CLAIMS

We claim:

1. ~~A method for active booster immunization against tetanus, diphtheria, and pertussis, comprising:~~

~~administering to a human subject~~Use of an immunogenic composition in the manufacture of a medicament for active booster immunization of a human subject against tetanus, diphtheria, and pertussis,

wherein the immunogenic composition comprises ~~comprising~~ tetanus, diphtheria, and acellular pertussis (Tdap) antigens, an aluminum salt adjuvant to which the Tdap antigens are adsorbed, and a toll-like receptor 9 (TLR9) agonist, ~~wherein~~

wherein the TLR9 agonist is an oligonucleotide of from 10 to 35 nucleotides in length comprising an unmethylated cytidine-phospho-guanosine (CpG) motif,

wherein the immunogenic composition comprises from about 750 µg to about 6000 µg of the oligonucleotide,

wherein the human subject is three years of age or older, and

wherein the Tdap antigens comprise a tetanus toxoid (TT), a diphtheria toxoid (DT), a pertussis toxoid (PT), a pertussis filamentous haemagglutinin (FHA), and pertussis pertactin (PRN)~~, and~~

~~the Tdap antigens and the oligonucleotide are present in the immunogenic composition in amounts effective to stimulate an immune response against the Tdap antigens~~.

2. The ~~method~~ use of claim 1, wherein the oligonucleotide comprises the sequence 5’-AACGTTCGAG-3’ (SEQ ID NO:3).

3. The ~~method~~ use of claim 1, wherein the oligonucleotide comprises the sequence 5’-TGACTGTGAA CGTTCGAGAT GA-3’(SEQ ID NO:1).

4. The ~~method~~ use of any one of claims 1-3, wherein the oligonucleotide comprises a modified nucleoside~~, optionally wherein the modified nucleoside is~~ selected from the group consisting of 2’-deoxy-7-deazaguanosine, 2’-deoxy-6-thioguanosine, arabinoguanosine, 2’-deoxy-2’substituted-arabinoguanosine, and 2’-O-substituted-arabinoguanosine.

5. The ~~method~~ use of claim 4, wherein the oligonucleotide comprises the sequence 5'-TCG$_1$AACG$_1$TTCG$_1$-3' (SEQ ID NO:2) in which G$_1$ is 2'-deoxy-7-deazaguanosine, ~~optionally~~ or wherein the oligonucleotide comprises the sequence 5'-TCG$_1$AACG$_1$TTCG$_1$-X-G$_1$CTTG$_1$CAAG$_1$CT-5', and in which G$_1$ is 2'-deoxy-7-deazaguanosine and X is glycerol (5'-SEQ ID NO:2-3'-X-3'-SEQ ID NO:2-5').

6. The ~~method~~ use of any one of claims 1-~~53~~, wherein the oligonucleotide comprises at least one phosphorothioate linkage, or wherein all nucleotide linkages are phosphorothioate linkages.

7. The ~~method~~ use of any one of claims 1-6, wherein the oligonucleotide is a single-stranded oligodeoxynucleotide.

8. The ~~method~~ use of any one of claims 1-~~76~~, wherein ~~a 0.5 ml dose of~~ the immunogenic composition comprises ~~from about 375 µg to about 6000 µg of the oligonucleotide or~~ from about 1000~~750~~ µg to about 4000~~3000~~ µg of the oligonucleotide~~, or wherein a 0.5 ml dose of the immunogenic composition comprises about 375 µg, about 750 µg, about 1500 µg, about 3000 µg, or 6000 µg about of the oligonucleotide~~.

9. The ~~method~~ use of any one of claims 1-8, wherein the aluminum salt adjuvant comprises one or more of the group consisting of amorphous aluminum hydroxyphosphate sulfate, aluminum hydroxide, aluminum phosphate, and potassium aluminum sulfate.

10. The ~~method~~ use of claim 8, wherein the aluminum salt adjuvant comprises aluminum hydroxide.

11. The ~~method~~ use of claim 9 or claim 10, wherein ~~a 0.5 ml dose of~~ the immunogenic composition comprises from about 0.25 to about 0.50 mg Al^{3+}~~, or wherein a 0.5 ml dose of the immunogenic composition comprises from about 0.30 to about 0.40 mg Al^{3+}~~.

12. The ~~method~~ use of any one of claims 1-11, wherein ~~a 0.5 ml dose~~ of the immunogenic composition comprises about 5 Lf TT, about 2.5 Lf DT, about 8 µg PT, about 8 µg FHA, and about 2.5 µg PRN.

13. The ~~method~~ use of any one of claims 1-12, wherein the immunogenic composition further comprises at least one additional antigen.

14. The ~~method~~ use of claim 13, wherein the at least one additional antigen comprises one or both of a pertussis fimbriae (FIM) antigen and a pertussis adenylate cyclase (AC) antigen.

