WE CLAIM:

1. A modified factor H binding protein (fHbp) comprising wild type fHbp variant and at least one exogenous loop(s), wherein
    - the modified factor H binding protein (fHbp) is selected from amino acid sequence with at least 75% identity with any one of sequences of SEQ ID Nos 6 to 10,
    - the at least one exogenous peptide loop(s) is immunogenic,
    - the at least one exogenous peptide loop(s) is derived from a bacterial membrane protein,
    - the modified fHbp is a fusion protein,
    - the fHbp variant is selected from v1, v2 and v3, modified with at least one PorA loop comprising at least 10 amino acids inserted into a β-turn region in fHbp; and
    - the PorA loop is selected from VR1, and VR2,

    optionally, wherein the modified fHbp is modified to reduce factor H binding activity.

2. A nucleic acid sequence encoding the modified fHbp, wherein the nucleic acid sequence is with at least 75% identity with any one of sequences of SEQ ID NO. 1 to 5.

3. An immunogenic composition comprising at least one modified fHbp as claimed in any one of the claims 1 or the nucleic acid sequence encoding the modified fHbp as claimed in claim 2.

4. The immunogenic composition as claimed in claim 3, wherein the composition comprises:
    a pharmaceutically acceptable carrier;
    an adjuvant;
    at least one other prophylactically or therapeutically active molecule comprising a monovalent carrier protein: capsule polysaccharide conjugate vaccine;
    or combination thereof.

5. The immunogenic composition as claimed in any one of the claims 3 or 4, wherein the fHbp scaffold bearing exogenous peptide loops is incorporated as the protein carrier molecule in the conjugate vaccine.

6. The immunogenic composition as claimed in any one of the claims 3 to 5 comprising a recombinant protein/ modified fHbp fusion protein in combination with at least one additional antigen selected from:

- a protein antigen from PorB, Fet A, OmpC, NHBA, NadA, meningococcal antigen 287, NspA, HmbR, NhhA, App, 936,
- a saccharide or conjugate antigen from N. meningitidis serogroup A, C, W, Y and/or X,
- a saccharide or conjugate antigen from Streptococcus pneumoniae,
- a diphtheria antigen, such as a diphtheria toxoid e.g. the CRM197 mutant,
- a tetanus antigen, such as a tetanus toxoid,
- an antigen from Bordetella pertussis, acellular or whole cell pertussis antigens,
- a saccharide or conjugate antigen from Haemophilus influenzae B,
- polio antigen(s) such as IPV,
- measles, mumps and/or rubella antigens,
- influenza antigen(s), such as the haemagglutinin and/or neuraminidase surface proteins,
- antigen (protein or saccharide or conjugate) from Streptococcus agalactiae (group B streptococcus),
- antigen (protein or saccharide or conjugate) from Streptococcus pyogenes (group A streptococcus),
- antigen (protein or saccharide or conjugate) from Staphylococcus aureus,
- antigen (protein or saccharide or conjugate) from Salmonella Spp.

7. The immunogenic composition as claimed in any one of the claims 3 to 6, wherein the fHbp scaffold bearing exogenous peptide loops is incorporated as the protein carrier molecule in the polysaccharide conjugate vaccine selected from Monovalent (A, X, C, W, Y), Bivalent (A-C, A-B, X-B, C-B), Trivalent (AC-B, AC-Hib), Quadrivalent (AC-Hib-B), Pentavalent (ACWYX), or Hexavalent (ACWYX-B).

8. A combination of the modified fHbp as claimed in any one of claims 1, a nucleic acid as claimed in claim 2, or a composition as claimed in any one of claims 3 or 4, and at least one other prophylactically or therapeutically active molecule,
optionally, wherein the at least one other prophylactically or therapeutically active molecule comprises a conjugate vaccine, comprising any of serogroup capsular polysaccharides selected from A, C, Y, W, or X strains, or combinations thereof,
optionally, wherein the protein: capsule polysaccharide vaccine comprises any of serogroup C or A capsule with bacterial toxoids, bivalent vaccines (with serogroup C and A capsular polysaccharide conjugated to bacterial toxoids), quadrivalent (serogroups A, C, Y, W polysaccharides conjugated to bacterial toxoids) or pentavalent (serogroups A, C, Y, W, X polysaccharides conjugated to bacterial toxoid) conjugate vaccines.

