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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
12/227,458	10/23/2009	Ian Mcniece	PROTEO-32848/US-2/PCT	2285

7590 09/04/2015

MCNIECE COHEN FOUNDATION
438 MINORCA AVE
CORAL GABLES, FL 33134

EXAMINER
MOSS, NATALIE M

ART UNIT	PAPER NUMBER
1653	

MAIL DATE	DELIVERY MODE
09/04/2015	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of Abandonment	Application No.	Applicant(s)
	12/227,458	MCNIECE, IAN
	Examiner	Art Unit
	NATALIE MOSS	1653

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address--

This application is abandoned in view of:

1. ☒ Applicant's failure to timely file a proper reply to the Office letter mailed on *09 January 2015*.
   (a) ☐ A reply was received on _____ (with a Certificate of Mailing or Transmission dated _____), which is after the expiration of the period for reply (including a total extension of time of _____ month(s)) which expired on _____.
   (b) ☐ A proposed reply was received on _____, but it does not constitute a proper reply under 37 CFR 1.113 to the final rejection.
   (A proper reply under 37 CFR 1.113 to a final rejection consists only of: (1) a timely filed amendment which places the application in condition for allowance; (2) a timely filed Notice of Appeal (with appeal fee); or (3) if this is utility or plant application, a timely filed Request for Continued Examination (RCE) in compliance with 37 CFR 1.114. Note that RCEs are not permitted in design applications.)
   (c) ☐ A reply was received on _____ but it does not constitute a proper reply, or a bona fide attempt at a proper reply, to the non-final rejection. See 37 CFR 1.85(a) and 1.111. (See explanation in box 7 below).
   (d) ☒ No reply has been received.

2. ☐ Applicant's failure to timely pay the required issue fee and publication fee, if applicable, within the statutory period of three months from the mailing date of the Notice of Allowance (PTOL-85).
   (a) ☐ The issue fee and publication fee, if applicable, was received on _____ (with a Certificate of Mailing or Transmission dated _____), which is after the expiration of the statutory period for payment of the issue fee (and publication fee) set in the Notice of Allowance (PTOL-85).
   (b) ☐ The submitted fee of $_____ is insufficient. A balance of $_____ is due.
   The issue fee required by 37 CFR 1.18 is $_____. The publication fee, if required by 37 CFR 1.18(d), is $_____.
   (c) ☐ The issue fee and publication fee, if applicable, has not been received.

3. ☐ Applicant's failure to timely file corrected drawings as required by, and within the three-month period set in, the Notice of Allowability (PTO-37).
   (a) ☐ Proposed corrected drawings were received on _____ (with a Certificate of Mailing or Transmission dated _____), which is after the expiration of the period for reply.
   (b) ☐ No corrected drawings have been received.

4. ☐ The letter of express abandonment which is signed by the attorney or agent of record or other party authorized under 37 CFR 1.33(b). See 37 CFR 1.138(b).

5. ☐ The letter of express abandonment which is signed by an attorney or agent (acting in a representative capacity under 37 CFR 1.34) upon the filing of a continuing application.

6. ☐ The decision by the Board of Patent Appeals and Interference rendered on _____ and because the period for seeking court review of the decision has expired and there are no allowed claims.

7. ☐ The reason(s) below:

/KAREN COCHRANE CARLSON/
Primary Examiner, Art Unit 1656

Petitions to revive under 37 CFR 1.137, or requests to withdraw the holding of abandonment under 37 CFR 1.181, should be promptly filed to minimize any negative effects on patent term.

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Office Action Summary	Application No. 12/227,458	Applicant(s) MCNIECE, IAN	
	Examiner NATALIE MOSS	Art Unit 1653	AIA (First Inventor to File) Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *11/24/2014*.

☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

5)☒ Claim(s) *1-16,19 and 20* is/are pending in the application.

5a) Of the above claim(s) *2,13-16,19 and 20* is/are withdrawn from consideration.

6)☐ Claim(s) _____ is/are allowed.

7)☒ Claim(s) *1 and 3-12* is/are rejected.

8)☐ Claim(s) _____ is/are objected to.

9)☐ Claim(s) _____ are subject to restriction and/or election requirement.

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to PPHfeedback@uspto.gov.

Application Papers

10)☐ The specification is objected to by the Examiner.

11)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

a)☐ All b)☐ Some** c)☐ None of the:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)

2) ☒ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b) Paper No(s)/Mail Date _____.

3) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .

4) ☐ Other: _____.

DETAILED ACTION

This Office Action is in response to the papers filed on 27 February 2013.

APPLICANT'S ELECTION

A requirement for restriction was issued by Examiner Lora Driscoll on 04 January 2012. Applicants' election without traverse of Group II (Claims 1 and 3-12; drawn to a method for propagation of a non-adherent culture of mesenchymal stem cells) in the reply filed on 27 February 2013 is acknowledged.

Upon further consideration, Examiner has rejoined Claim 2.

Claims 13-16 and 19-20 are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected invention, there being no allowable generic or linking claim.

PRIORITY

Acknowledgement of Provisional Application 60/801661, filed on 19 May 2006, is made.

CLAIMS UNDER EXAMINATION

Claims 1-20 are pending. Claims 1-12 have been examined on their merits.

Claim Rejections - 35 USC § 112

The following is a quotation of 35 U.S.C. 112(b):

> (b) CONCLUSION.—The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the inventor or a joint inventor regards as the invention.

> The following is a quotation of 35 U.S.C. 112 (pre-AIA), second paragraph:
> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 2-3, 8 and 10-11 are rejected under 35 U.S.C. 112(b) or 35 U.S.C. 112 (pre-AIA), second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which the inventor or a joint inventor, or for pre-AIA the applicant regards as the invention.

Claims 2-3 recites Matrigel™ and Teflon® respectively. Because the trademarks are used in the cited claims as a limitation to identify or describe a particular material or product, the claim does not comply with the requirements of the 35 U.S.C. 112(b) or pre-AIA 35 U.S.C. 112, second paragraph. *Ex parte Simpson*, 218 USPQ 1020 (Bd. App. 1982).

Claim 8 recites the term "substantially". The term "substantially" is a relative term which renders the claim indefinite. The term "substantially" is not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised of the scope of the invention.

Claims 10-11 are drawn to an in vivo method of treatment. Claim 1 recites an in vitro method of cell propagation. Hence, an in vivo method of treatment lacks antecedent basis in an in vitro method of culture. Appropriate correction is required.

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of pre-AIA 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.
>
> e) the invention was described in (1) an application for patent, published under section 122(b), by another filed in the United States before the invention by the applicant for patent or (2) a patent granted on an application for patent by another filed in the United States before the invention by the applicant for patent, except that an international application filed under the treaty defined in section 351(a) shall have the effects for purposes of this subsection of an application filed in the United States only if the international application designated the United States and was published under Article 21(2) of such treaty in the English language.

Claims 1, 3, 5-8 and 10-12 are rejected under pre-AIA 35 U.S.C. 102b as being anticipated by Huang et al. (Chondrogenesis of Human Bone Marrow-Derived Mesenchymal Stem Cells in Agarose Culture. (2004) The Anatomical Record Part A 278A:428-436).

Huang et al. teaches a method that comprises culturing human mesenchymal stem cells in agarose constructs (See Abstract). Therefore Claims 1 and 3 are included in this rejection (**Claims 1 and 3**).

Huang teaches that said cells are harvested from human patients by filtering and centrifugation (See page 429, right "Materials and Methods"). Therefore the art teaches mechanical manipulation of said cells. Claim 5 is rejected (**Claim 5**). The art teaches that said cells are isolated from bone marrow (hence, a biological sample containing mesenchymal stem cells) (See page 429, right "Materials and Methods").Therefore Claims 6-7 are rejected (**Claims 6-7**). Said cells are interpreted to be substantially purified. Therefore Claim 8 is rejected (**Claim 8**).

Because Huang anticipates the mesenchymal stem cells of Claim 1, they are suitable for administration to a subject, wherein the subject is human. Therefore Claims 10-11 are included in this rejection (**Claims 10-11**).

Huang teaches that said cells are cultured for 21 days (See page 430, right column, first paragraph). Therefore Claim 12 is included in this rejection (**Claim 12**).

Therefore Huang et al. anticipates Applicant's invention as claimed.

Claims 1, 3-5 and 10-12 are rejected under pre-AIA 35 U.S.C. 102b as being anticipated by Wang et al. (Ex vivo expansions and transplantations of mouse bone marrow-derived hematopoietic stem/progenitor cells. J Zhejiang Univ SCI 2004 5(2):157-163).

Wang et al. disclose a method of culturing mesenchymal stem cells (See Abstract). Mesenchymal stem cells are grown in Teflon culture bags (See page 158 right column, second paragraph). Therefore Claims 1 and 3 are rejected (**Claims 1 and 3**).

Claim 4 is included in this rejection because the art does not teach the use of trypsin in said propagation (**Claim 4**).

Culture of said cells in a Teflon bag anticipates Claim 5 (**Claim 5**).

Because Wang anticipates the mesenchymal stem cells of Claim 1, they are suitable for administration to a subject, wherein the subject is human. Therefore Claims 10-11 are included in this rejection (**Claims 10-11**).

The art teaches culture for 7 days (See page 158 right column, second paragraph). Therefore Claim 12 is rejected (**Claim 12**).

Therefore Wang et al. anticipates Applicant's invention as claimed.

Claims 1-2, 5-8 and 10-12 are rejected under pre-AIA 35 U.S.C. 102(e) as being anticipated by Lin et al. (Human Mesenchymal Stem Cells And Culturing Methods Thereof. US 2007/0128722, filed on 05 December 2005).

Lin et al teach a method that comprises culturing mesenchymal stem cells obtained from cord blood on Matrigel (See [0072]). Therefore Claims 1-2 are anticipated (**Claims 1-2**). Said cells are isolated from cord blood (See [0076]). Said isolation anticipates mechanical manipulation of said cells. Therefore Claim 5 is rejected (**Claim 5**). Further, isolation form cord blood (a biological sample) anticipates Claims 6-7 (**Claims 6-7**). Said isolated cells anticipate Claim 8

(**Claim 8**). Because Lin anticipates the mesenchymal stem cells of Claim 1, they are suitable for administration to a subject, wherein the subject is human. Therefore Claims 10-11 are included in this rejection (**Claims 10-11**).

Lin teaches culturing cells for 7-10 days (See [0057]). Therefore Claim 12 is included in this rejection (**Claim 12**).

Claim Rejections - 35 USC § 103

The following is a quotation of pre-AIA 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The factual inquiries set forth in *Graham v. John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under pre-AIA 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.

2. Ascertaining the differences between the prior art and the claims at issue.

3. Resolving the level of ordinary skill in the pertinent art.

4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

Claims 1 and 9 are rejected under pre-AIA 35 U.S.C. 103(a) as being unpatentable over Huang et al.

Claim 1 is rejected as recited supra.

Huang teaches that said cells are cultured for 21 days (See page 430, right column, first paragraph).

While the art teaches that said cells are cultured in the agarose, Huang does not teach the final concentration of said cells at the end of the culture period.

The amount final concentration of cells obtained depends on the total culture time, which is a results effective variable. Culturing cells for a defined period of time to obtain a desired concentration of cells is well known in the art. The final concentration would be arrived at through experimental optimization. Therefore Claim 9 Is rendered obvious (**Claim 9**).

Therefore Huang et al. renders obvious Applicant's Invention as claimed.

Conclusion

No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to NATALIE MOSS whose telephone number is (571) 270-7439. The examiner can normally be reached on Monday-Friday, 8am-5pm EST.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Sharmila Landau can be reached on (571) 272-0614. The fax phone number for the organization where this application or proceeding is assigned is (571) 270-8439.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the APIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/NATALIE MOSS/
Examiner
Art Unit 1653

/KAREN COCHRANE CARLSON/

Primary Examiner, Art Unit 1656

Notice of References Cited	Application/Control No. 12/227,458	Applicant(s)/Patent Under Reexamination MCNIECE, IAN	
	Examiner NATALIE MOSS	Art Unit 1653	Page 1 of 1

U.S. PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Name	Classification
*	A	US-2007/0128722	06-2007	Lin et al.	435/366
	B	US-			
	C	US-			
	D	US-			
	E	US-			
	F	US-			
	G	US-			
	H	US-			
	I	US-			
	J	US-			
	K	US-			
	L	US-			
	M	US-			

FOREIGN PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Country	Name	Classification
	N					
	O					
	P					
	Q					
	R					
	S					
	T					

NON-PATENT DOCUMENTS

*		Include as applicable: Author, Title Date, Publisher, Edition or Volume, Pertinent Pages)
	U	Huang et al. (Chondrogenesis of Human Bone Marrow-Derived Mesenchymal Stem Cells in Agarose Culture. (2004) The Anatomical Record Part A 278A:428-436).
	V	Wang et al. (Ex vivo expansions and transplantations of mouse bone marrow-derived hematopoietic stem/progenitor cells. J Zhejiang Univ SCI 2004 5(2):157-163).
	W	
	X	

*A copy of this reference is not being furnished with this Office action. (See MPEP § 707.05(a).)
Dates in MM-YYYY format are publication dates. Classifications may be US or foreign.

12/227458 1653

IAP20Rec'd PCT 17 NOV 2008

PTO/SB/08a (09-08)
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INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Use as many sheets as necessary)

Sheet	1	of	2

Complete if Known	
Application Number	Not Yet Assigned
Filing Date	Herewith
First Named Inventor	Ian Mcniece
Art Unit	N/A
Examiner Name	Not Yet Assigned
Attorney Docket Number	68324(71699)

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document	Pages, Columns, Lines, Where Relevant Passages or Relevant Figures Appear
	AA*	US-20050265980	12-01-2005	Chen et al.	
	AB*	US-20040092011	05-13-2004	Wilkison et al.	
	AC*	US-20050013804	01-20-2005	Kato et al.	

FOREIGN PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Foreign Patent Document Country Code[3]-Number[4]-Kind Code[5] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document	Pages, Columns, Lines, Where Relevant Passages Or Relevant Figures Appear	T[6]

Examiner Signature	/Natalie Moss/	Date Considered	01/05/2015

*EXAMINER: Initial if reference considered, whether or not citation is in conformance with MPEP 609. Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. * CITE NO.: Those application(s) which are marked with an single asterisk (*) next to the Cite No. are not supplied (under 37 CFR 1.98(a)(2)(iii)) because that application was filed after June 30, 2003 or is available in the IFW. [1] Applicant's unique citation designation number (optional). [2] See Kinds Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English language Translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /N.M./

12/227458

PTO/SB/08b (09-06)
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INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Use as many sheets as necessary)

Sheet	2	of	2

Complete if Known	
Application Number	Not Yet Assigned
Filing Date	Herewith
First Named Inventor	Ian Mcniece
Art Unit	N/A
Examiner Name	Not Yet Assigned
Attorney Docket Number	68324(71699)

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.), date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]

Examiner Signature	/Natalie Moss/	Date Considered	01/05/2015

*EXAMINER: Initial if reference considered, whether or not citation is in conformance with MPEP 609. Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant.

[1]Applicant's unique citation designation number (optional). [2]Applicant is to place a check mark here if English language Translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /N.M./

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NEWS 8	FEB 03	DWPI: New coverage of Singapore PCT-transfers and grants
NEWS 9	FEB 24	INFULL and DEFULL databases Now Available via STN Viewer
NEWS 10	MAR 28	New STN Platform Enhancements Available, Increase Efficiency of Search Workflow.
NEWS 11	APR 25	New Format Adopted for Taiwanese Granted Patent Numbers in CAS Databases and INPADOC.
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NEWS 13	MAY 9	STN AnaVist, Version 2.1, Improves Operating System Compatibility and Performance
NEWS 14	MAY 19	Availability of Digital Object Identifiers (DOIs) Enhanced in STN Databases
NEWS 15	MAY 20	New Cluster NPS available for all Databases with the Numeric Property Search feature
NEWS 16	MAY 29	CAS REGISTRY BLAST Upgrade Improves Search Capabilities and Results Ranking
NEWS 17	JUN 10	MEDLINE on STN Now Updated Daily
NEWS 18	AUG 18	Latest Version of Emtree Introduces 811 New Terms
NEWS 19	JUL 1	CHEMCATS (Chemical Catalogs Online) on STN Enhanced with New Search and Display Fields and More Frequent Updates
NEWS 20	JUL 24	Batch search results for DGENE, USGENE and PCTGEN now available for 30 days
NEWS 21	JUL 28	Latest release of new STN now available, expands global patent coverage and enhances search capabilities
NEWS 22	SEP 4	KRFULL: New Full-text Database for Korean Patent Publications Now Available on new STN
NEWS 23	OCT 1	Cooperative Patent Classification (CPC) Combination Set Data

Now Available in CAplus, INPADOCDB and USPAT Databases
NEWS 24 OCT 23 CPC Thesaurus based on official CPC Scheme
NEWS 25 DEC 22 2015 MeSH Thesaurus Installed in MEDLINE with a Special Message for Customers Doing Pharmacovigilance Research
NEWS 26 DEC 24 CAS Expands Coverage of Reactions from Dissertations in CASREACT
NEWS 27 DEC 24 Additional Experimental Spectra Now Available in CAS REGISTRY in STN

NEWS EXPRESS 27 MAY 2014 CURRENT WINDOWS VERSION IS V8.5.2.1,
AND CURRENT DISCOVER FILE IS DATED 29 SEPTEMBER 2014.

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FILE COVERS 1907 - 5 Jan 2015 VOL 162 ISS 3
FILE LAST UPDATED: 4 Jan 2015 (20150104/ED)
REVISED CLASS FIELDS (/NCL) LAST RELOADED: Nov 2014
USPTO MANUAL OF CLASSIFICATIONS THESAURUS ISSUE DATE: Sep 2014

CAplus includes complete International Patent Classification (IPC) reclassification data for the fourth quarter of 2014.

CAplus now includes the comprehensive Cooperative Patent Classification (CPC). See HELP CPC for details.

CAS Information Use Policies apply and are available at:

http://www.cas.org/legal/infopolicy

This file contains CAS Registry Numbers for easy and accurate substance identification.

