(WO01/34809). As a refinement of this approach, others have identified proteins that are recognized by hyperimmune sera from patients who have suffered staphylococcal infection (WO01/98499, WO02/059148).

[0012] *S. aureus* secretes a plethora of virulence factors into the extracellular milieu (Archer, 1998; Dinges *et al.*, 2000; Foster, 2005; Shaw *et al.*, 2004; Sibbald *et al.*, 2006). Like most secreted proteins, these virulence factors are translocated by the Sec machinery across the plasma membrane. Proteins secreted by the Sec machinery bear an N-terminal leader peptide that is removed by leader peptidase once the pre-protein is engaged in the Sec translocon (Dalbey and Wickner, 1985; van Wely *et al.*, 2001). Recent genome analysis suggests that Actinobacteria and members of the Firmicutes encode an additional secretion system that recognizes a subset of proteins in a Sec-independent manner (Pallen, 2002). ESAT-6 (early secreted antigen target 6 kDa) and CFP-10 (culture filtrate antigen 10 kDa) of *Mycobacterium tuberculosis* represent the first substrates of this novel secretion system termed ESX-1 or Snm in *M. tuberculosis* (Andersen *et al.*, 1995; Hsu *et al.*, 2003; Pym *et al.*, 2003; Stanley *et al.*, 2003). In *S. aureus,* two ESAT-6 like factors designated EsxA and EsxB are secreted by the Ess pathway (ESAT-6 secretion system) (Burts *et al.*, 2005).

[0013] The first generation of vaccines targeted against *S. aureus* or against the exoproteins it produces have met with limited success (Lee, 1996). There remains a need to develop effective vaccines against staphylococcal infections. Additional compositions for treating staphylococcal infections are also needed.

SUMMARY ~~OF THE INVENTION~~

[0013a] In a first aspect, there is provided an isolated polypeptide comprising a variant Protein A (SpA) having (a) at least one amino acid substitution that disrupts Fc binding and (b) at least a second amino acid substitution that disrupts VH3 binding and (c) an amino acid sequence that is at least 70% identical to the amino acid sequence of SEQ ID NO:2, wherein the variant SpA comprises a variant domain D segment having a glycine residue amino acid substitution at amino acids corresponding to position 9 and 10 of SEQ ID NO: 2.

[0013b] In a second aspect, there is provided a peptide composition comprising, in a pharmaceutically acceptable composition, a non-toxigenic variant Protein A (SpA) peptide having mutations that attenuate the binding of the Protein A domain D to IgG, Fcγ, VH3 $F(ab)_2$, von Willebrand factor (vWF), and tumor necrosis factor α receptor 1 (TNFR1), wherein the composition is capable of stimulating an immune response in a subject in need thereof, wherein the SpA peptide comprises glycine residue amino acid substitutions at amino acids corresponding to position 9 and 10 of SEQ ID NO: 2.

[0013c] In a third aspect, there is provided an immunogenic composition comprising an isolated peptide comprising a Protein A (SpA) variant having an amino acid substitution at amino acids corresponding to positions 9, 10, 36, and 37 of SEQ ID NO:2, wherein the substitutions at amino acids corresponding to positions 9 and 10 are with a glycine residue and the substitutions at amino acids corresponding to positions 36 and 37 are with a serine residue.

[0013d] In a fourth aspect, there is provided a vaccine comprising the composition as defined herein and a pharmaceutically acceptable excipient.

[0013e] In a fifth aspect, there is provided a method of making a vaccine comprising the steps of mixing antigens to make the composition as defined herein and adding a pharmaceutically acceptable excipient.

[0013f] In a sixth aspect, there is provided a use of the vaccine as defined herein in the manufacture of a medicament for preventing or treating staphylococcal infection.

[0013g] In a seventh aspect, there is provided a use of the composition as defined herein in the manufacture of a vaccine for treatment or prevention of staphylococcal infection.

[0013h] In an eighth aspect, there is provided a method of preparing an immunoglobulin for use in prevention or treatment of staphylococcal infection comprising the steps of immunizing a recipient with the vaccine as defined herein and isolating immunoglobulin from the recipient.

[0013i] In a ninth aspect, there is provided a use of a composition comprising a Protein A (SpA) variant having a glycine residue amino acid substitution at amino acids corresponding to positions 9 and 10 of SEQ ID NO:2, in the manufacture of a medicament for eliciting an immune response against a staphylococcus bacterium in a subject.