9. The factor H binding protein (fHbp) as claimed in claim 1 as and when used as an epitope display scaffold.

10. A vaccine formulation comprising at least one recombinant protein/ modified fHbp fusion protein, an adjuvant and one or more pharmaceutically acceptable excipient, wherein in the recombinant protein/ modified fHbp fusion protein the one or more exogenous loops is selected from but not limited to group consisting of Transferrin Binding Protein, Neisserial Heparin Binding Protein, Neisserial Surface Protein A, PorA, meningococcal enterobactin receptor FetA, Neisserial Adhesin A, the fHbp-fHbp fusion protein as claimed in any one of the previous claims or a combination thereof,
optionally, wherein the recombinant protein/ modified fHbp fusion protein is fHbp as claimed in any one of claims 1, or the nucleic acid sequence encoding the modified fHbp as claimed in claim 2, and wherein the fHbp is derived from Neisseria meningitidis serogroup A, B, C, H, I, K, L, 29E, W135, X, Y and Z.

11. The vaccine formulation as claimed in claim 10, wherein the recombinant protein/ modified fHbp fusion protein is derived from Neisseria meningitidis serogroup B, optionally, wherein the recombinant protein/ modified fHbp fusion protein is fHbp has molecular weight in the range of 10 kDa to 200 kDa, preferably up to 50 kDa.

12. The vaccine formulation as claimed in any one of the claims 10 or 11, wherein the adjuvant is selected from the group consisting of aluminum hydroxide, aluminum phosphate, aluminum hydroxyphosphate, and potassium aluminum sulfate, MF-59, a liposome, a lipopolysaccharide, a saponin, lipid A, lipid A derivatives, Monophosphoryl lipid A, GLA, 3–deacylated monophosphoryl lipid A, AS01, AS03, AF3, IL-2, RANTES, GM- CSF, TNF-a, IFN-g, G-CSF, LFA-3, CD72, B7-1, B7-2, OX-40L and 41 BBL, an oligonucleotide, an oligonucleotide comprising at least one unmethylated CpG and/or a liposome, Freund's adjuvant, Freund's complete adjuvant, Freund's incomplete adjuvant, polymers, co-polymers such as polyoxyethylene-polyoxypropylene copolymers, including block co-polymers, polymer p 1005, CRL-8300 adjuvant, muramyl dipeptide, such as an agonist of TLR1/2 TLR2, TLR3, TLR-4 agonists, TLR5, TLR7, TLR7/8, TLR8, TLR9, ODN 2216 (type A), TLR11/12, TLR-4 agonists, flagellin, flagellins derived from gram negative bacteria, TLR-5 agonists, fragments of flagellins capable of binding to TLR-5 receptors, Alpha-C-galactosylceramide, Chitosan, Interleukin-2, QS-21, squalene, Quil A, cholera toxin B subunit, polyphosphazene and derivatives, mycobacterium cell wall preparations, mycolic acid derivatives, non-ionic block copolymer surfactants, OMV, fHbp, saponin combination with sterols and lipids, dmLT, 1,25-dihydroxyvitamin D3, CAF01, poly [di (carboxylatophenoxy)- phosphazene] (PCPP) and Venezuelan equine encephalitis (VEE) replicon particles or a combination thereof.

13. The vaccine formulation as claimed in any one of the claims 10 to 12, wherein the adjuvant is aluminium hydroxide having particle size > 500 nm.

14. The vaccine formulation as claimed in any one of the claims 10 to 13, wherein the one or more pharmaceutically acceptable excipient is

a. a buffering agent selected from carbonate, phosphate, acetate, HEPES, Succinate, TRIS, borate, citrate, lactate, gluconate, tartrate, or a combination thereof;

b. a sugar selected from trehalose, mannose, raffinose, lactobionic acid, glucose, maltulose, iso- maltulose, maltose, lactose, dextrose, fructose, or a combination thereof;

c. a sugar alcohol or polyol selected from mannitol, lactitol, sorbitol, glycerol, xylitol, maltitol, lactitol, erythritol, isomalt and hydrogenated starch hydrolysates or a combination thereof;

