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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))*

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:
24 February 2022 (24.02.2022)

(54) Title: BIOMARKERS AND CLASSIFIER OF PSORIASIS AND METHODS OF TREATMENT

(57) Abstract: The present invention features biomarkers, for, e.g., in diagnosis and treatment of psoriasis. The invention provides a minimally invasive method of determining levels of biomarkers (e.g., NOS2/iNOS) in surface skin samples.

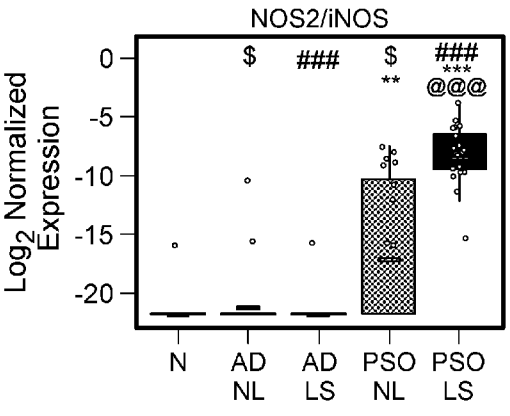

FIG. 1A

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US21/41796

A. CLASSIFICATION OF SUBJECT MATTER

IPC - G01N 33/573; G01N 33/68; C12Q 1/26 (2021.01)

CPC - G01N 33/573; G01N 33/6893; G01N 2333/90254; G01N 2800/205; G01N 33/50; G01N 33/6881; G01N 33/5044; G01N 2800/52; C12Q 2600/158

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 --- Y	US 2019/0316201 A1 (HELMHOLTZ ZENTRUM MüNCHEN - DEUTSCHES FORSCHUNGSZENTRUM FüR GESUNDHEIT UND UMWEL) 17 October 2019; figure 1A and 1B; paragraphs [0010]-[0011], [0016], [0021], [0032], [0035], [0050], [0057], [0088]	1-5, 24/1-2 --- 6/1-5, 7/6/1-5, 8/7/6/1-5, 9/6/1-5, 10/6/1-5, 14-20, 25
X --- Y	US 2011/0318741 A1 (SCHAFER PETER H) 29 December 2011; paragraphs [0007]-[0009], [0059], [0073], [0076]-[0077], [0101], [0148], [0150], [0560], [0591]	21-22 --- 25
Y	US 7,183,057 B2 (BENSON NICHOLAS R) 27 February 2007; column 2, lines 3-7, 37-41, and 60-67; column 3, lines 13-17; column 4, lines 63-66; column 18, lines 16-27	6/1-5, 7/6/1-5, 8/7/6/1-5, 9/6/1-5, 10/6/1-5
Y	WO 2019/073477 A1 (MANKIND PHARMA LTD.) 18 April 2019; abstract; page 4, lines 1-6; page 11, lines 6-11	14, 20
Y	WO 2020/018032 A2 (TA KIN TAMBAY) 23 January 2020; page 6, lines 19-28	14-15, 18
Y	WO 2008/065514 A2 (GLENMARK PHARMACEUTICALS LIMITED) 05 June 2008; paragraphs [0017], [0031]-[0032]	14-16
Y	US 2020/0093939 A1 (REGENXBIO INC.) 26 March 2020; paragraphs [0058]-[0062], [0632]	14, 19
Y	CN 105534996 B (CHANGSHA BAISHUN BIOTECHNOLOGY CO LTD) 01 May 2018 (English machine translation); abstract; paragraphs [0009]-0021]	14-15, 17

☐ 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
22 November 2021 (22.11.2021)	DEC 28 2021
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. 571-273-8300	Authorized officer Shane Thomas Telephone No. PCT Helpdesk: 571-272-4300

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US21/41796

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:

      ☒ in the form of an Annex C/ST.25 text file.

      ☐ on paper or in the form of an image file.

   b. ☐ furnished together with the international application under PCT Rule 13*ter*.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.

   c. ☒ furnished subsequent to the international filing date for the purposes of international search only:

      ☒ in the form of an Annex C/ST.25 text file (Rule 13*ter*.1(a)).

      ☐ on paper or in the form of an image file (Rule 13*ter*.1(b) and Administrative Instructions, Section 713).

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 forming part of the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US21/41796

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.: 11-13, 23
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 1-10, 14-22; 24-25 and NOS2/iNOS (biomarker)

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.

-***-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 1-10, 14-22, 24-25 and NOS2/iNOS (biomarker) are directed toward methods directed toward psoriasis and treatment thereof; and kits associated therewith.

The methods and kits will be searched to the extent they encompass NOS2/iNOS (first exemplary biomarker). Applicant is invited to elect additional biomarker(s) to be searched. Additional biomarker(s) will be searched upon the payment of additional fees. It is believed that claims 1-10 (each in-part), 14-20 (each in-part), and 21-22 encompass this first named invention and thus these claims will be searched without fee to the extent that they encompass NOS2/iNOS (biomarker). Applicants must specify the searchable claims that encompass any additionally elected biomarker(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 IL-17C (biomarker).