[0013j] In a tenth aspect, there is provided a use of an isolated peptide comprising a Protein A variant having a glycine residue amino acid substitution at amino acids corresponding to positions 9 and 10 of SEQ ID NO:2, in the manufacture of a medicament for treating a staphylococcal infection in a subject having, suspected of having or at risk of developing a staphylococcal infection.

[0014] Protein A (SpA)(SEQ ID NO:33), a cell wall anchored surface protein of *Staphylococcus aureus*, provides for bacterial evasion from innate and adaptive immune responses. Protein A binds immunoglobulins at their Fc portion, interacts with the VH3 domain of B cell receptors inappropriately stimulating B cell proliferation and apotosis, binds to von Willebrand factor A1 domains to activate intracellular clotting, and also binds to the TNF Receptor-1 to contribute to the pathogenesis of staphylococcal pneumonia. Due to the fact that Protein A captures immunoglobulin and displays toxic attributes, the possibility that this surface molecule may function as a vaccine in humans has not been rigorously pursued. Here the inventors demonstrate that Protein A variants no longer able to bind to immunoglobulins, which are thereby removed of their toxigenic potential, *i.e.*, are non-toxigenic, stimulate humoral immune responses that protect against staphylococcal disease.

[0015] In certain embodiments the SpA variant is a full length SpA variant comprising a variant A, B, C, D, and/or E domain. In certain ~~aspects~~embodiments, the SpA variant comprises or consists of the amino acid sequence that is 80, 90, 95, 98, 99, or 100% identical to the amino acid sequence of SEQ ID NO:34 In other embodiments the SpA variant comprises a segment of SpA. The SpA segment can comprise at least or at most 1, 2, 3, 4, 5 or more IgG binding domains. The IgG domains can be at least or at most 1, 2, 3, 4, 5 or more variant A, B, C, D, or E domains. In certain ~~aspects~~embodiments the SpA variant comprises at least or at most 1, 2, 3, 4, 5, or more variant A domains. In a further ~~aspect~~embodiment the SpA variant comprises at least or at most 1, 2, 3, 4, 5, or more variant B domains. In still a further ~~aspect~~embodiment the SpA variant comprises at least or at most 1, 2, 3, 4, 5, or more variant C domains. In yet a further ~~aspect~~embodiment the SpA variant comprises at least or at most 1, 2, 3, 4, 5, or more variant D domains. In certain ~~aspects~~embodiments the SpA variant comprises at least or at most 1, 2, 3, 4, 5, or more variant E domains. In a further ~~aspect~~embodiment the SpA variant comprises a combination of A, B, C, D, and E domains in various combinations and permutations. The combinations can include all or part of a SpA signal peptide segment, a SpA region X segment, and/or a SpA sorting signal segment. In other ~~aspects~~embodiments the SpA variant does not include a SpA signal peptide segment, a SpA region X segment, and/or a SpA sorting signal segment. In certain ~~aspects~~embodiments a variant A domain comprises a substitution at position(s) 7, 8, 34, and/or 35 of SEQ ID NO:4. In another ~~aspect~~embodiment a variant B domain comprises a substitution at position(s) 7, 8, 34, and/or 35 of SEQ ID NO:6. In still anther ~~aspect~~embodiment a variant C domain comprises a substitution at position(s) 7, 8, 34, and/or 35 of SEQ ID NO:5. In certain ~~aspects~~embodiments a variant D domain comprises a substitution at position(s) 9, 10, 36, and/or 37 of SEQ ID NO:2. In a further ~~aspect~~embodiment a variant E domain comprises a substitution at position(s) 6, 7, 33, and/or 34 of SEQ ID NO:3.

[0016] In certain ~~aspects~~embodiments, an SpA domain D variant or its equivalent can comprise a mutation at position 9 and 36; 9 and 37; 9 and 10; 36 and 37; 10 and 36; 10 and 37; 9, 36, and 37; 10, 36, and 37, 9, 10 and 36; or 9,10 and 37 of SEQ ID NO:2. In a further ~~aspect~~embodiment, analogous mutations can be included in one or more of domains A, B, C, or E.