d. a surfactant selected from polysorbate 20, polysorbate 40, polysorbate 60, polysorbate 65, polysorbate 85, nonylphenoxypolyethoxethanol, octylphenoxypolyethoxethanol, oxtoxynol 40, nonoxynol- 9, triethanolamine, triethanolamine polypeptide oleate, polyoxyethylene- 660 hydroxystearate, polyoxyethylene- 35 ricinoleate, soy lecithin, a poloxamer, copolymers of ethylene oxide (EO), propylene oxide (PO), and/or butylene oxide (BO), octoxynols, phospholipids, nonylphenol ethoxylates, polyoxyethylene fatty ethers derived from lauryl, cetyl, stearyl and oleyl alcohols, sorbitan esters or a combination thereof;

e. a polymer selected from dextran, carboxymethylcellulose, hyaluronic acid, cyclodextrin or a combination thereof;

f. a salt selected from NaCl, KCl, KH_2PO_4, $Na_2HPO_4.2H_2O$, $CaC1_2$, $MgC1_2$, or a combination thereof;

g. an amino acid selected from tricine, leucine, iso-leucine, glycine, glutamine, L-arginine, L-arginine hydrochloride, lysine, L-alanine, Tryptophan, Phenylalanine, Tyrosine, Valine, Cysteine, Glycine, Methionine, Proline, Serine, Threonine, or a combination thereof;

h. a hydrolysed protein selected from gelatin, lactalbumin hydrolysate, monosodium glutamate, collagen hydrolysate, keratin hydrolysate, peptides, Casein hydrolysate, whey protein hydrolysate, serum albumin or a combination thereof;

i. a preservative selected from phenoxyethanol, Benzethonium chloride (Phemerol), Phenol, m-cresol, Thiomersal, Formaldehyde, paraben esters, benzalkonium chloride, benzyl alcohol, chlorobutanol, p-chlor-m-cresol, benzyl alcohol or a combination thereof; and

j. a liquid carrier selected from water for injection (WFI) or saline.

15. The vaccine formulation as claimed in any one of the claims 10 to 14, wherein the vaccine formulation comprises:
    - at least one at least one recombinant protein/ at least one modified fHbp as claimed in any one of previous claims
    - aluminium hydroxide;
    - mannitol;
    - phosphate; and
    - polysorbate.

16. The vaccine formulation as claimed in any one of the claims 10 to 15, wherein the vaccine formulation comprises:
    - at least one modified fHbp of the amino acid sequence with at least 75% identity with from any one of amino acid sequences SEQ ID NO. 6 to 10; or
    - at least one modified fHbp encoded the nucleic acid sequence is with at least 75% identity with by any one of nucleic acid sequence SEQ ID NO. 1 to 5; and
    - aluminium hydroxide;
    - mannitol;
    - phosphate; and
    - polysorbate.

17. The vaccine formulation as claimed in any one of the claims 10 to 16, comprising (i) at least one recombinant protein/ modified fHbp fusion protein; (ii) aluminium hydroxide in an amount in the range of 0.5 mg/ml to 4.5 mg/ml; (iii) mannitol in an amount in the range of 5 mg/ml to 100 mg/ml; (iv) phosphate buffer in an amount in the range of 1 mM to 10 mM; and (v) polysorbate 20 in an amount in the range of 0.01 mg/ml to 2 mg/ml; wherein each recombinant protein/ modified fHbp fusion protein is present in an amount in the range of 15 µg/ml to 150 µg/ml.

18. The vaccine formulation as claimed in any one of the claims 10 to 17, comprising (i) at least four recombinant proteins/ modified fHbp fusion proteins; (ii) aluminium hydroxide in an amount in the range of 0.5 mg/ml to 4.5 mg/ml; (iii) mannitol in an

amount in the range of 5 mg/ml to 100 mg/ml; (iv) phosphate buffer in an amount in the range of 1 mM to 10 mM; and (v) polysorbate 20 in an amount in the range of 0.01 mg/ml to 2 mg/ml; wherein each recombinant protein/ modified fHbp fusion protein is present in an amount in the range of 15 µg/ml to 150 µg/ml.

