Claims

We claim:

1. A vaccine comprising an immunogenic complex that comprises:

(a) a biotinylated polysaccharide; and

(b) a fusion protein comprising an amino acid sequence at least 90% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59, wherein the fusion protein comprises:

(i) a biotin-binding moiety;

(ii) a non-hemolytic pneumolysin (Ply) polypeptide comprising mutations at amino acid residues corresponding to amino acid residues 294, 385, 428, and 433 of wild-type *Streptococcus pneumoniae* pneumolysin (SEQ ID NO: 3) wherein the mutations are G294P, D385N, C428G and W433F; and

(iii) an SP0435 polypeptide;

wherein the biotinylated polysaccharide antigen is non-covalently associated with the biotin-binding moiety of the fusion protein to form the immunogenic complex.

2. The vaccine of claim 1, wherein the biotinylated polysaccharide antigen comprises a polysaccharide of *Streptococcus pneumoniae.*

3. The vaccine of claim 1, wherein the biotinylated polysaccharide antigen comprises a polysaccharide of *Streptococcus pneumoniae* having a serotype selected from 1, 2, 3, 4, 5, 6A, 6B, 6C, 6D, 6E, 6F, 6G, 6H, 7A, 7B, 7C, 7F, 8, 9A, 9L, 9N, 9V, 10A, 10B, 10C, 10F, 11A, 11B, 11C, 11D, 11E, 11F, 12A, 12B, 12F, 13, 14, 15A, 15B, 15C, 15F, 16A, 16F, 17A, 17F, 18A, 18B, 18C, 18F, 19A, 19B, 19C, 19F, 20A, 20B, 21, 22A, 22F, 23A, 23B, 23F, 24A, 24B, 24F, 25A, 25F, 27, 28A, 28F, 29, 31, 32A, 32F, 33A, 33B, 33C, 33D, 33E, 33F, 34, 35A, 35B, 35C, 35F, 36, 37, 38, 39, 40, 41A, 41F, 42, 43, 44, 45, 46, 47A, 47F, and 48.

4. The vaccine of claim 1, wherein the biotinylated polysaccharide antigen comprises a polysaccharide of *Streptococcus pneumoniae* having a serotype selected from 1, 2, 3, 4, 5, 6A, 6B, 6C, 7C, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15A, 15B, 16F, 17F, 18C, 19A, 19F, 20B, 22F, 23A, 23B, 23F, 24F, 31, 33F, 35B, and 38.

5. The vaccine of any one of claims 1-3, wherein the biotinylated polysaccharide antigen comprises a polysaccharide of *Streptococcus pneumoniae* having a serotype selected from 6C, 7C, 15A, 16F, 23A, 23B, 24F, 31, 35B, and 38.

6. The vaccine of claim 1, comprising:

a plurality of different species of immunogenic complexes comprising:

(a) a plurality of first biotinylated polysaccharide antigens, wherein the plurality comprises polysaccharide antigens of one or more of *Streptococcus pneumoniae* serotypes 1, 2, 3, 4, 5, 6A, 6B, 6C, 6D, 6E, 6F, 6G, 6H, 7A, 7B, 7C, 7F, 8, 9A, 9L, 9N, 9V, 10A, 10B, 10C, 10F, 11A, 11B, 11C, 11D, 11E, 11F, 12A, 12B, 12F, 13, 14, 15A, 15B, 15C, 15F, 16A, 16F, 17A, 17F, 18A, 18B, 18C, 18F, 19A, 19B, 19C, 19F, 20A, 20B, 21, 22A, 22F, 23A, 23B, 23F, 24A, 24B, 24F, 25A, 25F, 27, 28A, 28F, 29, 31, 32A, 32F, 33A, 33B, 33C, 33D, 33E, 33F, 34, 35A, 35B, 35C, 35F, 36, 37, 38, 39, 40, 41A, 41F, 42, 43, 44, 45, 46, 47A, 47F, and 48; and

(b) a plurality of first fusion proteins, wherein each first fusion protein comprises an amino acid sequence at least 90% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59, and wherein each first fusion protein comprises:

(i) a biotin-binding moiety;

(ii) a non-hemolytic pneumolysin (Ply) polypeptide comprising mutations at amino acid residues corresponding to amino acid residues 294, 385, 428, and 433 of wild-type *Streptococcus pneumoniae* pneumolysin (SEQ ID NO: 3), wherein the non-hemolytic pneumolysin (Ply) polypeptide 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%, at least 99.5%, or 100% identical to SEQ ID NO: 6; and

(iii) an SP0435 polypeptide, wherein the SP0435 polypeptide 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%, at least 99.5%, or 100% identical to SEQ ID NO: 8;

wherein each of the plurality of first biotinylated polysaccharide antigens is non-covalently associated with the biotin-binding moiety of one or more of the plurality of first fusion proteins to form one of the immunogenic complexes.