[0017] In further ~~aspects~~embodiments, the amino acid glutamine (Q) at position 9 of SEQ ID NO:2 (or its analogous amino acid in other SpA domains) can be replaced with an alanine (A), an asparagine (N), an aspartic acid (D), a cysteine (C), a glutamic acid (E), a phenylalanine (F), a glycine (G), a histidine (H), an isoleucine (I), a lysine (K), a leucine (L), a methionine (M), a proline (P), a serine (S), a threonine (T), a valine (V), a tryptophane (W), or a tyrosine (Y).

In some ~~aspects~~embodiments the glutamine at position 9 can be substituted with an arginine (R). In a further ~~aspect~~embodiment, the glutamine at position 9 of SEQ ID NO:2, or its equivalent, can be substituted with a lysine or a glycine. Any 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more of the substitutions can be explicitly excluded.

[0018] In another ~~aspect~~embodiment, the amino acid glutamine (Q) at position 10 of SEQ ID NO:2 (or its analogous amino acid in other SpA domains) can be replaced with an alanine (A), an asparagine (N), an aspartic acid (D), a cysteine (C), a glutamic acid (E), a phenylalanine (F), a glycine (G), a histidine (H), an isoleucine (I), a lysine (K), a leucine (L), a methionine (M), a proline (P), a serine (S), a threonine (T), a valine (V), a tryptophane (W), or a tyrosine (Y). In some ~~aspects~~embodiments the glutamine at position 10 can be substituted with an arginine (R). In a further ~~aspect~~embodiment, the glutamine at position 10 of SEQ ID NO:2, or its equivalent, can be substituted with a lysine or a glycine. Any 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more of the substitutions can be explicitly excluded.

[0019] In certain ~~aspects~~embodiments, the aspartic acid (D) at position 36 of SEQ ID NO:2 (or its analogous amino acid in other SpA domains) can be replaced with an alanine (A), an asparagine (N), an arginine (R), a cysteine (C), a phenylalanine (F), a glycine (G), a histidine (H), an isoleucine (I), a lysine (K), a leucine (L), a methionine (M), a proline (P), a glutamine (Q), a serine (S), a threonine (T), a valine (V), a tryptophane (W), or a tyrosine (Y). In some ~~aspects~~embodiments the aspartic acid at position 36 can be substituted with a glutamic acid (E). In certain ~~aspects~~embodiments, an aspartic acid at position 36 of SEQ ID NO:2, or its equivalent, can be substituted with an alanine or a serine. Any 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more of the substitutions can be explicitly excluded.

[0020] In another ~~aspect~~embodiment, the aspartic acid (D) at position 37 of SEQ ID NO:2 (or its analogous amino acid in other SpA domains) can be replaced with an alanine (A), a an asparagine (N), an arginine (R), a cysteine (C), a phenylalanine (F), a glycine (G), a histidine (H), an isoleucine (I), a lysine (K), a leucine (L), a methionine (M), a proline (P), a glutamine (Q), a serine (S), a threonine (T), a valine (V), a tryptophane (W), or a tyrosine (Y). In some ~~aspects~~embodiments the aspartic acid at position 37 can be substituted with a glutamic acid (E). In certain ~~aspects~~embodiments, an aspartic acid at position 37 of SEQ ID NO:2, or its equivalent, can be substituted with an alanine or a serine. Any 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more of the substitutions can be explicitly excluded.

[0021] In a particular embodiment the amino at position 9 of SEQ ID NO:2 (or an analogous amino acid in another SpA domain) is replaced by an alanine (A), a glycine (G), an isoleucine (I), a leucine (L), a proline (P), a serine (S), or a valine (V), In certain ~~aspects~~embodiments the amino acid at position 9 of SEQ ID NO:2 is replaced by a glycine. In a further ~~aspect~~embodiment the amino acid at position 9 of SEQ ID NO:2 is replaced by a lysine.

[0022] In a particular embodiment the amino at position 10 of SEQ ID NO:2 (or an analogous amino acid in another SpA domain) is replaced by an alanine (A), a glycine (G), an isoleucine (I), a leucine (L), a proline (P), a serine (S), or a valine (V), In certain ~~aspects~~embodiments the amino acid at position 10 of SEQ ID NO:2 is replaced by a glycine. In a further ~~aspect~~embodiment the amino acid at position 10 of SEQ ID NO:2 is replaced by a lysine.

