The invention relates to compositions and methods for the preparation, manufacture and therapeutic use of polynucleotide molecules encoding at least one polypeptide of interest to modulate the immune response. Described herein are compositions, methods, processes, kits and devices for the design, preparation, manufacture and/or formulation of polynucleotides encoding at least one polypeptide of interest which modulates the activity of the immune system. In one non-limiting embodiment, such polynucleotides take the form or function as modified mRNA molecules which encode at least one polypeptide of interest or variants thereof which modulates the activity of the immune system.
INTERNATIONAL SEARCH REPORT

A. CLASSIFICATION OF SUBJECT MATTER
   IPC(8) - A61K 31/7088 (2015.01)
   CPC - A61K 31/7088 (2015.01)

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
   IPC(8) - A61K 31/7088, 31/7115, 39/00; C12N 15/85, C12N 15/87 (2015.01)
   CPC - A61K 39/00, 31/7088, 31/7115; C12N 15/85, 15/87 (2015.01)

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
   CPC - A61K 39/00, 3.1/7088, 31/7115; C12N 15/85, 15/87 (2015.01) (keyword delimited)
   US Classes - 514/44A, 44R, 536/23.1

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
PUBBase, Orbit, Google Patents, PubMed
Search terms used: 5'UTR, 3'UTR, calreticulin, vector, IRES, vector, modified

C. DOCUMENTS CONSIDERED TO BE RELEVANT

<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y</td>
<td>WO 2013/090186 A1 (MODERNERA THERAPEUTICS et al) 20 June 2013 (20.06.2013)</td>
<td>1, 4, 5, 16-20</td>
</tr>
</tbody>
</table>

Further documents are listed in the continuation of Box C.

Date of the actual completion of the international search: 11 March 2015
Date of mailing of the international search report: 08 APR 2015

Name and mailing address of the ISA/US
Mail Stop PCT, Attn: ISA/US, Commissioner for Patents
P.O. Box 1450, Alexandria, Virginia 22313-1450
Facsimile No. 571-273-3201

Form PCT/ISA/2 10 (second sheet) (July 2009)

Authorized officer: Blaine R. Copenheaver
1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing filed or furnished:

   a. (means)
      - □ on paper
      - □ in electronic form

   b. (time)
      - □ in the international application as filed
      - □ together with the international application in electronic form
      - □ subsequently to this Authority for the purposes of search

2. □ In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

   **SEQ ID NOs: 20, 39-41, and 115-132 were searched.**
This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. Claims Nos.: because they relate to subject matter not required to be searched by this Authority, namely:

2. Claims Nos.: because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. Claims Nos.: because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

This International Searching Authority found multiple inventions in this international application, as follows:

See Extra Sheet.

1. As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.

3. As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.: 1-5 and 16-20 restricted to SEQ ID NOs: 20, 39, and 41.

Remark on Protest

- [ ] The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- [ ] The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- [ ] No protest accompanied the payment of additional search fees.
This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees need to be paid.

Group I: claims 1-20 are drawn to a polynucleotide for the expression of a polypeptide of interest, and a composition and method comprising the same.