15. The use of any one of claims 8-14, wherein the immunogenic composition comprises about 1500 µg of the oligonucleotide~~method of claim 13, wherein the at least one additional antigen comprises an inactivated poliovirus, optionally wherein the inactivated poliovirus comprises one or more of a type 1 virus, a type 2 virus, and a type 3 virus~~.

16. The use of any one of claims 8-14, wherein the immunogenic composition comprises about 3000 µg of the oligonucleotide~~method of any one of claims 1-15, wherein the human subject is at least 10 years of age or older~~.

17. The use of any one of claims 1-16~~method of claim 16~~, wherein the human subject is at least 10 years of age or older and has not received a pediatric vaccine or a booster vaccine within the previous 3 years, wherein

the pediatric vaccine comprises diphtheria, tetanus, and acellular pertussis (DTaP) antigens; or diphtheria, tetanus, and whole cell pertussis (DTwP) antigens; and

the booster vaccine comprises tetanus and diphtheria (Td) antigens; or tetanus, diphtheria, and acellular pertussis (Tdap) antigens.

18. The ~~method~~ use of claim 17, wherein the human subject is from 10-18 years of age.

19. The ~~method~~ use of claim 17, wherein the human subject is 19 years of age or older, or wherein the human subject is from 19-64 years of age, or 65 years of age or older.

20. The ~~method~~ use of any one of claims 17-19, wherein the Tdap antigens and the oligonucleotide are present in the immunogenic composition in amounts effective to stimulate an immune response against Tdap antigens by one month after administration, wherein the immune response comprises:

concentrations of anti-TT antibody, and anti-DT antibody of at least 0.4 international units/ml (IU/ml); and

concentrations of anti-PT antibody, anti-FHA antibody, and anti-PRN antibody of at least 2-fold higher in international units/ml (IU/ml) than pre-administration levels or limit of quantitation levels.

21. The ~~method~~ use of claim 20, wherein the immune response comprises:

(i) one or both of: an anti-TT antibody concentration and an anti-DT antibody concentration of at least 1.0 IU/ml by one month post-administration; and/or

(ii) one or more of: an anti-PT antibody concentration, an anti-FHA antibody concentration, and an anti-PRN antibody concentration of at least 4-fold higher by one month post-administration, wherein the antibody concentrations are relative to either the pre-administration levels or limit of quantitation levels in international units/ml (IU/ml).

22. The ~~method~~ use of claim 20, wherein the immune response comprises a sustained tetanus and/or diphtheria immune response comprising:

one or both of an anti-TT antibody concentration and an anti-DT antibody concentration of at least 1.5~~, 2.0 or 2.5~~ times the pre-administration levels at about 6 months ~~or about 12 months~~ post-administration, or wherein one or both of the anti-TT antibody concentration and the anti-DT antibody concentration is at least 2~~, 3 or 4~~ times the pre-administration levels at about 6 months ~~or about 12 months~~ post-administration.

23. The ~~method~~ use of claim 20, wherein the immune response comprises a sustained pertussis immune response comprising:

one or more of an anti-PT antibody concentration, an anti-FHA antibody concentration, and an anti-PRN antibody concentration of at least 1.5~~, 2.0 or 2.5~~ times the pre-administration level at about 6 months ~~or about 12 months~~ post-administration, or wherein one or more of the anti-PT antibody concentration, the anti-FHA antibody concentration, and the anti-PRN antibody concentration is at least 2~~, 3 or 4~~ times the pre-administration level at about 6 months ~~or about 12 months~~ post-administration.

24. The ~~method~~ use of any one of claims 20-23, wherein the immunogenic composition has a satisfactory safety profile.

25. An immunogenic composition ~~for active booster immunization against tetanus, diphtheria, and pertussis~~, comprising: tetanus, diphtheria, and acellular pertussis (Tdap) antigens, an aluminum salt adjuvant to which the Tdap antigens are adsorbed, and a toll-like receptor 9 (TLR9) agonist, wherein

the TLR9 agonist is an oligonucleotide of from 10 to 35 nucleotides in length comprising an unmethylated cytidine-phospho-guanosine (CpG) motif,

the immunogenic composition comprises from about 750 µg to about 6000 µg of the oligonucleotide, and

the Tdap antigens comprise a tetanus toxoid (TT), a diphtheria toxoid (DT), a pertussis toxoid (PT), a pertussis filamentous haemagglutinin (FHA), and pertussis pertactin (PRN)~~, and~~

~~the Tdap antigens and the oligonucleotide are present in the immunogenic composition in amounts effective to stimulate an immune response against the Tdap antigens in a human subject~~.

26. The immunogenic composition of claim 25, wherein the oligonucleotide comprises the sequence
5'-AACGTTCGAG-3' (SEQ ID NO:3).