[0023] In a particular embodiment the amino at position 36 of SEQ ID NO:2 (or an analogous amino acid in another SpA domain) is replaced by an alanine (A), a glycine (G), an isoleucine (I), a leucine (L), a proline (P), a serine (S), or a valine (V), In certain ~~aspects~~embodiments the amino acid at position 36 of SEQ ID NO:2 is replaced by a serine. In a further ~~aspect~~embodiment the amino acid at position 36 of SEQ ID NO:2 is replaced by an alanine.

[0024] In a particular embodiment the amino at position 37 of SEQ ID NO:2 (or an analogous amino acid in another SpA domain) is replaced by an alanine (A), a glycine (G), an isoleucine (I), a leucine (L), a proline (P), a serine (S), or a valine (V), In certain ~~aspects~~embodiments the amino acid at position 37 of SEQ ID NO:2 is replaced by a serine. In a further ~~aspect~~embodiment the amino acid at position 37 of SEQ ID NO:2 is replaced by an alanine.

[0025] In certain ~~aspects~~embodiments the SpA variant includes a substitution of (a) one or more amino acid substitution in an IgG Fc binding sub-domain of SpA domain A, B, C, D, and/or E that disrupts or decreases binding to IgG Fc, and (b) one or more amino acid substitution in a V_H3 binding sub-domain of SpA domain A, B, C, D, and/or E that disrupts or decreases binding to V_H3. In still further ~~aspects~~embodiments the amino acid sequence of a SpA variant comprises an amino acid sequence that is at least 50%, 60%, 70%, 80%, 90%, 95%, or 100% identical, including all values and ranges there between, to the amino acid sequence of SEQ ID NOs:2-6.

[0026] In a further ~~aspect~~embodiment the SpA variant includes (a) one or more amino acid substitution in an IgG Fc binding sub-domain of SpA domain D, or at a corresponding amino acid position in other IgG domains, that disrupts or decreases binding to IgG Fc, and (b) one or more amino acid substitution in a V_H3 binding sub-domain of SpA domain D, or at a

corresponding amino acid position in other IgG domains, that disrupts or decreases binding to V_H3. In certain ~~aspects~~embodiments amino acid residue F5, Q9, Q10, S11, F13, Y14, L17, N28, I31, and/or K35 (SEQ ID NO:2, QQNNFNKDQQSAFYEILNMPNLNEAQRNGFIQSLKDDPSQSTNVLGEAKKLNES) of the IgG Fc binding sub-domain of domain D are modified or substituted. In certain ~~aspects~~embodiments amino acid residue Q26, G29, F30, S33, D36, D37, Q40, N43, and/or E47 (SEQ ID NO:2) of the V_H3 binding sub-domain of domain D are modified or substituted such that binding to Fc or V_H3 is attenuated. In further ~~aspects~~embodiments corresponding modifications or substitutions can be engineered in corresponding positions of the domain A, B, C, and/or E. Corresponding positions are defined by alignment of the domain D amino acid sequence with one or more of the amino acid sequences from other IgG binding domains of SpA, for example see FIG. 2A. In certain ~~aspects~~embodiments the amino acid substitution can be any of the other 20 amino acids. In a further ~~aspect~~embodiment conservative amino acid substitutions can be specifically excluded from possible amino acid substitutions. In other ~~aspects~~embodiments only non-conservative substitutions are included. In any event, any substitution or combination of substitutions that reduces the binding of the domain such that SpA toxicity is significantly reduced is contemplated. The significance of the reduction in binding refers to a variant that produces minimal to no toxicity when introduced into a subject and can be assessed using in vitro methods described herein.

[0027] In certain embodiments, a variant SpA comprises at least or at most 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more variant SpA domain D peptides. In certain ~~aspects~~embodiments 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, or 19 or more amino acid residues of the variant SpA are substituted or modified - including but not limited to amino acids F5, Q9, Q10, S11, F13, Y14, L17, N28, I31, and/or K35 (SEQ ID NO:2) of the IgG Fc binding sub-domain of domain D and amino acid residue Q26, G29, F30, S33, D36, D37, Q40, N43, and/or E47 (SEQ ID NO:2) of the V_H3 binding sub-domain of domain D. In one ~~aspect~~embodiment of the invention glutamine residues at position 9 and/or 10 of SEQ ID NO:2 (or corresponding positions in other domains) are mutated. In another ~~aspect~~embodiment, aspartic acid residues 36 and/or 37 of SEQ ID NO:2 (or corresponding positions in other domains) are mutated. In a further ~~aspect~~embodiment, glutamine 9 and 10, and aspartic acid residues 36 and 37 are mutated. Purified non-toxigenic SpA or SpA-D mutants/variants described herein are no longer able to significantly bind (i.e., demonstrate attenuated or disrupted binding affinity) Fcγ or $F(ab)_2$ V_H3 and also do not stimulate B cell apoptosis. These non-toxigenic Protein A variants can be used as subunit

