

We claim:

1. A composition comprising an MHC ligand peptide covalently attached to an MHC class II molecule comprising an MHC class II α chain or a portion thereof and an MHC class II β chain or a portion thereof,

wherein the MHC ligand peptide is covalently attached to the MHC class II molecule by a peptide linker,

wherein the MHC ligand peptide or the peptide linker comprises a first cysteine and the MHC class II molecule comprises a second cysteine, and

wherein the first cysteine and the second cysteine form a disulfide bond such that the MHC ligand peptide is bound in a peptide-binding groove formed by the MHC class II α chain or the portion thereof and the MHC class II β chain or the portion thereof.

2. The composition of claim 1, wherein the MHC class II α chain or the portion thereof comprises an $\alpha 1$ domain, and the MHC class II β chain or the portion thereof comprises a $\beta 1$ domain.

3. The composition of claim 2, wherein the MHC class II α chain or the portion thereof comprises an MHC class II α chain extracellular domain, and the MHC class II β chain or the portion thereof comprises an MHC class II β chain extracellular domain.

4. The composition of claim 2 or 3, wherein:

(1) the MHC class II α chain or the portion thereof comprises the $\alpha 1$ domain, an $\alpha 2$ domain, a transmembrane domain, and a cytoplasmic domain; and

(2) the MHC class II β chain or the portion thereof comprises the $\beta 1$ domain, a $\beta 2$ domain, a transmembrane domain, and a cytoplasmic domain.

5. The composition of any preceding claim, wherein the composition is membrane-anchored.

6. The composition of any one of claims 1-3, wherein the composition is soluble.

7. The composition of claim 6, wherein:

(1) the MHC class II α chain or the portion thereof comprises the $\alpha 1$ domain and an $\alpha 2$ domain but does not comprise a transmembrane domain or a cytoplasmic domain; and

(2) the MHC class II β chain or the portion thereof comprises the $\beta 1$ domain and a $\beta 2$ domain but does not comprise a transmembrane domain or a cytoplasmic domain.

8. The composition of claim 6 or 7, wherein the MHC class II α chain or the portion thereof and the MHC class II β chain or the portion thereof are linked by a Jun-Fos zipper, electrostatic engineering, knobs-into-holes, an immunoglobulin scaffold, an immunoglobulin Fc region, or a linker.

9. The composition of claim 8, wherein the MHC class II α chain or the portion thereof and the MHC class II β chain or the portion thereof are linked by a Jun-Fos zipper comprising a Jun leucine zipper dimerization motif and a Fos leucine zipper dimerization motif, and wherein

the MHC class II α chain or the portion thereof is linked to the Jun leucine zipper dimerization motif and the MHC class II β chain or the portion thereof is linked to the Fos leucine zipper dimerization motif, or the MHC class II α chain or the portion thereof is linked to the Fos leucine zipper dimerization motif and the MHC class II β chain or the portion thereof is linked to the Jun leucine zipper dimerization motif.

10. The composition of claim 9, wherein the C-terminal end of the MHC class II α chain or the portion thereof is linked to the Jun leucine zipper dimerization motif and the C-terminal end of the MHC class II β chain or the portion thereof is linked to the Fos leucine zipper dimerization motif, or

wherein the C-terminal end of the MHC class II α chain or the portion thereof is linked to the Fos leucine zipper dimerization motif and the C-terminal end of the MHC class II β chain or the portion thereof is linked to the Jun leucine zipper dimerization motif.

11. The composition of claims 9 or 10, wherein the MHC class II α chain or the portion thereof is linked to the Jun leucine zipper dimerization motif by an MHC-Jun linker, and the MHC class II β chain or the portion thereof is linked to the Fos leucine zipper dimerization motif by an MHC-Fos linker, or

wherein the MHC class II α chain or the portion thereof is linked to the Fos leucine zipper dimerization motif by the MHC-Fos linker, and the MHC class II β chain or the portion thereof is linked to the Jun leucine zipper dimerization motif by the MHC-Jun linker.

