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(54) **Title:** COMPOSITIONS AND METHODS FOR PROMOTING PAYLOAD DELIVERY IN CELLS

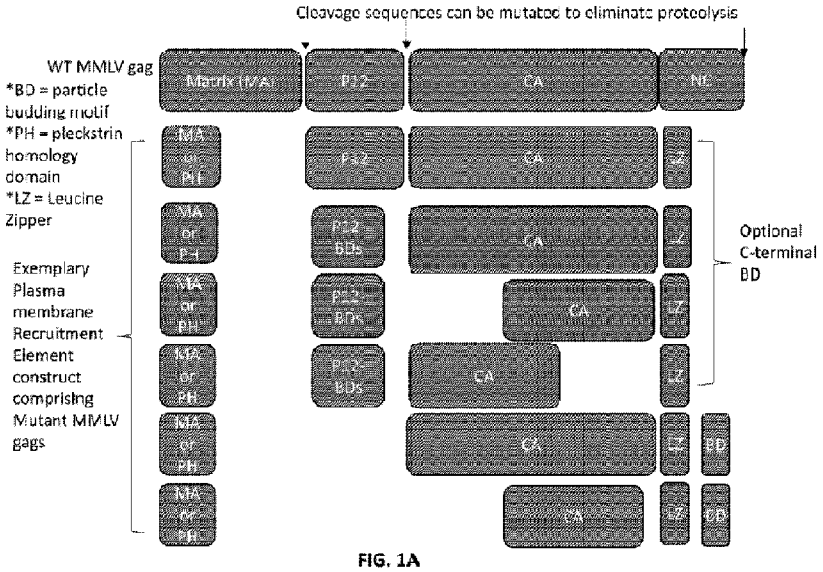

FIG. 1A

(57) **Abstract:** Disclosed herein, in some aspects, are compositions of lipid delivery particles used to deliver a payload to a target cell and methods of producing the like. Lipid delivery particles can comprise ectosomes, exosomes, nanoparticles, or other classes of extracellular vesicles suitable for transporting a payload. Lipid delivery particles can comprise a plasma membrane recruitment element comprising a mutant gag polyprotein.

[Continued on next page]

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Declarations under Rule 4.17:

— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*

— *of inventorship (Rule 4.17(iv))*

Published:

— *with international search report (Art. 21(3))*

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments (Rule 48.2(h))*

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06 March 2025 (06.03.2025)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2024/033130

A. CLASSIFICATION OF SUBJECT MATTER

IPC: ***C12N 15/88*** (2024.01); ***A61K 35/76*** (2024.01); ***A61K 47/69*** (2024.01); ***A61K 48/00*** (2024.01); ***C07K 14/15*** (2024.01); ***C12N 7/00*** (2024.01); ***C12N 15/48*** (2024.01); *C12N 15/86* (2024.01)

CPC: ***C12N15/88; A61K35/76; A61K47/6901; A61K48/005; C07K14/15; C12N7/00; C12N15/113;*** *C07K2319/035; C12N15/86; C12N2740/10042*

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)

See Search History Document

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

See Search History Document

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

See Search History Document

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2012/0213817 A1 (HAYNES JOEL R) 23 August 2012 (23.08.2012) par. [0007], [0072], [0075], [0077], [0130], [0147], [0301], [0303]; claims 1, 3, 5, 7-9	1-2, 4, 5/1-2, 5/4, 47-48, 50, 101-102, 104, 105/101-102, 105/104, 106/101-102, 106/104, 142-143, 145
Y		3, 5/3, 49, 103, 105/103, 106/103, 144
Y	US 2022/0389451 A1 (THE BROAD INSTITUTE INC. et al.) 08 December 2022 (08.12.2022) par. [0028], [0164]	3, 5/3, 49, 103, 105/103, 106/103, 144

☑ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"D" document cited by the applicant in the international application
"E" earlier application or patent but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
25 November 2024 (25.11.2024)	**29 January 2025 (29.01.2025)**

Name and mailing address of the ISA/US	Authorized officer
COMMISSIONER FOR PATENTS **MAIL STOP PCT, ATTN: ISA/US** **P.O. Box 1450** **Alexandria, VA 22313-1450** **UNITED STATES OF AMERICA**	**SHANE THOMAS**
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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2024/033130

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first 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.: **Group I+, claims 1 (in part), 2-3, 4 (in part), 5, 47-49, 50 (in part), 51, 101 (in part), 102-103, 104 (in part), 105-106, 142-144, and 145 (in part), SEQ ID NO: 285 (wild type gag polyprotein), full-length capsid protein (additional plasma membrane recruitment element).**

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.

INTERNATIONAL SEARCH REPORT

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Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

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.: **6-46, 52-100, 107-141, 148-200**
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

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

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 must be paid.

Group I+: claims 1-5, 47-51, 101-106, 142-147, SEQ ID NO: 285 (wild type gag polyprotein), full-length capsid protein (additional plasma membrane recruitment element), are direction to a lipid delivery vehicle comprising a recombinant protein.

The methods of Claims 1 (in part), 2-3, 4 (in part), 5, 47-49, 50 (in part), 51, 101 (in part), 102-103, 104 (in part), 105-106, 142-144, and 145 (in part), are believed to encompass the first named invention of Groups I + and are the claims that will be searched to the extent that they comprise a sequence and plasma membrane recruitment element encompassing SEQ ID NO: 285 (first exemplary wild type gag polyprotein), full-length capsid protein (first exemplary additional plasma membrane recruitment element). This first named invention of Group I+ has been selected to encompass the first species of each of the genera found in claims 1, 4, 50, 101, 104, 145, are based on the guidance set forth in section 10.54 of the PCT International Search and Preliminary Examination Guidelines.