27. The immunogenic composition of claim 25, wherein the oligonucleotide comprises the sequence of 5'-TGACTGTGAA CGTTCGAGAT GA-3'(SEQ ID NO:1).

28. The immunogenic composition of any one of claims 25-27, wherein the oligonucleotide comprises a modified nucleoside~~, optionally wherein the modified nucleoside is~~ selected from the group consisting of 2'-deoxy-7-deazaguanosine, 2'-deoxy-6-thioguanosine, arabinoguanosine, 2'-deoxy-2'substituted-arabinoguanosine, and 2'-O-substituted-arabinoguanosine.

29. The immunogenic composition of claim 28, wherein the oligonucleotide comprises the sequence
5'-TCG$_1$AACG$_1$TTCG$_1$-3' (SEQ ID NO:2) in which G$_1$ is 2'-deoxy-7-deazaguanosine, or wherein the oligonucleotide comprises the sequence 5'-TCG$_1$AACG$_1$TTCG$_1$-X-

$G_1CTTG_1CAAG_1CT$-5', and in which G_1 is 2'-deoxy-7-deazaguanosine and X is glycerol (5'-SEQ ID NO:2-3'-X-3'-SEQ ID NO:2-5').

30. The immunogenic composition of any one of claims 25-2927, wherein the oligonucleotide comprises at least one phosphorothioate linkage, or wherein all nucleotide linkages are phosphorothioate linkages.

31. The immunogenic composition of any one of claims 25-30, wherein the oligonucleotide is a single-stranded oligodeoxynucleotide.

32. The immunogenic composition of any one of claims 25-31~~claim 30~~, wherein ~~a 0.5 ml dose of~~ the immunogenic composition comprises from ~~about 375 µg to about 6000 µg of the oligonucleotide or from~~ about 1000~~750~~ µg to about 4000~~3000~~ µg of the oligonucleotide~~, or wherein a 0.5 ml dose of the immunogenic composition comprises about 375 µg, about 750 µg, about 1500 µg, about 3000 µg, or 6000 µg about of the oligonucleotide~~.

33. The immunogenic composition of any one of claims 25-32, wherein the aluminum salt adjuvant comprises one or more of the group consisting of amorphous aluminum hydroxyphosphate sulfate, aluminum hydroxide, aluminum phosphate, and potassium aluminum sulfate.

34. The immunogenic composition of any one of claims 25-32, wherein the aluminum salt adjuvant comprises aluminum hydroxide.

35. The immunogenic composition of claims 33 or claim 34~~claim 32~~, wherein ~~a 0.5 ml dose of~~ the immunogenic composition comprises from about 0.25 to about 0.50 mg Al^{3+}~~, or wherein a 0.5 ml dose of the immunogenic composition comprises from about 0.30 to about 0.40 mg Al^{3+}~~.

36. The immunogenic composition of any one of claims 25-35, wherein ~~a 0.5 ml dose of~~ the immunogenic composition comprises about 5 Lf TT, about 2.5 Lf DT, about 8 µg PT, about 8 µg FHA, and about 2.5 µg PRN.

37. The immunogenic composition of any one of claims 25-36, wherein the immunogenic composition further comprises at least one additional antigen.

38. The immunogenic composition of claim 37, wherein the at least one additional antigen comprises one or both of a pertussis fimbriae (FIM) antigen and a pertussis adenylate cyclase (AC) antigen.

39. The immunogenic composition of any one of claims 25-38, wherein the immunogenic composition comprises about 1500 µg of the oligonucleotide~~claim 37, wherein the at least one additional antigen comprises an inactivated poliovirus, optionally wherein the inactivated poliovirus comprises one or more of a type 1 virus, a type 2 virus, and a type 3 virus~~.

40. The immunogenic composition of any one of claims 25-38, wherein the immunogenic composition comprises about 3000 µg of the oligonucleotide.

41~~40~~. A kit comprising the immunogenic composition of any one of claims 25-40~~39~~, and instructions for administration of the immunogenic composition to stimulate ~~the~~ an immune response against the Tdap antigens in ~~the~~ a human subject.

42~~41~~. A kit comprising:

i) a first composition comprising tetanus, diphtheria, and acellular pertussis (Tdap) antigens and an aluminum salt adjuvant to which the Tdap antigens are adsorbed;

ii) a second composition comprising a TLR9 agonist; and

iii) instructions for mixing the first composition with the second composition to prepare the immunogenic composition of any one of claims 25-40~~39~~.

43~~42~~. The kit of claim 42~~41~~, further comprising:

iv) a further set of instructions for administration of the immunogenic composition to stimulate an immune response against the Tdap antigens in a human subject.

44~~43~~. The kit of claim 43~~42~~, further comprising a syringe and needle for intramuscular injection of the immunogenic composition.