

date. Other genetically encoded systems for enabling spontaneous amide bond formation include SnoopTag/ SnoopTagJr and SnoopCatcher; RrgATag/RrgATag2/DogTag and RrgACatcher, IsopepTag/ IsopepTag-N and Pilin-C or Pilin-N, PsCsTag and PsCsCatcher; and SnoopTagJr and DogTag (mediated by SnoopLigase), and variants of all these systems.

5 The present inventors have proven that use of large/multicomponent antigens in vaccine compositions is possible using genetically-encoded systems for enabling amide bond formation, which may improve the response to the large/multicomponent antigen. This is a surprising result.

SUMMARY

10 In one aspect of the present disclosure, there is provided a composition comprising a virus-like particle displaying an antigenic component, wherein said composition comprises:

- i) an antigenic component comprising a first peptide tag, and
- ii) a ~~particulate~~ moiety comprising a second peptide tag,

wherein the antigenic component and the moiety are linked via an isopeptide bond between said first and second peptide tags, and wherein the antigenic component is over 50 kDa, and

15 wherein the moiety is a surface antigen of the hepatitis B virus (HBsAg).

In another aspect of the present disclosure, there is provided a vaccine comprising a composition disclosed herein for use in the prophylaxis and/or treatment of a disease.

20 In a further aspect of the present disclosure, there is provided a method of producing a composition disclosed herein, or a vaccine comprising a composition disclosed herein, said method comprising:

- introducing a first nucleic acid which encodes a first genetic fusion of a first protein to a first peptide tag into a first host cell;
- 25 - incubating said first host cell under conditions for expressing said first genetic fusion;
- introducing a second nucleic acid which encodes a second genetic fusion of a second protein to a second peptide tag into a second host cell;
- incubating said second host cell under conditions for expressing said second genetic fusion;
- optionally purifying the expressed components;
- 30 - incubating the expressed components under conditions for formation of an isopeptide bond between the first peptide tag and the second peptide tag; and purifying the resultant composition

wherein the first protein comprises an antigenic component and the second protein comprises a moiety.

~~In another aspect of the present disclosure, there is provided a nucleic acid molecule for use in a method disclosed herein wherein the nucleic molecule encodes an amino sequence as set out in any of SEQ ID NOs: 27 to 41.~~

5 ~~In a further aspect of the present disclosure, there is provided a nucleic acid molecule for use in a method disclosed herein wherein the nucleic molecule encodes an amino sequence as set out in any of SEQ ID NOs: 50 to 58.~~

10 ~~In another aspect of the present disclosure, there is provided a vector comprising a nucleic acid molecule disclosed herein.~~

~~In a further aspect of the present disclosure, there is provided a host cell comprising a nucleic acid molecule disclosed herein, or a vector disclosed herein.~~

15 In one example of the present disclosure there is provided a composition comprising a particle displaying a protein component, wherein said composition comprises:

- i) i) a protein component comprising a first peptide tag, and
- ii) ii) a moiety comprising a second peptide tag,

20 wherein the protein component and the moiety are linked via an isopeptide bond between said first and second peptide tags, and wherein the protein component is over 50 kDa.

In another example of the present disclosure there is provided a composition comprising a particle displaying a protein component, wherein said composition comprises:

- i) a protein component comprising a first peptide tag, and
- ii) a moiety comprising a second peptide tag,

25 wherein the protein component and the moiety are linked via an isopeptide bond between said first and second peptide tags, and wherein the protein component is multimeric.

30 The protein component may have any function e.g. it may be an enzyme or have enzymatic properties. The protein component may be a full-length protein, or it may be a part, segment, domain or truncation of a full-length protein. The protein component may be an antigen or an immunogen. The protein component may also be called the antigenic component.

In another example of the present disclosure there is provided a composition comprising a particle displaying an antigenic component, wherein said composition comprises:

- iii) an antigenic component comprising a first peptide tag, and
- iv) a moiety comprising a second peptide tag,

~~In another example of the present disclosure there is provided a composition comprising a particle displaying an antigenic component, wherein said composition comprises:~~

~~iii) — an antigenic component comprising a first peptide tag, and~~

~~iv) — a moiety comprising a second peptide tag,~~

- 5 wherein the antigenic component and the moiety are linked via an isopeptide bond between said first and second peptide tags, and wherein the antigenic component is over approximately 50 kDa.

In some examples of any example of the present disclosure the protein component or the antigenic component may be over 60 kDa, 70 kDa, 80 kDa, 90 kDa, 100 kDa, 110 kDa, 120 kDa, 130

Claims

1. A composition comprising a virus-like particle displaying an antigenic component, wherein said composition comprises:

i) an antigenic component comprising a first peptide tag, and

5 ii) a ~~particulate~~ moiety comprising a second peptide tag,

wherein the antigenic component and the moiety are linked via an isopeptide bond between said first and second peptide tags, and wherein the antigenic component is over 50 kDa, and wherein the moiety is a surface antigen of the hepatitis B virus (HBsAg).

