CLAIMS

1. A fusion protein, consisting of or comprising a first amino acid sequence and a second amino acid sequence, wherein:

   a) the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 874-1850 of a *C. difficile* Toxin A sequence or residues 1444-1851 of a *C. difficile* Toxin B sequence; and

   b) the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of a long repeat unit located within amino acid residues 1851-2710 of a *C. difficile* Toxin A sequence or within amino acid residues 1852-2366 of a *C. difficile* Toxin B sequence;

   with the proviso that the fusion protein is not a polypeptide comprising amino acid residues 543-2710 of a *C. difficile* Toxin A;

   with the proviso that the fusion protein is not a polypeptide comprising amino acid residues 543-2366 of a *C. difficile* Toxin B; and

   wherein part or all of the amino acid sequence of the cysteine protease domain is absent.

2. The fusion protein according to Claim 1, wherein the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 770-1850 or 774-1850 or 794-1850 or 814-1850 or 834-1850 or 854-1850 of a *C. difficile* Toxin A sequence; or residues 1164-1851 or 1204-1851 or 1244-1851 or 1284-1851 or 1324-1851 or 1364-1851 of a *C. difficile* Toxin B sequence.

3. The fusion protein according to Claim 1, wherein the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence amino acid sequence consisting of residues 770-1850 of a *C. difficile* Toxin A sequence or residues 767-1851 of a *C. difficile* Toxin B sequence.

4. The fusion protein according to any preceding claim, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from one or more of the amino acid sequences 1851-2389 or 1851-2007 or 2008-2141 or 2142-2253 or 2254-2389 or 2390-2502 or 2503-2594 or 2595-2710 of a *C. difficile* Toxin A sequence.

5. The fusion protein according to Claim 4, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from the amino acid sequences 1851-2007 or 1851-2141 or 1851-2253 or 1851-2389 or 1851-2502 or 1851-2594 or 1851-2710 of a *C. difficile* Toxin A sequence.

6. The fusion protein according to any of Claims 1-3, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from amino acid sequences 1852-2007 or 2008-2139 or 2140-2273 or 2274-2366 of a *C. difficile* Toxin B sequence.

7. The fusion protein according to Claim 6, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from amino acid sequences 1852-2007 or 1852-2139 or 1852-2273 or 1852-2366 of a *C. difficile* Toxin B sequence.

8. The fusion protein according to any of Claims 1-3, wherein the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 767-1850 of a *C. difficile* Toxin A sequence or residues 767-1851 of a *C. difficile* Toxin B sequence, and wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 1851-2710 of a *C. difficile* Toxin A sequence or residues 1852-2366 of a *C. difficile* Toxin B sequence; or wherein the fusion protein comprises or consists of an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 770-2710 or 770-2389 of a *C. difficile* Toxin A sequence.

9. The fusion protein according to any preceding claim, wherein other than the above-defined Toxin A and/ or Toxin B amino acid sequences, said fusion protein does not include any additional Toxin A and/ or Toxin B amino acid sequence.

10. A fusion protein according to any preceding claim, for use as an antigen in a method of generating an antibody that binds to a *C. difficile* Toxin A and/ or to a *C. difficile* Toxin B;

such as wherein said antibody comprises whole IgG, an Fab antibody or an F(ab')2 antibody.

11. An *in vitro* method for isolating antibodies that bind to *C. difficile* Toxin A and/ or to *C. difficile* Toxin B, said method comprising

a) immobilising on a surface (for example on a matrix within a column) one or more *C. difficile* fusion protein(s) according to any of Claims 1-9;
b) contacting the immobilised fusion protein(s) with a solution containing antibodies that bind to *C. difficile* Toxin A and/ or Toxin B;
c) allowing said antibodies to bind to said fusion protein(s), thereby forming a bound complex of antibody and fusion protein(s);
d) washing away any unbound antibody or protein; and
e) eluting the bound antibodies from the surface, thereby providing affinity-purified antibodies.

12. An antibody that binds to and neutralises a *C. difficile* Toxin A and / or a *C. difficile* Toxin B, wherein said antibody is obtainable by a method according to Claim 10 or Claim 11 and wherein said antibody does not substantially bind to the effector domain and/ or to the cysteine protease domain of a *C. difficile* Toxin A or Toxin B;

such as wherein said antibody comprises whole IgG, an Fab antibody or an F(ab')2 antibody.

13. An antibody according to Claim 12, wherein said antibody binds to an epitope presented by residues 1500-1850 of a *C. difficile* Toxin A and/ or by residues 1500-1851 of a *C. difficile* Toxin B.

14. A fusion protein according to any of Claims 1-9 and/ or an antibody according to Claim 12 or Claim 13, for use in the prevention, treatment or suppression of CDI; preferably for use in the prevention, treatment or suppression of CDI in a human.

