.

   (b) a mutation of amino acid residue S on position 215 into P.

5. Pre-fusion RSV F polypeptide according to claim 4, wherein the polypeptide comprises a heterologous trimerization domain linked to said truncated F1 domain.

6. Pre-fusion RSV F polypeptide according to any one of the claims 1-5, wherein the polypeptide comprises at least one further mutation, wherein said mutation is selected from the group consisting of:

   (a) a mutation of the amino acid residue S on position 46 into G;

   (b) a mutation of the amino acid residue K on position 77 into E;

   (c) a mutation of the amino acid residue K on position 80 into E;

   (d) a mutation of the amino acid residue E on position 92 into D;

   (e) a mutation of the amino acid residue N on position 175 into P;

   (f) a mutation of the amino acid residue G on position 184 into N;

   (g) a mutation of the amino acid residue V on position 185 into N;

   (h) a mutation of the amino acid residue K on position 201 into Q;

(i) a mutation of the amino acid residue K on position 209 into Q;

(j) a mutation of the amino acid residue K on position 421 into N;

(k) a mutation of the amino acid residue N on position 426 into S;

(l) a mutation of the amino acid residue K on position 465 into E or Q;

(m) a mutation of the amino acid residue D on position 486 into N;

(n) a mutation of the amino acid residue E on position 487 into Q, N or I; and

(o) a mutation of the amino acid residue K on position 508 into E.

7. Pre-fusion RSV F polypeptide according to any of the preceding claims 1-6, wherein the polypeptide comprises at least two mutations, wherein the at least two mutations comprise a mutation of amino acid residue N/T on position 67 into I; and a mutation of amino acid residue S on position 215 into P.

8. Pre-fusion RSV F polypeptide according to claim 7, wherein the polypeptide comprises at least one further mutation selected from the group consisting of:

(a) a mutation of the amino acid residue S on position 46 into G;

(b) a mutation of the amino acid residue K on position 77 into E;

(c) a mutation of the amino acid residue K on position 80 into E;

(d) a mutation of the amino acid residue E on position 92 into D;

(e) a mutation of the amino acid residue N on position 175 into P;

(f) a mutation of the amino acid residue G on position 184 into N;

(g) a mutation of the amino acid residue V on position 185 into N;

(h) a mutation of the amino acid residue K on position 201 into Q;

(i) a mutation of the amino acid residue K on position 209 into Q;

(j) a mutation of the amino acid residue K on position 421 into N;

(k) a mutation of the amino acid residue N on position 426 into S;

(l) a mutation of the amino acid residue K on position 465 into E or Q;

(m) a mutation of the amino acid residue D on position 486 into N;

(n) a mutation of the amino acid residue E on position 487 into Q, N or I; and

(o) a mutation of the amino acid residue K on position 508 into E.

9. Pre-fusion RSV F polypeptide according to any one of the preceding claims 5-8, wherein the heterologous trimerization domain comprises the amino acid sequence EKKIEAIEKKIEAIEKKIEA (SEQ ID NO: 3).

10. Pre-fusion RSV F polypeptide according to claim 9, wherein the trimerization domain is linked to amino acid residue 495 of the RSV F protein.

11. Pre-fusion RSV F polypeptide according to any one of the claims 5-8, wherein the heterologous trimerization domain comprises the amino acid sequence GYIPEAPRDGQAYVRKDGEWVLLSTFL (SEQ ID NO: 4).

12. Pre-fusion RSV F polypeptide according to claim 11, wherein the trimerization domain is linked to amino acid residue 513 of the RSV F protein.

13. Pre-fusion RSV F polypeptide according to any one of the preceding claims 3-12, wherein the linking sequence between the F1 and the F2 domain comprises 5 amino acid residues.

14. Pre-fusion RSV F polypeptide according to claim 13, wherein the linking sequence comprises the amino acid sequence GSGSG (SEQ ID NO: 5).

15. Pre-fusion RSV F polypeptide according to any of the preceding claims 3-14, wherein the F1 domain and/or the F2 domain are from an RSV A strain.

16. Pre-fusion RSV F polypeptide according to any of the preceding claims 3-14, wherein the F1 domain and/or the F2 domain are from an RSV B strain.

17. Pre-fusion RSV F polypeptide according to any of the preceding claims, wherein the polypeptide is stable for at least 30 minutes at 55° C, preferably at 58° C, more preferably at 60° C.

18. Pre-fusion RSV F polypeptide according to any of the preceding claims, wherein the polypeptide is stable after storage at 4°C for at least 30 days, preferably at least 60 days, preferably at least 6 months, even more preferably at least 1 year.

19. Pre-fusion RSV F polypeptide according to any one of the preceding claims, wherein the polypeptide comprises an amino acid sequence selected from the group consisting

of SEQ ID NO: 21 – SEQ ID NO: 48, SEQ ID NO: 51 and SEQ ID NO: 72- SEQ ID NO: 89.

20. Pre-fusion RSV F polypeptide according to any of the preceding claims, wherein the polypeptide does not comprise a HIS-Tag.

21. Nucleic acid molecule encoding a pre-fusion RSV F polypeptide according to any one of the preceding claims 1-20.

22. Nucleic acid molecule according to claim 21, wherein the nucleic acid molecule has been codon-optimized for expression in mammalian cells.

23. Vector comprising a nucleic acid molecule according to claim 21 or claim 22.

24. Composition comprising a pre-fusion RSV F polypeptide according to any of the claims 1-20, a nucleic acid molecule according to claim 21 or claim 22 and/or a vector according to claim 23.

25. Pre-fusion RSV F polypeptide according to any of the claims 1-20, a nucleic acid molecule according to claim 21 or claim 22 and/or a vector according to claim 23 for use in inducing an immune response against RSV F protein.

26. Pre-fusion RSV F polypeptide according to any of the claims 1-20, a nucleic acid molecule according to claim 21 or claim 22 and/or a vector according to claim 23 for use as a vaccine.

27. Pre-fusion RSV F polypeptide according to any of the claims 1-20, a nucleic acid molecule according to claim 21 or claim 22 and/or a vector according to claim 23 for use in the prophylaxis and/or treatment of RSV infection.