vaccines and raise humoral immune responses and confer protective immunity against *S. aureus* challenge. Compared to wild-type full-length Protein A or the wild-type SpA-domain D, immunization with SpA-D variants resulted in an increase in Protein A specific antibody. Using a mouse model of staphylococcal challenge and abscess formation, it was observed that immunization with the non-toxigenic Protein A variants generated significant protection from staphylococcal infection and abscess formation. As virtually all *S. aureus* strains express Protein A, immunization of humans with the non-toxigenic Protein A variants can neutralize this virulence factor and thereby establish protective immunity. In certain ~~aspects~~embodiments the protective immunity protects or ameliorates infection by drug resistant strains of Staphylococcus, such as USA300 and other MRSA strains.

[0028] Embodiments include the use of Protein A variants in methods and compositions for the treatment of bacterial and/or staphylococcal infection. This application also provides an immunogenic composition comprising a Protein A variant or immunogenic fragment thereof. In certain ~~aspects~~embodiments, the immunogenic fragment is a Protein A domain D segment. Furthermore, the present ~~invention~~disclosure provides methods and compositions that can be used to treat (*e.g.*, limiting staphylococcal abscess formation and/or persistence in a subject) or prevent bacterial infection. In some cases, methods for stimulating an immune response involve administering to the subject an effective amount of a composition including or encoding all or part of a Protein A variant polypeptide or antigen, and in certain ~~aspects~~embodiments other bacterial proteins. Other bacterial proteins include, but are not limited to (i) a secreted virulence factor, and/or a cell surface protein or peptide, or (ii) a recombinant nucleic acid molecule encoding a secreted virulence factor, and/or a cell surface protein or peptide.

[0029] In other ~~aspects~~embodiments, the subject can be administered all or part of a Protein A variant, such as a variant Protein A domain D segment. The polypeptide of the ~~invention~~disclosure can be formulated in a pharmaceutically acceptable composition. The composition can further comprise one or more of at least or at most 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, or 19 additional staphylococcal antigen or immunogenic fragment thereof (*e.g.*, Eap, Ebh, Emp, EsaB, EsaC, EsxA, EsxB, SdrC, SdrD, SdrE, IsdA, IsdB, ClfA, ClfB, Coa, Hla (*e.g.*, H35 mutants), IsdC, SasF, vWbp, or vWh). Additional staphylococcal antigens that can be used in combination with a Protein A variant include, but are not limited to 52kDa vitronectin binding protein (WO 01/60852), Aaa (GenBank CAC80837), Aap (GenBank accession AJ249487), Ant (GenBank accession NP_372518), autolysin glucosaminidase,

autolysin amidase, Cna, collagen binding protein (US6288214), EFB (FIB), Elastin binding protein (EbpS), EPB, FbpA, fibrinogen binding protein (US6008341), Fibronectin binding protein (US5840846), FnbA, FnbB, GehD (US 2002/0169288), HarA, HBP, Immunodominant ABC transporter, IsaA/PisA, laminin receptor, Lipase GehD, MAP, Mg2+ transporter, MHC II analogue (US5648240), MRPII, Npase, RNA III activating protein (RAP), SasA, SasB, SasC, SasD, SasK,SBI, SdrF(WO 00/12689), SdrG / Fig (WO 00/12689), SdrH (WO 00/12689), SEA exotoxins (WO 00/02523), SEB exotoxins (WO 00/02523), SitC and Ni ABC transporter, SitC/MntC/saliva binding protein (US5,801,234), SsaA, SSP-1, SSP-2, and/or Vitronectin binding protein (see PCT publications WO2007/113222, WO2007/113223, WO2006/032472, WO2006/032475, WO2006/032500, each of which is incorporated herein by reference in their entirety). The staphylococcal antigen or immunogenic fragment can be administered concurrently with the Protein A variant. The staphylococcal antigen or immunogenic fragment and the Protein A variant can be administered in the same composition. The Protein A variant can also be a recombinant nucleic acid molecule encoding a Protein A variant. A recombinant nucleic acid molecule can encode the Protein A variant and at least one staphylococcal antigen or immunogenic fragment thereof. As used herein, the term "modulate" or "modulation" encompasses the meanings of the words "enhance," or "inhibit." "Modulation" of activity may be either an increase or a decrease in activity. As used herein, the term "modulator" refers to compounds that effect the function of a moiety, including up-regulation, induction, stimulation, potentiation, inhibition, down-regulation, or suppression of a protein, nucleic acid, gene, organism or the like.