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=> s mesenchymal stem cells
          58109 MESENCHYMAL
              3 MESENCHYMALS
          58109 MESENCHYMAL
                  (MESENCHYMAL OR MESENCHYMALS)
         318793 STEM
          48648 STEMS
         351230 STEM
                  (STEM OR STEMS)
        3174152 CELLS
              2 CELLSES
        3174153 CELLS
                  (CELLS OR CELLSES)
L1        23398 MESENCHYMAL STEM CELLS
                  (MESENCHYMAL(W)STEM(W)CELLS)

=> s L1 and "mesenchymal stem"
          58109 "MESENCHYMAL"
              3 "MESENCHYMALS"
          58109 "MESENCHYMAL"
                  ("MESENCHYMAL" OR "MESENCHYMALS")
         318793 "STEM"
          48648 "STEMS"
         351230 "STEM"
                  ("STEM" OR "STEMS")
          32322 "MESENCHYMAL STEM"
                  ("MESENCHYMAL"(W)"STEM")
L2        23398 L1 AND "MESENCHYMAL STEM"

=> s L2 and agarose
          49595 AGAROSE
            170 AGAROSES
          49622 AGAROSE
                  (AGAROSE OR AGAROSES)
L3          112 L2 AND AGAROSE

=> s L3 and culture
         663462 CULTURE
         278958 CULTURES
         829108 CULTURE
                  (CULTURE OR CULTURES)
L4           68 L3 AND CULTURE

=> d 1-68
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L4 ANSWER 1 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2014:1953262 CAPLUS Full-text
TI Chitosan-pDNA nanoparticle characteristics determine the transfection efficacy of gene delivery to human mesenchymal stem cells
AU Malakooty Poor, Elham; Baghaban Eslaminejad, Mohamadreza; Gheibi, Nematollah; Bagheri, Fatemeh; Atyabi, Fatemeh
CS Department of Stem Cells and Developmental Biology at Cell Science Research Center, Royan Institute for Stem Cell Biology and Technology, ACECR, Tehran, Iran
SO Artificial Cells, Nanomedicine, and Biotechnology (2014), 42(6), 376-384
CODEN: ACNBCI; ISSN: 2169-141X
DOI 10.3109/21691401.2013.832685
PB Informa Healthcare
DT Journal; (online computer file)
LA English
RE.CNT 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 2 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2014:1477975 CAPLUS Full-text
DN 161:424996
TI Cell structure for cell transplantation, biocompatible polymer block, and methods for producing same
IN Nakamura, Kentaro; Iwazawa, Reiko; Miyoshi, Hayato; Yamaguchi, Kazuhiro; Fushimi, Hideo
PA Fujifilm Corporation, Japan
SO PCT Int. Appl., 68pp.
CODEN: PIXXD2
DT Patent
LA Japanese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2014133081	A1	20140904	WO 2014-JP54882	20140227

W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW
RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, RU, TJ, TM

PRAI JP 2013-36942 A 20130227
RE.CNT 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 3 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2014:1285071 CAPLUS Full-text
TI Poly(ethyleneimine) functionalized carbon nanotubes as efficient nano-vector for transfecting mesenchymal stem cells
AU Moradian, Hanieh; Fasehee, Hamidreza; Keshvari, Hamid; Faghihi, Shahab

CS Tissue Engineering and Biomaterials Division, National Institute of Genetic Engineering and Biotechnology (NIGEB), Tehran, Iran
SO Colloids and Surfaces, B: Biointerfaces (2014), 122, 115-125
CODEN: CSBBEQ; ISSN: 0927-7765
DOI 10.1016/j.colsurfb.2014.06.056
PB Elsevier B.V.
DT Journal; (online computer file)
LA English
RE.CNT 96 THERE ARE 96 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 4 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2014:443635 CAPLUS Full-text
DN 160:512513
TI method for preparing stem cells from human amniotic membrane
IN Xu, Zhiguo; Gong, Bo; Xue, Fei; Yu, Yanzhi; Liu, Yongjun; Lu, Jianwei; Zhu, Delin; Zhang, Jing; Wang, Xuejun
PA Shanghai Tongze Heji Biotechnology Co., Ltd., Peop. Rep. China
SO Faming Zhuanli Shenqing, 34pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	CN 103642751	A	20140319	CN 2013-10650384	20131206
PRAI	CN 2013-10650384		20131206		

L4 ANSWER 5 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2014:70972 CAPLUS Full-text
DN 161:603569
TI Purinergic responses of chondrogenic stem cells to dynamic loading
AU Gadjanski, Ivana; Vunjak-Novakovic, Gordana
CS Department of Biomedical Engineering, Columbia University, New York, NY, USA
SO Journal of the Serbian Chemical Society (2013), 78(12), 1865-1874
CODEN: JSCSEN; ISSN: 0352-5139
DOI 10.2298/JSC131118141G
PB Serbian Chemical Society
DT Journal
LA English
RE.CNT 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 6 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:1704556 CAPLUS Full-text
DN 161:108624
TI Expression and purification of human HMGB1 A-box and identification of its induction-promoting effects on mesenchymal stem cells
AU Cao, Xiaofang; Wang, Hengxiang; He, Ziming; Wang, Fang; Yang, Yang; Guo, Zikuan
CS Hebei North University, Shijiazhuang, 075000, Peop. Rep. China
SO Zuzhi Gongcheng Yu Chongjian Waike Zazhi (2012), 8(6), 301-304
CODEN: ZGYCA3; ISSN: 1673-0364
DOI 10.3969/j.issn.1673-0364.2012.06.001
PB Shanghai Jiaotong Daxue Yixueyuan Fushu Dijiu Renmin Yiyuan
DT Journal

LA Chinese

L4 ANSWER 7 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:1519024 CAPLUS Full-text
DN 161:294916
TI Three-dimensional printing of stem cell-laden hydrogels submerged in a hydrophobic high-density fluid
AU Campos, Daniela F. Duarte; Blaeser, Andreas; Weber, Michael; Jaekel, Joerg; Neuss, Sabine; Jahnen-Dechent, Wilhelm; Fischer, Horst
CS Department of Dental Materials and Biomaterials Research, RWTH Aachen University Hospital, Aachen, D-52074, Germany
SO Biofabrication (2013), 5(1), 015003, 11 pp.
CODEN: BIOFFN; ISSN: 1758-5090
DOI 10.1088/1758-5082/5/1/015003
PB IOP Publishing Ltd.
DT Journal; (online computer file)
LA English
OSC.G 2 THERE ARE 2 CAPLUS RECORDS THAT CITE THIS RECORD (2 CITINGS)
RE.CNT 43 THERE ARE 43 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 8 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:1474779 CAPLUS Full-text
TI Exploring the roles of integrin binding and cytoskeletal reorganization during mesenchymal stem cell mechanotransduction in soft and stiff hydrogels subjected to dynamic compression
AU Steward, Andrew J.; Wagner, Diane R.; Kelly, Daniel J.
CS Trinity Centre for Bioengineering, Trinity College Dublin, Dublin, 2, Ire.
SO Journal of the Mechanical Behavior of Biomedical Materials (2014), 38, 174-182
CODEN: JMBBCP; ISSN: 1878-0180
DOI 10.1016/j.jmbbm.2013.07.020
PB Elsevier Ltd.
DT Journal; (online computer file)
LA English
RE.CNT 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 9 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:1401715 CAPLUS Full-text
DN 161:252314
TI A Comparison of Three-Dimensional Culture Systems to Evaluate In Vitro Chondrogenesis of Equine Bone Marrow-Derived Mesenchymal Stem Cells
AU Watts, Ashlee E.; Ackerman-Yost, Jeremy C.; Nixon, Alan J.
CS Comparative Orthopaedics Laboratory, Department of Clinical Sciences, Cornell University, Ithaca, NY, USA
SO Tissue Engineering, Part A (2013), 19(19-20), 2275-2283
CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2012.0479
PB Mary Ann Liebert, Inc.
DT Journal; (online computer file)
LA English
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 38 THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 10 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN

AN 2013:1401707 CAPLUS Full-text
DN 161:236065
TI Supplementation of Exogenous Adenosine 5'-Triphosphate Enhances Mechanical Properties of 3D Cell-Agarose Constructs for Cartilage Tissue Engineering
AU Gadjanski, Ivana; Yodmuang, Supansa; Spiller, Kara; Bhumiratana, Sarindr; Vunjak-Novakovic, Gordana
CS Department of Biomedical Engineering, Columbia University, New York, NY, USA
SO Tissue Engineering, Part A (2013), 19(19-20), 2188-2200
CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2012.0352
PB Mary Ann Liebert, Inc.
DT Journal; (online computer file)
LA English
OSC.G 2 THERE ARE 2 CAPLUS RECORDS THAT CITE THIS RECORD (2 CITINGS)
RE.CNT 88 THERE ARE 88 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 11 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:1286642 CAPLUS Full-text
DN 161:517543
TI Magnetic resonance contrast and biological effects of intracellular superparamagnetic iron oxides on human mesenchymal stem cells with long-term culture and hypoxic exposure
AU Rosenberg, Jens T.; Sellgren, Katelyn L.; Sachi-Kocher, Afi; Calixto Bejarano, Fabian; Baird, Michelle A.; Davidson, Michael W.; Ma, Teng; Grant, Samuel C.
CS Chemical & Biomedical Engineering, FAMU-FSU College of Engineering, The Florida State University, Tallahassee, FL, USA
SO Cytotherapy (2013), 15(3), 307-322
CODEN: CYTRF3; ISSN: 1465-3249
DOI 10.1016/j.jcyt.2012.10.013
PB Elsevier Inc.
DT Journal; (online computer file)
LA English
OSC.G 2 THERE ARE 2 CAPLUS RECORDS THAT CITE THIS RECORD (2 CITINGS)
RE.CNT 84 THERE ARE 84 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 12 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:872081 CAPLUS Full-text
DN 161:59314
TI Human mesenchymal stem cell culture on heparin-based hydrogels and the modulation of interactions by gel elasticity and heparin amount
AU Kim, Mihye; Kim, Young Ha; Tae, Giyoong
CS School of Materials Science and Engineering, Department of Nanobio Materials and Electronics, Gwangju Institute of Science and Technology, Gwangju, 500-712, S. Korea
SO Acta Biomaterialia (2013), 9(8), 7833-7844
CODEN: ABCICB; ISSN: 1742-7061
DOI 10.1016/j.actbio.2013.04.041
PB Elsevier Ltd.
DT Journal; (online computer file)
LA English
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 45 THERE ARE 45 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 13 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:795298 CAPLUS Full-text
DN 158:718473
TI Nerve implants based on a compacted biomaterial containing fibrin, a polysaccharide and stem cells
IN Garrido Gomez, Juan; Gonzalez Andrades, Miguel; Alaminos Mingorance, Miguel; Campos Munoz, Antonio; Carriel Araya, Victor Sebastian
PA Servicio Andaluz de Salud, Spain; Universidad de Granada
SO Eur. Pat. Appl., 33pp.
CODEN: EPXXDW
DT Patent
LA English
FAN.CNT 2

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI EP 2594295	A1	20130522	EP 2011-382349	20111116
R: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LI, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR, BA, ME				
WO 2013072409	A1	20130523	WO 2012-EP72709	20121115
W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, RU, TJ, TM				
PRAI EP 2011-382349	A	20111116		

RE.CNT 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 14 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:795097 CAPLUS Full-text
DN 158:718472
TI Nerve implants based on a compacted biomaterial containing fibrin, a polysaccharide and stem cells
IN Garrido Gomez, Juan; Gonzalez Andrades, Miguel; Alaminos Mingorance, Miguel; Campos Munoz, Antonio; Carriel Araya, Victor Sebastian
PA Servicio Andaluz de Salud, Spain; Universidad de Granada
SO PCT Int. Appl., 68pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 2

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2013072409	A1	20130523	WO 2012-EP72709	20121115
W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS,				

JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY,
MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM,
PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SC, SD, SE, SG, SK,
SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ,
VC, VN, ZA, ZM, ZW
RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR,
HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS,
SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML,
MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD,
SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, RU, TJ, TM
EP 2594295 A1 20130522 EP 2011-382349 20111116
R: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR,
HU, IE, IS, IT, LI, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO,
RS, SE, SI, SK, SM, TR, BA, ME
PRAI EP 2011-382349 A 20111116
RE.CNT 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 15 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:665890 CAPLUS Full-text
DN 160:213366
TI Determination and validation of reference gene stability for qPCR analysis in polysaccharide hydrogel-based 3D chondrocytes and mesenchymal stem cell cultural models
AU Chooi, Wai Hon; Zhou, Ruijie; Yeo, Suan Siong; Zhang, Feng; Wang, Dong-An
CS Division of Bioengineering, School of Chemical and Biomedical Engineering, Nanyang Technological University, Singapore, 637457, Singapore
SO Molecular Biotechnology (2013), 54(2), 623-633
CODEN: MLBOEO; ISSN: 1073-6085
DOI 10.1007/s12033-012-9604-x
PB Springer
DT Journal; (online computer file)
LA English
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 16 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:622043 CAPLUS Full-text
DN 159:155084
TI Effect of cell seeding concentration on the quality of in vitro generated tissue-engineered cartilage constructed by the technique of self-assembly
AU Jia, Jie; Yang, Shu-hua; Zhang, Yu-kun; Sun, Zhi-bo
CS Department of Orthopedics, Union Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, 430022, Peop. Rep. China
SO Zhongguo Jiaoxing Waike Zazhi (2012), 20(11), 1030-1033
CODEN: ZJWZAF; ISSN: 1005-8478
DOI 10.3977/j.issn.1005-8478.2012.11.20
PB Zhongguo Jiaoxing Waike Zazhishe
DT Journal
LA Chinese

L4 ANSWER 17 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:543612 CAPLUS Full-text
DN 158:529512
TI In vitro culture of human cartilage endplate stem cells derived from

human degenerative intervertebral disc cartilage endplate for therapy of degenerative intervertebral disc diseases
IN Huang, Bo; Wang, Hai; Zhou, Yue
PA Second Affiliated Hospital of Third Military Medical University, PLA, Peop. Rep. China
SO Faming Zhuanli Shenqing, 11pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI CN 103013910	A	20130403	CN 2012-10418889	20121026
PRAI CN 2012-10418889		20121026		

L4 ANSWER 18 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:402138 CAPLUS Full-text
DN 158:413635
TI Method for providing the synergistic growth of the cultured multiple types of cells
IN Ma, Xiaojun; Li, Nan; Yu, Weiting; Sun, Guangwei; Wang, Wei
PA Dalian Institute of Chemical Physics, Chinese Academy of Sciences, Peop. Rep. China
SO Faming Zhuanli Shenqing, 9pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI CN 102965330	A	20130313	CN 2011-10257323	20110901
CN 102965330	B	20140709		
PRAI CN 2011-10257323		20110901		

L4 ANSWER 19 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2013:70474 CAPLUS Full-text
DN 160:165991
TI Recapitulating Aspects of the Oxygen and Substrate Environment of the Damaged Joint Milieu for Stem Cell-Based Cartilage Tissue Engineering
AU O'Heireamhoin, Sven; Buckley, Conor T.; Jones, Elena; McGonagle, Dennis; Mulhall, Kevin J.; Kelly, Daniel J.
CS Trinity Centre for Bioengineering, Trinity Biomedical Sciences Institute, Trinity College Dublin, Dublin, Ire.
SO Tissue Engineering, Part C: Methods (2013), 19(2), 117-127
CODEN: TEPCAE; ISSN: 1937-3384
DOI 10.1089/ten.tec.2012.0142
PB Mary Ann Liebert, Inc.
DT Journal; (online computer file)
LA English
OSC.G 2 THERE ARE 2 CAPLUS RECORDS THAT CITE THIS RECORD (2 CITINGS)
RE.CNT 87 THERE ARE 87 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 20 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1897299 CAPLUS Full-text
DN 159:249781
TI Enhanced Adenovirus Transduction of hMSCs Using 3D Hydrogel Cell Carriers

AU Neumann, Alexander J.; Schroeder, Josh; Alini, Mauro; Archer, Charles W.; Stoddart, Martin J.
CS Musculoskeletal Regeneration Program, AO Research Institute Davos, Davos Platz, 7270, Switz.
SO Molecular Biotechnology (2013), 53(2), 207-216
CODEN: MLBOEO; ISSN: 1073-6085
DOI 10.1007/s12033-012-9522-y
PB Springer
DT Journal; (online computer file)
LA English
OSC.G 6 THERE ARE 6 CAPLUS RECORDS THAT CITE THIS RECORD (6 CITINGS)
RE.CNT 60 THERE ARE 60 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 21 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1884394 CAPLUS Full-text
DN 159:758244
TI The Effects of Cyclic Hydrostatic Pressure on Chondrogenesis and Viability of Human Adipose- and Bone Marrow-Derived Mesenchymal Stem Cells in Three-Dimensional Agarose Constructs
AU Puetzer, Jennifer; Williams, John; Gillies, Allison; Bernacki, Susan; Loboa, Elizabeth G.
CS Joint Department of Biomedical Engineering, University of North Carolina at Chapel Hill and North Carolina State University, Raleigh, NC, USA
SO Tissue Engineering, Part A (2013), 19(1-2), 299-306
CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2012.0015
PB Mary Ann Liebert, Inc.
DT Journal; (online computer file)
LA English
OSC.G 4 THERE ARE 4 CAPLUS RECORDS THAT CITE THIS RECORD (4 CITINGS)
RE.CNT 32 THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 22 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1806456 CAPLUS Full-text
DN 160:238422
TI Engineering osteochondral constructs through spatial regulation of endochondral ossification
AU Sheehy, Eamon J.; Vinardell, Tatiana; Buckley, Conor T.; Kelly, Daniel J.
CS Trinity Centre for Bioengineering, Trinity Biomedical Sciences Institute, Trinity College Dublin, Dublin, Ire.
SO Acta Biomaterialia (2013), 9(3), 5484-5492
CODEN: ABCICB; ISSN: 1742-7061
DOI 10.1016/j.actbio.2012.11.008
PB Elsevier Ltd.
DT Journal; (online computer file)
LA English
OSC.G 14 THERE ARE 14 CAPLUS RECORDS THAT CITE THIS RECORD (14 CITINGS)
RE.CNT 66 THERE ARE 66 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 23 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1623929 CAPLUS Full-text
DN 157:644593
TI Biological material suitable for the therapy of osteoarthrosis, ligament damage and for the treatment of joint disorders

