ANNEX A

Description

[0001] The present invention relates to a pharmaceutical composition containing an mRNA which is stabilised by sequence modifications in the translated region and is optimised for translation. The pharmaceutical composition according to the invention is particularly suitable as a vaccine against viral infectious diseases, excluding AIDS (HIV). In addition, a method for identification of sequence modifications in the translated region that serve for optimising the stabilisation and translation of mRNA is disclosed.

[0017] In particular, a modified mRNA and at least one mRNA modified in this manner in combination with a pharmaceutically acceptable vehicle and/or vehicle-containing pharmaceutical composition, wherein the modified mRNA codes for at least one antigenic viral peptide or polypeptide, with the exception of an HIV peptide or HIV polypeptide, wherein the sequence of the mRNA, in particular in the region coding for the at least one peptide or polypeptide, has the following modifications, which may be present either alternatively or in combination, compared with the wild-type mRNA, will be provided.

[0049] Furthermore, the pharmaceutical composition according to the invention is used against viral infectious diseases such as ~~AIDS (HIV);~~ hepatitis A, B or C, herpes; herpes zoster (varicella); measles (rubeola virus); yellow fever; dengue, etc. (flaviviruses); flu (influenza viruses); haemorrhagic infectious diseases (Marburg or Ebola viruses). Preferably in the case of infectious diseases as well, the corresponding surface antigens with the strongest antigenic potential are coded by the modified mRNA. In the genes of pathogenic viral organisms mentioned, this is typically a secreted form of a surface antigen. In addition, according to the invention, mRNAs coding for polypeptides is preferably used, wherein the polypeptides are polyepitopes, for example, of the aforementioned antigens, especially surface antigens of pathogenic viral organism, preferably secreted protein forms.

[0050] Moreover, the modified mRNA according to the invention, in addition to the antigenically or gene therapeutically effective peptide or polypeptide, with the exception of an HIV peptide or HIV polypeptide, may also contain at least one other functional section, which, for example, codes for an immune response-promoting cytokine (monokine, lymphokine, interleukin or chemokine, such as IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-12, INF-α, INF-γ, GM-CSF, LT-α or growth factors such as hGH.

Claims

1. Modified mRNA coding for at least one antigenic viral peptide or polypeptide, with the exception of an HIV peptide or HIV polypeptide, **characterised in that** the G/C content of the region of the modified mRNA coding for the peptide or polypeptide is increased compared to the G/C content of the coding region of the wild type mRNA coding for the peptide or polypeptide, and the encoded amino acid sequence is unchanged as compared to the wild type.

2. Modified mRNA according to claim 1, **characterised in that** the G/C content of the region of the modified mRNA coding for the peptide or polypeptide is increased by at least 7 % points, preferably at least 15 % points, compared to the G/C content of the coding region of the wild type mRNA coding for the peptide or polypeptide.

3. Modified mRNA according to one of claims 1 to 2, **characterised in that** the modified mRNA comprises a 5' cap structure and/or a poly-A tail of at least 70 nucleotides and/or an IRES and/or a 5' stabilisation sequence and/or a 3' stabilisation sequence.

4. Modified mRNA according to one of claims 1 to 3, **characterised in that** the modified mRNA comprises at least one analogue of naturally occurring nucleotides.

5. Modified mRNA according to claim 4, **characterised in that** the analogue is selected from the group consisting of phosphorus thioates, phosphorus amidates, peptide nucleotides, methylphosphonates, 7-deazaguanosine, 5-methylcytosine and inosine.

6. Modified mRNA according to one of claims 1 to 5, **characterised in that** the viral antigen derives from the secreted form of a surface antigen.

7. Modified mRNA according to one of claims 1 to 5, **characterised in that** the mRNA codes for a surface antigen of pathogenic viral germs.

8. Modified mRNA according to one of claims 1 to 7, **characterised in that** the mRNA is associated with a cationic peptide or protein or is bound thereto.

9. Modified mRNA according to claim 8, **characterised in that** the cationic peptide or protein is selected from the group consisting of protamine, poly-L lysine, and histones.

10. Modified mRNA according to one of claims 1 to 9, **characterised in that** the polypeptide is a

polyepitope of viral antigens.

11. Modified mRNA according to one of claims 1 to 10, **characterised in that** the modified mRNA is a multicistronic mRNA.

12. Modified mRNA according to one of claims 1 to 11, **characterised in that** the mRNA in addition codes for at least one cytokine.

13. Modified mRNA according to one of claims 1 to 12, **characterised in that** the mRNA comprises one or more IRES sequence(s), wherein the IRES sequences are in particular selected from picorna viruses (e.g. FMDV), plague viruses (CFFV), polio viruses (PV), encephalo myocarditis viruses (ECMV), foot-and-mouth disease viruses (FMDV), hepatitis C viruses (HCV), classic swine fever viruses (CSFV), murine leukemia virus (MLV), simian immunodefiency viruses (SIV), or cricket paralysis viruses (CrPV).

14. Pharmaceutical composition **characterized in that** it contains a modified mRNA according to one of claims 1 to 13 in combination with a pharmaceutically acceptable carrier and/or vehicle.

15. Pharmaceutical composition according to claim 14, **characterised in that** the pharmaceutical composition contains at least one the immune response stimulating adjuvant.

16. Pharmaceutical composition according to one of claims 14 or 15, which in addition contains at least one cytokine.

17. Use of a pharmaceutical composition according to one of claims 14 to 16 or a modified mRNA according to one of claims 1 to 13 for the preparation of a vaccine for inoculation against viral infectious diseases, excluding AIDS (HIV).

18. Use of a pharmaceutical composition according to claim 17 for the preparation of a vaccine for inoculation against ~~AIDS,~~ hepatitis A, B or C, herpes, herpes zoster, rubella, dengue, haemorrhagic infectious diseases, yellow fever and influenza.