2. A composition as claimed in claim 1 wherein the antigenic component is over 60 kDa, 70 kDa, 80
10 kDa, 90 kDa, 100 kDa, 110 kDa, 120 kDa, 130 kDa, 140 kDa, 150 kDa, 160 kDa, 170 kDa, 180 kDa, 190 kDa, 200 kDa, 300k Da or 400 kDa.

3. A composition as claimed in claim 1 or claim 2 wherein the antigenic component is a monomer or multimer, optionally wherein the multimer is a trimer, tetramer, pentamer, hexamer, septamer, octamer, nonamer or decamer.

15 ~~4. A composition as claimed in any preceding claim wherein the moiety is a virus, bacteria, multimerisation scaffold for vaccination, a protein component which multimerises to form a virus-like particle (VLP), a viral structural protein, a multimerisation domain which forms nanoparticles, a synthetic nanoparticle or a synthetic VLP.~~

~~5. A composition as claimed in any preceding claim wherein the moiety is an HBsAg particle.~~

20 ~~46.~~ A composition as claimed in any preceding claim wherein the first and second peptide tag are selected from any one of a SpyTag and SpyCatcher pair; a SnoopTag or SnoopTagJr and SnoopCatcher pair; a RrgATag, RrgATag2 or DogTag and RrgACatcher pair, an IsopepTag Pilin-C pair; an IsopepTag-N and Pilin-N pair; a PsCsTag and PsCsCatcher pair; and SnoopTagJr and DogTag pair mediated by SnoopLigase; ~~or variants, derivatives or modifications thereof~~, optionally wherein the
25 first and second peptide tags are a SpyTag/SpyCatcher pair.

57. A composition as claimed in any preceding claim wherein the antigenic component comprises an immunogenic component of an HCMV pentamer, wherein the immunogenic component of an HCMV pentamer comprises one or more of the gH, gL, pUL128, pUL130 or pUL131 subunits..

30 68. A composition as claimed in claim 57 wherein the immunogenic component of an HCMV pentamer comprises each of the gH, gL, pUL128, pUL130 or pUL131 subunits.

79. A composition as claimed in claim 57 or claim 68 wherein the immunogenic component of an HCMV pentamer comprises a gH subunit with a truncated transmembrane domain.

~~810.~~ A composition as claimed in any of claims ~~57~~ to claim 7-9, wherein the first peptide tag is attached to the gH subunit.

~~911~~. A composition as claimed in any one of claim 1 to claim ~~46~~ wherein the antigenic component comprises an immunogenic component of an RSV-F protein, wherein the immunogenic component of an RSV-F protein is a pre-fusion F protein.

5 ~~1012~~. A composition as claimed in claim ~~911 wherein said RSV-F protein~~ wherein the first peptide tag is attached to the C-terminus of the pre-fusion F protein.

~~1113~~. A composition as claimed in any preceding claim wherein the first peptide tag is a SpyTag.

~~1214~~. A composition as claimed in any preceding claim wherein the ~~moiety is HBsAg, and optionally wherein the~~ second peptide tag is a SpyCatcher.

10 ~~1315~~. A composition as claimed in any preceding claim wherein the composition is an immunogenic or vaccine composition.

~~1416~~. A vaccine comprising a composition as claimed in any of claims 1 to ~~claim 1315~~ for use in the prophylaxis and/or treatment of a disease.

~~1517~~. A method of producing a composition in accordance with any of claims 1 to ~~claim 14, or a vaccine comprising a composition in accordance with claim 1416~~, said method comprising:

- 15 - introducing a first nucleic acid which encodes a first genetic fusion of a first protein to a first peptide tag into a first host cell;
- incubating said first host cell under conditions for expressing said first genetic fusion;
- introducing a second nucleic acid which encodes a second genetic fusion of a second protein to a second peptide tag into a second host cell;
- 20 - incubating said second host cell under conditions for expressing said second genetic fusion;
- optionally purifying the expressed components;
- incubating the expressed components under conditions for formation of an isopeptide bond between the first peptide tag and the second peptide tag; and purifying the resultant composition

25 wherein the first protein comprises an antigenic component and the second protein comprises a moiety.

~~18. A nucleic acid molecule for use in a method as claimed in claim 17 wherein the nucleic molecule encodes an amino sequence as set out in any of SEQ ID NOs: 27 to 41.~~

30 ~~19. A nucleic acid molecule for use in a method as claimed in claim 17 wherein the nucleic molecule encodes an amino sequence as set out in any of SEQ ID NOs: 50 to 58.~~

~~20. A vector comprising a nucleic acid molecule as claimed in claim 18 or 19.~~

~~21. A host cell comprising a nucleic acid molecule as claimed in claim 18 or 19, or a vector as claimed in claim 20.~~