CLAIMS

1. A vaccine comprising an immunogenic complex, in which the immunogenic complex comprises:

(a) a biotinylated polysaccharide antigen; and

(b) a fusion protein comprising:

(ii) a biotin-binding moiety; and

(iii) at least one polypeptide antigen;

in which the biotinylated polysaccharide antigen comprises a polysaccharide of *Salmonella enterica*, and further in which the biotinylated polysaccharide antigen is non-covalently associated with the biotin-binding moiety of the fusion protein to form an immunogenic complex.

2. A vaccine according to Claim 1, in which:

the biotinylated polysaccharide antigen comprises a polysaccharide of *Salmonella enterica* selected from Vi polysaccharide of Typhi, and O-specific polysaccharides of Typhimurium, Enteritidis, and Paratyphi;

the at least one polypeptide antigen of the fusion protein is or comprises a polypeptide antigen from *Salmonella*, *Shigella*, or *Streptococcus pneumoniae*;

the at least one polypeptide antigen of the fusion protein comprises a *Salmonella* SseB polypeptide or antigenic fragment thereof;

the at least one polypeptide antigen of the fusion protein comprises a *Shigella* IpaB polypeptide or antigenic fragment thereof; or

the at least one polypeptide antigen of the fusion protein comprises a *Streptococcus pneumoniae* SP1500 polypeptide or antigenic fragment thereof; a *Streptococcus pneumoniae* SP0785 polypeptide or antigenic fragment thereof, or both.

3. A vaccine according to Claim 1 or 2, in which:

the at least one polypeptide antigen of the fusion protein is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least

95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4;

the at least one polypeptide antigen of the fusion protein is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 5;

the at least one polypeptide antigen of the fusion protein is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to either SEQ ID NO: 8 or SEQ ID NO: 9, or a combination of SEQ ID NO: 8 and SEQ ID NO: 9; or

the biotin-binding moiety is a polypeptide comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 3 or a biotin-binding fragment thereof.

4. An immunogenic composition (e.g., a vaccine) comprising a plurality of different species of immunogenic complexes, in which the different species comprise:

a plurality of biotinylated polysaccharide antigens comprising polysaccharide antigens of one or more *Salmonella enterica* serotypes; and

a plurality of fusion proteins, each fusion protein comprising:

a biotin-binding moiety; and

a polypeptide antigen,

in which each of the plurality of biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of one or more of the plurality of fusion proteins to form an immunogenic complex.

5. An immunogenic composition according to Claim 4, in which:

the different species each comprise a distinct polysaccharide antigen of one or more *Salmonella enterica* serotypes and/or a distinct polypeptide antigen;

the one or more *Salmonella enterica* serotypes is or comprises S. Typhimurium, S. Enteritidis, S. Typhi Vi, S. Paratyphi A, or combinations thereof;

the polypeptide antigen is or comprises a polypeptide antigen from *Salmonella*, *Shigella*, and/or *Streptococcus pneumoniae*; or

the polypeptide antigen is or comprises: an SseB polypeptide antigen of *Salmonella*, an IpaB polypeptide antigen of *Shigella*, and/or a polypeptide antigen comprising an SP1500 polypeptide and/or an SP0785 polypeptide, of *S. pneumoniae*.

6. An immunogenic composition according to Claim 4 or 5, comprising at least two different species of immunogenic complexes, in which the different species comprise:

a first biotinylated polysaccharide antigen comprising a polysaccharide of S. Typhimurium non-covalently complexed with a first fusion protein; and

a second biotinylated polysaccharide antigen comprising a polysaccharide of S. Enteritidis non-covalently complexed with a second fusion protein,

in which the first fusion protein and the second fusion protein each independently comprise:

a biotin-binding moiety; and

a polypeptide antigen,

in which each of the first and second biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of the respective fusion protein to form an immunogenic complex.

7. An immunogenic composition according to Claim 6, wherein the polypeptide antigen of the first fusion protein and of the second fusion protein is or comprises an SseB polypeptide antigen of *Salmonella*, preferably in which the SseB polypeptide antigen of Salmonella is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4 or an immunogenic fragment thereof.

8. An immunogenic composition according to Claim 4 or 5, comprising at least two different species of immunogenic complexes, in which the different species comprise:

a first biotinylated polysaccharide antigen comprising a polysaccharide of S. Typhi Vi non-covalently complexed with a first fusion protein; and

a second biotinylated polysaccharide antigen comprising a polysaccharide of S. Paratyphi A non-covalently complexed with a second fusion protein,

in which the first fusion protein and the second fusion protein each independently comprise:

a biotin-binding moiety; and

a polypeptide antigen,

in which each of the first and second biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of the respective fusion protein to form an immunogenic complex.

9. An immunogenic composition according to Claim 8, in which the polypeptide antigen of the first fusion protein and of the second fusion protein is or comprises an SP1500 polypeptide, an SP0785 polypeptide, or both, of *S. pneumoniae*, and/or the fusion protein is CP1, preferably in which the SP1500 polypeptide antigen of *S. pneumoniae* is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 8, or an immunogenic fragment thereof; the SP0785 polypeptide antigen of *S. pneumoniae* is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 8, or an immunogenic fragment thereof; and the CP1 fusion protein is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 6, or an immunogenic fragment thereof.

