

CLAIMS

1. A method for predicting the usefulness of a protein or a fragment thereof expressed by diseased cells for immunotherapy, the method comprising ascertaining the distribution or
5 localization of the protein or a nucleic acid coding therefor, or of a fragment of the protein.
2. The method of claim 1, which comprises ascertaining whether the protein or a nucleic acid coding therefor, or a fragment of the protein is located or abundant in the cytosol and/or within exosomes *in vivo*.
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3. The method of claim 1 or 2, wherein localization or abundance of the protein or a nucleic acid coding therefor, or of a fragment of the protein in the cytosol and/or within exosomes indicates that the protein or a fragment thereof is useful for immunotherapy.
- 15 4. A method for predicting the usefulness of a protein or a fragment thereof expressed by diseased cells for immunotherapy, the method comprising ascertaining whether the protein or a fragment thereof is cross-presented by antigen presenting cells, preferably professional antigen presenting cells.
- 20 5. The method of claim 4, wherein cross-presentation of the protein or a fragment thereof by antigen presenting cells indicates that the protein or fragment thereof is useful for immunotherapy.
6. The method of claim 4 or 5, wherein ascertaining whether the protein or a fragment
25 thereof is cross-presented by antigen presenting cells comprises ascertaining whether the protein or a nucleic acid coding therefor, or a fragment of the protein is located or abundant in the cytosol and/or within exosomes *in vivo*.
7. The method of any one of claims 4 to 6, wherein localization or abundance of the
30 protein or a nucleic acid coding therefor, or of a fragment of the protein in the cytosol and/or within exosomes indicates that the protein or a fragment thereof is cross-presented by antigen presenting cells.

8. The method of any one of claims 4 to 7, wherein ascertaining whether the protein or a fragment thereof is cross-presented by antigen presenting cells comprises ascertaining an existing antibody response to the protein or a fragment thereof.
- 5 9. The method of any one of claims 4 to 8, wherein an existing antibody response to the protein or a fragment thereof indicates that the protein or a fragment thereof is cross-presented by antigen presenting cells.
- 10 10. The method of any one of claims 4 to 9, wherein ascertaining whether the protein or a fragment thereof is cross-presented by antigen presenting cells comprises ascertaining whether the protein or a fragment thereof binds to F actin.
- 15 11. The method of any one of claims 4 to 10, wherein binding of the protein or a fragment thereof to F actin indicates that the protein or a fragment thereof is cross-presented by antigen presenting cells.
- 20 12. The method of any one of claims 4 to 11, wherein ascertaining whether the protein or a fragment thereof is cross-presented by antigen presenting cells comprises ascertaining whether the protein or a fragment thereof binds to RNA.
- 25 13. The method of any one of claims 4 to 12, wherein binding of the protein or a fragment thereof to RNA indicates that the protein or a fragment thereof is cross-presented by antigen presenting cells.
- 30 14. The method of any one of claims 2, 3, 6 to 13, wherein the protein fragment is present within exosomes as MHC peptide complex, preferably on the surface of exosomes.
15. The method of any one of claims 2, 3, 6 to 13, wherein localization or abundance of the protein or a nucleic acid coding therefor, or of a fragment of the protein in the cytosol indicates processing and presentation of the protein in the MHC I pathway, preferably of diseased cells.

16. The method of claim 15, wherein processing and presentation of the protein in the MHC I pathway results in recognition of complexes formed by MHC I and fragments of the protein by CD8+ T cells.
- 5 17. The method of any one of claims 2, 3, 6 to 13, wherein localization or abundance of the protein or a nucleic acid coding therefor, or of a fragment of the protein within exosomes indicates accumulation of the protein or a nucleic acid coding therefor, or of a fragment of the protein in antigen presenting cells, preferably professional antigen presenting cells.
- 10 18. The method of claim 17, wherein accumulation of the protein or a nucleic acid coding therefor, or of a fragment of the protein in antigen presenting cells indicates processing and presentation of the protein in the MHC I and/or MHC II pathway, preferably of the antigen presenting cells.
- 15 19. The method of claim 18, wherein processing and presentation of the protein in the MHC I pathway results in recognition of complexes formed by MHC I and fragments of the protein by CD8+ T cells.
- 20 20. A method for predicting the usefulness of a protein or a fragment thereof expressed by diseased cells for immunotherapy, the method comprising ascertaining one or more of the following:
- (a) ascertaining an existing antibody response to the protein or a fragment thereof,
 - (b) ascertaining whether the protein or a fragment thereof binds to F actin, and/or
 - (c) ascertaining whether the protein or a fragment thereof binds to RNA.
- 25 21. The method of claim 20, wherein an existing antibody response to the protein or a fragment thereof indicates that the protein or a fragment thereof is useful for immunotherapy.
22. The method of claim 20 or 21, wherein binding of the protein or a fragment thereof to
- 30 F actin indicates that the protein or a fragment thereof is useful for immunotherapy.
23. The method of any one of claims 20 to 22, wherein binding of the protein or a fragment thereof to RNA indicates that the protein or a fragment thereof is useful for immunotherapy.

