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(54) Title: DEFINED CULTURE CONDITIONS OF HUMAN EMBRYONIC STEM CELLS

(57) Abstract: The present invention relates to compositions and methods for culturing stem cells, particularly embryonic stem cells. Specifically, the invention relates to a culture medium that supports proliferation of substantially undifferentiated stem cells, while maintaining potency of the cells. In an embodiment, the culture medium is defined and supports proliferation of substantially undifferentiated embryonic stem cells in essentially serum free and feeder cell free conditions. Compositions for making the medium and methods using the culture medium are also provided.



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INTERNATIONAL SEARCH REPORT

International application No.

PCT/US06/29473

A. CLASSIFICATION OF SUBJECT MATTER

IPC: C12N 5/00(2006.01),5/02(2006.01)

USPC: 435/404,405,407,383,384,388,389;424/93.7

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. : 435/404,405,407,383,384,388,389;424/93.7

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)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2004/0219136 A1 (HARIR) 4 November 2004 (04.11.2004), whole document.	1, 2, 12, 13, 17, 18, 28-32, 41, 42, 47-50, 53, 54
X	US 2004/0244806 A1 (FERREE) 9 December 2004 (09.12.2004), especially paragraph 0036.	17, 18, 28
X	US 6,713,057 B1 (CHATTERJEE) 30 March 2004 (30.03.2004), whole document, especially Examples.	30
A	WO 03/1044442 A1 (PEBAY et al.) 18 December 2003 (18.12.2003), whole document.	1, 2, 12, 13, 17, 18, 28-32, 41, 42, 47-50, 53, 54

☐ 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

16 May 2008 (16.05.2008)

Date of mailing of the international search report

24 JUN 2008

Name and mailing address of the ISA/US

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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.: 3-11,14-16,19-27,33-40,43-46,51,52 and 55
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.:

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 1, claim(s) 1 and 2, drawn to a cell culture media for stem cells.

Group 2, claim(s) 31 and, 32, drawn to a composition of matter comprising the cell culture medium of claim 1, and stem cells.

Group 3, claim(s) 17, 18 and 28, drawn to a first composition for preparing a culture medium.

Group 4, claim(s) 29, drawn to a second composition for preparing a culture medium.

Group 5, claim(s) 30, drawn to a third composition for preparing a cell culture medium.

Group 6, claim(s) 12, 13, 47 and 48, drawn to a first culture system for stem cells, comprising the cell culture medium of claim 1, plus a matrix.

Group 7, claim(s) 49 and 50, drawn to a second culture system for stem cells, comprising the composition of claim 29, plus a matrix.

Group 8, claim(s) 41 and 42, drawn to a method of culturing stem cells using the cell culture system of claim 12.

Group 9, claim(s) 53 and 54, drawn to a cell therapy treatment method, comprising a step of culturing stem cells using the cell culture system of claim 49.

The inventions listed as Groups 1-9 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 only feature shared by all claims is the presence of a TNF protein, leukocytes naturally produce TNF-alpha (see, Ferree, paragraph 0036); thus a composition comprising a protein from the TNF family does not provide a contribution over the prior art, so unity of invention is lacking.

Alternatively, if one were to consider the combination of a protein from the TNF family and a protein from the Wnt family to be the special technical feature, linking at least Groups 1-4 and 6-9, leukocytes still possess both proteins TNF-alpha and TGF-beta (disclosed as species of proteins from the Wnt family at page 23, line 1 of current specification); thus even this shared technical feature does not provide a contribution over the prior art, so unity of invention is lacking. Claim 30 (Inventive Group 5) lacks this feature entirely.