12. The composition of claim 11, wherein the MHC-Jun linker and the MHC-Fos linker each comprise the sequence set forth in SEQ ID NO: 1.

13. The composition of any preceding claim, wherein the MHC ligand peptide is about 10 to about 18 amino acids in length, is about 10 to about 15 amino acids in length, or is about 10 to about 12 amino acids in length, or

wherein the MHC ligand peptide comprises residues P-1 to P9 or residues P-3 to P9.

14. The composition of any preceding claim, wherein the MHC ligand peptide is an antigenic MHC ligand peptide.

15. The composition of any preceding claim, wherein the MHC ligand peptide is associated with a T-cell-mediated disease.

16. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is a flexible linker.

17. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule comprises one or more flexible amino acids and one or more polar amino acids.

18. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule does not comprise any charged amino acids.

19. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule comprises a cleavage site.

20. The composition of claim 19, wherein the cleavage site is a tobacco etch virus (TEV) protease cleavage site.

21. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is non-immunogenic.
22. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is connected to the N-terminal end of the MHC class II β chain or the portion thereof.
23. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is connected to the N-terminal end of the MHC class II α chain or the portion thereof.
24. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is at least about 9 amino acids in length.
25. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is between about 9 and about 50 amino acids in length.
26. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule comprises 2-4 repeats of the sequence set forth in SEQ ID NO: 4.
27. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule comprises the first cysteine.
28. The composition of claim 27, wherein the first cysteine is the only cysteine in the peptide linker linking the MHC ligand peptide to the MHC class II molecule.
29. The composition of claim 27 or 28, wherein the first cysteine is in the first four amino acids of the peptide linker linking the MHC ligand peptide to the MHC class II molecule.
30. The composition of any preceding claim, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule comprises 2-4 repeats of the

sequence set forth in SEQ ID NO: 4, wherein one amino acid in one of the repeats is mutated to cysteine.

31. The composition of claim 30, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule comprises the sequence set forth in SEQ ID NO: 21.

32. The composition of any one of claims 1-26, wherein the MHC ligand peptide comprises the first cysteine.

33. The composition of claim 32, wherein the first cysteine faces away from an epitope formed by the composition.

34. The composition of any preceding claim, wherein the second cysteine is in the MHC class II α chain or the portion thereof.

35. The composition of claim 34, wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is connected to the N-terminal end of the MHC class II β chain or the portion thereof.

36. The composition of any preceding claim, wherein the second cysteine is not present in a wild type MHC class II molecule corresponding to the MHC class II molecule in the composition.

37. The composition of claim 36, wherein the second cysteine is in place of a non-cysteine amino acid in the corresponding wild type MHC class II molecule.

38. The composition of claim 37, wherein the second cysteine is in the MHC class II α chain or the portion thereof, and

wherein the second cysteine is at a position corresponding to position 101 in the sequence set forth in SEQ ID NO: 49 when the MHC class II α chain or the portion thereof is optimally aligned with SEQ ID NO: 49.

39. The composition of any preceding claim, wherein the MHC class II molecule lacks a cysteine present in a corresponding wild type MHC class II molecule.

40. The composition of claim 39, wherein the cysteine present in the corresponding wild type MHC class II molecule has been replaced with an alanine or a glutamine in the MHC class II molecule in the composition.

41. The composition of any one of claims 1-38, wherein the MHC class II α chain or the portion thereof lacks a cysteine present in a corresponding wild type MHC class II α chain.

42. The composition of claim 41, wherein the cysteine present in the corresponding wild type MHC class II α chain has been replaced with an alanine or a glutamine in the MHC class II α chain or the portion thereof in the composition.

43. The composition of claim 41 or 42, wherein the cysteine in the corresponding wild type MHC class II α chain is at a position corresponding to position 70 in the sequence set forth in SEQ ID NO: 49 when the MHC class II α chain or the portion thereof is optimally aligned with SEQ ID NO: 49.

44. The composition of any preceding claim, wherein the composition further comprises one or more immunostimulatory molecules.

45. The composition of claim 44, wherein the one or more immunostimulatory molecules comprise a pan-DR-binding epitope (PADRE) and/or a peptide from lymphocytic choriomeningitis virus (LCMV).