10. An immunogenic composition according to Claim 4 or 5, comprising at least four different species of immunogenic complexes, in which the different species comprise:

a first biotinylated polysaccharide antigen comprising a polysaccharide of S. Typhimurium non-covalently complexed with a first fusion protein; and

a second biotinylated polysaccharide antigen comprising a polysaccharide of S. Enteritidis non-covalently complexed with a second fusion protein,

a third biotinylated polysaccharide antigen comprising a polysaccharide of S. Typhi Vi non-covalently complexed with a third fusion protein; and

a fourth biotinylated polysaccharide antigen comprising a polysaccharide of S. Paratyphi A non-covalently complexed with a fourth fusion protein,

in which the first fusion protein, the second fusion protein, the third fusion protein, and the fourth fusion protein each independently comprise:

a biotin-binding moiety; and

a polypeptide antigen,

in which each of the first, second, third, and fourth biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of the respective fusion protein to form an immunogenic complex.

11. An immunogenic composition according to Claim 10, in which the polypeptide antigen of the first fusion protein and of the second fusion protein is or comprises an SseB polypeptide antigen of *Salmonella*, and the polypeptide antigen of the third fusion protein and of the fourth fusion protein is or comprises an SP1500 polypeptide, an SP0785 polypeptide, or both, of *S. pneumoniae*, and/or the fusion protein is CP1, preferably in which the SseB polypeptide antigen of *Salmonella* is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4 or an immunogenic fragment thereof; the SP1500 polypeptide antigen of *S. pneumoniae* is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 8, or an immunogenic fragment thereof; the SP0785 polypeptide antigen of *S. pneumoniae* is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 9, or an immunogenic fragment thereof; and the CP1 fusion protein is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 6, or an immunogenic fragment thereof.

12. An immunogenic composition according to Claim 4 or 5, comprising at least four different species of immunogenic complexes, in which the different species comprise:

a first biotinylated polysaccharide antigen comprising a polysaccharide of S. Typhimurium non-covalently complexed with a first fusion protein; and

a second biotinylated polysaccharide antigen comprising a polysaccharide of S. Enteritidis non-covalently complexed with a second fusion protein,

a third biotinylated polysaccharide antigen comprising a polysaccharide of S. Typhi Vi non-covalently complexed with a third fusion protein; and

a fourth biotinylated polysaccharide antigen comprising a polysaccharide of S. Paratyphi A non-covalently complexed with a fourth fusion protein,

in which the first fusion protein, the second fusion protein, the third fusion protein, and the fourth fusion protein each independently comprise:

a biotin-binding moiety; and

a polypeptide antigen,

in which each of the first, second, third, and fourth biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of the respective fusion protein to form an immunogenic complex.

13. An immunogenic composition according to Claim 12, in which the polypeptide antigen of the first fusion protein, the second fusion protein, the third fusion protein, and the fourth fusion protein is or comprises an SseB polypeptide antigen of *Salmonella*, preferably in which the SseB polypeptide antigen of *Salmonella* is or comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4 or an immunogenic fragment thereof.

14. A vaccine comprising a plurality of different species of immunogenic complexes, in which the different species comprise:

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Typhimurium non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Enteritidis non-covalently complexed with a fusion protein;

in which each fusion protein comprises

a biotin-binding moiety; and

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4, SEQ ID NO: 5, or either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9, or an antigenic fragment thereof; and

in which each of the biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of at least one fusion protein to form an immunogenic complex.

15. A vaccine according to Claim 14, in which:

the first polypeptide antigen comprises an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4;

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 5; or

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9.

16. A vaccine comprising a plurality of different species of immunogenic complexes, in which the different species comprise:

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Typhi non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Paratyphi non-covalently complexed with a fusion protein;

in which each fusion protein comprises

a biotin-binding moiety; and

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4, SEQ ID NO: 5, or either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9, or an antigenic fragment thereof; and

in which each of the biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of at least one fusion protein to form an immunogenic complex.

17. A vaccine according to Claim 16, in which:

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4;

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 5; or

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9.

18. A vaccine comprising a plurality of different species of immunogenic complexes, in which the different species comprise:

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Typhimurium non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Enteritidis non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Typhi non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Paratyphi non-covalently complexed with a fusion protein;

in which each fusion protein comprises

a biotin-binding moiety; and

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% SEQ ID NO: 4, SEQ ID NO: 5, or either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9, or an antigenic fragment thereof; and

in which each of the biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of at least one fusion protein to form an immunogenic complex.

19. A vaccine according to Claim 18, in which:

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4;

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 5; or

the first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9.

20. A vaccine comprising a plurality of different species of immunogenic complexes, in which the different species comprise:

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Typhimurium non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Enteritidis non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Typhi non-covalently complexed with a fusion protein;

a biotinylated polysaccharide antigen of *Salmonella enterica* serotype Paratyphi non-covalently complexed with a fusion protein;

in which each fusion protein comprises

a biotin-binding moiety;

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4, SEQ ID NO: 5, or either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9, or an antigenic fragment thereof; and

a second polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4, SEQ ID NO: 5, or either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9, or an antigenic fragment thereof; and

in which each of the biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of at least one fusion protein to form an immunogenic complex.

