

Claims

1. A method for providing an individualized cancer vaccine comprising the steps:
 - (a) identifying cancer specific somatic mutations in a tumor specimen of a cancer patient to provide a cancer mutation signature of the cancer patient, comprising
 - (aa) obtaining nucleic acid sequence information by sequencing genomic DNA and/or RNA of the tumor specimen of the cancer patient,
 - (bb) obtaining reference nucleic acid sequence information by sequencing DNA and/or RNA of normal non-cancerous cells, and
 - (cc) comparing the nucleic acid sequence information from the tumor specimen obtained in step (aa) with the reference nucleic acid sequence information obtained in step (bb); and
 - (b) providing an RNA vaccine featuring the cancer mutation signature obtained in step (a), wherein the RNA vaccine featuring the mutation signature of the patient comprises RNA encoding a recombinant polyepitopic polypeptide comprising mutation based neo-epitopes fused together by peptide bonds or linkers.
2. The method according to claim 1, wherein the step of identifying cancer specific somatic mutations comprises identifying the cancer mutation signature of the exome of one or more cancer cells.
3. The method according to claim 1 or 2, wherein the step of identifying cancer specific somatic mutations comprises single cell sequencing of one or more cancer cells.
4. The method according to claim 3, wherein the cancer cells are circulating tumor cells.
5. The method according to any one of claims 1 to 4, wherein the step of identifying cancer specific somatic mutations involves using next generation sequencing (NGS).
6. The method according to any one of claims 1 to 5, wherein the normal non-cancerous cells are obtained from the cancer patient.

7. The method according to any one of claims 1 to 6, wherein the reference nucleic acid sequence information is obtained from germline cells.
8. The method according to any one of claims 1 to 6, wherein the reference nucleic acid sequence information is obtained from genomic germline DNA obtained from peripheral blood mononuclear cells (PBMCs).
9. The method according to claim 7 or 8, wherein the tumor specimen is from a primary tumor and wherein the step of identifying cancer specific somatic mutations in the tumor specimen further comprises the steps:
 - (dd) providing a phylogenetic tree of cancer specific somatic mutations, wherein the reference nucleic acid sequence information obtained in step (bb) is used to root the tree,
 - (ee) reproducing ancestral sequences, wherein the ancestral sequences are sequences of nodes near the root of the phylogenetic tree containing primary basal mutations, wherein the primary basal mutations are the earliest mutations predicted to exist in the primary tumor; and
 - (ff) selecting the primary basal mutations from the ancestral sequences identified in step (ee).
10. The method according to any one of claims 1 to 9, wherein the step of identifying cancer specific somatic mutations is replicated at least in duplicates.
11. The method according to any one of claims 1 to 10, comprising the further step of determining the usability of the identified mutations in epitopes for cancer vaccination.
12. The method according to any one of claims 1 to 11, wherein the polypeptide preferably comprises up to 30 mutation based neo-epitopes.
13. The method according to any one of claims 1 to 12, wherein the polypeptide further comprises epitopes not containing cancer specific somatic mutations which are expressed by cancer cells.

14. The method according to any one of claims 1 to 13, wherein the polypeptide comprises neo-epitopes that are based on primary basal mutations.
15. The method according to any one of claims 1 to 14, wherein the neo-epitopes and/or epitopes are in their natural sequence context so as to form a vaccine sequence.
16. The method according to claim 15, wherein the vaccine sequence comprises 20 or more amino acids.
17. The method according to any one of claims 1 to 16, wherein the neo-epitopes, epitopes and/or vaccine sequences are lined up head-to-tail and/or are spaced by linkers.
18. The method according to any one of claims 1 to 17, wherein the vaccine is a prophylactic and/or therapeutic vaccine.
19. An RNA vaccine obtained by the method according to any one of claims 1 to 18, wherein the vaccine, to be administered to a patient, provides a collection of MHC presented epitopes incorporating sequence changes based on the identified mutations.
20. The RNA vaccine according to claim 19, wherein the MHC presented epitopes are MHC class II-presented epitopes that are capable of eliciting a CD4⁺ helper T cell response against cells expressing antigens from which the MHC presented epitopes are derived and optionally MHC class I-presented epitopes that are capable of eliciting a CD8⁺ T cell response against cells expressing antigens from which the MHC presented epitopes are derived.
21. An RNA vaccine comprising RNA encoding a recombinant polypeptide comprising 5 or more mutation based neo-epitopes fused together by peptide bonds or linkers, the neo-epitopes resulting from cancer specific somatic mutations in a tumor specimen of a cancer patient.
22. The RNA vaccine according to claim 21, wherein the polypeptide comprises 10 or more mutation based neo-epitopes.

23. The RNA vaccine according to claim 21 or 22, wherein the polypeptide comprises 20 or more mutation based neo-epitopes.
24. The RNA vaccine according to any one of claims 21 to 23, wherein the polypeptide further comprises epitopes not containing cancer specific somatic mutations which are expressed by cancer cells.
25. The RNA vaccine according to any one of claims 21 to 24, wherein the polypeptide comprises neo-epitopes that are based on primary basal mutations.