IN Callegaro, Lanfranco; Zanellato, Anna Maria
PA Fidia Farmaceutici S.p.A., Italy
SO Ital., 28pp.; Chemical Indexing Equivalent to 154:95686 (WO)
CODEN: ITXXBY
DT Patent
LA Italian
FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	IT 1394570	B1	20120705	IT 2009-MI1171	20090702
	CA 2763945	A1	20110106	CA 2010-2763945	20100629
	WO 2011000820	A2	20110106	WO 2010-EP59183	20100629
	WO 2011000820	A3	20110407		
	W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
	RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AP, EA, EP, OA				
	EP 2448606	A2	20120509	EP 2010-729841	20100629
	EP 2448606	B1	20130515		
	R: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LI, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR				
	CN 102470190	A	20120523	CN 2010-80026688	20100629
	PT 2448606	E	20130712	PT 2010-729841	20100629
	ES 2421300	T3	20130830	ES 2010-729841	20100629
	RU 2529803	C2	20140927	RU 2012-103465	20100629
	US 20120114609	A1	20120510	US 2012-13380971	20120104
	US 8771672	B2	20140708		
	IN 2012CN00865	A	20130329	IN 2012-CN865	20120125
	HK 1165336	A1	20130906	HK 2012-106123	20120621
PRAI	IT 2009-MI1171	A	20090702		
	WO 2010-EP59183	W	20100629		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT

L4 ANSWER 24 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1443673 CAPLUS Full-text
DN 159:336832
TI Sequential differentiation of mesenchymal stem cells in an agarose scaffold promotes a physis-like zonal alignment of chondrocytes
AU Schmitt, Jacqueline Frida; Hua, See Kwee; Zheng, Yang; Po, James Hui Hoi; Hin, Lee Eng
CS Department of Orthopaedic Surgery, Yong Loo Lin School of Medicine, National University of Singapore, Singapore, 119260, Singapore
SO Journal of Orthopaedic Research (2012), 30(11), 1753-1759
CODEN: JOREDR; ISSN: 0736-0266
DOI 10.1002/jor.22123
PB John Wiley & Sons, Inc.
DT Journal
LA English

OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 25 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1160023 CAPLUS Full-text
DN 157:352298
TI Methods for producing and isolating organ-specific stem cells from trophoblast-encased matrix
IN Gu, Yansong
PA Empire Technology Development LLC, USA
SO PCT Int. Appl., 36pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2012105979	A1	20120809	WO 2011-US23591	20110203
W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
US 20120202261	A1	20120809	US 2011-13257949	20110920
PRAI WO 2011-US23591	W	20110203		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
RE.CNT 6 THERE ARE 6 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 26 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:1070689 CAPLUS Full-text
DN 159:129362
TI Conditioned Media from Mesenchymal Stem Cells Enhanced Bone Regeneration in Rat Calvarial Bone Defects
AU Osugi, Masashi; Katagiri, Wataru; Yoshimi, Ryoko; Inukai, Takeharu; Hibi, Hideharu; Ueda, Minoru
CS Department of Oral and Maxillofacial Surgery, Nagoya University Graduate School of Medicine, Aichi, Japan
SO Tissue Engineering, Part A (2012), 18(13-14), 1479-1489
CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2011.0325
PB Mary Ann Liebert, Inc.
DT Journal
LA English
OSC.G 22 THERE ARE 22 CAPLUS RECORDS THAT CITE THIS RECORD (22 CITINGS)
RE.CNT 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 27 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN

AN 2012:1019591 CAPLUS Full-text
DN 157:256312
TI Method for effectively inducing epidermal stem cells to teeth with fibroblast growth factor 8 and sonic hedgehog homolog
IN Wang, Bingmei; Zhang, Yanding; Huang, Yide; Chen, Yiping
PA Fujian Normal University, Peop. Rep. China
SO Faming Zhuanli Shenqing, 12pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI CN 102559582	A	20120711	CN 2012-10024734	20120206
PRAI CN 2012-10024734		20120206		

L4 ANSWER 28 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:995001 CAPLUS Full-text
DN 159:30600
TI High mesenchymal stem cell seeding densities in hyaluronic acid hydrogels produce engineered cartilage with native tissue properties
AU Erickson, Isaac E.; Kestle, Sydney R.; Zellars, Kilief H.; Farrell, Megan J.; Kim, Minwook; Burdick, Jason A.; Mauck, Robert L.
CS McKay Orthopaedic Research Laboratory, Department of Orthopaedic Surgery, 424 Stemmler Hall, The University of Pennsylvania, Philadelphia, PA, 19104, USA
SO Acta Biomaterialia (2012), 8(8), 3027-3034
CODEN: ABCICB; ISSN: 1742-7061
DOI 10.1016/j.actbio.2012.04.033
PB Elsevier Ltd.
DT Journal; (online computer file)
LA English
OSC.G 24 THERE ARE 24 CAPLUS RECORDS THAT CITE THIS RECORD (25 CITINGS)
RE.CNT 50 THERE ARE 50 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 29 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:977502 CAPLUS Full-text
DN 157:483086
TI Perpetual phenotype of self-assembled tissue-engineered cartilages transferred with lentiviral-mediated C-1-1
AU Sun, Zhibo; Yang, Shuhua; Zhang, Yukun; Zhang, Bo
CS Department of Orthopedics, Union Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, 430022, Peop. Rep. China
SO Zhonghua Shiyan Waike Zazhi (2012), 29(4), 726-728
CODEN: ZSWZAA; ISSN: 1001-9030
DOI 10.3760/cma.j.issn.1001-9030.2012.04.057
PB Zhonghua Shiyan Waike Zazhi Bianjibu
DT Journal
LA Chinese

L4 ANSWER 30 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2012:601626 CAPLUS Full-text
DN 156:529922
TI The three-dimensional tissue construction method under pseudo microgravity environment

IN Uemura, Hisakimi; Nishi, Masanobu
PA National Institute of Advanced Industrial Science and Technology AIST, Japan
SO Jpn. Kokai Tokkyo Koho, 22pp.
CODEN: JKXXAF
DT Patent
LA Japanese
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	JP 2012080874	A	20120426	JP 2011-83789	20110405
PRAI	JP 2010-206215	A	20100915		

L4 ANSWER 31 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:1440803 CAPLUS Full-text
DN 157:192077
TI Characteristics of stem cells derived from the degenerated human intervertebral disc cartilage endplate
AU Liu, Lan-Tao; Huang, Bo; Li, Chang-Qing; Zhuang, Ying; Wang, Jian; Zhou, Yue
CS Department of Orthopedics, Xinqiao Hospital, Third Military Medical University, Chongqing, Peop. Rep. China
SO PLoS One (2011), 6(10), e26285
CODEN: POLNCL; ISSN: 1932-6203
DOI 10.1371/journal.pone.0026285
PB Public Library of Science
DT Journal; (online computer file)
LA English
OSC.G 8 THERE ARE 8 CAPLUS RECORDS THAT CITE THIS RECORD (8 CITINGS)
RE.CNT 47 THERE ARE 47 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 32 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:1332892 CAPLUS Full-text
DN 157:23707
TI Chondrogenesis of bone marrow mesenchymal stem cells in co-culture system with chondrocyte
AU Sun, Ming-lin; Lv, Dan; Zhu, Lei; Zhang, Chun-qiu
CS Department of Orthopaedics, Affiliated Hospital of Medical College of Chinese People's Armed Police Forces, Tianjin, 300162, Peop. Rep. China
SO Zhonghua Guke Zazhi (2011), 31(9), 976-982
CODEN: ZGZAE6; ISSN: 0253-2352
DOI 10.3760/cma.j.issn.0253-2352.2011.09.011
PB Zhonghua Yixue Zazhishe Youxian Zeren Gongsi
DT Journal
LA Chinese

L4 ANSWER 33 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:1281647 CAPLUS Full-text
DN 157:337526
TI Chondrocytes and bone marrow-derived mesenchymal stem cells undergoing chondrogenesis in agarose hydrogels of solid and channelled architectures respond differentially to dynamic culture conditions
AU Sheehy, Eamon J.; Buckley, Conor T.; Kelly, Daniel J.
CS Trinity Centre for Bioengineering, School of Engineering, Trinity College Dublin, Ire.
SO Journal of Tissue Engineering and Regenerative Medicine (2011), 5(9),

747-758
CODEN: JTERAX; ISSN: 1932-6254
URL: http://onlinelibrary.wiley.com/doi/10.1002/term.385/pdf
DOI 10.1002/term.385
PB Wiley-Blackwell
DT Journal; (online computer file)
LA English
OSC.G 14 THERE ARE 14 CAPLUS RECORDS THAT CITE THIS RECORD (15 CITINGS)
RE.CNT 70 THERE ARE 70 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 34 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:1281640 CAPLUS Full-text
DN 157:337522
TI Composition - function relations of cartilaginous tissues engineered from chondrocytes and mesenchymal stem cells isolated from bone marrow and infrapatellar fat pad
AU Vinardell, T.; Buckley, C. T.; Thorpe, S. D.; Kelly, D. J.
CS Trinity Centre for Bioengineering, School of Engineering, Trinity College Dublin, Ire.
SO Journal of Tissue Engineering and Regenerative Medicine (2011), 5(9), 673-683
CODEN: JTERAX; ISSN: 1932-6254
URL: http://onlinelibrary.wiley.com/doi/10.1002/term.357/pdf
DOI 10.1002/term.357
PB Wiley-Blackwell
DT Journal; (online computer file)
LA English
OSC.G 16 THERE ARE 16 CAPLUS RECORDS THAT CITE THIS RECORD (16 CITINGS)
RE.CNT 66 THERE ARE 66 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 35 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:1230552 CAPLUS Full-text
DN 158:168721
TI The effect of cyclic hydrostatic pressure on the functional development of cartilaginous tissues engineered using bone marrow derived mesenchymal stem cells
AU Meyer, E. G.; Buckley, C. T.; Steward, A. J.; Kelly, D. J.
CS Trinity Centre for Bioengineering, School of Engineering, Trinity College, Dublin, Ire.
SO Journal of the Mechanical Behavior of Biomedical Materials (2011), 4(7), 1257-1265
CODEN: JMBBCP; ISSN: 1751-6161
DOI 10.1016/j.jmbbm.2011.04.012
PB Elsevier B.V.
DT Journal
LA English
OSC.G 19 THERE ARE 19 CAPLUS RECORDS THAT CITE THIS RECORD (19 CITINGS)
RE.CNT 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 36 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:910998 CAPLUS Full-text
DN 155:202127
TI Extracellular matrixes of mesenchymal stem cells derived from umbilical cord blood for cancer treatment

IN Kang, Kyung Sun
PA Kang Stem Holdings Co., Ltd., S. Korea
SO PCT Int. Appl., 24pp.
CODEN: PIXXD2
DT Patent
LA Korean
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2011087299	A2	20110721	WO 2011-KR250	20110113
	WO 2011087299	A3	20111201		
	W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
	RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
	KR 2012101695	A	20120914	KR 2012-7016665	20110113
	KR 1415809	B1	20140708		
PRAI	KR 2010-3150	A	20100113		
	WO 2011-KR250	W	20110113		

L4 ANSWER 37 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:827968 CAPLUS Full-text
DN 156:240173
TI Hydrogel for cell housing in the brain and in the spinal cord
AU Perale, Giuseppe; Giordano, Carmen; Bianco, Fabio; Rossi, Filippo; Tunesi, Marta; Daniele, Francesco; Crivelli, Filippo; Matteoli, Michela; Masi, Maurizio
CS Department of Chemistry, Materials and Chemical Engineering, Polytechnic University of Milan, Milan, Italy
SO International Journal of Artificial Organs (2011), 34(3), 295-303
CODEN: IJAODS; ISSN: 0391-3988
DOI 10.5301/IJAO.2011.6488
PB Wichtig Editore
DT Journal
LA English
OSC.G 11 THERE ARE 11 CAPLUS RECORDS THAT CITE THIS RECORD (11 CITINGS)
RE.CNT 44 THERE ARE 44 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 38 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:328661 CAPLUS Full-text
DN 154:330052
TI Separation of mesenchymal stem cells using CD146-recognizing binding molecules
IN Aicher, Wilhelm; Pilz, Gregor-Alexander; Ulrich, Christine
PA Eberhard-Karls-Universitaet Tuebingen Universitaetsklinikum, Germany
SO PCT Int. Appl., 36pp.; Chemical Indexing Equivalent to 154:278841 (DE)
CODEN: PIXXD2
DT Patent

LA German
FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2011029877	A1	20110317	WO 2010-EP63247	20100909
	W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
	RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
	DE 102009041885	A1	20110310	DE 2009-102009041885	20090909
	DE 102009041885	B4	20120322		
PRAI	DE 2009-102009041885	A	20090909		

RE.CNT 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 39 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:308473 CAPLUS Full-text
DN 156:198968
TI Chondrogenesis of bone mesenchymal stem cells with different states of chondrocyte in co-culture system
AU Sun, Ming-lin; Lu, Dan; Zhu, Lei
CS Department of Orthopaedics, Affiliated Hospital, Medical College of Chinese People's Armed Police Force, Tianjin, 300162, Peop. Rep. China
SO Zhongguo Jiaoxing Waike Zazhi (2011), 19(3), 233-237
CODEN: ZJWZAF; ISSN: 1005-8478
DOI 10.3977/j.issn.1005-8478.2011.03.16
PB Zhongguo Jiaoxing Waike Zazhishe
DT Journal
LA Chinese

L4 ANSWER 40 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2011:289003 CAPLUS Full-text
DN 154:278841
TI Separation of mesenchymal stem cells using CD146-recognizing binding molecules
IN Aicher, Wilhelm; Pilz, Gregor-Alexander; Ulrich, Christine
PA Eberhard-Karls-Universitaet Tuebingen Universitaetsklinikum, Germany
SO Ger. Offen., 13pp.; Chemical Indexing Equivalent to 154:330052 (WO)
CODEN: GWXXBX
DT Patent
LA German
FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	DE 102009041885	A1	20110310	DE 2009-102009041885	20090909
	DE 102009041885	B4	20120322		
	WO 2011029877	A1	20110317	WO 2010-EP63247	20100909
	W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG,				

ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW
RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

PRAI DE 2009-102009041885 A 20090909

RE.CNT 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 41 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN

AN 2011:17861 CAPLUS Full-text

DN 154:95686

TI Biological material suitable for the therapy of osteoarthrosis, ligament damage and for the treatment of joint disorders

IN Callegaro, Lanfranco; Zanellato, Anna Maria

PA Fidia Farmaceutici S.p.A., Italy

SO PCT Int. Appl., 21pp.; Chemical Indexing Equivalent to 157:644593 (IT)
CODEN: PIXXD2

DT Patent

LA English

FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2011000820	A2	20110106	WO 2010-EP59183	20100629
	WO 2011000820	A3	20110407		
	W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
	RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AP, EA, EP, OA				
	IT 1394570	B1	20120705	IT 2009-MI1171	20090702
	CA 2763945	A1	20110106	CA 2010-2763945	20100629
	EP 2448606	A2	20120509	EP 2010-729841	20100629
	EP 2448606	B1	20130515		
	R: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LI, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR				
	CN 102470190	A	20120523	CN 2010-80026688	20100629
	ES 2421300	T3	20130830	ES 2010-729841	20100629
	RU 2529803	C2	20140927	RU 2012-103465	20100629
	US 20120114609	A1	20120510	US 2012-13380971	20120104
	US 8771672	B2	20140708		
	IN 2012CN00865	A	20130329	IN 2012-CN865	20120125
	HK 1165336	A1	20130906	HK 2012-106123	20120621
PRAI	IT 2009-MI1171	A	20090702		

WO 2010-EP59183 W 20100629
ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)

L4 ANSWER 42 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:1436950 CAPLUS Full-text
DN 153:613401
TI Detection of changes in cell populations and mixed cell populations
IN Shamah, Steven; Laing, Lance G.; Yuzhakov, Alexander; Wagner, Rick; Abodeely, Marla; Rockney, Bennet; Schulz, Stephen C.; Padalia, Zinkal; Getman, Michael; Sandberg, Eric
PA SRU Biosystems, Inc, USA
SO PCT Int. Appl., 114 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 3

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2010132890	A1	20101118	WO 2010-US35152	20100517
	WO 2010132890	A9	20110324		
	W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
	RW: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AP, EA, EP, OA				
	CA 2761114	A1	20101118	CA 2010-2761114	20100517
	AU 2010248784	A1	20111201	AU 2010-248784	20100517
	KR 2012026551	A	20120319	KR 2011-7030075	20100517
	EP 2430448	A1	20120321	EP 2010-720106	20100517
	R: AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LI, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR				
	CN 102460171	A	20120516	CN 2010-80033792	20100517
	JP 2012526998	T	20121101	JP 2012-511070	20100517
PRAI	US 2009-61178787	P	20090515		
	US 2009-61257345	P	20091102		
	US 2010-61296099	P	20100119		
	US 2010-61315144	P	20100318		
	US 2010-61323070	P	20100412		
	WO 2010-US35152	W	20100517		

RE.CNT 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 43 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:1085472 CAPLUS Full-text
DN 155:611055
TI Matrix compositions and the development of breast acini and ducts in 3D cultures
AU Swamydas, Muthulekha; Eddy, Jill M.; Burg, Karen J. L.; Dreau, Didier

CS Cellular and Molecular Biology Division, Department of Biology, University of North Carolina, Charlotte, NC, 28223, USA
SO In Vitro Cellular & Developmental Biology: Animal (2010), 46(8), 673-684
CODEN: IVCAED; ISSN: 1071-2690
DOI 10.1007/s11626-010-9323-1
PB Springer
DT Journal; (online computer file)
LA English
OSC.G 4 THERE ARE 4 CAPLUS RECORDS THAT CITE THIS RECORD (4 CITINGS)
RE.CNT 53 THERE ARE 53 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 44 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:1071999 CAPLUS Full-text
DN 154:582932
TI Evaluation of the Complex Transcriptional Topography of Mesenchymal Stem Cell Chondrogenesis for Cartilage Tissue Engineering
AU Huang, Alice H.; Stein, Ashley; Mauck, Robert L.
CS McKay Orthopaedic Research Laboratory, Department of Orthopaedic Surgery, University of Pennsylvania, Philadelphia, PA, USA
SO Tissue Engineering, Part A (2010), 16(9), 2699-2708
CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2010.0042
PB Mary Ann Liebert, Inc.
DT Journal
LA English
OSC.G 26 THERE ARE 26 CAPLUS RECORDS THAT CITE THIS RECORD (26 CITINGS)
RE.CNT 40 THERE ARE 40 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 45 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:736014 CAPLUS Full-text
DN 153:126548
TI Bone-cartilage composite implant activated by two genes inducing differentiation of stem cells to chondrocytes and osteoblasts and its preparation method and application in combined repair of bone and cartilage at joint
IN Zhang, Junfeng; Chen, Jiangning; Diao, Huajia; Chen, Huan; Li, Pei
PA Nanjing University, Peop. Rep. China
SO Faming Zhuanli Shenqing, 9pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	CN 101721748	A	20100609	CN 2009-10234615	20091125
	CN 101721748	B	20130410		
PRAI	CN 2009-10234615		20091125		

OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)