19. The vaccine formulation as claimed in any one of the claims 10 to 18, wherein the vaccine formulation comprises:

- at least one modified fHbp of the amino acid sequence with at least 75% identity with from any one of amino acid sequences SEQ ID NO. 6 to 10; or
- at least one modified fHbp encoded the nucleic acid sequence is with at least 75% identity with by any one of nucleic acid sequence SEQ ID NO. 1 to 5;
  and
- aluminium hydroxide in an amount in the range of 0.5 mg/ml to 4.5 mg/ml;
- mannitol in an amount in the range of 5 mg/ml to 100 mg/ml;
- phosphate buffer in an amount in the range of 1 mM to 10 mM; and
- polysorbate 20 in an amount in the range of 0.01 mg/ml to 2 mg/ml.

20. The vaccine formulation as claimed in any one of the claims 10 to 19, comprising:
    a. fHbpV3.45 M5:PorA316-320/exP1.14 (of SEQ ID NO. 6 or encoded by SEQ ID NO. 1) in an amount in the range of 15 µg/ml to 150 µg/ml; or
    b. fHbpV2.19 M6:PorA316-320/exP1.4 (of SEQ ID NO. 7 or encoded by SEQ ID NO. 2) in an amount in the range of 15 µg/ml to 150 µg/ml; or
    c. fHbpV1.14:PorA307-311/exP1.9 (of SEQ ID NO. 8 or encoded by SEQ ID NO. 3) in an amount in the range of 15 µg/ml to 150 µg/ml; or
    d. fHbpV1.1:PorA307-311/exP1.4 (of SEQ ID NO. 9 or encoded by SEQ ID NO. 4); in an amount in the range of 15 µg/ml to 150 µg/ml; and
    e. aluminium hydroxide in an amount in the range of 0.5 mg/ml to 4.5 mg/ml;
    f. mannitol in an amount in the range of 5 mg/ml to 100 mg/ml;
    g. phosphate buffer in an amount in the range of 1 mM to 10 mM; and
    h. polysorbate 20 in an amount in the range of 0.01 mg/ml to 2 mg/ml.

21. The vaccine formulation as claimed in any one of the claims 10 to 20, comprising:

a. fHbpV3.45 M5:PorA316-320/exP1.14 (of SEQ ID NO. 6 or encoded by SEQ ID NO. 1) in an amount in the range of 15 µg/mL to 150 µg/mL; or
b. fHbpV2.19 M6:PorA316-320/exP1.4 (of SEQ ID NO. 7 or encoded by SEQ ID NO. 2) in an amount in the range of 15 µg/mL to 150 µg/mL; or
c. fHbpV1.1:PorA307-311/exP1.9 (of SEQ ID NO. 10 or encoded by SEQ ID NO. 5) in an amount in the range of 15 µg/mL to 150 µg/mL; or
d. fHbpV1.14:PorA307-311/exP1.9 (of SEQ ID NO. 8 or encoded by SEQ ID NO. 3) in an amount in the range of 15 µg/mL to 150 µg/mL; and
e. aluminium hydroxide in an amount in the range of 0.5 mg/mL to 4.5 mg/mL;
f. mannitol in an amount in the range of 5 mg/mL to 100 mg/mL;
g. phosphate buffer in an amount in the range of 1 mM to 10 mM; and
h. polysorbate 20 in an amount in the range of 0.01 mg/mL to 2 mg/mL.

22. The vaccine formulation as claimed in any one of the claims 10 to 21, optionally wherein the formulation comprises 2-phenoxyethanol in an amount in range of 1 mg/mL to 10 mg/ml,
optionally, wherein the vaccine composition is stable at 2-8°C, 25°C and 40°C for over a period of six months,
optionally, wherein the formulation has Zeta potential in the range of -16 mV to -30 mV; osmolality in the range of 200 mOsmol/kg to 500 mOsmol/kg.