[0030] In certain embodiments the methods and compositions use or include or encode all or part of the Protein A variant or antigen. In other ~~aspects~~embodiments, the Protein A variant may be used in combination with secreted factors or surface antigens including, but not limited to one or more of an isolated Eap, Ebh, Emp, EsaB, EsaC, EsxA, EsxB, SdrC, SdrD, SdrE, IsdA, IsdB, ClfA, ClfB, Coa, Hla, IsdC, SasF, vWbp, or vWh polypeptide or immunogenic segment thereof. Additional staphylococcal antigens that can be used in combination with a Protein A variant include, but are not limited to 52kDa vitronectin binding protein (WO 01/60852), Aaa, Aap, Ant, autolysin glucosaminidase, autolysin amidase, Cna, collagen binding protein (US6288214), EFB (FIB), Elastin binding protein (EbpS), EPB, FbpA, fibrinogen binding protein (US6008341), Fibronectin binding protein (US5840846), FnbA, FnbB, GehD (US 2002/0169288), HarA, HBP, Immunodominant ABC transporter, IsaA/PisA, laminin

receptor, Lipase GehD, MAP, Mg2+ transporter, MHC II analogue (US5648240), MRPII, Npase, RNA III activating protein (RAP), SasA, SasB, SasC, SasD, SasK,SBI, SdrF(WO 00/12689), SdrG / Fig (WO 00/12689), SdrH (WO 00/12689), SEA exotoxins (WO 00/02523), SEB exotoxins (WO 00/02523), SitC and Ni ABC transporter, SitC/MntC/saliva binding protein (US5,801,234), SsaA, SSP-1, SSP-2, and/or Vitronectin binding protein. In certain embodiments, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 or more of Eap, Ebh, Emp, EsaB, EsaC, EsxA, EsxB, SdrC, SdrD, SdrE, IsdA, IsdB, ClfA, ClfB, Coa, Hla, IsdC, SasF, vWbp, vWh, 52kDa vitronectin binding protein (WO 01/60852), Aaa, Aap, Ant, autolysin glucosaminidase, autolysin amidase, Cna, collagen binding protein (US6288214), EFB (FIB), Elastin binding protein (EbpS), EPB, FbpA, fibrinogen binding protein (US6008341), Fibronectin binding protein (US5840846), FnbA, FnbB, GehD (US 2002/0169288), HarA, HBP, Immunodominant ABC transporter, IsaA/PisA, laminin receptor, Lipase GehD, MAP, Mg2+ transporter, MHC II analogue (US5648240), MRPII, Npase, RNA III activating protein (RAP), SasA, SasB, SasC, SasD, SasK,SBI, SdrF(WO 00/12689), SdrG / Fig (WO 00/12689), SdrH (WO 00/12689), SEA exotoxins (WO 00/02523), SEB exotoxins (WO 00/02523), SitC and Ni ABC transporter, SitC/MntC/saliva binding protein (US5,801,234), SsaA, SSP-1, SSP-2, and/or Vitronectin binding protein. can be specifically excluded from a formulation of the ~~invention~~disclosure.

[0031] The following table lists the various combinations of SpA variants and various other Staphyloccal antigens

Table 1. SpA and staphylococcal antigen combinations.

Eap	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Ebh		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Emp			+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
EsaB				+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
EsaC					+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
EsxA						+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
EsxB							+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SdrC								+	+	+	+	+	+	+	+	+	+	+	+	+	+