L4 ANSWER 46 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:728805 CAPLUS Full-text
DN 154:226350
TI In vitro study of micro-dystrophin gene-modified mesenchymal stem cells
AU Zhao, Daidl; Lian, Zhiyun; Liu, Libin; Luo, Li; Li, Huiying; Liu, Ju; Zhou, Hongyu

CS Department of Neurology, West China Hospital, Sichuan University, Chengdu, Sichuan Province, 610041, Peop. Rep. China
SO Neural Regeneration Research (2010), 5(7), 496-501
CODEN: NRREBM; ISSN: 1673-5374
DOI 10.3969/j.issn.1673-5374.2010.07.003
PB Publishing House of Neural Regeneration Research
DT Journal
LA English
RE.CNT 32 THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 47 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:607631 CAPLUS Full-text
DN 153:198842
TI Isolation of human amniotic mesenchymal stem cells and its differentiation potential
AU Piao, Zhengfu; Ding, Shuqin; Zhang, Haiyan; Kobayashi, Mamoru; Kamo, Isao; Sakuragawa, Norio; Li, Ning
CS Institute of Hepatitis, Beijing Youan Hospital, Capital Medical University, Beijing, 100069, Peop. Rep. China
SO Shengwu Yixue Gongcheng Yu Linchuang (2010), 14(1), 15-19, C2
CODEN: SYGYAS; ISSN: 1009-7090
DOI 10.3969/j.issn.1009-7090.2010.01.004
PB Shengwu Yixue Gongcheng Yu Linchuang Bianjibu
DT Journal
LA Chinese

L4 ANSWER 48 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2010:413550 CAPLUS Full-text
DN 154:302581
TI Transfection of mouse bone marrow mesenchymal stem cells with Lipofectamine-mediated cytosine deaminase genes
AU Song, Fei; Xing, Qi; Ji, Guangchun; Ma, Yufang; Ma, Xuehu
CS College of Environmental Life Science, Dalian University of Technology, Dalian, Liaoning Province, 116027, Peop. Rep. China
SO Zhongguo Zuzhi Gongcheng Yanjiu Yu Linchuang Kangfu (2009), 13(49), 9775-9778
CODEN: ZZGYAA; ISSN: 1673-8225
DOI 10.3969/j.issn.1673-8225.2009.49.037
PB Zhongguo Zuzhi Gongcheng Yanjiu Yu Linchuang Kangfu Zazhishe
DT Journal
LA English
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 49 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:1591994 CAPLUS Full-text
DN 152:542211
TI Transient exposure to transforming growth factor beta 3 improves the mechanical properties of mesenchymal stem cell-laden cartilage constructs in a density-dependent manner
AU Huang, Alice H.; Stein, Ashley; Tuan, Rocky S.; Mauck, Robert L.
CS McKay Orthopaedic Research Laboratory, Department of Orthopaedic Surgery, Department of Bioengineering, University of Pennsylvania, Philadelphia, PA, USA
SO Tissue Engineering, Part A (2009), 15(11), 3461-3472

CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2009.0198
PB Mary Ann Liebert, Inc.
DT Journal
LA English
OSC.G 51 THERE ARE 51 CAPLUS RECORDS THAT CITE THIS RECORD (51 CITINGS)
RE.CNT 58 THERE ARE 58 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 50 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:1569892 CAPLUS Full-text
DN 152:83300
TI Solutions for tissue engineering and methods of use
IN Hopkins, Richard
PA The Children's Mercy Hospital, USA
SO PCT Int. Appl., 49pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2009152384	A1	20091217	WO 2009-US47115	20090611
W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
AU 2009257400	A1	20091217	AU 2009-257400	20090611
AU 2009257400	B2	20140501		
CA 2727625	A1	20091217	CA 2009-2727625	20090611
US 20100035344	A1	20100211	US 2009-483196	20090611
EP 2300495	A1	20110330	EP 2009-763672	20090611
R: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LI, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR, AL, BA, RS				
AU 2014208230	A1	20140821	AU 2014-208230	20140730
AU 2014208231	A1	20140821	AU 2014-208231	20140730
PRAI US 2008-61060790	P	20080611		
US 2008-61060796	P	20080611		
AU 2009-257400	A3	20090611		
WO 2009-US47115	W	20090611		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 2 THERE ARE 2 CAPLUS RECORDS THAT CITE THIS RECORD (2 CITINGS)
RE.CNT 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 51 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:1540543 CAPLUS Full-text
DN 152:30639

TI Method for preserving proliferation and differentiation potential of undifferentiated cells
IN Huang, Lynn L.H.
PA National Cheng Kung University, Taiwan
SO U.S. Pat. Appl. Publ., 13 pp.
CODEN: USXXCO
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 20090305415	A1	20091210	US 2008-155487	20080605
PRAI US 2008-155487		20080605		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 2 THERE ARE 2 CAPLUS RECORDS THAT CITE THIS RECORD (2 CITINGS)

L4 ANSWER 52 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:1470998 CAPLUS Full-text
DN 151:577405
TI Compositions and methods for generating musculoskeletal tissue
IN Apple, Aliza Hanna; Lotz, Jeffrey Charles; Schneider, Richard Alan
PA University of California, USA
SO PCT Int. Appl., 60pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2009142770	A2	20091126	WO 2009-US3189	20090522
WO 2009142770	A3	20100114		
W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW			
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AP, EA, EP, OA			
US 20110177132	A1	20110721	US 2011-993668	20110407
US 8603819	B2	20131210		
PRAI US 2008-61055834	P	20080523		
WO 2009-US3189	W	20090522		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 3 THERE ARE 3 CAPLUS RECORDS THAT CITE THIS RECORD (3 CITINGS)

L4 ANSWER 53 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:1229124 CAPLUS Full-text
DN 153:227320
TI Dynamic Compression Stimulates Proteoglycan Synthesis by Mesenchymal Stem Cells in the Absence of Chondrogenic Cytokines
AU Kisiday, John D.; Frisbie, David D.; McIlwraith, C. Wayne; Grodzinsky, Alan J.

CS Orthopaedic Research Center, Department of Clinical Science, Colorado State University, Fort Collins, CO, USA
SO Tissue Engineering, Part A (2009), 15(10), 2817-2824
CODEN: TEPAB9; ISSN: 1937-3341
DOI 10.1089/ten.tea.2008.0357
PB Mary Ann Liebert, Inc.
DT Journal
LA English
OSC.G 25 THERE ARE 25 CAPLUS RECORDS THAT CITE THIS RECORD (25 CITINGS)
RE.CNT 45 THERE ARE 45 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 54 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:239266 CAPLUS Full-text
DN 150:268177
TI A method for improvement of differentiation of mesenchymal stem cells using a double-structured tissue implant
IN Shortkroff, Sonya; Khoury, Joseph; Tarrant, Laurence J. B.; Claesson, Hans P. I.; Smith, Robert Lane
PA Histogenics Corporation, USA
SO PCT Int. Appl., 80 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 5

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2009026392	A1	20090226	WO 2008-US73762	20080820
W: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW				
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
US 20090012627	A1	20090108	US 2007-894124	20070820
US 8685107	B2	20140401		
AU 2008288882	A1	20090226	AU 2008-288882	20080820
AU 2008288882	B2	20140109		
CA 2696486	A1	20090226	CA 2008-2696486	20080820
US 20090069903	A1	20090312	US 2008-195255	20080820
EP 2182887	A1	20100512	EP 2008-798300	20080820
R: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LI, LT, LU, LV, MC, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR, AL, BA, MK, RS				
PRAI US 2007-894124	A	20070820		
US 2007-60967886	P	20070906		
US 2007-60958401	P	20070703		
WO 2008-US73762	W	20080820		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 6 THERE ARE 6 CITED REFERENCES AVAILABLE FOR THIS RECORD

ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 55 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2009:174235 CAPLUS Full-text
DN 153:241628
TI Cyclic compression maintains viability and induces chondrogenesis of human mesenchymal stem cells in fibrin gel scaffolds
AU Pelaez, Daniel; Huang, Chun-Yuh Charles; Cheung, Herman S.
CS Research Service and Geriatrics Research, Education, and Clinical Center, Veterans Affairs Medical Center, Miami, FL, USA
SO Stem Cells and Development (2009), 18(1), 93-102
CODEN: SCDTAE; ISSN: 1547-3287
DOI 10.1089/scd.2008.0030
PB Mary Ann Liebert, Inc.
DT Journal
LA English
OSC.G 33 THERE ARE 33 CAPLUS RECORDS THAT CITE THIS RECORD (33 CITINGS)
RE.CNT 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 56 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2008:1387546 CAPLUS Full-text
DN 149:571013
TI Dynamic compression can inhibit chondrogenesis of mesenchymal stem cells
AU Thorpe, S. D.; Buckley, C. T.; Vinardell, T.; O'Brien, F. J.; Campbell, V. A.; Kelly, D. J.
CS Trinity Centre for Bioengineering, School of Engineering, Trinity College, Dublin, Ire.
SO Biochemical and Biophysical Research Communications (2008), 377(2), 458-462
CODEN: BBRCA9; ISSN: 0006-291X
DOI 10.1016/j.bbrc.2008.09.154
PB Elsevier Inc.
DT Journal
LA English
OSC.G 36 THERE ARE 36 CAPLUS RECORDS THAT CITE THIS RECORD (36 CITINGS)
RE.CNT 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 57 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2008:1164989 CAPLUS Full-text
DN 149:409495
TI Osteogenic differentiation of mesenchymal stem cells in defined protein beads
AU Lund, Amanda W.; Bush, Jeff A.; Plopper, George E.; Stegemann, Jan P.
CS Department of Biology, Rensselaer Polytechnic Institute, Troy, NY, 12180, USA
SO Journal of Biomedical Materials Research, Part B: Applied Biomaterials (2008), 87B(1), 213-221
CODEN: JBMRGL; ISSN: 1552-4973
DOI 10.1002/jbm.b.31098
PB John Wiley & Sons, Inc.
DT Journal
LA English
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)
RE.CNT 41 THERE ARE 41 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 58 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2008:343909 CAPLUS Full-text
DN 148:523432
TI Evaluation of adult equine bone marrow- and adipose-derived progenitor cell chondrogenesis in hydrogel cultures
AU Kisiday, John D.; Kopesky, Paul W.; Evans, Christopher H.; Grodzinsky, Alan J.; McIlwraith, C. Wayne; Frisbie, David D.
CS Orthopaedic Research Center, Department of Clinical Science, Colorado State University, Fort Collins, CO, 80523, USA
SO Journal of Orthopaedic Research (2008), 26(3), 322-331
CODEN: JOREDR; ISSN: 0736-0266
DOI 10.1002/jor.20508
PB John Wiley & Sons, Inc.
DT Journal
LA English
OSC.G 63 THERE ARE 63 CAPLUS RECORDS THAT CITE THIS RECORD (63 CITINGS)
RE.CNT 52 THERE ARE 52 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 59 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2007:1486332 CAPLUS Full-text
DN 148:299190
TI Effect of caspase-3 inhibitor on anoikis of mesenchymal stem cells of rats in vitro
AU Feng, Jianjun; Yang, Shuhua; Xu, Liang; Tian, Hongtao
CS Affiliated Union Hospital, Huazhong University of Science and Technology, Wuhan, Hubei Province, 430022, Peop. Rep. China
SO Zhongguo Linchuang Kangfu (2006), 10(41), 7-9
CODEN: ZLKHAH; ISSN: 1671-5926
PB Zhongguo Linchuang Kangfu Zazhishe
DT Journal
LA Chinese

L4 ANSWER 60 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2007:1364220 CAPLUS Full-text
DN 148:24846
TI Treatment of disc degenerative disease using cells able to increase angiogenesis alone or in combination with growth factors or a matrix and compositions for same
IN Ichim, Thomas E.
PA Medistem Laboratories, Inc., USA
SO PCT Int. Appl., 76pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2007136673	A2	20071129	WO 2007-US11778	20070518
	WO 2007136673	A3	20080320		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SV, SY, TJ, TM, TN, TR,

TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AP, EA, EP, OA

	US 20100008992	A1	20100114	US 2009-301597	20090930
PRAI	US 2006-60801957	P	20060519		
	WO 2007-US11778	W	20070518		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 1 THERE ARE 1 CAPLUS RECORDS THAT CITE THIS RECORD (1 CITINGS)

L4 ANSWER 61 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2007:1029696 CAPLUS Full-text
DN 147:360025
TI Isolation and identification of mesenchymal stem cells using Frizzled-9
IN Buehring, Hans-Joerg
PA Eberhard-Karls-Universitaet Tuebingen Universitaetsklinikum, Germany
SO Ger. Offen., 21pp.
CODEN: GWXXBX
DT Patent
LA German
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	DE 102006011911	A1	20070913	DE 2006-102006011911	20060308
	DE 102006011911	B4	20091126		
PRAI	DE 2006-102006011911		20060308		

RE.CNT 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 62 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2007:860469 CAPLUS Full-text
DN 147:336213
TI Role of caspase-3 inhibitor in induced anoikis of mesenchymal stem cells in vitro
AU Feng, Jianjun; Yang, Shuhua; Xu, Liang; Tian, Hongtao; Sun, Li; Tang, Xin
CS Department of Orthopaedics, Union Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, 430022, Peop. Rep. China
SO Journal of Huazhong University of Science and Technology, Medical Sciences (2007), 27(2), 183-185
CODEN: JHUSAW; ISSN: 1672-0733
DOI 10.1007/s11596-007-0220-0
PB Huazhong University of Science and Technology
DT Journal
LA English
OSC.G 4 THERE ARE 4 CAPLUS RECORDS THAT CITE THIS RECORD (4 CITINGS)
RE.CNT 6 THERE ARE 6 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 63 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2007:661219 CAPLUS Full-text
DN 148:164789
TI Differential Effects on Messenger Ribonucleic Acid Expression by Bone Marrow-Derived Human Mesenchymal Stem Cells Seeded in Agarose Constructs Due to Ramped and Steady Applications of Cyclic Hydrostatic

Pressure
AU Finger, Allison R.; Sargent, Carolyn Y.; Dulaney, Katherine O.; Bernacki, Susan H.; Loboa, Elizabeth G.
CS Joint Department of Biomedical Engineering, University of North Carolina at Chapel Hill, NC, USA
SO Tissue Engineering (2007), 13(6), 1151-1158
CODEN: TIENFP; ISSN: 1076-3279
DOI 10.1089/ten.2006.0290
PB Mary Ann Liebert, Inc.
DT Journal
LA English
OSC.G 24 THERE ARE 24 CAPLUS RECORDS THAT CITE THIS RECORD (24 CITINGS)
RE.CNT 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 64 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2005:1206676 CAPLUS Full-text
DN 144:148945
TI Encapsulation of adult human mesenchymal stem cells within collagen-agarose microenvironments
AU Batorsky, Anna; Liao, Jiehong; Lund, Amanda W.; Plopper, George E.; Stegemann, Jan P.
CS Department of Biology, Rensselaer Polytechnic Institute, Troy, NY, USA
SO Biotechnology and Bioengineering (2005), 92(4), 492-500
CODEN: BIBIAU; ISSN: 0006-3592
DOI 10.1002/bit.20614
PB John Wiley & Sons, Inc.
DT Journal
LA English
OSC.G 56 THERE ARE 56 CAPLUS RECORDS THAT CITE THIS RECORD (56 CITINGS)
RE.CNT 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 65 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2005:1147154 CAPLUS Full-text
DN 144:188757
TI Temporal expression patterns and corresponding protein inductions of early responsible genes in rabbit bone marrow-derived mesenchymal stem cells under cyclic compressive loading
AU Huang, C.-Y. Charles; Reuben, Paul M.; Cheung, Herman S.
CS Research Service and Geriatrics Research, Education, and Clinical Center, Veterans Affairs Medical Center, Miami, FL, USA
SO Stem Cells (Durham, NC, United States) (2005), 23(8), 1113-1121
CODEN: STCEEJ; ISSN: 1066-5099
DOI 10.1634/stemcells.2004-0202
PB AlphaMed Press
DT Journal
LA English
OSC.G 47 THERE ARE 47 CAPLUS RECORDS THAT CITE THIS RECORD (47 CITINGS)
RE.CNT 58 THERE ARE 58 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 66 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2005:259816 CAPLUS Full-text
DN 142:322683
TI Biological engineering of articular structures containing both cartilage and bone

IN Mao, Jeremy Jian
PA The Board of Trustees of the University of Illinois, USA
SO PCT Int. Appl., 48 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2005025493	A2	20050324	WO 2004-US24068	20040728
	WO 2005025493	A3	20050818		
	W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VC, VN, YU, ZA, ZM, ZW				
	RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
	US 20050074877	A1	20050407	US 2004-899964	20040727
	EP 1648389	A2	20060426	EP 2004-816170	20040728
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, FI, RO, CY, TR, BG, CZ, EE, HU, PL, SK				
PRAI	US 2003-60490640	P	20030728		
	US 2004-899964	A	20040727		
	WO 2004-US24068	W	20040728		