15. A therapeutically effective amount of a fusion protein according to any of Claims 1-9 and/ or an antibody according to Claim 12 or Claim 13, for use in a method for preventing, treating or suppressing CDI in a human, said method comprising administering said therapeutically effective amount of a fusion protein and / or antibody to a human.

16. A therapeutically effective amount of a fusion protein according to Claim 15, wherein said fusion protein is administered as part of a combination therapy with one or more additional antigen selected from a *C. difficile* non-Toxin antigen, an inactivated or attenuated whole cell *C. difficile* bacterium, and a *C. difficile* cell extract; and
    a. optionally one or more antigen selected from 1) an antigen from a bacterium that causes nosocomial infection, and 2) an inactivated or attenuated whole cell bacterium that causes nosocomial infection; and
    b. optionally one or more antibiotic, such as an antibiotic effective against a bacterium that causes nosocomial infection.

17. A therapeutically effective amount of an antibody according to Claim 15, wherein said antibody is administered as part of a combination therapy with one or more additional antibodies selected from an antibody that binds to a *C. difficile* non-Toxin antigen, and an antibody that binds to a whole cell *C. difficile* bacterium; and
    a. optionally one or more an antibody selected from 1) an antibody that binds to antigen from a bacterium that causes nosocomial infection, and 2) an antibody that binds to a whole cell bacterium that causes nosocomial infection; and

b. optionally one or more antibiotic, such as an antibiotic effective against a bacterium that causes nosocomial infection.

16. Use of an antibody according to Claim 12 or Claim 13 in an *in vitro* immunoassay method for confirming the presence or absence of a *C. difficile* infection in a patient sample, wherein the presence of a *C. difficile* infection is confirmed by detecting the binding of said antibody to a *C. difficile* Toxin A and/ or Toxin B present in said sample, and wherein failure to detect the binding of said antibody to a *C. difficile* Toxin A and/ or Toxin B present in said sample confirms the absence of a *C. difficile* infection.

19. Use of a fusion protein according to any of Claims 1-10 in an *in vitro* immunoassay method for confirming the presence or absence of a *C. difficile* infection in a patient sample, wherein the presence of a *C. difficile* infection is confirmed by detecting the binding of said fusion protein to an antibody present in said sample, and wherein failure to detect the binding of said fusion protein to an antibody present in said sample confirms the absence of a *C. difficile* infection.

CLAIMS

1. A fusion protein, consisting of or comprising a first amino acid sequence and a second amino acid sequence, wherein:

   a) the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 874-1850 of a *C. difficile* Toxin A sequence or residues 1444-1851 of a *C. difficile* Toxin B sequence; and

   b) the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of a long repeat unit located within amino acid residues 1851-2710 of a *C. difficile* Toxin A sequence or within amino acid residues 1852-2366 of a *C. difficile* Toxin B sequence;

   with the proviso that the fusion protein is not a polypeptide comprising amino acid residues 543-2710 of a *C. difficile* Toxin A;

   with the proviso that the fusion protein is not a polypeptide comprising amino acid residues 543-2366 of a *C. difficile* Toxin B; and

   the fusion protein lacks cysteine protease activity, wherein part or all of the amino acid sequence providing said activity is absent.

2. The fusion protein according to Claim 1, wherein the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 770-1850 or 774-1850 or 794-1850 or 814-1850 or 834-1850 or 854-1850 of a *C. difficile* Toxin A sequence; or residues 1164-1851 or 1204-1851 or 1244-1851 or 1284-1851 or 1324-1851 or 1364-1851 of a *C. difficile* Toxin B sequence.

3. The fusion protein according to Claim 1, wherein the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence amino acid sequence consisting of residues 770-1850 of a *C. difficile* Toxin A sequence or residues 767-1851 of a *C. difficile* Toxin B sequence.

4. The fusion protein according to any preceding claim, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from one or more of the amino acid sequences 1851-2389 or 1851-2007 or 2008-2141 or 2142-2253 or 2254-2389 or 2390-2502 or 2503-2594 or 2595-2710 of a *C. difficile* Toxin A sequence.

5. The fusion protein according to Claim 4, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from the amino acid sequences 1851-2007 or 1851-2141 or 1851-2253 or 1851-2389 or 1851-2502 or 1851-2594 or 1851-2710 of a *C. difficile* Toxin A sequence.

6. The fusion protein according to any of Claims 1-3, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from amino acid sequences 1852-2007 or 2008-2139 or 2140-2273 or 2274-2366 of a *C. difficile* Toxin B sequence.

7. The fusion protein according to Claim 6, wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence selected from amino acid sequences 1852-2007 or 1852-2139 or 1852-2273 or 1852-2366 of a *C. difficile* Toxin B sequence.