23. The vaccine formulation as claimed in any one of the claims 10 to 22, further comprising one or more antigen selected from Diphtheria toxoid (D), Tetanus toxoid (T), Whole cell pertussis (wP), hepatitis B virus surface antigen (HbsAg), Haemophilus influenzae b PRP-Carrier protein conjugate (Hib), Haemophilus influenzae (a, c, d, e, f serotypes and the unencapsulated strains), Neisseria meningitidis A antigen(s), Neisseria meningitidis C antigen(s), Neisseria meningitidis W-135 antigen(s), Neisseria meningitidis Y antigen(s), Neisseria meningitidis X antigen(s), Streptococcus Pneumoniae antigen(s), Neisseria meningitidis B bleb or purified antigen(s), Staphylococcus aureus antigen(s), Anthrax, BCG, Hepatitis (A, C, D, E, F and G strains) antigen(s), Human papilloma virus, HIV, Salmonella typhi antigen(s), acellular pertussis, modified adenylate cyclase, Malaria Antigen (RTS,S), Measles, Mumps, Rubella, Dengue, Zika, Ebola, Chikungunya, Japanese encephalitis,

rotavirus, Diarrheal antigens, Flavivirus, smallpox, yellow fever, Shingles, Varicella virus antigens, and combinations thereof.

24. The vaccine formulation as claimed in any one of the claims 10 to 23 comprising (i) at least one fusion protein comprising stable non-functional/ non-lipidated fHbp and PorA VR2 loop, and (ii) at least one polysaccharide-protein conjugate.

25. The vaccine formulation as claimed in any one of the claims 10 to 24, wherein the recombinant protein/ modified fHbp fusion protein is co-administered with one or more vaccine comprising at least one polysaccharide-protein conjugates of at least one of N. meningitidis serogroups (selected from A, C, Y, W, X or B) or Haemophilus influenzae type B (Hib), or combination thereof.

26. The vaccine formulation as claimed in any one of the claims 10 to 25 comprising i) at least one fusion protein comprising stable non-functional /non-lipidated fHbp and PorA VR2 loop and at least one conjugate selected from (a) a conjugate of (i) the capsular saccharide of serogroup A N. meningitidis and (ii) tetanus toxoid; (b) a conjugate of (i) capsular saccharide of serogroup C N. meningitidis and (ii) CRM197; (c) a conjugate of (i) capsular saccharide of serogroup Y N. meningitidis and (ii) CRM197; (d) a conjugate of (i) capsular saccharide of serogroup W135 N. meningitidis and (ii) CRM197; and (d) a conjugate of (i) capsular saccharide of serogroup X N. meningitidis and (ii) tetanus toxoid.

27. The vaccine formulation as claimed in any one of the claims 10 to 26 comprising i) at least four fusion proteins, each consisting of one fHbp variant type coupled to three PorA VR2 loop and at least one conjugate selected from (a) a conjugate of (i) the capsular saccharide of serogroup A N. meningitidis and (ii) tetanus toxoid; (b) a conjugate of (i) capsular saccharide of serogroup C N. meningitidis and (ii) CRM197; (c) a conjugate of (i) capsular saccharide of serogroup Y N. meningitidis and (ii) CRM197; (d) a conjugate of (i) capsular saccharide of serogroup W135 N. meningitidis and (ii) CRM197; and (d) a conjugate of (i) capsular saccharide of serogroup X N. meningitidis and (ii) tetanus toxoid.

28. The vaccine formulation as claimed in any one of the claims 10 to 27 for use in the treatment or prevention of infection and/or disease caused by Neisseria meningitidis serogroup B,
optionally wherein the vaccine formulation elucidates cross protection against Neisseria gonorrhea strains and Neisseria meningitidis serogroups ACWYX.

29. The vaccine formulation as claimed in any one of the claims 10 to 28, wherein the percent adsorption of the recombinant protein/ modified fHbp fusion protein on to an adjuvant is in the range of 70 % to 100 %.

30. The vaccine formulation as claimed in claim 29, wherein percent adsorption of fHbp V3.45 M5 PorA $^{316\text{-}320\ exP1.14}$ on to an adjuvant is in the range of 80 % to 100 %,
optionally, wherein the percent adsorption of fHbp V1.14 PorA $^{307\text{-}311\ exP1.9}$ on to an adjuvant is in the range of 80 % to 90 %,
wherein the percent adsorption of fHbp V2.19 PorA $^{316\text{-}320\ exP1.4}$ on to an adjuvant is in the range of 80 % to 90 %,
optionally, wherein the percent adsorption of fHbp V1.1 PorA $^{307\text{-}311\ exP1.4}$ on to an adjuvant is in the range of 80 % to 90 %,
optionally, wherein the percent adsorption of fHbp V1.1 PorA $^{307\text{-}311\ exP1.9}$ on to an adjuvant is in the range of 70 % to 80 %.