ASSIGNMENT HISTORY FOR US PATENT AVAILABLE IN LSUS DISPLAY FORMAT
OSC.G 5 THERE ARE 5 CAPLUS RECORDS THAT CITE THIS RECORD (5 CITINGS)
RE.CNT 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 67 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2004:554395 CAPLUS Full-text
DN 141:120844
TI Effects of cyclic compressive loading on chondrogenesis of rabbit bone-marrow derived mesenchymal stem cells
AU Huang, C.-Y. Charles; Hagar, Kristen L.; Frost, Lauren E.; Sun, Yubo; Cheung, Herman S.
CS Research Service and Geriatrics Research, Education, Clinical Center, Veterans Affairs Medical Center, Miami, FL, USA
SO Stem Cells (Miamisburg, OH, United States) (2004), 22(3), 313-323
CODEN: STCEEJ; ISSN: 1066-5099
DOI 10.1634/stemcells.22-3-313
PB AlphaMed Press
DT Journal
LA English
OSC.G 138 THERE ARE 138 CAPLUS RECORDS THAT CITE THIS RECORD (139 CITINGS)
RE.CNT 45 THERE ARE 45 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 68 OF 68 CAPLUS COPYRIGHT 2015 ACS on STN
AN 2002:391853 CAPLUS Full-text
DN 136:382534
TI Engineered tissues from hair follicle derived mesenchymal stem cells

```
     and their usage for transplants and screening
IN   Daig, Rosemarie
PA   Germany
SO   PCT Int. Appl., 16 pp.
     CODEN: PIXXD2
DT   Patent
LA   German
FAN.CNT 1
     PATENT NO.          KIND  DATE        APPLICATION NO.          DATE
     ---------------     ----  --------    ---------------------    --------
PI   WO 2002040645        A2   20020523    WO 2001-EP12852          20011107
     WO 2002040645        A3   20021205
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
             CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
             GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
             LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL,
             PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG,
             US, UZ, VN, YU, ZA, ZW
         RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,
             DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF,
             BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
     DE 10056465          A1   20020718    DE 2000-10056465         20001114
     AU 2002021814        A    20020527    AU 2002-21814            20011107
     EP 1337624           A2   20030827    EP 2001-996595           20011107
     EP 1337624           B1   20060920
         R:  AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
             IE, SI, LT, LV, FI, RO, MK, CY, AL, TR
     AT 340253            T    20061015    AT 2001-996595           20011107
PRAI DE 2000-10056465     A    20001114
     WO 2001-EP12852      W    20011107
OSC.G   3     THERE ARE 3 CAPLUS RECORDS THAT CITE THIS RECORD (3 CITINGS)
RE.CNT  5     THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD
              ALL CITATIONS AVAILABLE IN THE RE FORMAT


=> FIL STNGUIDE
COST IN U.S. DOLLARS                                SINCE FILE      TOTAL
                                                      ENTRY    SESSION
FULL ESTIMATED COST                                  143.86     144.11

FILE 'STNGUIDE' ENTERED AT 11:15:33 ON 05 JAN 2015
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FILE CONTAINS CURRENT INFORMATION.
LAST RELOADED: Jan  2, 2015 (20150102/UP).

=> log y

     (FILE 'HOME' ENTERED AT 11:13:51 ON 05 JAN 2015)

     FILE 'CAPLUS' ENTERED AT 11:14:10 ON 05 JAN 2015
L1        23398 SEA FILE=CAPLUS SPE=ON  ABB=ON  PLU=ON  MESENCHYMAL STEM CELLS

L2        23398 SEA FILE=CAPLUS SPE=ON  ABB=ON  PLU=ON  L1 AND "MESENCHYMAL
                STEM"
L3          112 SEA FILE=CAPLUS SPE=ON  ABB=ON  PLU=ON  L2 AND AGAROSE
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L4            68 SEA FILE=CAPLUS SPE=ON  ABB=ON  PLU=ON  L3 AND CULTURE
                 D 1-68

     FILE 'STNGUIDE' ENTERED AT 11:15:33 ON 05 JAN 2015
COST IN U.S. DOLLARS                            SINCE FILE      TOTAL
                                                     ENTRY    SESSION
FULL ESTIMATED COST                                   0.63     144.74

STN INTERNATIONAL LOGOFF AT 11:19:28 ON 05 JAN 2015
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Search Notes	Application/Control No.	Applicant(s)/Patent Under Reexamination
	12227458	MCNIECE, IAN
	Examiner	Art Unit
	NATALIE MOSS	1653

CPC- SEARCHED		
Symbol	Date	Examiner

CPC COMBINATION SETS - SEARCHED		
Symbol	Date	Examiner

US CLASSIFICATION SEARCHED			
Class	Subclass	Date	Examiner

SEARCH NOTES		
Search Notes	Date	Examiner
See STN Search Notes	01/05/2015	NMM
See EAST Search Notes	01/05/2015	NMM

INTERFERENCE SEARCH			
US Class/ CPC Symbol	US Subclass / CPC Group	Date	Examiner

/NATALIE MOSS/ Examiner.Art Unit 1653	

Organization TC2800 Bldg/Room Jeff

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0008003330 NOV 12 2014
MAILED FROM ZIP CODE 22206

REFUSED

"RETURN TO SENDER"

IFW

Transmittal Communication on Petition	Application No. 12/227,458	Applicant/Patent Under Reexamination MCNIECE, IAN	
	Deciding Official ANDREA SMITH	Office of Petitions OPET	

--The MAILING DATE of this communication appears on the cover sheet with the correspondence address. --

(ADDITIONAL PARTY'S CORRESPONDENCE ADDRESS)

Michael Cohen
Proteonomix, Inc.
140 East Ridgewood Ave.
Suite 415
Paramus, NJ 07652

Enclosed is a copy of the latest communication from the United States Patent and Trademark Office in the above-identified Application/Patent.

UNITED STATES PATENT AND TRADEMARK OFFICE

Commissioner for Patents
United States Patent and Trademark Office
P.O. Box 1450
Alexandria, VA 22313-1450
www.uspto.gov

In re Application of	:	
Ian McNiece	:	DECISION ON PETITION
Application No. 12/227,458	:	TO WITHDRAW FROM
Filed: October 23, 2009	:	RECORD
Attorney Docket No. PROTEO-32848/US-2/PCT	:	

This is a decision on the Request to Withdraw as attorney or agent under 37 CFR § 1.36(b) filed May 24, 2013.

The request is **NOT APPROVED**.

The requested change in the correspondence address is improper.

The Office will only accept correspondence address changes to the most current address information provided for the assignee of the entire interest that properly became of record under 37 CFR 3.71, or, if no assignee of the entire interest has properly been made of record, the most current address information provided for the first named inventor.

37 CFR 3.71(c) states:

> *An assignee becomes of record either in a national patent application or a reexamination proceeding by filing a statement in compliance with § 3.73(b) that is signed by a party who is authorized to act on behalf of the assignee.*

As there is currently no Statement under 37 CFR 3.73(b) with the current assignee information of record in the present application, and since the current address information for the first named inventor was not provided, the Office cannot change the correspondence address to the address listed in the Request to Withdraw. [1]

Additionally, if the correspondence address is that of an assignee, then the assignee of the entire right, title and interest must also comply with 37 CFR 1.31.

[1] *See* USPTO Form No. PTO/SB/96.

37 CFR 1.31 states:

> *An applicant for patent may file and prosecute the applicant's own case, or the applicant may give power of attorney so as to be represented by one or more patent practitioners or joint inventors, except that a juristic entity (e.g., organizational assignee) must be represented by a patent practitioner even if the juristic entity is the applicant. The Office cannot aid in the selection of a patent practitioner.*

Further, the Office will no longer change the correspondence address to that of a new practitioner unless the Request is accompanied by a power of attorney to a new practitioner (e.g., Form PTO/SB/81).

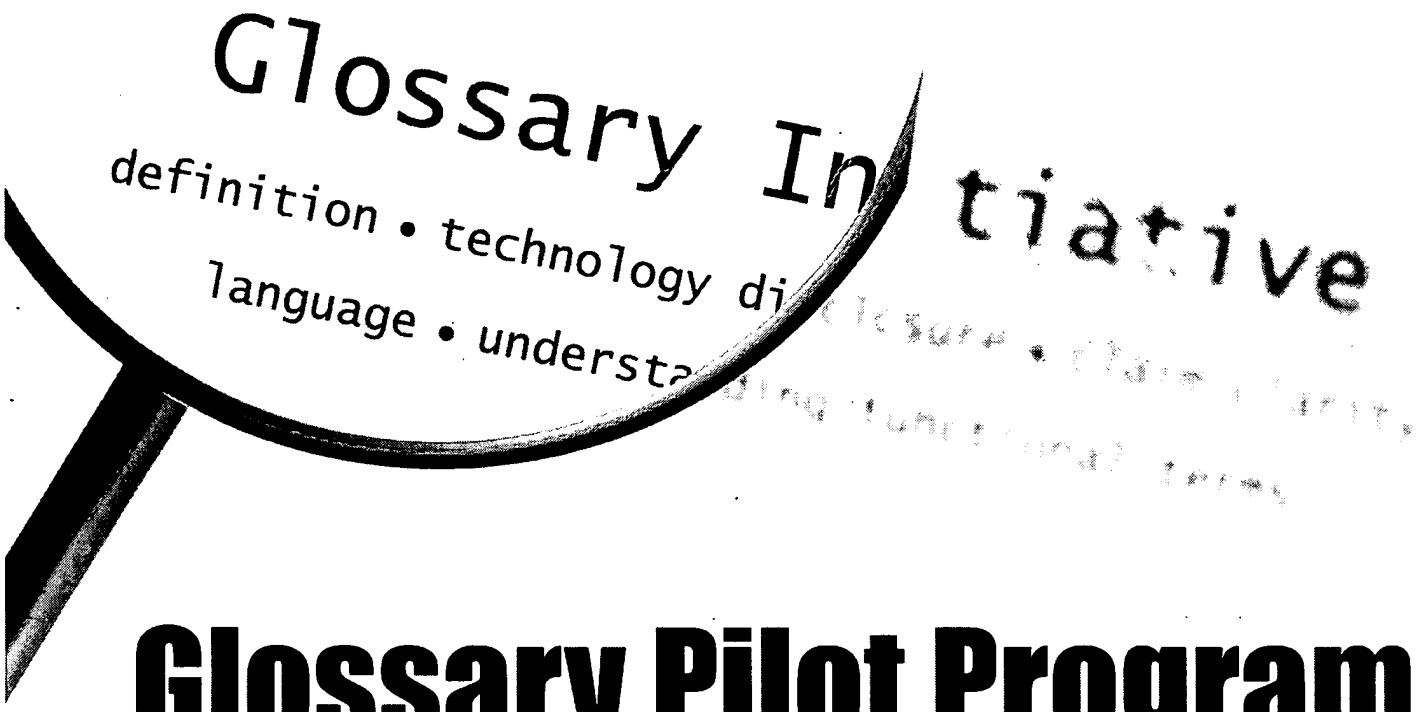
In view of the above, all future communications from the Office will continue to be directed to the above-listed address unless properly notified by the applicant.

This application file is being referred to Technology Center Art Unit 1653 for review of the response filed February 27, 2013.

Telephone inquiries concerning this decision should be directed to the undersigned at (571) 272-3226. Telephone inquiries regarding the examination of the application should be directed to the Technology Center at (571) 272-1600.

/Andrea Smith/
Andrea Smith
Paralegal Specialist
Office of Petitions

cc: Michael Cohen
Proteonomix, Inc.
140 East Ridgewood Ave.
Suite 415
Paramus, NJ 07652



Glossary Pilot Program

A NEW STRATEGY FOR IMPROVING CLAIM CLARITY USING GLOSSARIES

Still Accepting Applications!

The Glossary Pilot Program began June 2, 2014 and will run for six months or until 200 petitions are granted for participation into the program.

Benefits of Participation – Expedited Examination

Applications accepted into this pilot program will receive expedited processing up to the issuance of a first Office action.

Pilot Program Eligibility

Acceptance into the pilot program requires that the application be classified in software-related technological fields that fall under the examination jurisdiction of USPTO Technology Centers 2100, 2400, and 2600 or the Business Methods area of Technology Center 3600.

Applications must be filed electronically using EFS-Web system and include a petition to make special using Form PTO/SB/436 (no petition fee is required).

For complete information, please visit:
www.uspto.gov/patents/init_events/glossary_initiative.jsp

For questions and additional information, please contact:

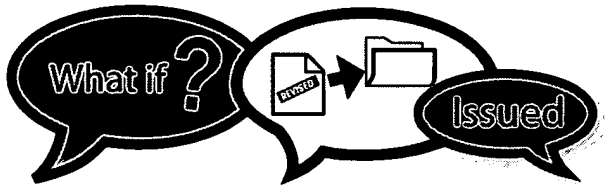
Seema Rao • Director, Technology Center 2100
571-272-0800 • E-mail: Glossary@uspto.gov

EXTENDED USPTO PATENT APPLICATION INITIATIVES

AFCP 2.0 and QPIDS have been extended **through Sept. 30, 2015**

EXTENDED

AFCP 2.0



After Final Consideration Pilot 2.0 (AFCP 2.0)

AFCP 2.0 is ideal for situations where an examiner and applicant may be close to an agreement.

Examiner Interview

Under AFCP 2.0, for an accepted submission, the examiner will schedule and conduct an interview with you to discuss the results of a limited search and consideration, if the submission does not place the application in condition for allowance. You will benefit in an interview from the additional search and consideration afforded by the pilot, even when the results do not lead to allowance.

Take advantage of a new feature in AFCP 2.0

AFCP 2.0 now incorporates a new form to more prominently point out an examiner's decision and rationale regarding the after-final response. In applications that include an AFCP 2.0 request, an examiner will complete and attach the AFCP 2.0 Decision form in the next action mailed to the applicant. This new form will start being mailed after Nov. 1, 2014.

For information on the AFCP 2.0 Pilot program, please visit: www.uspto.gov/patents/init_events/afcp.jsp
Questions regarding AFCP 2.0 can be sent to afterfinalconsiderationpilotafcp20@uspto.gov
For an application-specific issue with AFCP 2.0, contact Tariq Hafiz, Director, Technology Center 2600, at 571-272-4550

EXTENDED

QPIDS Quick Path Information Disclosure Statement

The QPIDS pilot program may eliminate the need for a Request for Continued Examination (RCE) with an information disclosure statement (IDS) filed after payment of the issue fee.

For complete information on the QPIDS program, please visit: www.uspto.gov/patents/init_events/qpids.jsp
For an application-specific issue with QPIDS, contact Remy Yucel by telephone at (571) 272-0700 or irem.yucel@uspto.gov.

Please visit the Patent Application Initiatives webpage for information on these and additional Patent Application Initiatives.
www.uspto.gov/patents/init_events/patapp-initiatives-timeline.jsp

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OPIR
Sept. 2014

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
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Alexandria, Virginia 22313-1450
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APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
12/227,458	10/23/2009	Ian Mcniece	PROTEO-32848/US-2/PCT

CONFIRMATION NO. 2285

72960
Casimir Jones, S.C.
2275 DEMING WAY, SUITE 310
MIDDLETON, WI 53562

POWER OF ATTORNEY NOTICE

OC000000071949718

Date Mailed: 11/20/2014

NOTICE REGARDING CHANGE OF POWER OF ATTORNEY

This is in response to the Power of Attorney filed 11/19/2014.

- The withdrawal as attorney in this application has been accepted. Future correspondence will be mailed to the new address of record. 37 CFR 1.33.

/eefswuser/

Office of Data Management, Application Assistance Unit (571) 272-4000, or (571) 272-4200, or 1-888-786-0101

Doc Code: PET.AUTO **Document Description: Petition automatically granted by EFS-Web**	PTO/SB/83 U.S. Patent and Trademark Office Department of Commerce

Electronic Petition Request	**REQUEST FOR WITHDRAWAL AS ATTORNEY OR AGENT AND CHANGE OF CORRESPONDENCE ADDRESS**
Application Number	12227458
Filing Date	23-Oct-2009
First Named Inventor	Ian Mcniece
Art Unit	1653
Examiner Name	LORA DRISCOLL
Attorney Docket Number	PROTEO-32848/US-2/PCT
Title	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

◉ Please withdraw me as attorney or agent for the above identified patent application and the practitioners of record associated with Customer Number: 72960

The reason(s) for this request are those described in 37 CFR:

11.116(b)(6)

Certifications

☒ I/We have given reasonable notice to the client, prior to the expiration of the response period, that the practitioner(s) intend to withdraw from employment

☒ I/We have delivered to the client or a duly authorized representative of the client all papers and property (including funds) to which the client is entitled

☒ I/We have notified the client of any responses that may be due and the time frame within which the client must respond

Change the correspondence address and direct all future correspondence to the first named inventor or assignee that has properly made itself of record pursuant to 37 CFR 3.71(for applications filed before September 16, 2012) or the applicant (for applications filed on or after September 16, 2012):

Name	MCNIECE COHEN FOUNDATION
Address	438 MINORCA AVE
City	CORAL GABLES
State	FL
Postal Code	33134
Country	US

I am authorized to sign on behalf of myself and all withdrawing practitioners.	
Signature	/Tanya A. Arenson/
Name	/Tanya A. Arenson/
Registration Number	47391

UNITED STATES PATENT AND TRADEMARK OFFICE

Commissioner for Patents
United States Patent and Trademark Office
P.O. Box 1450
Alexandria, VA 22313-1450
www.uspto.gov

Decision Date : November 19, 2014

In re Application of :

Ian Mcniece

Application No : 12227458

Filed : 23-Oct-2009

Attorney Docket No : PROTEO-32848/US-2/PCT

DECISION ON REQUEST TO WITHDRAW AS ATTORNEY/AGENTOF RECORD

This is an electronic decision on the Request to Withdraw as attorney or agent of record under 37 CFR § 1.36(b), filed November 19, 2014

The request is **APPROVED.**

The request was signed by /Tanya A. Arenson/ (registration no. 47391) on behalf of all attorneys/agents associated with Customer Number 72960 . All attorneys/agents associated with Cusotmer Number 72960 have been withdrawn.

Since there are no remaining attorneys of record, all future communications from the Office will be directed to the first named inventor or assignee that has properly made itself of record pursuant to 37 CFR 3.71(for applications filed before September 16, 2012) or the applicant (for applications filed on or after September 16, 2012), with correspondence address:

Name	MCNIECE COHEN FOUNDATION
Name2	
Address 1	438 MINORCA AVE
Address 2	
City	CORAL GABLES
State	FL
Postal Code	33134
Country	US

As a reminder, requester is required to inform the first named inventor or assignee that has properly made itself of record pursuant to 37 CFR 3.71 (for applications filed before September 16, 2012) or the applicant (for applications filed on or after September 16, 2012)of the electronically processed petition.

Telephone inquiries concerning this decision should be directed to the Patent Electronic Business Center (EBC) at 866-217-9197.

Office of Petitions

Electronic Acknowledgement Receipt

EFS ID:	20741220
Application Number:	12227458
International Application Number:	
Confirmation Number:	2285
Title of Invention:	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS
First Named Inventor/Applicant Name:	Ian Mcniece
Customer Number:	72960
Filer:	Tanya A Arenson/Diana Yang
Filer Authorized By:	Tanya A Arenson
Attorney Docket Number:	PROTEO-32848/US-2/PCT
Receipt Date:	19-NOV-2014
Filing Date:	23-OCT-2009
Time Stamp:	15:04:36
Application Type:	U.S. National Stage under 35 USC 371

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Petition automatically granted by EFS	petition-request.pdf	34825 b2fdcb698066f2716369d5e1958f65333102 29b3	no	2

Warnings:	
Information:	

Total Files Size (in bytes):	34825

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

Office of Petitions: Routing Sheet

Application No. 12/227,458

This application is being forwarded to your office for further processing. A decision has been rendered on a petition filed in this application.

- [ ] **GRANTED**
- [X] **DISMISSED**
- [ ] **DENIED**

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
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Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
12/227,458	10/23/2009	Ian Mcniece	PROTEO-32848/US-2/PCT	2285

72960 7590 11/12/2014

Casimir Jones, S.C.
2275 DEMING WAY, SUITE 310
MIDDLETON, WI 53562

EXAMINER	
DRISCOLL, LORA E BARNHART	

ART UNIT	PAPER NUMBER
1653	

MAIL DATE	DELIVERY MODE
11/12/2014	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

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United States Patent and Trademark Office
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In re Application of	:	
Ian McNiece	:	DECISION ON PETITION
Application No. 12/227,458	:	TO WITHDRAW FROM
Filed: October 23, 2009	:	RECORD
Attorney Docket No. PROTEO-32848/US-2/PCT	:	

This is a decision on the Request to Withdraw as attorney or agent under 37 CFR § 1.36(b) filed May 24, 2013.

The request is **NOT APPROVED**.

The requested change in the correspondence address is improper.

The Office will only accept correspondence address changes to the most current address information provided for the assignee of the entire interest that properly became of record under 37 CFR 3.71, or, if no assignee of the entire interest has properly been made of record, the most current address information provided for the first named inventor.

37 CFR 3.71(c) states:

> *An assignee becomes of record either in a national patent application or a reexamination proceeding by filing a statement in compliance with § 3.73(b) that is signed by a party who is authorized to act on behalf of the assignee.*

As there is currently no Statement under 37 CFR 3.73(b) with the current assignee information of record in the present application, and since the current address information for the first named inventor was not provided, the Office cannot change the correspondence address to the address listed in the Request to Withdraw. [1]

Additionally, if the correspondence address is that of an assignee, then the assignee of the entire right, title and interest must also comply with 37 CFR 1.31.

[1] *See* USPTO Form No. PTO/SB/96.

37 CFR 1.31 states:

