

CLAIMS

What is claimed is:

1. A polynucleotide comprising:
a sequence encoding a rotavirus (RV) NSP3 protein; and
a heterologous polynucleotide, wherein the heterologous polynucleotide encodes a peptide or protein and is in frame with the NSP3, and wherein the peptide or protein comprises a viral peptide or protein.
2. The polynucleotide of claim 1, wherein the polynucleotide is operably linked to a promoter.
3. The polynucleotide of claim 1, wherein the sequence encoding an RV NSP3 is a sequence encoding SEQ ID NO: 1, or a sequence with at least about 90% identity to SEQ ID NO: 1.
4. The polynucleotide of claim 1, wherein the viral peptide or protein comprises a norovirus (NoV) peptide or protein.
5. The polynucleotide of claim 4, wherein the NoV peptide or protein comprises NoV VP1 protein.
6. The polynucleotide of claim 5, wherein the NoV peptide or protein is selected from SEQ ID NOs: 24, 26 or 80-84.
7. The polynucleotide of claim 1, wherein the viral peptide or protein comprises a SARS-CoV-2 protein or peptide.

8. The polynucleotide of claim 7, wherein the SARS-CoV-2 protein or peptide is selected from an N protein, an S protein, or fragment of either an N protein or an S protein.
9. The polynucleotide of claim 1, wherein the peptide or protein comprises a cleavage site.
10. The polynucleotide of claim 9, wherein the cleavage site is a protease cleavage site.
11. The polynucleotide of claim 9, wherein the cleavage site is a self-cleaving peptide sequence.
12. The polynucleotide of claim 11, wherein the self-cleaving peptide sequence is porcine teschovirus 2A element (SEQ ID NO: 29).
13. The polynucleotide of claim 1, wherein the heterologous polynucleotide is about 2.6 kb or less in length.
14. The polynucleotide of claim 1, wherein the peptide or protein is a glycoprotein.
15. The polynucleotide of claim 1, wherein the peptide or protein comprises one or more glycosylation site.
16. The polynucleotide of claim 1, wherein the polynucleotide encodes a protein selected from SEQ ID NOs: 2-11 or a sequence with at least about 90% identity to a sequence selected from SEQ ID NOs: 2-11.
17. The polynucleotide of claim 1, wherein the polynucleotide comprises any one of SEQ ID NOs: 13-22 or a sequence with at least about 90% identity to any one of SEQ ID NOs: 13-22.
18. An infectious particle comprising the polynucleotide of any one of claims 1-17.

19. A pharmaceutical composition comprising the infectious particle of claim 18, optionally, further comprising a pharmaceutically acceptable carrier or excipient.
20. Use of the pharmaceutical composition of claim 19 in the manufacture of a medicament for eliciting an immune response to one or more microorganism in a subject, wherein an effective amount of the pharmaceutical composition is to be administered to the subject to elicit an immune response to the one or more microorganism.
21. Use of the pharmaceutical composition of claim 19 in the manufacture of a medicament for vaccinating a subject against one or more pathogens, wherein an effective amount of the pharmaceutical composition is to be administered to the subject to vaccinate said subject against the one or more pathogens.
22. A method of generating recombinant rotavirus (RV) *in vitro* comprising:
introducing the polynucleotide of any one of claims 1-17 into a cell; allowing the cell to express the polynucleotide; incubating the cells for a sufficient time to produce RV; and harvesting virus produced by the cells to generate RV *in vitro*.
23. The method of claim 22, wherein the method further comprises introducing one or more additional polynucleotides into the cell before the allowing step, wherein the one or more additional polynucleotides comprise a sequence encoding an RV protein selected from VP1, VP2, VP3, VP4, VP6, VP7, NSP1, NSP2, NSP3, NSP4, and NSP5, wherein each sequence encoding an RV protein is operably linked to a promoter.
24. The method of claim 23, wherein the one or more additional polynucleotides comprise a sequence encoding an RV protein selected from VP1, VP2, VP3, VP4, VP6, VP7, NSP1, NSP2, NSP4, and NSP5.
25. A system, platform, or kit for generating recombinant rotavirus (RV) comprising:
(a) the polynucleotide of any one of claims 1-17; and
(b) cells capable of expressing the polynucleotides of (a).

26. The pharmaceutical composition of claim 19 for use in therapy.