The first invention of Group I is restricted to a polynucleotide, and a composition and method comprising the same, for the expression of a polypeptide of interest comprising: (a) a first region of linked nucleosides, wherein the first region of linked nucleosides comprises at least an open reading frame of a nucleic acid sequence, wherein the nucleic acid sequence selected to be SEQ ID NO: 41, said first region encoding said polypeptide of interest, said polypeptide is selected to be SEQ ID NO: 39; (b) a first flanking region located 5' relative to said first region comprising a 5' untranslated region (5'UTR) a 5' terminal cap; (c) a second flanking region located 3' relative to said first region comprising a 3' untranslated region (3'UTR), wherein the 3'UTR is selected to be SEQ ID NO: 20; and a 3' tailing sequence of linked nucleosides; and wherein said polynucleotide comprises at least one chemically modified nucleoside, wherein at least one chemically modified nucleoside is selected to be a single modified 2-methylthio-N-6-cis-hydroxyisopentenyl) adenosine (ms26A); wherein the 3'UTR is heterologous to the 5'UTR. Applicant is invited to elect sequences to be searched in a specific combination by paying additional fee for each set of election. An exemplary election would be a polynucleotide for the expression of a polypeptide of interest comprising: (a) a first region of linked nucleosides, wherein the first region of linked nucleosides comprises at least an open reading frame of a nucleic acid sequence, wherein the nucleic acid sequence selected to be SEQ ID NO: 42, said first region encoding said polypeptide of interest, said polypeptide of interest has an amino acid sequence selected to be SEQ ID NO: 39; (b) a first flanking region located 5' relative to said first region comprising a 5' untranslated region (5'UTR) a 5' terminal cap; (c) a second flanking region located 3' relative to said first region comprising a 3' untranslated region (3'UTR), wherein the 3'UTR is selected to be SEQ ID NO: 20; and a 3' tailing sequence of linked nucleosides; and wherein said polynucleotide comprises at least one chemically modified nucleoside, wherein the at least one chemically modified nucleoside is selected to be a single modified 2-methylthio-N-6-cis-hydroxyisopentenyl) adenosine (ms26A); wherein the 3'UTR is heterologous to the 5'UTR. Additional sequences will be searched upon the payment of additional fees. Applicants must specify the claims that read on any additional elected inventions. Applicants must further indicate, if applicable, the claims which read on the first named invention if different than what was indicated above for this group. Failure to clearly identify how any paid additional invention fees are to be applied to the "+" group(s) will result in only the first claimed invention to be searched/examined.

The inventions listed in Groups I do not relate to a single general inventive concept under PCT Rule 13.1, because under PCT Rule 13.2 they lack the same or corresponding special technical features for the following reasons:

The Groups I formulas do not share a significant structural element, requiring the selection of alternatives for the nucleic acid sequences, where the nucleic acid sequence selected from the group consisting of SEQ ID NO: 41-50, 179-499, 520-838, 855-910, 1015-1274, 1291-1330, 1405-1591 and 1606-1640, "said polypeptide of interest has an amino acid sequence selected from the group consisting of SEQ ID NO: 39, 40, 115-178, 510-519, 847-854, 963-1014, 1283-1290, 1368-1404, 1599-1605," and "a 3' tailing sequence of linked nucleosides; and wherein said polynucleotide comprises at least one chemically modified nucleoside."

The Groups I share the technical features of a polynucleotide for the expression of a polypeptide of interest comprising: (a) a first region of linked nucleosides, said first region encoding said polypeptide of interest; a first flanking region located 5' relative to said first region comprising a 5' untranslated region (5'UTR) and at least one 5' terminal cap; (c) a second flanking region located 3' relative to said first region comprising a 3' untranslated region (3'UTR) and a 3' tailing sequence of linked nucleosides; and wherein said polynucleotide comprises at least one chemically modified nucleoside. However, these shared technical features do not represent a contribution over the prior art. Specifically, WO 2013/090186 A1 to Moderna Therapeutics et al. discloses a polynucleotide for the expression of a polypeptide of interest (a synthetic isolated RNA comprising a first region of linked nucleosides encoding a polypeptide of interest, Para. [0007]) comprising: (a) a first region of linked nucleosides (RNA comprising a first region of linked nucleosides, Para. [0007]), said first region encoding said polypeptide of interest (linked nucleosides encoding a polypeptide of interest, Para. [0007]); a first flanking region located 5' relative to said first region comprising a 5' untranslated region (5'UTR) (a first terminal region located at the 5' terminus of said first region comprising a 5' untranslated region (UTR), Para. [0007]) at least one 5' terminal cap (The first terminal region may comprise at least one 5' cap structure, Para. [0009]); (c) a second flanking region located 3' relative to said first region comprising a 3' untranslated region (3'UTR) and a 3' tailing sequence of linked nucleosides (second terminal region located at the 3' terminus of said first region comprising a 3'UTR and a 3' tailing sequence of linked nucleosides, Para. [0007]); and wherein said polynucleotide comprises at least one chemically modified nucleoside (the 3' tailing region may comprise at least one modified nucleoside, Para. [0007]).

The inventions listed in Groups I therefore lack unity under Rule 13 because they do not share a same or corresponding special technical features.