> *An applicant for patent may file and prosecute the applicant's own case, or the applicant may give power of attorney so as to be represented by one or more patent practitioners or joint inventors, except that a juristic entity (e.g., organizational assignee) must be represented by a patent practitioner even if the juristic entity is the applicant. The Office cannot aid in the selection of a patent practitioner.*

Further, the Office will no longer change the correspondence address to that of a new practitioner unless the Request is accompanied by a power of attorney to a new practitioner (e.g., Form PTO/SB/81).

In view of the above, all future communications from the Office will continue to be directed to the above-listed address unless properly notified by the applicant.

This application file is being referred to Technology Center Art Unit 1653 for review of the response filed February 27, 2013.

Telephone inquiries concerning this decision should be directed to the undersigned at (571) 272-3226. Telephone inquiries regarding the examination of the application should be directed to the Technology Center at (571) 272-1600.

/Andrea Smith/
Andrea Smith
Paralegal Specialist
Office of Petitions

cc: Michael Cohen
Proteonomix, Inc.
140 East Ridgewood Ave.
Suite 415
Paramus, NJ 07652

Transmittal Communication on Petition	Application No. 12/227,458	Applicant/Patent Under Reexamination MCNIECE, IAN	
	Deciding Official ANDREA SMITH	Office of Petitions OPET	

--The MAILING DATE of this communication appears on the cover sheet with the correspondence address. --

(ADDITIONAL PARTY'S CORRESPONDENCE ADDRESS)

Michael Cohen
Proteonomix, Inc.
140 East Ridgewood Ave.
Suite 415
Paramus, NJ 07652

Enclosed is a copy of the latest communication from the United States Patent and Trademark Office in the above-identified Application/Patent.

Office of Petitions: Decision Count Sheet

Mailing Month: 11

Application No.: 12227458

12227458

For US serial numbers: enter number only, no slashes or commas. Ex: 10123456
For PCT: enter "51+single digit of year of filing+last 5 numbers", Ex. for PCT/US05/12345, enter 51512345

Deciding Official: SMITH, ANDREA

Count (1) - Palm Credit 12/227,458

Decision: DISMISSED

FINANCE WORK NEEDED ☐ Select Check Box for YES

DISMISSED

Decision Type: 307 - WITHDRAWAL OF ATTORNEY (37 CFR 1.36)

307

Notes: **PLS. NOTE THAT THE CORRECT FILING DATE OF THE PETITION IS 5/24/13 NOT 5/24/14. THANKS!**

Count (2)

Decision: n/a

FINANCE WORK NEEDED ☐ Select Check Box for YES

Decision Type: NONE

Notes:

Count (3)

Decision: n/a

FINANCE WORK NEEDED ☐ Select Check Box for YES

Decision Type: NONE

Notes:

Initials of Approving Official (if required)

If more than 3 decisions, attach 2nd count sheet & mark this box ☐

Printed on: *11/10/2014*

Office of Petitions: Routing Sheet

Application No. 12/227,458

This application is being forwarded to your office for further processing. A decision has been rendered on a petition filed in this application.

- [X] **GRANTED**
- [ ] **DISMISSED**
- [ ] **DENIED**

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
12/227,458	10/23/2009	Ian Mcniece	PROTEO-32848/US-2/PCT	2285

72960 7590 10/03/2014

Casimir Jones, S.C.
2275 DEMING WAY, SUITE 310
MIDDLETON, WI 53562

EXAMINER
DRISCOLL, LORA E BARNHART

ART UNIT	PAPER NUMBER
1653	

MAIL DATE	DELIVERY MODE
10/03/2014	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

UNITED STATES PATENT AND TRADEMARK OFFICE

Commissioner for Patents
United States Patent and Trademark Office
P.O. Box 1450
Alexandria, VA 22313-1450
www.uspto.gov

In re Application of	:	
Ian McNiece	:	
Application No. 12/227,458	:	DECISION ON PETITION
Filed: October 23, 2009	:	
Attorney Docket No. PROTEO-32848/US-2/PCT	:	

This application was recently referred to the Office of Petitions for a decision on the petition under 37 CFR 1.137(b)[1], filed February 27, 2013, to revive the above-identified application.

The petition is **GRANTED**.

This application became abandoned for failure to timely reply to the Restriction and/or Election Requirement mailed on January 4, 2012. A Notice of Abandonment was mailed August 7, 2012. The petition satisfies the requirements of 37 CFR 1.137(b) in that petitioner has supplied (1) the reply in the form of an election; (2) the petition fee of $945; and (3) a proper statement of unintentional delay.

The Office acknowledges $1,365 for a five (5) months extension of time filed on February 27, 2013. However, an extension of time under 37 CFR 1.136 must be filed prior to the expiration of the maximum extendable period for reply. See In re Application of S., 8 USPQ2d 1630, 1631 (Comm'r. Pats. 1988). Accordingly, since the $1,365 extension of time fee was subsequent to the maximum extendable period for reply, this fee is unnecessary and will be credited to petitioner's deposit account.

This application file is being referred to Technology Center Art Unit 1653 for further processing in accordance with this decision.

Telephone inquiries concerning this decision should be directed to the undersigned at (571) 272-3226.

/Andrea Smith/
Andrea Smith
Paralegal Specialist
Office of Petitions

[1] Since the present petition was filed prior to the rule change of December 18, 2013, it is properly treated under the unintentional standards of 37 CFR 1.137(b).

Office of Petitions: Decision Count Sheet

Mailing Month: 10

Application No.: 12227458

For US serial numbers: enter number only, no slashes or commas. Ex: 10123456
For PCT: enter "51+single digit of year of filing+last 5 numbers", Ex. for PCT/US05/12345, enter 51512345

Deciding Official: SMITH, ANDREA

Count (1) - Palm Credit 12/227,458

Decision: GRANT

FINANCE WORK NEEDED
☐ Select Check Box for YES

Decision Type: 502 - 37 CFR 1.137(b) - REVIVAL BASED ON UNINTENTIC

Notes:

Count (2)

Decision: n/a

FINANCE WORK NEEDED
☐ Select Check Box for YES

Decision Type: NONE

Notes:

Count (3)

Decision: n/a

FINANCE WORK NEEDED
☐ Select Check Box for YES

Decision Type: NONE

Notes:

Initials of Approving Official (if required)

If more than 3 decisions, attach 2nd count sheet & mark this box ☐

Printed on: *10/2/2014*

Doc Code: PET.POA.WDRW
Document Description: Petition to withdraw attorney or agent (SB83)

PTO/SB/83 (11-08)
Approved for use through 11/30/2011. OMB 0651-0035
U.S. Patent and Trademark Office, U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

REQUEST FOR WITHDRAWAL AS ATTORNEY OR AGENT AND CHANGE OF CORRESPONDENCE ADDRESS

Application Number	12/227,458
Filing Date	2009-10-23
First Named Inventor	Ian McNiece
Art Unit	1653
Examiner Name	Lora E. Barnhart Driscoll
Attorney Docket Number	PROTEO-32848/US-2/PCT

To: Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Please withdraw me as attorney or agent for the above identified patent application, and

- ☐ all the practitioners of record;
- ☐ the practitioners (with registration numbers) of record listed on the attached paper(s); or
- ☑ the practitioners of record associated with Customer Number: 72960

NOTE: The immediately preceding box should only be marked when the practitioners were appointed using the listed Customer Number.

The reason(s) for this request are those described in 37 CFR :

☐ 10.40(b)(1)	☐ 10.40(b)(2)	☐ 10.40(b)(3)	☐ 10.40(b)(4)
☐ 10.40(c)(1)(i)	☐ 10.40(c)(1)(ii)	☐ 10.40(c)(1)(iii)	☐ 10.40(c)(1)(iv)
☐ 10.40(c)(1)(v)	☑ 10.40(c)(1)(vi)	☐ 10.40(c)(2)	☐ 10.40(c)(3)
☐ 10.40(c)(4)	☐ 10.40(c)(5)	☐ 10.40(c)(6) Please explain below:	

Certifications

Check each box below that is factually correct. WARNING: If a box is left unchecked, the request will likely not be approved.

1. ☑ I/We have given reasonable notice to the client, prior to the expiration of the response period, that the practitioner(s) intend to withdraw from employment.

2. ☑ I/We have delivered to the client or a duly authorized representative of the client all papers and property (including funds) to which the client is entitled.

3. ☑ I/We have notified the client of any responses that may be due and the time frame within which the client must respond.

Please provide an explanation, if necessary:

This collection of information is required by 37 CFR 1.36. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

PTO/SB/83 (11-08)
Approved for use through 11/30/2011. OMB 0651-0035
U.S. Patent and Trademark Office, U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

REQUEST FOR WITHDRAWAL AS ATTORNEY OR AGENT AND CHANGE OF CORRESPONDENCE ADDRESS

Complete the following section only when the correspondence address will change. *Changes of address will only be accepted to an inventor or an assignee that has properly made itself of record pursuant to 37 CFR 3.71.*

Change the correspondence address and direct all future correspondence to:

A. ☐ The address of the inventor or assignee associated with Customer Number: _____

OR

B. ☑ Inventor or Assignee name	Michael Cohen, Proteonomix, Inc.

Address 140 East Ridgewood Ave., Suite 415			
City Paramus	State NJ	Zip 07652	Country US
Telephone (855) 467-7682		Email michael.cohen@proteonomix.com	

I am authorized to sign on behalf of myself and all withdrawing practitioners.

Signature	/David A. Casimir/		
Name	David A. Casimir	Registration No. 42,395	
Address 2275 Deming Way, Suite 310			
City Middleton	State WI	Zip 53562	Country US
Date	May 23, 2013	Telephone No. (608) 662-1277	

NOTE: ***Withdrawal is effective when approved rather than when received.***

This collection of information is required by 37 CFR 1.36. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (*i.e.*, GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

Electronic Acknowledgement Receipt

EFS ID:	15854631
Application Number:	12227458
International Application Number:	
Confirmation Number:	2285
Title of Invention:	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS
First Named Inventor/Applicant Name:	Ian Mcniece
Customer Number:	72960
Filer:	David Alan Casimir/Diana Yang
Filer Authorized By:	David Alan Casimir
Attorney Docket Number:	PROTEO-32848/US-2/PCT
Receipt Date:	24-MAY-2013
Filing Date:	23-OCT-2009
Time Stamp:	18:13:17
Application Type:	U.S. National Stage under 35 USC 371

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Petition to withdraw attorney or agent (SB83)	32848US2PCT_WithdrawalAttorneyAgent.pdf	802446 4ae59244cf6ae82ae44760f90e11ac3a8b4b8bc8	no	3

Warnings:	
Information:	

Total Files Size (in bytes):	802446

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
12/227,458	10/23/2009	Ian Mcniece	PROTEO-32848/US-2/PCT

CONFIRMATION NO. 2285

72960
Casimir Jones, S.C.
2275 DEMING WAY, SUITE 310
MIDDLETON, WI 53562

POA ACCEPTANCE LETTER

OC00000059857182

Date Mailed: 03/14/2013

NOTICE OF ACCEPTANCE OF POWER OF ATTORNEY

This is in response to the Power of Attorney filed 02/27/2013.

The Power of Attorney in this application is accepted. Correspondence in this application will be mailed to the above address as provided by 37 CFR 1.33.

/deelliott/

Office of Data Management, Application Assistance Unit (571) 272-4000, or (571) 272-4200, or 1-888-786-0101

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
12/227,458	10/23/2009	Ian Mcniece	68324(71699)

CONFIRMATION NO. 2285

POWER OF ATTORNEY NOTICE

49383
EDWARDS WILDMAN PALMER LLP
P.O. BOX 55874
BOSTON, MA 02205

OC00000059857156

Date Mailed: 03/14/2013

NOTICE REGARDING CHANGE OF POWER OF ATTORNEY

This is in response to the Power of Attorney filed 02/27/2013.

- The Power of Attorney to you in this application has been revoked by the assignee who has intervened as provided by 37 CFR 3.71. Future correspondence will be mailed to the new address of record(37 CFR 1.33).

/deelliott/

Office of Data Management, Application Assistance Unit (571) 272-4000, or (571) 272-4200, or 1-888-786-0101

Doc Code: PET.OP
Document Description: Petition for Review by the Office of Petitions

PTO/SB/64 (07-09)
Approved for use through 07/31/2012. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PETITION FOR REVIVAL OF AN APPLICATION FOR PATENT ABANDONED UNINTENTIONALLY UNDER 37 CFR 1.137(b)	Docket Number (Optional) PROTE-32848/US-2/PCT

First named inventor: Ian McNiece

Application No.: 12/227,458 Art Unit: 1653

Filed: October 23, 2009 Examiner: Lora E. Barnhart Driscoll

Title: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL A

Attention: Office of Petitions
Mail Stop Petition
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450
FAX (571) 273-8300

NOTE: If information or assistance is needed in completing this form, please contact Petitions Information at (571) 272-3282.

The above-identified application became abandoned for failure to file a timely and proper reply to a notice or action by the United States Patent and Trademark Office. The date of abandonment is the day after the expiration date of the period set for reply in the office notice or action plus any extensions of time actually obtained.

APPLICANT HEREBY PETITIONS FOR REVIVAL OF THIS APPLICATION

NOTE: A grantable petition requires the following items:
(1) Petition fee;
(2) Reply and/or issue fee;
(3) Terminal disclaimer with disclaimer fee - required for all utility and plant applications filed before June 8, 1995; and for all design applications; and
(4) Statement that the entire delay was unintentional

1. Petition Fee

☑ Small entity-fee $ 945 (37 CFR 1.17(m)). Application claims small entity status. See 37 CFR 1.27.

☐ Other than small entity-fee $ _____ (37 CFR 1.17(m))

2. Reply and/or fee

A. The reply and/or fee to the above-noted Office action in the form of Response to Restriction Requirement (identify type of reply):

☐ has been filed previously on _____.

☑ is enclosed herewith.

B. The issue fee and publication fee (if applicable) of $_____.

☐ has been paid previously on _____.

☐ is enclosed herewith.

This collection of information is required by 37 CFR 1.137(b). The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 1.0 hour to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Mail Stop Petition, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

3. Terminal disclaimer with disclaimer fee

☐ Since this utility/plant application was filed on or after June 8, 1995, no terminal disclaimer is required.

☐ A terminal disclaimer (and disclaimer fee (37 CFR 1.20(d)) of $_____for a small entity or $_____ for other than a small entity) disclaiming the required period of time is enclosed herewith (see PTO/SB/63).

4. STATEMENT: The entire delay in filing the required reply from the due date for the required reply until the filing of a grantable petition under 37 CFR 1.137(b) was unintentional. [NOTE: The United States Patent and Trademark Office may require additional information if there is a question as to whether either the abandonment or the delay in filing a petition under 37 CFR 1.137(b) was unintentional (MPEP 711.03(c), subsections (III)(C) and (D)).]

WARNING:

Petitioner/applicant is cautioned to avoid submitting personal information in documents filed in a patent application that may contribute to identity theft. Personal information such as social security numbers, bank account numbers, or credit card numbers (other than a check or credit card authorization form PTO-2038 submitted for payment purposes) is never required by the USPTO to support a petition or an application. If this type of personal information is included in documents submitted to the USPTO, petitioners/applicants should consider redacting such personal information from the documents before submitting them to the USPTO. Petitioner/applicant is advised that the record of a patent application is available to the public after publication of the application (unless a non-publication request in compliance with 37 CFR 1.213(a) is made in the application) or issuance of a patent. Furthermore, the record from an abandoned application may also be available to the public if the application is referenced in a published application or an issued patent (see 37 CFR 1.14). Checks and credit card authorization forms PTO-2038 submitted for payment purposes are not retained in the application file and therefore are not publicly available.

/Tanya A. Arenson/	February 27, 2013
Signature	Date
Tanya A. Arenson	47,391
Type or Printed name	Registration Number, If applicable
Casimir Jones, S.C.	608-662-1277
Address	Telephone Number
2275 Deming Way, Ste 310, Middleton, WI 53562	
Address	

Enclosures:

☑ Fee Payment

☑ Reply

☐ Terminal Disclaimer Form

☐ Additional sheets containing statements establishing unintentional delay

☐ Other: _____

CERTIFICATE OF MAILING OR TRANSMISSION [37 CFR 1.8(a)]

I hereby certify that this correspondence is being:

☐ Deposited with the United States Postal Service on the date shown below with sufficient postage as first class mail in an envelope addressed to: Mail Stop Petition, Commissioner for Patents, P. O. Box 1450, Alexandria, VA 22313-1450.

☐ Transmitted by facsimile on the date shown below to the United States Patent and Trademark Office at (571) 273-8300.

Date	Signature
	Typed or printed name of person signing certificate

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these records.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

PTO/SB/81 (01-09)
Approved for use through 11/30/2011. OMB 0651-0035
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

POWER OF ATTORNEY OR REVOCATION OF POWER OF ATTORNEY WITH A NEW POWER OF ATTORNEY AND CHANGE OF CORRESPONDENCE ADDRESS

Application Number	12/227,458
Filing Date	23-Oct-2009
First Named Inventor	Ian McNiece
Title	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER
Art Unit	1653
Examiner Name	Dirscoll, Lora E Barnhart
Attorney Docket Number	PROTEO-32848/US-2/PCT

I hereby revoke all previous powers of attorney given in the above-identified application.

☐ A Power of Attorney is submitted herewith.

OR

☒ I hereby appoint Practitioner(s) associated with the following Customer Number as my/our attorney(s) or agent(s) to prosecute the application identified above, and to transact all business in the United States Patent and Trademark Office connected therewith:

72960

OR

☐ I hereby appoint Practitioner(s) named below as my/our attorney(s) or agent(s) to prosecute the application identified above, and to transact all business in the United States Patent and Trademark Office connected therewith:

Practitioner(s) Name	Registration Number

Please recognize or change the correspondence address for the above-identified application to:

☒ The address associated with the above-mentioned Customer Number.

OR

☐ The address associated with Customer Number:

OR

☐ Firm or Individual Name					
Address					
City		State		Zip	
Country					
Telephone		Email			

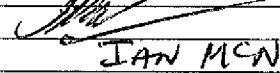
I am the:

☐ Applicant/Inventor.

OR

☒ Assignee of record of the entire interest. See 37 CFR 3.71.
Statement under 37 CFR 3.73(b) (Form PTO/SB/96) submitted herewith or filed on _____.

SIGNATURE of Applicant or Assignee of Record

Signature		Date	2/21/13
Name	IAN MCNIECE	Telephone	305-510-7057
Title and Company	Director, McNiece Cohen Foundation		

NOTE: Signatures of all the inventors or assignees of record of the entire interest or their representative(s) are required. Submit multiple forms if more than one signature is required, see below*.

☐ *Total of _____ forms are submitted.

This collection of information is required by 37 CFR 1.31, 1.32 and 1.33. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 3 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

PTO/SB/96 (07-09)
Approved for use through 07/31/2012. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

STATEMENT UNDER 37 CFR 3.73(b)

Applicant/Patent Owner: McNiece Cohen Foundation

Application No./Patent No.: 12227458 Filed/Issue Date: 10/23/2009