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

Groups I+ share the technical features including: a method of diagnosing psoriasis in a human subject in need thereof, comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion; (b) determining the level of NOS2/iNOS biomarker in the sample; (d) comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control; wherein the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control; a method of treating psoriasis, comprising identifying a human subject having psoriasis who is likely to be responsive to psoriasis therapy, further comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion; (b) determining the level of NOS2/iNOS biomarker in the sample; (d) comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control; (f) determining that the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference level of NOS2/iNOS biomarkers; and (g) administering a therapeutically effective amount of a psoriasis therapy to the subject identified as likely to be responsive to the psoriasis therapy; a method of discriminating between atopic dermatitis and psoriasis in a human subject exhibiting an indeterminate lesion, the method comprising: (a) obtaining a surface skin sample from the subject exhibiting the skin lesion; (b) measuring the level of NOS2/iNOS biomarker in the sample; (d) comparing the level of the NOS2/iNOS biomarker to a reference level of NOS2/iNOS biomarker in a control sample; (e) determining that the subject has psoriasis if the NOS2/iNOS level in the sample, are higher than the reference levels of NOS2/iNOS biomarker and the one or more additional biomarkers in the control; and (f) determining that the subject has atopic dermatitis if the NOS2/iNOS level, in the sample are similar to or lower than the reference levels in the control; a method of predicting whether a human subject with psoriasis will be responsive to a psoriasis treatment, comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment, (b) measuring the level of NOS2/iNOS biomarker in the samples; and (c) comparing the level of the NOS2/iNOS biomarker in the samples with each other; wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates the likelihood of an effective response to the psoriasis treatment in the subject; a method of monitoring subject response to a psoriasis treatment in a human psoriasis subject in need thereof, comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment, (b) measuring the level of NOS2/iNOS biomarker in the samples; and (c) comparing the level of the NOS2/iNOS biomarker in the samples with each other; wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates an effective response to the psoriasis treatment; and a kit comprising means for quantifying the level of iNOS/NOS2, and one or more additional biomarkers.

However, these shared technical features are previously disclosed by the article 'A specific molecular signature for psoriasis and eczema' by Coimbra, et al. (hereinafter 'Coimbra'), in view of US 2011/0318741 A1 to Schafer, et al. (hereinafter 'Schafer').

Coimbra discloses a method of diagnosing psoriasis in a human subject in need thereof (a method of diagnosing psoriasis in a human subject in need thereof; page 2, second column, fifth paragraph), comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion (regulation of genes in psoriasis skin (comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion); page 2, second column, fourth paragraph); (b) determining the level of NOS2/iNOS biomarker in the sample (determining the level of NOS2/iNOS biomarker in the sample; page 2, second column, fourth paragraph); (d) comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control (wherein NOS2 was upregulated in psoriasis (comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control); page 2, second column, fourth paragraph; wherein the term 'upregulated' indicates a comparison to a control sample); wherein the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control (wherein NOS2 is higher in psoriasis than in controls (the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control); page 2, second column, fourth paragraph); a method of treating psoriasis (a method of treating psoriasis; page 1, first column, second paragraph; page 2, fourth paragraph - fifth paragraph), comprising identifying a human subject having psoriasis who is likely to be responsive to psoriasis therapy (comprising identifying a human subject having psoriasis and selecting a therapy for psoriasis based on said identification (who is likely to be responsive to psoriasis therapy); page 1, first column, second paragraph; page 2, fourth paragraph - fifth paragraph), further comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion (regulation of genes in psoriasis skin (comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion); page 2, second column, fourth paragraph); (b) determining the level of NOS2/iNOS biomarker in the sample (determining the level of NOS2/iNOS biomarker in the sample; page 2, second column, fourth paragraph); (d) comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control (wherein NOS2 was upregulated in psoriasis (comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control); page 2, second column, fourth paragraph; wherein the term 'upregulated' indicates a comparison to a control sample); wherein the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control (wherein NOS2 is higher in psoriasis than in controls (the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control); page 2, second column, fourth paragraph); and (g)

