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(71) Applicants (for all designated States except US): **QUEST DIAGNOSTICS INVESTMENTS INCORPORATED** [US/US]; 300 Delaware Avenue, Wilmington, DE 19899 (US). **U.S. GENOMICS** [US/US]; 12 Gill Street, Suite 4700, Woburn, MA 01801 (US).

(72) Inventors; and

(75) Inventors/Applicants (for US only): **HUANG, Donghui** [CN/US]; 15 Darlington, Irvine, CA 92620 (US). **STROM, Charles, M.** [US/US]; 2939 Calle Gaucho, San Clemente, CA 92673 (US). **POTTS, Steven, J.** [US/US]; 13480 Turlock Ct., San Diego, CA 92129 (US). **ROOKE, Jenny, Ellen** [US/US]; 1 Fitchburg St. #F192, Somerville, MA 02143 (US).

(74) Agents: **WARBURG, Richard, J.** et al.; Foley & Lardner LLP, P.O. Box 80278, San Diego, CA 92138-0278 (US).

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11 December 2008

(54) Title: NUCLEIC ACID SIZE DETECTION METHOD

(57) Abstract: The present invention provides methods of determining the size of a particular nucleic acid segment of interest in a sample of nucleic acids through fragmentation of DNA, size fractionation, an optional second fragmentation, and identification using a marker sequence. In particular aspects, an expansion or reduction of tandem repeat sequences can be detected. In further aspects, carriers and individuals afflicted with fragile X syndrome or other diseases associated with tandem repeats can be distinguished from normal individuals.



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INTERNATIONAL SEARCH REPORT

International application No.

PCT/US07/08985

A. CLASSIFICATION OF SUBJECT MATTER

IPC: C12P 19/34(2006.01);B01D 57/02(2006.01)

USPC: 435/91.2;204/450

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)

U.S. : 435/91.2; 204/450

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

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

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- Y	Petek et al. (1999) American Journal of Medical Genetics 84:229-232	1-7, 10-14, 17-23 ----- 8, 9, 15, 16
Y	Enzelberger et al. (US pat. 6, 960,437 issued Nov 1, 2005)	8, 9, 15, 16



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

"E" earlier application or patent 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

21 August 2008 (21.08.2008)

Date of mailing of the international search report

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Mail Stop PCT, Attn: ISA/US
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450

Facsimile No. (571) 273-3201

Authorized officer

SUCHIRA PANDE

Telephone No. 571-272-0700

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US07/08985

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 Continuation 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 any 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.: 1-23

- 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.

BOX 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, claim(s) 1-71, drawn to a method.

Group II, claim(s) 72-75, drawn to product (kit).

The inventions listed as Groups I and II 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:

Petek et al. (1999) American Journal of Medical Genetics 84:229-232 teach all the components of the kit (product) of claim 72. Claim 72 recites a kit for detecting the size of a particular nucleic acid segment in a sample comprising:

a primer pair for amplifying a marker nucleotide sequence upstream or downstream of the particular nucleic acid segment (see Fig. 2 Panel A where primer pair WK195 and WK196 are taught), and one or more restriction endonucleases (Enzyme PstI is taught as an enzyme) for cleaving the nucleic acid sample to generate a fragment of the nucleic acid sample which contains the particular nucleic acid segment and the upstream or downstream marker sequence, wherein said particular nucleic acid segment is a tandem repeat sequence (see fig. 2 Panel B CGG tandem repeat is taught). Thus all the components of the kit namely the primer pair, the restriction enzyme and the template DNA recited in claim 72 are taught by Prior art to one of ordinary skill at the time the invention was made. Hence the kit of claim 72 lacks the same or corresponding special technical features of the method of group I invention.

Species of method of group I invention:

1. A method for determining the size of a nucleic acid segment in a nucleic acid sample (Claims 1-23)
2. A method of detecting a mutation in a tandem repeat segment of a gene in a nucleic acid sample (claims 24-45)
3. A method of identifying FMR1 alleles having a normal number of tandem repeats, a premutation, or a full mutation in the nucleic acid of an individual (claims 46-61)
4. A method of determining the size of a tandem repeat segment in a sample of nucleic acids (claims 62-64)
5. A method for screening male and female individuals for carrier status of mutations in the tandem repeat region of the FMR1 gene (Claims 65-71)

The following claim(s) are generic: Claim 1 is generic.

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

Claim 1 is generic. Rest of the methods recited in claims 24-71 are species of this generic method. They are directed to achieving specific different end results and consist of different steps in addition to the steps claimed in claim 1.

If no additional fee is paid the search will be restricted to the first invention recited in the claims i.e. the method (claims 1-71). Within the method the search will be further restricted to the species of method first recited in the claims i.e. claims 1-23.