46. The composition of claim 44 or 45, wherein the one or more immunostimulatory molecules are directly or indirectly covalently linked to the MHC class II molecule.

47. The composition of any one of claims 44-46, wherein the one or more immunostimulatory molecules are directly or indirectly covalently linked to the MHC class II α chain or the portion thereof and/or the MHC class II β chain or the portion thereof.

48. The composition of any preceding claim, wherein the MHC class II molecule is a human MHC class II molecule.

49. The composition of claim 48, wherein the human MHC class II molecule is selected from the group consisting of HLA-DQ, HLA-DR, and HLA-DP.

50. The composition of claim 49, wherein the human MHC class II molecule is an HLA-DQ2 molecule.

51. The composition of claim 49, wherein the human MHC class II molecule is an HLA-DR2 molecule.

52. The composition of any preceding claim, wherein the MHC class II α chain or the portion thereof comprises an MHC class II α chain extracellular domain, and the MHC class II β chain or the portion thereof comprises an MHC class II β chain extracellular domain,

wherein the peptide linker linking the MHC ligand peptide to the MHC class II molecule is a flexible linker between about 9 and about 50 amino acids in length that comprises the first cysteine and is connected to the N-terminal end of the MHC class II β chain or the portion thereof,

wherein the second cysteine is in the MHC class II α chain or the portion thereof and is not present in a wild type MHC class II molecule corresponding to the MHC class II molecule in the composition, and

wherein the MHC class II molecule lacks a cysteine present in a corresponding wild type MHC class II molecule.

53. The composition of claim 52, wherein the composition is soluble,
wherein the MHC class II α chain or the portion thereof comprises the $\alpha 1$ domain and an $\alpha 2$ domain but does not comprise a transmembrane domain or a cytoplasmic domain,

wherein the MHC class II β chain or the portion thereof comprises the $\beta 1$ domain and a $\beta 2$ domain but does not comprise a transmembrane domain or a cytoplasmic domain, and

wherein the MHC class II α chain or the portion thereof and the MHC class II β chain or the portion thereof are linked by a Jun-Fos zipper comprising a Jun leucine zipper dimerization motif and a Fos leucine zipper dimerization motif.

54. The composition of claim 52 or 53, wherein the second cysteine is at a position corresponding to position 101 in the sequence set forth in SEQ ID NO: 49 when the MHC class II α chain or the portion thereof is optimally aligned with SEQ ID NO: 49, and wherein the cysteine in the corresponding wild type MHC class II molecule is at a position corresponding to position 70 in the sequence set forth in SEQ ID NO: 49 when the MHC class II α chain or the portion thereof is optimally aligned with SEQ ID NO: 49.

55. The composition of any one of claims 52-54, wherein the MHC class II molecule is a human MHC class II molecule is selected from the group consisting of HLA-DQ, HLA-DP, and HLA-DR.

56. The composition of claim 55, wherein the human MHC class II molecule is HLA-DQ.

57. A nucleic acid encoding the composition of any preceding claim.

58. A method of eliciting an immune response in a subject, comprising administering to the subject an effective amount of the composition of any one of claims 1-56 or a nucleic acid encoding the composition.

59. A method of generating an antigen-binding protein that specifically binds an antigenic composition comprising an MHC ligand peptide covalently attached to an MHC class II molecule, comprising:

- (a) immunizing a non-human animal with the composition of any one of claims 1-56 or a nucleic acid encoding the composition; and
- (b) maintaining the non-human animal in conditions sufficient for the non-human animal to mount an immune response to the composition.

60. A method of generating an antigen-binding protein, comprising:

- (a) immunizing a non-human animal with the composition of any one of claims 1-56 or a nucleic acid encoding the composition; and
- (b) maintaining the non-human animal in conditions sufficient for the non-human animal to mount an immune response to the composition.

61. The method of claim 60, wherein the antigen-binding protein specifically binds an antigenic composition comprising an MHC ligand peptide covalently attached to an MHC class II molecule.