Claims

1. A method for selecting a number A of neoepitopes for an individual suffering from or suspected of suffering from cancer, said method comprising the steps of:

a. Obtaining one or more neoepitopes from said individual, each neoepitope comprising at least one minimal epitope compared to a reference sequence, wherein each neoepitope comprises at least one mutation, wherein the minimal epitope consists of a number of amino acids equal to or smaller than the number of amino acids of the neoepitope and comprises said at least one mutation; wherein obtaining the neoepitopes comprises the step of identifying mutations in nucleic acid sequences which are specific for the tumor;

b. Determining MHC I and/or MHC II binding affinity for at least one minimal epitope and a corresponding reference peptide, wherein the binding affinity is determined by *in silico* prediction by calculating the %Rank score, wherein a low %Rank score indicates a strong binding affinity and a high %Rank score indicates a weak binding affinity, and wherein the corresponding reference peptide is obtained from body fluid or healthy tissue of the same individual, or in a normal, healthy population;

c. Selecting neoepitopes comprising at least one minimal epitope predicted to bind to MHC I and/or to MHC II, thereby obtaining MHC binding neoepitopes;

d. Ranking the MHC binding neoepitopes with respect to the likelihood of clinical utility of said MHC binding neoepitopes, wherein said ranking comprises:

I. determining the number of minimal epitopes comprised in the MHC binding neoepitopes selected in step c, and wherein neoepitopes comprising a high number of minimal epitopes are ranked higher than neoepitopes comprising a lower number of minimal epitopes, preferably wherein the minimal epitope is an MHC I binding minimal epitope;

II. determining the MHC I binding differential of the MHC I binding neoepitopes selected in step c, wherein the MHC I binding differential is given by the formula (%Rank score (MHC I) for reference) / (%Rank score (MHC I) for neoepitope) wherein neoepitopes with a high MHC I binding differential are ranked higher than neoepitopes with a lower MHC I binding differential;

III. determining whether the mutation is in an anchoring position or a non-anchoring position of the minimal neoepitope for each minimal epitope

comprised within the neoepitopes, wherein minimal neoepitopes with the mutation in an anchoring position are prioritized; and

IV. prioritizing the MHC I binding neoepitopes selected in step c with respect to the MHCI rank of said MHC I binding neoepitopes, wherein neoepitopes with a low %Rank score (MHC I) are ranked higher than neoepitopes with a higher %Rank score, wherein the %Rank score (MHC I) of a neoepitope comprising one or more minimal epitopes predicted to bind to MHC I is equal to the lowest %Rank score (MHC I) of said one or more minimal epitopes; and

V. determining the BLOSUM score, wherein the MHC I binding neoepitopes selected in step c are prioritized with respect to their BLOSUM score in a descending order, wherein the BLOSUM score of a neoepitope is equal to the BLOSUM score of the best ranking minimal epitope it comprises, wherein a minimal epitope with a BLOSUM score < 2 is prioritized ; and

e. Selecting A neoepitopes among the highest ranking MHC binding neoepitopes of step d, thereby selecting A neoepitopes likely to have clinical utility.

2. The method according to claim 1, wherein A is an integer of from 1 to 100, such as from 5 to 50, such as from 5 to 30, such as from 10 to 20, such as 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24 or 25.

3. The method according to any one of the preceding claims, wherein step a comprises identifying mutations in nucleic acid sequences in one or more sample derived from the individual compared to a reference sample, preferably wherein the one or more sample is a sample derived from tumor cells of the individual.

4. The method according to claim 3, wherein the one or more sample is a sample derived from tumor cells of the individual and wherein step c further comprises selecting MHC I binding and/or MHC II binding neoepitopes which are found in at least two samples derived from tumor cells of the individual.

5. The method according to any one of the preceding claims, wherein the neoepitope comprises 1 to 50 minimal epitopes, such as 1 to 40 minimal epitopes, such as 1 to 30 minimal epitopes, such as 1 to 20 minimal epitopes.

6. The method according to any one of the preceding claims, wherein step c comprises selecting neoepitopes comprising at least one minimal epitope predicted to bind to MHC I and at least one minimal epitope predicted to bind to MHC II.

7. The method according to any one of the preceding claims, wherein step d IV comprises prioritizing neoepitopes having a %Rank score (MHC I) below 2.0, such as below 1.5, such as below 1, preferably equal to or below 0.5.

8. The method according to any one of the preceding claims, wherein MHC I binding neoepitopes having a %Rank score (MHC I) above 10 are excluded or down-prioritized.

9. The method according to any one of the preceding claims, wherein said neoepitopes have a length of from 7 to 40 amino acids, such as from 15 to 30 amino acids, such as from 25 to 30 amino acids, such as 27 amino acids.

10. The method according to any one of the preceding claims, wherein neoepitopes found in more than one sample derived from tumor cells are ranked higher than neoepitopes found in only one sample derived from tumor cells.

11. The method according to any one of the preceding claims, further comprising a step of determining the RNA expression levels of said neoepitopes, and selecting neoepitopes with a high RNA expression level.

12. The method according to any one of the preceding claims, further comprising a step of determining the allele frequency of mutations present in the MHC binding neoepitopes, and selecting neoepitopes with a high allele frequency.

13. The method according to any one of the preceding claims, wherein step d further comprises a step of down-prioritizing or excluding neoepitopes which do not comprise any minimal epitopes predicted to bind to MHC I.

14. The method according to any one of the preceding claims, wherein neoepitopes comprising at least 5 MHC binding minimal epitopes are ranked higher than neoepitopes comprising less than 5 MHC binding minimal epitopes

15. The method according to any one of the preceding claims, wherein the neoepitopes of step d, VI are ranked by assigning to each neoepitope the highest score of the minimal epitopes they comprise, and wherein the ranking of the neoepitopes is performed in a descending order 1) to 7) as follows:

1) if the mutation is in an anchoring position and the binding differential of the minimal epitope is equal to or greater than 20, the minimal epitope has the highest score;
2) if the mutation is in a non-anchoring position and the binding differential of the minimal epitope is equal to or greater than 20, the score of the minimal epitope is lower than the score for the minimal epitopes of 1);
3) if the mutation is in an anchoring position and the binding differential of the minimal epitope is lower than 20 and equal to or greater than 3, the score of the minimal epitope is lower than the score the minimal epitopes of 2);
4) if the mutation is in a non-anchoring position and the binding differential of the minimal epitope is lower than 20 and equal to or greater than 3, the score of the minimal epitope is lower than the score the minimal epitopes of 3);
5) if the mutation is in an anchoring position and the binding differential of the minimal epitope is less than 3 and equal to or greater than 1, the score of the minimal epitope is lower than the score of the minimal epitopes of 4);
6) if the mutation is in a non-anchoring position and the binding differential of the minimal epitope is less than 3 and equal to or greater than 1, the score of the minimal epitope is lower than the score of the minimal epitopes of 5;
7) if the minimal epitope has a mutation with a binding differential lower than 1, regardless of the position of the mutation, the minimal epitope has the lowest score.

16. A method of preparing a cancer vaccine comprising A neoepitopes, said method comprising a step of selecting said A neoepitopes using the method according to any one of claims 1 to 15.

17. The method of claim 16, wherein said cancer vaccine comprises a polynucleotide comprising a) a targeting unit, b) a dimerization unit, c) a first linker and d) an antigenic unit comprising said A neoepitopes.