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(54) Title: BIOCHEMICAL ANALYSIS OF PARTITIONED CELLS

(57) Abstract: The invention relates to compositions and methods for the analysis of biomolecules associated with cells, where the presence or absence of a particular biomolecule (e.g., an expressed protein or a nucleic acid gene expression product) associated with the cells is examined. The invention provides methods for single cell biochemical analysis, as well as instrumentation for the single-cell biochemical analysis. Most advantageously, the invention affords methods and instrumentation for high-throughput biochemical analysis of large numbers of single cells.

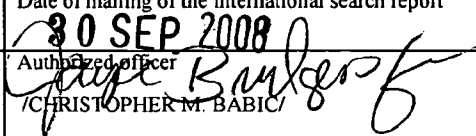


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INTERNATIONAL SEARCH REPORT

International application No.

PCT/US07/12727

A. CLASSIFICATION OF SUBJECT MATTER IPC: C12M 1/00(2006.01);C12Q 1/68(2006.01) USPC: 435/283.1,6 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. : 435/283.1, 6 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) EAST, PUBMED, SCIENCE DIRECT		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	U.S. 2002/0058332 A1 (QUAKE et al.) 16 May 2002 (16.05.2002), fig. 16-18, [0105-0118], [0174-0191].	1-4
Y	U.S. 2005/0227264 A1 (NOBILE et al.) 13 October 2005 (13.10.2005), whole document.	1-4
Y	U.S. 5,830,663 (EMBLETON et al.) 03 November 1998 (03.11.1998), col. 10-11, example 2.	1-4
A	U.S. 6,489,103 B1 (GRIFFITHS et al.) 03 December 2002 (03.12.2002).	1-4
<input type="checkbox"/> Further documents are listed in the continuation of Box C. <input type="checkbox"/> 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	"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
"E" earlier application or patent published on or after the international filing date	"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
"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)	"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
"O" document referring to an oral disclosure, use, exhibition or other means	"&"	document member of the same patent family
"P" document published prior to the international filing date but later than the priority date claimed		
Date of the actual completion of the international search 17 September 2008 (17.09.2008)	Date of mailing of the international search report 30 SEP 2008	
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) 273-3201	Authorized officer  CHRISTOPHER M. BABIC Telephone No. 571-272-1600	

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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.:
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.: 1-4

- 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.

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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) 1-4, drawn to a device that generates single cell aqueous reaction volumes.

Group II, claim(s) 5-60, drawn to drawn to a method for detecting a biomolecule associated with cells.

Group III, claim(s) 61-79, drawn to a composition.

This application contains claims directed to more than one species of the generic invention. These species are deemed to lack unity of invention because they are not so linked as to form a single general inventive concept under PCT Rule 13.1.

In order for more than one species to be examined, the appropriate additional examination fees must be paid. The species are as follows:

a) Nucleic acid

b) Polypeptide

The claims are deemed to correspond to the species listed above in the following manner:

Claims 15-24, 43-52, and 65-68 (nucleic acid)

Claims 25-32, 53-60, and 72-79 (polypeptide)

The following claim(s) are generic: 5, 33, and 61.

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:

A composition comprising: a) a plurality of aqueous reaction core solutions, each comprising: (i) a cell, and (ii) at least one reagent for detecting the presence or absence of a biomolecule associated with said cell, wherein the reagent is capable of generating or participating in generating an amplified signal corresponding to the presence or absence of the biomolecule; and b) an immiscible liquid shell that partitions the aqueous reaction core solution was known in the art at the time of invention (U.S. 4,353,888; col. 5-6, examples 1-4; Sefton teaches microencapsulation of red blood cells suspended in PBS; such a suspension solution is capable of participating in generating an amplified signal corresponding to the presence or absence of a biomolecule, for example). The said product cannot therefore be considered a special technical feature, as lack of unity rules hold that a feature known to a person of ordinary skill in the art makes no advance over the prior art.

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:

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Nucleic acids and polypeptides are structurally distinct molecules.