8. The fusion protein according to any of Claims 1-3, wherein the first amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 767-1850 of a *C. difficile* Toxin A sequence or residues 767-1851 of a *C. difficile* Toxin B sequence, and wherein the second amino acid sequence is provided by an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 1851-2710 of a *C. difficile* Toxin A sequence or residues 1852-2366 of a *C. difficile* Toxin B sequence; or wherein the fusion protein comprises or consists of an amino acid sequence that has at least 80% sequence identity with an amino acid sequence consisting of residues 770-2710 or 770-2389 of a *C. difficile* Toxin A sequence.

9. The fusion protein according to any preceding claim, wherein other than the above-defined Toxin A and/ or Toxin B amino acid sequences, said fusion protein does not include any additional Toxin A and/ or Toxin B amino acid sequence.

10. A fusion protein according to any preceding claim, for use as an antigen in a method of generating an antibody that binds to a *C. difficile* Toxin A and/ or to a *C. difficile* Toxin B.

11. An *in vitro* method for isolating antibodies that bind to *C. difficile* Toxin A and/ or to *C. difficile* Toxin B, said method comprising

    a) immobilising on a surface (for example on a matrix within a column) one or more *C. difficile* fusion protein(s) according to any of Claims 1-9;

    b) contacting the immobilised fusion protein(s) with a solution containing antibodies that bind to *C. difficile* Toxin A and/ or Toxin B;

    c) allowing said antibodies to bind to said fusion protein(s), thereby forming a bound complex of antibody and fusion protein(s);

    d) washing away any unbound antibody or protein; and

    e) eluting the bound antibodies from the surface, thereby providing affinity-purified antibodies.

12. Isolated polyclonal antibodies that bind to and neutralise a *C. difficile* Toxin A and / or a *C. difficile* Toxin B, wherein said antibodies are obtainable by a method according to Claim 10 or Claim 11 and wherein said polyclonal antibodies comprise antibodies that do not bind to the effector domain and/ or to the cysteine protease domain of a *C. difficile* Toxin A or Toxin B; and wherein said antibodies bind to an epitope presented by residues 1500-1850 of a *C. difficile* Toxin A and/ or by residues 1500-1851 of a *C. difficile* Toxin B.

13. The isolated antibodies according to Claim 12, wherein said antibodies comprise whole IgG, Fab antibodies or $F(ab')_2$ antibodies.

14. A fusion protein according to any of Claims 1-9 and/ or polyclonal antibodies according to Claim 12 or Claim 13, for use in the prevention, treatment or suppression of CDI; preferably for use in the prevention, treatment or suppression of CDI in a human.

15. A therapeutically effective amount of a fusion protein according to any of Claims 1-9 and/ or polyclonal antibodies according to Claim 12 or Claim 13, for use in a method for preventing, treating or suppressing CDI in a human, said method comprising administering said therapeutically effective amount of a fusion protein and / or polyclonal antibodies to a human.

16. A therapeutically effective amount of a fusion protein according to Claim 15, wherein said fusion protein is administered as part of a combination therapy with one or more additional antigen selected from a *C. difficile* non-Toxin antigen, an inactivated or attenuated whole cell *C. difficile* bacterium, and a *C. difficile* cell extract; and

    a) optionally one or more antigen selected from 1) an antigen from a bacterium that causes nosocomial infection, and 2) an inactivated or attenuated whole cell bacterium that causes nosocomial infection; and

    b) optionally one or more antibiotic, such as an antibiotic effective against a bacterium that causes nosocomial infection.

17. A therapeutically effective amount of polyclonal antibodies according to Claim 15, wherein said polyclonal antibodies are administered as part of a combination therapy with one or more additional antibodies selected from an antibody that binds to a *C. difficile* non-Toxin antigen, and an antibody that binds to a whole cell *C. difficile* bacterium; and

    a) optionally one or more an antibody selected from 1) an antibody that binds to antigen from a bacterium that causes nosocomial infection, and 2) an antibody that binds to a whole cell bacterium that causes nosocomial infection; and

    b) optionally one or more antibiotic, such as an antibiotic effective against a bacterium that causes nosocomial infection.

18. Use of polyclonal antibodies according to Claim 12 or Claim 13 in an *in vitro* immunoassay method for confirming the presence or absence of a *C. difficile* infection in a patient sample, wherein the presence of a *C. difficile* infection is confirmed by detecting the binding of said antibody to a *C. difficile* Toxin A and/ or Toxin B present in said sample, and wherein failure to detect the binding of said antibody to a *C. difficile* Toxin A and/ or Toxin B present in said sample confirms the absence of a *C. difficile* infection.

19. Use of a fusion protein according to any of Claims 1-10 in an *in vitro* immunoassay method for confirming the presence or absence of a *C. difficile* infection in a patient sample, wherein the presence of a *C. difficile* infection is confirmed by detecting the binding of said fusion protein to an antibody present in said sample, and wherein failure to detect the binding of said fusion protein to an antibody present in said sample confirms the absence of a *C. difficile* infection.