Titled: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

McNiece Cohen Foundation , a corporation

(Name of Assignee) (Type of Assignee, e.g., corporation, partnership, university, government agency, etc.

states that it is:

1. ☒ the assignee of the entire right, title, and interest in;

2. ☐ an assignee of less than the entire right, title, and interest in (The extent (by percentage) of its ownership interest is _____ %); or

3. ☐ the assignee of an undivided interest in the entirety of (a complete assignment from one of the joint inventors was made)

the patent application/patent identified above, by virtue of either:

A. ☐ An assignment from the inventor(s) of the patent application/patent identified above. The assignment was recorded in the United States Patent and Trademark Office at Reel _____, Frame _____, or for which a copy therefore is attached.

OR

B. ☒ A chain of title from the inventor(s), of the patent application/patent identified above, to the current assignee as follows:

1. From: Ian McNiece To: The John Hopkins University

The document was recorded in the United States Patent and Trademark Office at
Reel 023418 , Frame 0748 , or for which a copy thereof is attached.

2. From: The John Hopkins University To: Ian McNiece

The document was recorded in the United States Patent and Trademark Office at
Reel 029305 , Frame 0575 , or for which a copy thereof is attached.

3. From: Ian McNiece To: McNiece Cohen Foundation

The document was recorded in the United States Patent and Trademark Office at
Reel 029498 , Frame 0562 , or for which a copy thereof is attached.

☐ Additional documents in the chain of title are listed on a supplemental sheet(s).

☒ As required by 37 CFR 3.73(b)(1)(i), the documentary evidence of the chain of title from the original owner to the assignee was, or concurrently is being, submitted for recordation pursuant to 37 CFR 3.11.

[NOTE: A separate copy (*i.e.*, a true copy of the original assignment document(s)) must be submitted to Assignment Division in accordance with 37 CFR Part 3, to record the assignment in the records of the USPTO. See MPEP 302.08]

The undersigned (whose title is supplied below) is authorized to act on behalf of the assignee.

/Tanya A. Arenson/ February 27, 2013

Signature Date

Tanya A. Arenson Agent of Record

Printed or Typed Name Title

This collection of information is required by 37 CFR 3.73(b). The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (*i.e.*, GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:	McNiece	Conf. No:	2285
Application No:	12/227,458	Art Unit:	1653
Filed:	10/23/2009	Examiner:	Driscoll
Entitled:	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS		

RESPONSE TO RESTRICTION REQUIREMENT
MAILED JANUARY 4, 2012

EFS Web Filed
Commissioner for Patents
PO BOX 1450
Alexandria, VA 22313–1450

Examiner Driscoll:

This communication is responsive to the Office Communication mailed January 4, 2012, with a response due on or before February 4, 2012. A Petition to Revive an Unintentionally Abandoned Application and five month extension of time in which to respond to this communication is attached herewith. Applicants respectfully request reconsideration of the application in view of the following remarks.

The Commissioner is authorized by this paper to charge any fees required during the entire pendency of this application, including fees due under 37 C.F.R. §§ 1.16 and 1.17 and any extension of time fees, or credit any overpayment, to Deposit Account 50-4302, referencing Attorney Docket No. PROTE-32848/US-2/PCT. This paragraph is intended to be a CONSTRUCTIVE PETITION FOR EXTENSION OF TIME in accordance with 37 C.F.R. § 1.136(a)(3).

CLAIMS:

1. (original) A method for propagation of a non-adherent culture of mesenchymal stem cells (MSCs) comprising expanding MSCs in or on a non-adherent matrix.

2. (original) The method of claim 1, comprising encapsulation of MSCs in Matrigel™ or Hydrogel.

3. (original) The method of claim 1, comprising the cells propagated on agarose or on Teflon®.

4. (previously presented) The methods of claim 1, wherein the cells are propagated in the non-adherent culture without the use of trypsin.

5. (previously presented) The methods of claim 1, comprising mechanical manipulation of the MSCs.

6. (previously presented) The method of claim 1, further comprising a biological sample containing MSCs.

7. (original) The method of claim 6, further comprising isolating the MSCs from the biological sample containing the MSCs.

8. (original) The method of claim 7, wherein the isolated MSCs are substantially purified.

9. (previously presented) The method of claim 1, wherein the MSCs are expanded at least 2-fold, 10-fold, 100-fold, 1000-fold, 10,000-fold, or 100,000 fold.

10. (previously presented) The method of claim 1, wherein the MSCs are suitable for administration to a subject.

11. (original) The method of claim 10, wherein the subject is a human subject.

12. (previously presented) The method of claim 1 wherein the MSCs are propagated in non-adherent culture for at least a week, at least 2 weeks, at least a month, or at least 2 months.

13. (withdrawn) A method for treatment of a subject having a disease or condition susceptible to treatment with MSCs comprising administration of MSCs grown in a non-adherent culture of claim 1.

14. (withdrawn) The method of claim 13, wherein the disease or condition susceptible to treatment with MSCs is selected from the group consisting of muscle disease, neural disease, and vascular disease.

15. (withdrawn) The method of claim 13, wherein the MSCs are allogenic or autologous to the subject.

16. (withdrawn) The method of claim 13, wherein the subject is human.

17-18. (cancelled)

19. (withdrawn) A kit comprising an MSC of claim 1 and appropriate packing material.

20. (withdrawn) The kit of claim 19, further comprising reagents or supplies for propagation of MSCs under adherent or non-adherent conditions or both.

REMARKS

Applicants note that all amendments and cancellations of Claims presented herein are made without acquiescing to any of the Examiner's arguments or rejections, and solely for the purpose of expediting the patent application process in a manner consistent with the PTO's Patent Business Goals (PBG), and without waiving the right to prosecute the amended or cancelled Claims (or similar Claims) in the future.

In the Office Action mailed January 4, 2012, the Examiner required election to one of the following restriction groups: Group I, claim(s) 1-12, drawn to a method for propagating mesenchymal stem cells (MSCs) in a non-adherent matrix, e.g. MATRIGEL or hydrogel; Group II, claim(s) 1-12 drawn in part to a method for propagating mesenchymal stem cells (MSCs) on a non-adherent matrix, e.g. agarose or TEFLON; Group III, claim(s) 13-16, drawn to a method for treating a subject with MSCs; and Group IV, claim(s) 19 and 20, drawn to MSCs.

Applicants hereby elect the claims of Group II (claims 1-12, as drawn to a method for propagating mesenchymal stem cells (MSCs) on a non-adherent matrix, e.g. agarose or TEFLON) without traverse. Claims 13-16, 19 and 20 have been withdrawn without acquiescing to any of the Examiner's arguments or rejections, and solely for the purpose of expediting the patent application process in a manner consistent with the PTO's Patent Business Goals (PBG), and without waiving the right to prosecute the claims (or similar claims) in the future.

CONCLUSION

Should the Examiner believe that a telephone interview would aid in the prosecution of this application, the applicant encourages the Examiner to call the undersigned collect at (608) 662-1277.

Respectfully submitted,

Dated: February 27, 2013

/Tanya A. Arenson/
Tanya A. Arenson
Registration No. 47,391

Casimir Jones, s.c.
2275 Deming Way, Suite 310
Middleton, WI 53562

Electronic Patent Application Fee Transmittal

Application Number:	12227458
Filing Date:	23-Oct-2009
Title of Invention:	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS
First Named Inventor/Applicant Name:	Ian Mcniece
Filer:	Tanya A Arenson/Diana Yang
Attorney Docket Number:	68324(71699)

Filed as Small Entity

U.S. National Stage under 35 USC 371 Filing Fees

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Petition-revive unintent. abandoned appl	2453	1	945	945
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Extension - 5 months with $0 paid	2255	1	1365	1365
Miscellaneous:				
			Total in USD ($)	**2310**

Electronic Acknowledgement Receipt

EFS ID:	15069564
Application Number:	12227458
International Application Number:	
Confirmation Number:	2285
Title of Invention:	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS
First Named Inventor/Applicant Name:	Ian Mcniece
Customer Number:	49383
Filer:	Tanya A Arenson/Diana Yang
Filer Authorized By:	Tanya A Arenson
Attorney Docket Number:	68324(71699)
Receipt Date:	27-FEB-2013
Filing Date:	23-OCT-2009
Time Stamp:	17:57:14
Application Type:	U.S. National Stage under 35 USC 371

Payment information:

Submitted with Payment	yes
Payment Type	Deposit Account
Payment was successfully received in RAM	$2310
RAM confirmation Number	6225
Deposit Account	504302
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

Charge any Additional Fees required under 37 C.F.R. 1.492 (National application filing, search, and examination fees)

Charge any Additional Fees required under 37 C.F.R. Section 1.17 (Patent application and reexamination processing fees)

Charge any Additional Fees required under 37 C.F.R. Section 1.19 (Document supply fees)

Charge any Additional Fees required under 37 C.F.R. Section 1.20 (Post Issuance fees)

Charge any Additional Fees required under 37 C.F.R. Section 1.21 (Miscellaneous fees and charges)

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Petition for review by the PCT legal office.	32848US2PCT_PetitionReviveUnintentional.pdf	204932 824e7c8367e503be9802ab4f44c5f88a9234997f	no	3
Warnings:					
Information:					
2	Power of Attorney	32848US2PCT_POA_Executed.pdf	82818 d98e1989f9a8a7911336b0426fb47c4b5679d3c2	no	1
Warnings:					
Information:					
3	Assignee showing of ownership per 37 CFR 3.73.	32848US2PCT_373BStatement.pdf	429718 121fe8cc29343c1cdd6bda68c9024b41686c2467	no	2
Warnings:					
Information:					
4		32848US2PCT_ResponseRestriction.pdf	108904 94da3dc07b047cb2867355697f33c9587c3311e9	yes	4

Multipart Description/PDF files in .zip description

Document Description	Start	End
Response to Election / Restriction Filed	1	1
Claims	2	3
Applicant Arguments/Remarks Made in an Amendment	4	4

Warnings:					
Information:					
5	Fee Worksheet (SB06)	fee-info.pdf	32661 f6eaad3fa2f4e85b99b27f86a9ee4040155819b7	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	859033		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

Document code: WFEE

United States Patent and Trademark Office
Sales Receipt for Accounting Date: 10/02/2014

CKHLOK ADJ #00000003 Mailroom Dt: 02/27/2013
Seq No: 6225 Sales Acctg Dt: 02/28/2013 504302 12227458
02 FC : 2255 1365.00 CR

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
12/227,458	10/23/2009	Ian Mcniece	68324(71699)	2285

49383 7590 08/07/2012

EDWARDS WILDMAN PALMER LLP
P.O. BOX 55874
BOSTON, MA 02205

EXAMINER	
DRISCOLL, LORA E BARNHART	
ART UNIT	**PAPER NUMBER**
1653	
MAIL DATE	**DELIVERY MODE**
08/07/2012	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of Abandonment	Application No.	Applicant(s)
	12/227,458	MCNIECE, IAN
	Examiner	Art Unit
	Lora E. Barnhart Driscoll	1653

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address--

This application is abandoned in view of:

1. ☒ Applicant's failure to timely file a proper reply to the Office letter mailed on *03 January 2012*.
   (a) ☐ A reply was received on _____ (with a Certificate of Mailing or Transmission dated _____), which is after the expiration of the period for reply (including a total extension of time of _____ month(s)) which expired on _____.
   (b) ☐ A proposed reply was received on _____, but it does not constitute a proper reply under 37 CFR 1.113 (a) to the final rejection.
   (A proper reply under 37 CFR 1.113 to a final rejection consists only of: (1) a timely filed amendment which places the application in condition for allowance; (2) a timely filed Notice of Appeal (with appeal fee); or (3) a timely filed Request for Continued Examination (RCE) in compliance with 37 CFR 1.114).
   (c) ☐ A reply was received on _____ but it does not constitute a proper reply, or a bona fide attempt at a proper reply, to the non-final rejection. See 37 CFR 1.85(a) and 1.111. (See explanation in box 7 below).
   (d) ☒ No reply has been received.

2. ☐ Applicant's failure to timely pay the required issue fee and publication fee, if applicable, within the statutory period of three months from the mailing date of the Notice of Allowance (PTOL-85).
   (a) ☐ The issue fee and publication fee, if applicable, was received on _____ (with a Certificate of Mailing or Transmission dated _____), which is after the expiration of the statutory period for payment of the issue fee (and publication fee) set in the Notice of Allowance (PTOL-85).
   (b) ☐ The submitted fee of $_____ is insufficient. A balance of $_____ is due.
   The issue fee required by 37 CFR 1.18 is $_____. The publication fee, if required by 37 CFR 1.18(d), is $_____.
   (c) ☐ The issue fee and publication fee, if applicable, has not been received.

3. ☐ Applicant's failure to timely file corrected drawings as required by, and within the three-month period set in, the Notice of Allowability (PTO-37).
   (a) ☐ Proposed corrected drawings were received on _____ (with a Certificate of Mailing or Transmission dated _____), which is after the expiration of the period for reply.
   (b) ☐ No corrected drawings have been received.

4. ☐ The letter of express abandonment which is signed by the attorney or agent of record, the assignee of the entire interest, or all of the applicants.

5. ☐ The letter of express abandonment which is signed by an attorney or agent (acting in a representative capacity under 37 CFR 1.34(a)) upon the filing of a continuing application.

6. ☐ The decision by the Board of Patent Appeals and Interference rendered on _____ and because the period for seeking court review of the decision has expired and there are no allowed claims.

7. ☒ The reason(s) below:

Applicants submitted an amendment to the specification on 4/11/12, but that letter did not reply to the restriction.

/Lora E Barnhart Driscoll/
Primary Examiner, Art Unit 1653

Petitions to revive under 37 CFR 1.137(a) or (b), or requests to withdraw the holding of abandonment under 37 CFR 1.181, should be promptly filed to minimize any negative effects on patent term.

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants:	Ian Mcniece	Confirmation No.	2285
Application No.:	12/227,458	Attorney Docket No.:	68324(71699)
Filed:	October 23, 2009		

Title: **Method of Growth of Mesenchymal Cells Under Non-Adherent Conditions for Clinical Applications**

AMENDMENT

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

Please enter the following Amendment before examining this application.

Amendments to the Specification begin on page 2 of this paper.

Remarks begin on page 3 of this paper.

IN THE SPECIFICATION

On page 1, paragraph [0002], kindly replace the paragraph with the following replacement paragraph:

GOVERNMENT SUPPORT

[0002] This invention was made with government support under CA088878 awarded by the National Institute of Health. The U.S. Government has certain rights in the invention.

Remarks

This Amendment is to correct the government support clause in the originally filed application. The Amendment does not add new matter.

Respectfully submitted,

Date: April 11, 2012

/Guido J. Galvez/

Guido J. Galvez
Registration No. 52933
ph. 410-516-8300
fx. 410-516-0252

Johns Hopkins Technology Transfer
100 N. Charles Street, 5th Floor
Baltimore, MD 21201

Electronic Acknowledgement Receipt

EFS ID:	12518079
Application Number:	12227458
International Application Number:	
Confirmation Number:	2285
Title of Invention:	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS
First Named Inventor/Applicant Name:	Ian Mcniece
Customer Number:	49383
Filer:	Guido Joel Galvez/Cheryl Oliver
Filer Authorized By:	Guido Joel Galvez
Attorney Docket Number:	68324(71699)
Receipt Date:	11-APR-2012
Filing Date:	23-OCT-2009
Time Stamp:	15:06:08
Application Type:	U.S. National Stage under 35 USC 371

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Preliminary Amendment	P04683-03_Amendment.pdf	64572 3a11fd868c48b411b6b67ff61a2319d1e92d f54f	no	3

Warnings:	
Information:	

Total Files Size (in bytes):	64572

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
12/227,458	10/23/2009	Ian Mcniece	68324(71699)	2285

49383 7590 01/04/2012

EDWARDS WILDMAN PALMER LLP
P.O. BOX 55874
BOSTON, MA 02205

EXAMINER	
DRISCOLL, LORA E BARNHART	
ART UNIT	**PAPER NUMBER**
1653	

MAIL DATE	DELIVERY MODE
01/04/2012	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	12/227,458	MCNIECE, IAN	
	Examiner	Art Unit	
	Lora E. Barnhart Driscoll	1653	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *1* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☐ Responsive to communication(s) filed on _____.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

5)☒ Claim(s) *1-16,19 and 20* is/are pending in the application.

5a) Of the above claim(s) _____ is/are withdrawn from consideration.

6)☐ Claim(s) _____ is/are allowed.

7)☐ Claim(s) _____ is/are rejected.

8)☐ Claim(s) _____ is/are objected to.

9)☒ Claim(s) *1-16,19 and 20* are subject to restriction and/or election requirement.

Application Papers

10)☐ The specification is objected to by the Examiner.

11)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

12)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Applicants filed a preliminary amendment on 11/17/08 with the application.

Claims 1-16, 19, and 20 are currently pending.

Election/Restrictions

Restriction is required under 35 U.S.C. 121 and 372.

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 accordance with 37 CFR 1.499, applicant is required, in reply to this action, to elect a single invention to which the claims must be restricted.

Group I, claims 1-12, drawn to a method for propagating mesenchymal stem cells (MSCs) in a non-adherent matrix, e.g. MATRIGEL or hydrogel.

Group I, claims 1-12, drawn in part to a method for propagating mesenchymal stem cells (MSCs) on a non-adherent matrix, e.g. agarose or TEFLON.

Group III, claims 13-16, drawn to a method for treating a subject with MSCs.

Group IV, claims 19 and 20, drawn to MSCs.

The groups of inventions listed above 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 expression “special technical feature” refers to those features that define a contribution which each of the claimed inventions, considered as a whole, makes over the prior art. Thus, a feature found in the prior art cannot be considered to be a special technical feature.

In this case, MSCs were known in the art at the time of the invention. Kato et al. (2005, U.S. Patent Application Publication 2005/0013804; reference AC on 11/17/08 IDS) teach MSCs. (Paragraph 33.) Therefore, MSCs are not a special technical feature.

Election of Species

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.

The species are as follows:

Conditions in Group III: (a) muscle disease, (b) neural disease, and (c) vascular disease, as in claim 14; elect ONE if Group III is elected.