21. A vaccine according to Claim 20, in which:

the vaccine comprises a stoichiometrically equal ratio, by weight, of each of the polysaccharide antigens of the different species; or

the vaccine comprises at least one of the polysaccharide antigens of the different species at a stoichiometrically different ratio, by weight.

22. A vaccine according to any of Claims 14 to 21, in which the vaccine comprises a stoichiometrically different ratio, by weight, of each of the polysaccharide antigens of the different species.

23. A vaccine according to any preceding claim, in which the biotin-binding moiety is a polypeptide comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 3.

24. An immunogenic complex comprising a biotinylated polysaccharide antigen of *Salmonella enterica* non-covalently associated with a fusion protein, in which the fusion protein comprises a biotin-binding moiety and at least one polypeptide antigen.

25. An immunogenic complex according to Claim 24, in which:

the biotinylated polysaccharide antigen comprises a polysaccharide of *Salmonella enterica* having a serotype selected from Typhimurium, Enteritidis, Typhi, and Paratyphi;

the fusion protein comprises SseB, IpaB, or an SP1500 polypeptide, an SP0785 polypeptide, or both;

the fusion protein comprises:

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4 or an antigenic fragment thereof;

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 5 or an antigenic fragment thereof; or

a first polypeptide antigen comprising an amino acid sequence at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to SEQ ID NO: 4, SEQ ID NO: 5, or either SEQ ID NO: 8, SEQ ID NO: 9, or both SEQ ID NO: 8 and SEQ ID NO:9, or an antigenic fragment thereof; or

the immunogenic complex comprises a ratio of fusion protein to polysaccharide antigen of about 1:1, about 2:1, about 3:1, about 4:1, about 5:1, about 6:1, about 7:1, about 8:1, about 9:1, or about 10:1, by weight.

26. A vaccine comprising one or more immunogenic complexes according to any of Claims 24 or 25.

27. A pharmaceutical composition comprising:

a vaccine according to any of Claims 1 to 23 and 26 and a pharmaceutically acceptable carrier; or

an immunogenic complex according to Claim 24 or 25 and a pharmaceutically acceptable carrier;

preferably in which the pharmaceutical composition further comprises one or more adjuvants, preferably in which the one or more adjuvants is or comprises a co-stimulation factor, preferably in which the one or more adjuvants are selected from the group consisting of aluminium phosphate, aluminium hydroxide, and phosphated aluminium hydroxide, preferably in which the one or more adjuvants is or comprises aluminium phosphate, preferably in which the pharmaceutical composition is formulated for injection, preferably in which upon administration to a subject, the pharmaceutical composition induces an immune response, such as in which the immune response is to (i) at least one polysaccharide antigen of the vaccine or immunogenic complex, and/or (ii) at least one polypeptide antigen of the vaccine or immunogenic complex.

28. A method of making a vaccine, comprising non-covalently complexing a plurality of biotinylated polysaccharide antigens with a plurality of fusion proteins, in which each fusion protein comprises at least one polypeptide antigen selected SseB, IpaB, SP0785 or SP1500; in which the plurality of biotinylated polysaccharide antigens comprises polysaccharides of one or more *Salmonella enterica* serotypes selected from Typhimurium, Enteritidis, Typhi, and Paratyphi.

29. A vaccine according to any of Claims 1 to 23 and 26, an immunogenic complex according to Claim 24 or 25 or a pharmaceutical composition of according to Claim 27 for use in a method of immunizing a subject against *Salmonella enterica* infection and/or colonization, preferably in which the vaccine, immunogenic composition, or pharmaceutical composition induces an immune response, preferably in which the immune response is to at least one polysaccharide antigen or at least one polypeptide of a fusion protein, preferably in which the subject is immunized against *Salmonella enterica* infection and/or colonization with one dose of a vaccine, with two doses of a vaccine or with three doses of a vaccine.

30. A fusion protein comprising a rhizavidin protein and at least one peptide or polypeptide antigen, in which the rhizavidin protein comprises amino acids of SEQ ID NO: 3, or 85% sequence identity to amino acids of SEQ ID NO: 3, and *Salmonella* peptide or polypeptide comprises a fragment of at least 20 amino acids of the SseB protein, or the *Shigella* peptide or polypeptide comprises a fragment of at least 20 amino acids of the IpaB protein.

31. A fusion protein according to Claim 30, in which:

the SseB protein comprises at least SEQ ID NO: 4 or a protein of at least 20 amino acids that has at least 85% sequence identity to SEQ ID NO: 4;

the IpaB protein comprises at least SEQ ID NO: 5 or a protein of at least 20 amino acids that has at least 85% sequence identity to SEQ ID NO: 5;

the fusion protein comprises at least SEQ ID NO: 1; or

the fusion protein comprises at least SEQ ID NO: 2.