7. The vaccine of claim 6, wherein the plurality of different species of immunogenic complexes further comprises:

(a) a plurality of second biotinylated polysaccharide antigens, wherein the plurality comprises polysaccharide antigens of one or more of *Streptococcus pneumoniae* serotypes 1, 2, 3, 4, 5, 6A, 6B, 6C, 6D, 6E, 6F, 6G, 6H, 7A, 7B, 7C, 7F, 8, 9A, 9L, 9N, 9V, 10A, 10B, 10C, 10F, 11A, 11B, 11C, 11D, 11E, 11F, 12A, 12B, 12F, 13, 14, 15A, 15B, 15C, 15F, 16A, 16F, 17A, 17F, 18A, 18B, 18C, 18F, 19A, 19B, 19C, 19F, 20A, 20B, 21, 22A, 22F, 23A, 23B, 23F, 24A, 24B, 24F, 25A, 25F, 27, 28A, 28F, 29, 31, 32A, 32F, 33A, 33B, 33C, 33D, 33E, 33F, 34, 35A, 35B, 35C, 35F, 36, 37, 38, 39, 40, 41A, 41F, 42, 43, 44, 45, 46, 47A, 47F, and 48; and

(b) a plurality of second fusion proteins, wherein each second fusion protein comprises:

(i) a biotin-binding moiety;

(ii) 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%, at least 99.5%, or 100% identical to SEQ ID NO: 12; and

(iii) 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%, at least 99.5%, or 100% identical to SEQ ID NO: 10;

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

8. The vaccine of claim 7, wherein each of the plurality of second fusion proteins, 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%, at least 99.5%, or 100% identical to SEQ ID NO: 26 or SEQ ID NO: 61.

9. The vaccine of claim 7, wherein the plurality of different species of immunogenic complexes comprises:

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 16F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 24F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 31 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 35B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 38 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 1 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 2 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 3 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 4 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 5 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 8 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9N non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9V non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 10A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 11A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 12F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 14 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 17F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 18C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 20B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 22F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 33F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins.

10. The vaccine of claim 7, wherein the plurality of different species of immunogenic complexes comprises:

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 1 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 4 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9V non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 18C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 33F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 2 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 3 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 5 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 8 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9N non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 10A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 11A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 12F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 14 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 16F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 17F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 20B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 22F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 24F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 31 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 35B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 38 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins.

11. The vaccine of claim 7, wherein the plurality of different species of immunogenic complexes comprise:

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 1 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 4 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9V non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 18C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 33F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion protein;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 2 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 3 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 5 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 8 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9N non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 10A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 11A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 12F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 14 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 17F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 20B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 22F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 24F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 31 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 35B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 38 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins.

12. The vaccine of claim 7, wherein the plurality of different species of immunogenic complexes comprise:

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 1 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 4 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9V non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 18C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 33F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of first fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 2 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 5 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 6C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7C non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 7F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 8 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 9N non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 10A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 11A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 12F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 14 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 15A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 16F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 17F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 19F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 20B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 22F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23A non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 23B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 24F non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 31 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins;

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 35B non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins; and

a biotinylated polysaccharide antigen of *Streptococcus pneumoniae* serotype 38 non-covalently complexed with the biotin-binding moiety of one or more of the plurality of second fusion proteins.

13. The vaccine of claim 1, wherein the biotin-binding moiety is (i) 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%, at least 99.5%, or 100%

identical to SEQ ID NO: 1; or (ii) 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%, at least 99.5%, or 100% identical to SEQ ID NO: 2 or SEQ ID NO: 57.

14. A pharmaceutical composition comprising the vaccine of claim 1, and a pharmaceutically acceptable carrier.

15. A method of making a vaccine, comprising non-covalently complexing a biotinylated polysaccharide antigen with a fusion protein comprising an amino acid sequence at least 90% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59,

wherein the fusion protein comprises:

(a) a biotin-binding moiety;

(b) a non-hemolytic pneumolysin (Ply) polypeptide comprising mutations at amino acid residues corresponding to amino acid residues 294, 385, 428, and 433 of wild-type *Streptococcus pneumoniae* pneumolysin (SEQ ID NO: 3) wherein the mutations are G294P, D385N, C428G and W433F; and

(c) an SP0435 polypeptide.

16. A fusion protein comprising an amino acid sequence at least 90% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59,

wherein the fusion protein comprises:

(i) a biotin-binding moiety;

(ii) a non-hemolytic pneumolysin (Ply) polypeptide comprising mutations at amino acid residues corresponding to amino acid residues 294, 385, 428, and 433 of wild-type *Streptococcus pneumoniae* pneumolysin (SEQ ID NO: 3) wherein the mutations are G294P, D385N, C428G and W433F; and

(iii) an SP0435 polypeptide.

17. The vaccine of claim 1, wherein the fusion protein comprises an amino acid sequence 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%, at least 99.5%, or 100% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59.

18. The vaccine of claim 6, wherein each first fusion protein comprises an amino acid sequence 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%, at least 99.5%, or 100% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59.

19. The method of claim 15, wherein the fusion protein comprises an amino acid sequence 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%, at least 99.5%, or 100% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59.

20. The fusion protein of claim 16, wherein the fusion protein comprises an amino acid sequence 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%, at least 99.5%, or 100% identical to SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 58, or SEQ ID NO: 59.