-***-Continued Within the Next Supplemental Box-***-

-***-Continued from previous Supplemental Box-***-

administering a therapeutically effective amount of a psoriasis therapy to the subject identified as likely to be responsive to the psoriasis therapy (administering a therapeutically effective amount of a psoriasis therapy to the subject identified as likely to be responsive to the psoriasis therapy; page 1, second column, second paragraph, page 2, second column, fourth paragraph); a method of discriminating between atopic dermatitis and psoriasis in a human subject exhibiting an indeterminate lesion (a method of discriminating between atopic dermatitis and psoriasis in a human subject exhibiting an indeterminate lesion; page 1, first column, first paragraph; page 2, second column, fifth paragraph), the method comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion (regulation of genes in psoriasis skin (comprising: (a) obtaining a surface skin sample from the subject exhibiting a skin lesion); page 2, second column, fourth paragraph); (b) determining the level of NOS2/iNOS biomarker in the sample (determining the level of NOS2/iNOS biomarker in the sample; page 2, second column, fourth paragraph); (d) comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control (wherein NOS2 was upregulated in psoriasis (comparing the level of the NOS2/iNOS biomarker in the sample to a reference level of NOS2/iNOS biomarker in a control); page 2, second column, fourth paragraph; wherein the term 'upregulated' indicates a comparison to a control sample); wherein the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control (wherein NOS2 is higher in psoriasis than in controls (the subject has psoriasis if the levels of NOS2/iNOS biomarker is higher than the reference levels of the corresponding biomarkers in the control); page 2, second column, fourth paragraph); and (f) determining that the subject has atopic dermatitis if the NOS2/iNOS level in the sample are similar to or lower than the reference levels in the control (determining that the subject has atopic dermatitis if the NOS2/iNOS level in the sample are similar to or lower than the reference levels in the control, and the CCL27 level is upregulated relative to a control; page 2, second column, fourth paragraph-fifth paragraph).

Coimbra does not disclose a method of predicting whether a human subject with psoriasis will be responsive to a psoriasis treatment, comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment, (b) measuring the level of NOS2/iNOS biomarker in the samples; and (c) comparing the level of the NOS2/iNOS biomarker in the samples with each other; wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates the likelihood of an effective response to the psoriasis treatment in the subject; a method of monitoring subject response to a psoriasis treatment in a human psoriasis subject in need thereof, comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment, (b) measuring the level of NOS2/iNOS biomarker in the samples; and (c) comparing the level of the NOS2/iNOS biomarker in the samples with each other; wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates an effective response to the psoriasis treatment; and a kit comprising means for quantifying the level of iNOS/NOS2, and one or more additional biomarkers

Schafer discloses a method of predicting whether a human subject with psoriasis will be responsive to a psoriasis treatment (a method of predicting whether a human subject with psoriasis will be responsive to a psoriasis treatment; paragraph [0007]), comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment (comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment; paragraph [0007]), (b) measuring the level of NOS2/iNOS biomarker in the samples (measuring the level of NOS2/iNOS biomarker in the samples; paragraph [0008]); and (c) comparing the level of the NOS2/iNOS biomarker in the samples with each other (comparing the level of the NOS2/iNOS biomarker in the samples with each other; paragraph [0009]); wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates the likelihood of an effective response to the psoriasis treatment in the subject (wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates the likelihood of an effective response to the psoriasis treatment in the subject; paragraph [0009]); a method of monitoring subject response to a psoriasis treatment in a human psoriasis subject in need thereof (a method of monitoring subject response to a psoriasis treatment in a human psoriasis subject in need thereof; paragraph [0007]), comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment, (comprising: (a) obtaining one or more surface skin samples from the subject before, during, or after treatment; paragraph [0007]), (b) measuring the level of NOS2/iNOS biomarker in the samples (measuring the level of NOS2/iNOS biomarker in the samples; paragraph [0008]); and (c) comparing the level of the NOS2/iNOS biomarker in the samples with each other (comparing the level of the NOS2/iNOS biomarker in the samples with each other; paragraph [0009]); wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates the likelihood of an effective response to the psoriasis treatment in the subject (wherein a decreased level of NOS2/iNOS biomarker in the one or more samples taken during or after treatment compared to that in the one or more samples taken before treatment indicates an effective response to the psoriasis treatment in the subject; paragraph [0009]); and a kit comprising means for quantifying the level of iNOS/NOS2, and one or more additional biomarkers (a kit comprising means for quantifying the level of iNOS/NOS2, and one or more additional biomarkers; paragraphs [0008], [0010]).

It would have been obvious to a person of ordinary skill in the art at the time the invention was made to have modified the disclosure of Coimbra to have provided methods and kits for predicting whether a subject would respond to a particular treatment, for monitoring treatment of a subject with psoriasis, as disclosed by Schafer, in order to extend the use of the markers disclosed by Coimbra to enhance the effectiveness of these processes. It further would have been obvious to a person of ordinary skill in the art at the time the invention was made to have modified the disclosure of Coimbra to have provided a kit for diagnosis and treatment of psoriasis, such as a kit, as described by Schafer, in order to enable effective standardized diagnosis and treatment of psoriasis based on the detected markers.

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 a combination of the Coimbra and Schafer references, unity of invention is lacking.