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- 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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- (88) **Date of publication of the international search report:**
19 February 2015

(54) **Title:** MATHEMATICAL PROCESSES FOR DETERMINATION OF PEPTIDASE CLEAVAGE

(57) **Abstract:** The present invention provides a bioinformatic methodology for prediction of peptidase cleavage sites based on principal component analysis and based on training sets obtained by experimental protein cleavage. This invention is not limited to training sets derived from CSL approaches, nor to any other experimental determination of cleavage site. Undoubtedly there will be new approaches to developed for experimental measurement of cleavage sites and these too may be the source of training sources for the present invention.



INTERNATIONAL SEARCH REPORT

International application No.

PCT/US14/41525

A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - C12Q 1/37; G01N 33/48; C40B 50/02 (2014.01)

CPC - G06F 19/18, 19/24; C12Q 1/37

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 1/37; G01N 33/48, 33/50; C40B 50/02 (2014.01)

CPC: G06F 19/18, 19/24, 19/10; C12Q 1/37, 1/34; USPC: 435/24, 18, 4; 702/19, 1

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; Dialog ProQuest; PubMed/PubMed Central; Google/Google Scholar; predict, 'peptidase cleavage site,' equation, 'computer process,' peptidase, cleavage, cathepsin, cleaved, uncleaved, compare, intracellular, intranuclear, cytoplasmic, lysosome, macrophage

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- A	WO 2013/040142 A2 (IOGENETICS LLC) March 21, 2013; page 4, lines 10-34; page 6, lines 19-20; page 16, lines 9-24; page 42, lines 15-18; page 51, lines 27-29; page 64, lines 25-33; page 210, lines 14-25; page 214, lines 10-15	1, 2, 4/1, 5-36, 54-59, 75-83 ----- 3, 4/3, 37-53
Y	GUHA, S et al. Cathepsins: Fundamental Effectors Of Endolysosomal Proteolysis. Indian J Biochem Biophys. April 2008, Vol. 45, No. 2; pages 75-90; page 81, second column, second paragraph; page 82, first column, third paragraph; page 83, first column, fourth paragraph; PMID: 21086720.	10, 12-19, 21
Y	TEDELIND, et al. Nuclear Cysteine Cathepsin Variants In Thyroid Carcinoma Cells. Biol. Chem. August 2010, Vol. 391; pages 923-935; page 924, first column, second paragraph; DOI 10.1515/BC.2010.109.	11
Y	REISER, J et al. Specialized Roles For Cysteine Cathepsins In Health And Disease. J Clin Invest. October 2010, Vol. 120, No. 10; pages 3421-3431; page 3427, first column, first paragraph; DOI: 10.1172/JCI42918.	20
Y	ARTIGAS, G et al. Purification Process To Obtain Cathepsin D, B And L Suitable To Be Used In Food Industry. Food Biotechnology. 1996, Vol. 10, No. 1; pages 41-45; abstract; DOI: 10.1080/08905439609549900.	22
A	US 8455203 B2 (WANG, J et al.) June 4, 2013; column 42, lines 32-47; column 89, lines 55-66	3, 4/3, 37-53
A	WO 2011/119484 A1 (IOGENETICS LLC) September 29, 2011; abstract	1-59, 75-83

☒ 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

15 December 2014 (15.12.2014)

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INTERNATIONAL SEARCH REPORT

International application No.

PCT/US14/41525

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	BREMEL, R et al. An Integrated Approach To Epitope Analysis II: A System For Proteomic-Scale Prediction Of Immunological Characteristics. Immunome Res. 02 November 2010, Vol. 6, No. 8; pages 1-21; DOI: 10.1186/1745-7580-6-8.	1-59, 75-83
A	SHI, G et al. Molecular Cloning And Expression Of Human Alveolar Macrophage Cathepsin S, An Elastinolytic Cysteine Protease. J Biol Chem. 15 April 1992, Vol. 267, No. 11; pages 7258-7562; PMID: 1373132.	1-59, 75-83
A	SONG, J et al. PROSPER: An Integrated Feature-Based Tool For Predicting Protease Substrate Cleavage Sites. PLoS ONE. 29 November 2012, Vol. 7, No. 11, e50300; DOI: 10.1371/journal.pone.0050300.	1-59, 75-83

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US14/41525

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.:
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.:

Group I: Claims 1-59, 75-83

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/US14/41525

-***-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.

Group I: Claims 1-59 and 75-83 are directed toward a computer implemented process for generating an ensemble of peptidase cleavage prediction equations and a computer system or computer readable medium therefor.

Group II: Claims 60-74 and 86-99 are directed toward a synthetic polypeptide comprising peptide sequence that binds to at least one major histocompatibility complex (MHC) binding region.

Group III: Claims 84 and 85 are directed toward a graphical representation of the probability of peptidase cleavage, wherein the amino acid sequence forms one axis, and the probability of a peptide sequence comprising a peptidase cleavage site are charted against the amino acid sequence axis.

The inventions listed as Groups I-III 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 special technical features of Group I include a computer implemented process for generating an ensemble of peptidase cleavage prediction equations, which is not present in Groups II or III, the special technical features of Group II including a polypeptide comprising a peptide sequence that binds to at least one major histocompatibility complex (MHC) binding region with a predicted affinity of greater than about 10^6 M⁻¹, which is not present in either of Groups I or III, the special technical features of Group III including a graphical representation.

Groups I-III share the technical features including peptidase cleavage and an amino acid sequence. Groups I and II include a predicted peptidase cleavage site. Groups I and III share the technical features including predicting the probability of peptidase cleavage.

However, these shared technical features are previously disclosed by WO 2013/040142 A2 to Bremel, et al. (hereinafter 'Bremel'). Bremel discloses peptidase cleavage (cleavage by cathepsins (peptidase cleavage); page 210, lines 14-28); an amino acid sequence (a primary amino acid sequence; page 210, lines 14-28); a predicted peptidase cleavage site (predicting a peptidase cleavage pattern; page 210, lines 14-28); and the probability of peptidase cleavage (prediction of the probability of cleavage; page 210, lines 14-28).

Since none of the special technical features of the Groups I-III inventions is found in more than one of the inventions, and since all of the shared technical features are previously disclosed by the Bremel reference, unity of invention is lacking.