24. The method of any one of claims 1 to 23, wherein the protein or a fragment thereof comprises a disease specific amino acid modification.

5 25. The method of claim 24, wherein the amino acid modification is due to a disease specific somatic mutation.

26. The method of any one of claims 1 to 25, wherein the disease is cancer and the immunotherapy is anti-cancer immunotherapy.

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27. The method of any one of claims 1 to 26, wherein the protein fragment is a MHC binding peptide or a potential MHC binding peptide.

28. A method for selecting and/or ranking disease specific amino acid modifications for their usefulness in immunotherapy, the method comprising the steps of:
15 (i) identifying proteins expressed by diseased cells each protein comprising at least one disease specific amino acid modification, and
(ii) ascertaining the distribution or localization of a protein identified under (i) or a nucleic acid coding therefor, or of a fragment of the protein, and
20 (iii) repeating step (ii) for at least one further protein identified under (i).

29. The method of claim 28, wherein step (ii) comprises ascertaining whether the protein or a nucleic acid coding therefor, or a fragment of the protein is located or abundant in the cytosol and/or within exosomes *in vivo*.

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30. The method of claim 28 or 29, wherein localization or abundance of the protein or a nucleic acid coding therefor, or of a fragment of the protein in the cytosol and/or within exosomes indicates that the disease specific amino acid modification is useful for immunotherapy.

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31. A method for selecting and/or ranking disease specific amino acid modifications for their usefulness in immunotherapy, the method comprising the steps of:
(i) identifying proteins expressed by diseased cells each protein comprising at least one disease specific amino acid modification, and

- (ii) ascertaining whether a protein identified under (i) or a fragment of the protein is cross-presented by antigen presenting cells, preferably professional antigen presenting cells, and
- (iii) repeating step (ii) for at least one further protein identified under (i).

5 32. The method of claim 31, wherein cross-presentation of the protein or a fragment thereof by antigen presenting cells indicates that the disease specific amino acid modification is useful for immunotherapy.

33. A method for selecting and/or ranking disease specific amino acid modifications for their usefulness in immunotherapy, the method comprising the steps of:

- 10 (i) identifying proteins expressed by diseased cells each protein comprising at least one disease specific amino acid modification, and
- (ii) ascertaining for a protein identified under (i) or a fragment of the protein one or more of the following:
 - 15 (a) ascertaining an existing antibody response to the protein or a fragment thereof,
 - (b) ascertaining whether the protein or a fragment thereof binds to F actin, and/or
 - (c) ascertaining whether the protein or a fragment thereof binds to RNA, and
- (iii) repeating step (ii) for at least one further protein identified under (i).

20 34. The method of claim 33, wherein an existing antibody response to the protein or a fragment thereof indicates that the disease specific amino acid modification is useful for immunotherapy.

35. The method of claim 33 or 34, wherein binding of the protein or a fragment thereof to
25 F actin indicates that the disease specific amino acid modification is useful for immunotherapy.

36. The method of any one of claims 33 to 35, wherein binding of the protein or a fragment thereof to RNA indicates that the disease specific amino acid modification is useful
30 for immunotherapy.

37. The method of any one of claims 28 to 36, wherein the disease specific amino acid modification is comprised by a protein fragment which is a MHC binding peptide or a potential MHC binding peptide.

38. The method of any one of claims 1 to 37, which is used in the manufacture of a vaccine.
- 5 39. The method of claim 38, wherein the vaccine is derived from one or more proteins or fragments thereof or one or more disease specific amino acid modifications which are predicted as being useful for immunotherapy.
40. A method for providing a vaccine comprising the step:
- 10 identifying one or more proteins or fragments thereof or one or more disease specific amino acid modifications which are predicted as being useful for immunotherapy by the method of any one of claims 1 to 39.
41. The method of claim 40 further comprising the step:
- 15 providing a vaccine comprising a peptide or polypeptide comprising one or more proteins or fragments thereof or one or more disease specific amino acid modifications which are predicted as being useful for immunotherapy, or a nucleic acid encoding the peptide or polypeptide.
- 20 42. A vaccine produced according to the method of any one of claims 38 to 41.