Applicant is invited to elect additional sequences and plasma membrane recruitment elements, where available as an option within at least one searchable claim, to be searched. Additional sequence(s) and plasma membrane recruitment element(s) will be searched upon the payment of additional fees. Applicants must specify the searchable claims that encompass any additionally elected sequence(s) and plasma membrane recruitment element(s). Applicants must further indicate, if applicable, the claims which encompass 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. An exemplary election would be SEQ ID NO: 286 (wild type gag polyprotein), full length matrix protein (plasma membrane recruitment element).

Groups I+ share the technical features including: a lipid delivery particle comprising (a) a lipid membrane; (b) a plasma membrane recruitment element comprising a mutant gag polyprotein, and (c) a payload encapsulated by the lipid containing membrane, and wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein; a lipid delivery particle comprising (a) a lipid membrane; (b) a plasma membrane recruitment element comprising a mutant gag polyprotein that is not from HIV, and (c) a payload encapsulated by the lipid containing membrane, wherein the plasma membrane recruitment element lacks at least a fragment

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein; a recombinant protein comprising a plasma membrane recruitment element comprising a mutant gag polyprotein, wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein; a recombinant protein comprising a plasma membrane recruitment element comprising a mutant gag polyprotein that is not from HIV, and wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein.

However, these shared technical features are previously disclosed by WO 2021/202604 A1 to Sana Biotechnology, Inc. (hereinafter "Sana") WO2011062155A1 to Takara Bio (hereinafter "Takara").

Sana discloses a lipid delivery particle comprising (a) a lipid membrane; (b) a plasma membrane recruitment element, and (c) a payload encapsulated by the lipid containing membrane (lipid particles enclosing a lumen or cavity for antibody (payload), with a targeted envelope protein (plasma membrane recruitment element); abstract), and wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein (gag protein processed into MA, CA and nucleocapsid (lacks a gag-endogenous protease cleavage sequence); [0351]); a lipid delivery particle comprising (a) a lipid membrane; (b) a plasma membrane recruitment element comprising a mutant gag polyprotein that is not from HIV, and (c) a payload encapsulated by the lipid containing membrane (lipid particles enclosing a lumen or cavity for antibody (payload), with a targeted envelope protein (plasma membrane recruitment element) from henipavirus (not from HIV); abstract), wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein (gag protein processed into MA, CA and nucleocapsid (lacks a gag-endogenous protease cleavage sequence); [0351]); a recombinant protein comprising a plasma membrane recruitment element (recombinant protein encoding vector with envelope proteins, such as with a targeted envelope protein (plasma membrane recruitment element) from henipavirus (not from HIV); abstract, [0355]), wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein (gag protein processed into MA, CA and nucleocapsid (lacks a gag-endogenous protease cleavage sequence); [0351]); a recombinant protein comprising a plasma membrane recruitment element (recombinant protein encoding vector with envelope proteins, such as with a targeted envelope protein (plasma membrane recruitment element) from henipavirus (not from HIV); abstract, [0355]), and wherein the plasma membrane recruitment element lacks at least a fragment of a matrix protein, at least a fragment of a RNA-binding phosphoprotein, at least a fragment of a capsid protein, a gag-endogenous protease cleavage sequence, or at least a fragment of a nucleocapsid protein (gag protein processed into MA, CA and nucleocapsid (lacks a gag-endogenous protease cleavage sequence); [0351]). Sana does not disclose plasma membrane recruitment element comprising a mutant gag polyprotein. Takara discloses plasma membrane recruitment element comprising a mutant gag polyprotein (mutant gag protein encoded by a nucleic acid; abstract). It would have been obvious to a person of ordinary skill in the art, before the relevant date, to have modified the proteins and particles of Sana to have incorporated mutant GAG protein, as disclosed by Takara, because mutation introduced into GAG can enable effective gene transfer into a target cell (Takara; abstract).

Since none of the special technical features of the Groups I+ inventions is found in more than one of the inventions, and since all of the shared technical features are previously disclosed by the combination of the Sana and Takara references, unity of invention is lacking.

INTERNATIONAL SEARCH REPORT

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Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

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:
   a. ☐ forming part of the international application as filed.
   b. ☑ furnished subsequent to the international filing date for the purposes of international search (Rule 13*ter*.1(a)),
      ☑ accompanied by a statement to the effect that the sequence listing does not go beyond the disclosure in the international application as filed.

2. ☐ With regard to any nucleotide and/or amino acid sequence disclosed in the international application, this report has been established to the extent that a meaningful search could be carried out without a WIPO Standard ST.26 compliant sequence listing.

3. Additional comments:

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2024/033130

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 2021/202604 A1 (SANA BIOTECHNOLOGY INC. et al.) 07 October 2021 (07.10.2021) Entire Document	1-5, 47-50, 101-106, 142-145
P,X	WO 2023/225572 A2 (NVELOP THERAPEUTICS INC.) 23 November 2023 (23.11.2023) par. [0004]-[0005], [0114], [0171], [0181], [0201], [0203]; claims 1-2, 7	1-3, 5/1-3, 47-50, 101-104, 142-145
P,Y		4, 5/4, 105-106
E,X	WO 2024/138033 A2 (NVELOP THERAPEUTICS INC.) 27 June 2024 (27.06.2024) par. [0116]-[0118]; claims 1, 18, 22-23, 25-26	1-3, 5/1-3, 47-50, 101-104, 142-145
E,Y		4, 5/4, 105-106
P,X	WO 2023/102550 A2 (THE BROAD INSTITUTE, INC. et al.) 08 June 2023 (08.06.2023) Entire Document	1-3, 5/1-3, 47-50, 101-104, 142-145
P,Y		4, 5/4, 105-106