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(71) Applicant: **THOMAS JEFFERSON UNIVERSITY**
[US/US]; 1020 Walnut Street, Philadelphia, Pennsylvania
19107 (US).

(72) Inventors: **RIGOUTSOS, Isidore**; 30-30 36th Street, As-
toria, New York 11103 (US). **LOHER, Phillipe**; C/O
Computational Medicine Center, JAH, #M81, 1020 Locust
Street, Philadelphia, Pennsylvania 19107 (US). **LONDIN,**
Eric; 758 S. Hicks St., Philadelphia, Pennsylvania 19146
(US).

(74) Agents: **RESNICK, David S.** et al.; Nixon Peabody LLP,
100 Summer Street, Boston, Massachusetts 02110 (US).

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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))*
- *as to the applicant's entitlement to claim the priority of the
earlier application (Rule 4.17(iii))*

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))*
- *with sequence listing part of description (Rule 5.2(a))*

(88) Date of publication of the international search report:
2 April 2015

(54) Title: NOVEL HUMAN MIRNAS FOR USE IN DIAGNOSIS, PROGNOSIS AND THERAPY OF HUMAN CONDITIONS AND DISEASES

(57) Abstract: Disclosed herein are novel compositions, methods, and systems for determining whether a subject has, or is at risk of developing, or is at a given stage of a condition afflicting a tissue of interest, or determining the tissue or cell provenance of a biological sample, based on expression level of one or more of the novel miRNA and isomiR sequences disclosed herein. The compositions, methods, and systems described herein can be used to diagnose a disease or disorder, or prognose a given stage and/or progression of the disease or disorder, or determine the identity of the tissue or cell in a sample. In some embodiments, the compositions, methods, and systems described herein can be used to develop a treatment for the disease or disorder. For example, in some embodiments, the novel miRNAs can be used as therapeutics for treatment of a disease or disorder.



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INTERNATIONAL SEARCH REPORT

International application No.

PCT/US14/36690

A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - C12Q 1/68 (2015.01)

CPC - C12Q 1/6883

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): C12Q 1/68; C40B 30/00, 30/04; G01N 33/50; C07H 21/04 (2015.01)

CPC: C12Q 2600/158, 2600/106, 1/6809, 1/6883, 2600/156; G06F 19/20, 19/24; G01N 33/57407

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

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

PatSeer (US-G, US-A, EP-A, EP-B, WO, JP-bib, DE-C,B, DE-A, DE-T, DE-U, GB-A, FR-A); Google; Google Scholar; NCBI/BLAST/PubMed; Dialog ProQuest; subject, patient, 'microRNA,' 'miRNA,' expression, level, 'tissue sample,' isoform, disease, condition, cancer, stage, 'reference sample'

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 2011/0312520 A1 (KENNEDY, GC et al.); December 22, 2011; paragraphs [0004], [0010], [0040]	1-4
Y	US 7625699 B2 (DEVLIN, JJ et al.); December 1, 2009; column 5, lines 1-10; SEQ ID NO: 25799	1-4

☐ Further documents are listed in the continuation of Box C.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"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

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04 FEB 2015

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Shane Thomas

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INTERNATIONAL SEARCH REPORT

International application No.

PCT/US14/36690

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.: 5-38
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:

-Please See Supplemental Page-

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.:

Groups I+: Claims 1 (in-part), 2 (in-part), 3 (in-part), 4 (in-part), SEQ ID NO: 1 (Homo sapiens DNA sequence)

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

International application No.

PCT/US14/36690

****-Continued from Box No. III: Observations Where Unity of Invention Is Lacking:

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.

Groups I+: Claims 1-4 are directed toward a method of determining whether a subject has, or is at risk of developing, or is at a given stage of a condition afflicting a tissue of interest, comprising measuring in a biological sample from the tissue of interest expression level of one or more miRNAs.

The method of determining whether a subject has, or is at risk of developing, or is at a given stage of a condition afflicting a tissue of interest, comprising measuring in a biological sample from the tissue of interest expression level of one or more miRNAs will be searched to the extent that the one or more miRNAs encompasses SEQ ID NO: 1 (Homo sapiens DNA sequence). It is believed that Claims 1 (in-part), 2 (in-part), 3 (in-part) and 4 (in-part) encompass this first named invention and thus these claims will be searched without fee to the extent that they encompass SEQ ID NO: 1 (Homo sapiens DNA sequence). Additional miRNA sequence(s) will be searched upon the payment of additional fees. 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: 2 (Homo sapiens DNA sequence).

No technical features are shared between the miRNAs of Groups I+ and, accordingly, these claimed miRNAs lack unity a priori.

Groups I+ share the technical features including: a method of determining whether a subject has, or is at risk of developing, or is at a given stage of a condition afflicting a tissue of interest, comprising measuring in a biological sample from the tissue of interest expression level of one or more of the miRNAs, wherein the miRNAs comprise a sequence selected from SEQ ID NO: 1 to SEQ ID NO: 47,972, wherein the alteration of the level of said one or more miRNAs as compared to the level of the same one or more miRNA in a reference sample is indicative of the subject either having, or being at risk of developing, or is at a given stage of the condition.

However, these shared technical features are previously disclosed by US 2009/0124566 A1 to Chi, et al. (hereinafter 'Chi'). Chi discloses a method of determining whether a subject has, or is at risk of developing (a method of determining the susceptibility of a subject; abstract), or is at a given stage of (or to predict the stage of the disease; paragraph [0005]) a condition afflicting a tissue of interest (an erythrocyte disease; abstract, paragraph [0005]), comprising measuring in a biological sample from the tissue of interest (comprising measuring, in erythrocytes from a subject; paragraph [0005]) expression level of one or more of the miRNAs (expression level of one or more miRNAs; paragraph [0005]), wherein the miRNAs comprise a sequence selected from SEQ ID NO: 1 to SEQ ID NO: 47,972 (wherein the miRNAs comprise miR-144; paragraph [0055], Table 2, SEQ ID NO: 11; wherein SEQ ID NO: 11 of the Chi reference is 100% identical to SEQ ID NO: 34,951 of the instant PCT application), wherein the alteration of the level of said one or more miRNAs as compared to the level of the same one or more miRNA in a reference sample (paragraph [0061]) is indicative of the subject either having, or being at risk of developing (paragraph [0061]), or is at a given stage of the condition (or to predict stages of the disease; paragraph [0005]).

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 Chi reference, unity of invention is lacking.