31. A method for manufacturing a vaccine formulation as claimed in any one of the claims 10 to 30, the method comprising the following steps:

(a) growing host cells comprising the expression vector in nutrient medium;
(b) inducing the host cells for expressing protein;
(c) harvesting and separating the host cells;
(d) lysing the harvested cells and separating host cell debris to obtain tagged proteins;
(e) purifying the tagged proteins;
(f) removing tags from the tagged proteins to obtain recombinant proteins/ modified fHbp fusion proteins;
(g) purifying the recombinant proteins/ modified fHbp fusion proteins; and

(h) preparing the vaccine formulation comprising the purified recombinant proteins/ modified fHbp fusion proteins,

wherein the host cell is a bacterial expression host system,

optionally wherein the bacterial expression host system is an Escherichia coli strain selected from the group consisting of BL21 (DE3), BL21 (DE3) pLysS*, BL21 (DE3) pLysE*, BL21 star (DE3), BL21-A1, BLR (DE3), HMS174 (DE3)**, Tuner (DE3), Origami2 (DE3)**, Rosetta2 (DE3)*, Rosettagami (DE3), Lemo21 (DE3)*, T7 Express, m15 pREP4*, C41(DE3), C43(DE3) or B834(DE3).

32. The method as claimed in claim 31, wherein the concentration of L-methionine during growth of host cells in step (a) is maintained in the range of 1 mM to 10 mM, wherein the fermentation is in fed batch mode,

optionally, wherein the host cells are grown at a temperature in the range of 35℃ to 39℃; pH in the range of 5.0 to 9.0; dissolved oxygen in the range of 10 to 100%; agitation in the range of 100–1800 rpm; gas flow rate in the range of 0-2 volume of gas per unit volume of liquid per minute (VVM),

optionally wherein the host cells are induced using an inducer selected from lactose and its non-hydrolyzable analog isopropyl β-D-1-thiogalactopyranoside (IPTG),

optionally wherein the concentration of lactose is in the range of 1 g/L to 50 g/L and the concentration of IPTG is in the range of 1 mM to 10 mM.

33. The method as claimed in claim 31, wherein the tagged protein in step (e) is purified using chromatography step, followed by concentration and diafiltration.

34. The method as claimed in claim 31, wherein the tags are removed using TEV protease having a protein: TEV protease ratio in the range of 5:1 to 30:1.

35. The method as claimed in claim 31, wherein the recombinant protein/ modified fHbp fusion protein in step (g) is purified using chromatography step, followed by concentration and diafiltration.

36. The method as claimed in any claim 31, wherein the vaccine formulation is prepared by adsorbing individual recombinant protein/ modified fHbp fusion protein on to an adjuvant, followed by addition to an excipient mixture comprising a sugar alcohol, a buffering agent, a stabilizer, and a liquid carrier,

optionally, wherein the excipient mixture comprises a preservative.

37. The method as claimed in claim 34, wherein TEV protease is produced by a method comprising the following steps:

(a) growing host cells comprising the expression vector in nutrient medium;

(b) inducing the host cells for expressing TEV protease;

(c) harvesting and separating the host cells;

(d) lysing the harvested and separating host cells to obtain TEV protease;

(e) purifying the TEV protease; and

(f) concentrating, diafiltration and storing the purified TEV protease.

38. The method as claimed in claim 37, wherein the host cell for expressing TEV protease is an Escherichia coli strain selected from the group consisting of BL21 (DE3), BL21 (DE3) pLysS*, BL21 (DE3) pLysE*, BL21 star (DE3), BL21-A1, BLR (DE3), HMS174 (DE3)**, Tuner (DE3), Origami2 (DE3)**, Rosetta2 (DE3)*, Rosettagami (DE3), Lemo21 (DE3)*, T7 Express, m15 pREP4*, C41(DE3), C43(DE3), Rosetta™(DE3)pLysS or B834(DE3).

39. The method as claimed in any one of the claims 31 to 38, wherein the glucose feed is stopped, and glycerol feed is started when OD at 590/ 600 nm is 20-100 and inducing the culture by adding and/ or maintaining lactose at 1-50 g/L in fed batch mode.