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 because they are not clearly art-accepted substitutes for each other. The conditions affect nonoverlapping patient sets and are characterized by different pathologies and symptoms.

Applicant is required, in reply to this action, to elect a single species to which the claims shall be restricted if no generic claim is finally held to be allowable. The reply must also identify the claims readable on the elected species, including any claims subsequently added. An argument that a claim is allowable or that all claims are generic is considered non-responsive unless accompanied by an election.

Upon the allowance of a generic claim, applicant will be entitled to consideration of claims to additional species which are written in dependent form or otherwise require

all the limitations of an allowed generic claim. Currently, the following claim(s) are generic: 1, 13, and 19.

Applicant is advised that the reply to this requirement to be complete must include (i) an election of a species or invention to be examined even though the requirement may be traversed (37 CFR 1.143) and (ii) identification of the claims encompassing the elected invention.

The election of an invention or species may be made with or without traverse. To preserve a right to petition, the election must be made with traverse. If the reply does not distinctly and specifically point out supposed errors in the restriction requirement, the election shall be treated as an election without traverse. Traversal must be presented at the time of election in order to be considered timely. Failure to timely traverse the requirement will result in the loss of right to petition under 37 CFR 1.144. If claims are added after the election, applicant must indicate which of these claims are readable on the elected invention or species.

Should applicant traverse on the ground that the inventions have unity of invention (37 CFR 1.475(a)), applicant must provide reasons in support thereof. Applicant may submit evidence or identify such evidence now of record showing the inventions to be obvious variants or clearly admit on the record that this is the case. Where such evidence or admission is provided by applicant, if the examiner finds one of the inventions unpatentable over the prior art, the evidence or admission may be used in a rejection under 35 U.S.C. 103(a) of the other invention.

Applicant is reminded that upon the cancellation of claims to a non-elected invention, the inventorship must be amended in compliance with 37 CFR 1.48(b) if one or more of the currently named inventors is no longer an inventor of at least one claim remaining in the application. Any amendment of inventorship must be accompanied by a request under 37 CFR 1.48(b) and by the fee required under 37 CFR 1.17(i).

The examiner has required restriction between product and process claims. Where applicant elects claims directed to the product, and the product claims are subsequently found allowable, withdrawn process claims that depend from or otherwise require all the limitations of the allowable product claim will be considered for rejoinder. All claims directed to a nonelected process invention must require all the limitations of an allowable product claim for that process invention to be rejoined.

In the event of rejoinder, the requirement for restriction between the product claims and the rejoined process claims will be withdrawn, and the rejoined process claims will be fully examined for patentability in accordance with 37 CFR 1.104. Thus, to be allowable, the rejoined claims must meet all criteria for patentability including the requirements of 35 U.S.C. 101, 102, 103 and 112. Until all claims to the elected product are found allowable, an otherwise proper restriction requirement between product claims and process claims may be maintained. Withdrawn process claims that are not commensurate in scope with an allowable product claim will not be rejoined. See MPEP § 821.04(b). Additionally, in order to retain the right to rejoinder in accordance with the above policy, applicant is advised that the process claims should be amended during prosecution to require the limitations of the product claims. **Failure to do so may result**

in a loss of the right to rejoinder. Further, note that the prohibition against double patenting rejections of 35 U.S.C. 121 does not apply where the restriction requirement is withdrawn by the examiner before the patent issues. See MPEP § 804.01.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Lora E. Barnhart Driscoll, whose telephone number is (571)272-1928. The examiner can normally be reached on Monday-Thursday, 9:00am - 5:30pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Sue X. Liu, can be reached on 571-272-5539. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/Lora E Barnhart Driscoll/
Primary Examiner, Art Unit 1653

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APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
12/227,458	10/23/2009	Ian Mcniece	68324(71699)

CONFIRMATION NO. 2285

49383
EDWARDS ANGELL PALMER & DODGE LLP
P.O. BOX 55874
BOSTON, MA 02205

PUBLICATION NOTICE

OC000000040283961

Title:METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Publication No.US-2010-0047211-A1
Publication Date:02/25/2010

NOTICE OF PUBLICATION OF APPLICATION

The above-identified application will be electronically published as a patent application publication pursuant to 37 CFR 1.211, et seq. The patent application publication number and publication date are set forth above.

The publication may be accessed through the USPTO's publically available Searchable Databases via the Internet at www.uspto.gov. The direct link to access the publication is currently http://www.uspto.gov/patft/.

The publication process established by the Office does not provide for mailing a copy of the publication to applicant. A copy of the publication may be obtained from the Office upon payment of the appropriate fee set forth in 37 CFR 1.19(a)(1). Orders for copies of patent application publications are handled by the USPTO's Office of Public Records. The Office of Public Records can be reached by telephone at (703) 308-9726 or (800) 972-6382, by facsimile at (703) 305-8759, by mail addressed to the United States Patent and Trademark Office, Office of Public Records, Alexandria, VA 22313-1450 or via the Internet.

In addition, information on the status of the application, including the mailing date of Office actions and the dates of receipt of correspondence filed in the Office, may also be accessed via the Internet through the Patent Electronic Business Center at www.uspto.gov using the public side of the Patent Application Information and Retrieval (PAIR) system. The direct link to access this status information is currently http://pair.uspto.gov/. Prior to publication, such status information is confidential and may only be obtained by applicant using the private side of PAIR.

Further assistance in electronically accessing the publication, or about PAIR, is available by calling the Patent Electronic Business Center at 1-866-217-9197.

Office of Data Managment, Application Assistance Unit (571) 272-4000, or (571) 272-4200, or 1-888-786-0101

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U.S. APPLICATION NUMBER NO.	FIRST NAMED APPLICANT	ATTY. DOCKET NO.
12/227,458	Ian Mcniece	68324(71699)

49383
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P.O. BOX 55874
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INTERNATIONAL APPLICATION NO.	
PCT/US2007/011921	
I.A. FILING DATE	PRIORITY DATE
05/18/2007	05/19/2006

CONFIRMATION NO. 2285
371 ACCEPTANCE LETTER

OC000000038720585

Date Mailed: 11/16/2009

NOTICE OF ACCEPTANCE OF APPLICATION UNDER 35 U.S.C 371 AND 37 CFR 1.495

The applicant is hereby advised that the United States Patent and Trademark Office in its capacity as a Designated / Elected Office (37 CFR 1.495), has determined that the above identified international application has met the requirements of 35 U.S.C. 371, and is ACCEPTED for national patentability examination in the United States Patent and Trademark Office.

The United States Application Number assigned to the application is shown above and the relevant dates are:

10/23/2009	10/23/2009
DATE OF RECEIPT OF 35 U.S.C. 371(c)(1), (c)(2) and (c)(4) REQUIREMENTS	DATE OF COMPLETION OF ALL 35 U.S.C. 371 REQUIREMENTS

A Filing Receipt (PTO-103X) will be issued for the present application in due course. **THE DATE APPEARING ON THE FILING RECEIPT AS THE " FILING DATE" IS THE DATE ON WHICH THE LAST OF THE 35 U.S.C. 371 (c)(1), (c)(2) and (c)(4) REQUIREMENTS HAS BEEN RECEIVED IN THE OFFICE. THIS DATE IS SHOWN ABOVE.** *The filing date of the above identified application is the international filing date of the international application (Article 11(3) and 35 U.S.C. 363).* Once the Filing Receipt has been received, send all correspondence to the Group Art Unit designated thereon.

The following items have been received:

- Indication of Small Entity Status
- Copy of the International Application filed on 11/17/2008
- Copy of the International Search Report filed on 11/17/2008
- Preliminary Amendments filed on 11/17/2008
- Information Disclosure Statements filed on 11/17/2008
- Oath or Declaration filed on 10/23/2009
- U.S. Basic National Fees filed on 11/17/2008
- Priority Documents filed on 11/17/2008

Applicant is reminded that any communications to the United States Patent and Trademark Office must be mailed to the address given in the heading and include the U.S. application no. shown above (37 CFR 1.5)

ULYSSES G WALKER

Telephone: (703) 756-1401

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APPLICATION NUMBER	FILING or 371(c) DATE	GRP ART UNIT	FIL FEE REC'D	ATTY.DOCKET.NO	TOT CLAIMS	IND CLAIMS
12/227,458	10/23/2009	1636	390	68324(71699)	18	1

CONFIRMATION NO. 2285

49383
EDWARDS ANGELL PALMER & DODGE LLP
P.O. BOX 55874
BOSTON, MA 02205

FILING RECEIPT

OC00000038720584

Date Mailed: 11/16/2009

Receipt is acknowledged of this non-provisional patent application. The application will be taken up for examination in due course. Applicant will be notified as to the results of the examination. Any correspondence concerning the application must include the following identification information: the U.S. APPLICATION NUMBER, FILING DATE, NAME OF APPLICANT, and TITLE OF INVENTION. Fees transmitted by check or draft are subject to collection. Please verify the accuracy of the data presented on this receipt. **If an error is noted on this Filing Receipt, please submit a written request for a Filing Receipt Correction. Please provide a copy of this Filing Receipt with the changes noted thereon. If you received a "Notice to File Missing Parts" for this application, please submit any corrections to this Filing Receipt with your reply to the Notice. When the USPTO processes the reply to the Notice, the USPTO will generate another Filing Receipt incorporating the requested corrections**

Applicant(s)

Ian Mcniece, Lutherville, MD;

Assignment For Published Patent Application

THE JOHNS HOPKINS UNIVERSITY, Baltimore, MD

Power of Attorney: The patent practitioners associated with Customer Number 49383

Domestic Priority data as claimed by applicant

This application is a 371 of PCT/US2007/011921 05/18/2007
which claims benefit of 60/801,661 05/19/2006

Foreign Applications

If Required, Foreign Filing License Granted: 11/11/2009

The country code and number of your priority application, to be used for filing abroad under the Paris Convention, is **US 12/227,458**

Projected Publication Date: 02/25/2010

Non-Publication Request: No

Early Publication Request: No
** **SMALL ENTITY** **

Title

METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Preliminary Class

435

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Since the rights granted by a U.S. patent extend only throughout the territory of the United States and have no effect in a foreign country, an inventor who wishes patent protection in another country must apply for a patent in a specific country or in regional patent offices. Applicants may wish to consider the filing of an international application under the Patent Cooperation Treaty (PCT). An international (PCT) application generally has the same effect as a regular national patent application in each PCT-member country. The PCT process **simplifies** the filing of patent applications on the same invention in member countries, but **does not result** in a grant of "an international patent" and does not eliminate the need of applicants to file additional documents and fees in countries where patent protection is desired.

Almost every country has its own patent law, and a person desiring a patent in a particular country must make an application for patent in that country in accordance with its particular laws. Since the laws of many countries differ in various respects from the patent law of the United States, applicants are advised to seek guidance from specific foreign countries to ensure that patent rights are not lost prematurely.

Applicants also are advised that in the case of inventions made in the United States, the Director of the USPTO must issue a license before applicants can apply for a patent in a foreign country. The filing of a U.S. patent application serves as a request for a foreign filing license. The application's filing receipt contains further information and guidance as to the status of applicant's license for foreign filing.

Applicants may wish to consult the USPTO booklet, "General Information Concerning Patents" (specifically, the section entitled "Treaties and Foreign Patents") for more information on timeframes and deadlines for filing foreign patent applications. The guide is available either by contacting the USPTO Contact Center at 800-786-9199, or it can be viewed on the USPTO website at http://www.uspto.gov/web/offices/pac/doc/general/index.html.

For information on preventing theft of your intellectual property (patents, trademarks and copyrights), you may wish to consult the U.S. Government website, http://www.stopfakes.gov. Part of a Department of Commerce initiative, this website includes self-help "toolkits" giving innovators guidance on how to protect intellectual property in specific countries such as China, Korea and Mexico. For questions regarding patent enforcement issues, applicants may call the U.S. Government hotline at 1-866-999-HALT (1-866-999-4158).

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Title 35, United States Code, Section 184

Title 37, Code of Federal Regulations, 5.11 & 5.15

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This license is to be retained by the licensee and may be used at any time on or after the effective date thereof unless it is revoked. This license is automatically transferred to any related applications(s) filed under 37 CFR 1.53(d). This license is not retroactive.

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NOT GRANTED

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OCT 23 2009

IAP19 Rec'd PCT/PTO 23 OCT 2009

FAX TRANSMISSION

DATE: October 23, 2009

PTO IDENTIFIER: **Application Number** 12/227,458-Conf. #2285
Patent Number
Inventor: Ian Mcniece

MESSAGE TO: US Patent and Trademark Office

FAX NUMBER: (571) 273-8300

FROM: EDWARDS ANGELL PALMER & DODGE LLP
Jonathan M. Sparks, Ph.D.

PHONE: (617) 517-5543

Attorney Dkt. #: 68324(71699)

PAGES (Including Cover Sheet): 12

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Certificate of Transmission (1 page)
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Four Month Request for Extension of Time Under 37 CFR 1.136(a) (1 page)
Part 2 Copy of Notice (2 pages)
Response to Notifcation of Missing Requirements (2 pages)
Combined Declaration and Power of Attorney (4 pages)
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Telephone: (617) 239-0100 **Facsimile:** (617) 227-4420

PAGE 1/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

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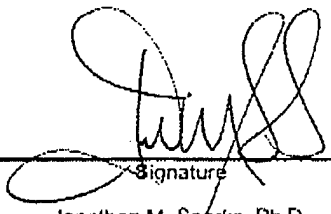
Application No. (if known): 12/227,458 Attorney Docket No.: 68324(71699)

Certificate of Transmission under 37 CFR 1.8

I hereby certify that this correspondence is being facsimile transmitted to the United States Patent and Trademark Office.

on October 23, 2009
Date



Signature

Jonathan M. Sparks, Ph.D.
Typed or printed name of person signing Certificate

53,624
Registration Number, if applicable

(617) 517-5543
Telephone Number

Note: Each paper must have its own certificate of transmission, or this certificate must identify each submitted paper.

Fee Transmittal (1 page)
Four Month Request for Extension of Time Under 37 CFR 1.136(a) (1 page)
Part 2 Copy of Notice (2 pages)
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PTO/SB/17 (10-08)
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U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
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Effective on 12/08/2004.
Fees pursuant to the Consolidated Appropriations Act, 2005 (H.R. 4818).

FEE TRANSMITTAL
For FY 2009

[x] Applicant claims small entity status. See 37 CFR 1.27

TOTAL AMOUNT OF PAYMENT ($) 930.00

Complete if Known	
Application Number	12/227,458-Conf. #2285
Filing Date	November 17, 2008
First Named Inventor	Ian McNiece
Examiner Name	Not Yet Assigned
Art Unit	N/A
Attorney Docket No.	68324(71699)

METHOD OF PAYMENT (check all that apply)

[ ] Check [ ] Credit Card [ ] Money Order [ ] None [ ] Other (please identify): _____

[x] Deposit Account Deposit Account Number: 04-1105 Deposit Account Name: Edwards Angell Palmer & Dodge LLP

For the above-identified deposit account, the Director is hereby authorized to: (check all that apply)

[x] Charge fee(s) indicated below [ ] Charge fee(s) indicated below, **except for the filing fee**

[x] Charge any additional fee(s) or underpayments of fee(s) under 37 CFR 1.16 and 1.17 [x] Credit any overpayments

FEE CALCULATION

1. BASIC FILING, SEARCH, AND EXAMINATION FEES

	FILING FEES		SEARCH FEES		EXAMINATION FEES		
Application Type	Fee ($)	Small Entity Fee ($)	Fee ($)	Small Entity Fee ($)	Fee ($)	Small Entity Fee ($)	Fees Paid ($)
Utility	330	165	540	270	220	110	
Design	220	110	100	50	140	70	
Plant	220	110	330	165	170	85	
Reissue	330	165	540	270	650	325	
Provisional	220	110	0	0	0	0	

2. EXCESS CLAIM FEES

Fee Description	Fee ($)	Small Entity Fee ($)
Each claim over 20 (including Reissues)	52	26
Each independent claim over 3 (including Reissues)	220	110
Multiple dependent claims	390	195

Total Claims	Extra Claims	Fee ($)	Fee Paid ($)
_____ - or HP = _____	x _____	=	_____

HP = highest number of total claims paid for, if greater than 20.

Indep. Claims	Extra Claims	Fee ($)	Fee Paid ($)
_____ - or HP = _____	x _____	=	_____

HP = highest number of independent claims paid for, if greater than 3.

Multiple Dependent Claims	
Fee ($)	Fee Paid ($)

3. APPLICATION SIZE FEE

If the specification and drawings exceed 100 sheets of paper (excluding electronically filed sequence or computer listings under 37 CFR 1.52(e)), the application size fee due is $270 ($135 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).

Total Sheets	Extra Sheets	Number of each additional 50 or fraction thereof	Fee ($)	Fee Paid ($)
_____ - 100 =	_____ /50 =	_____ (round up to a whole number) x	_____	= _____

4. OTHER FEE(S)

	Fees Paid ($)
Non-English Specification, $130 fee (no small entity discount)	
Other (e.g., late filing surcharge): 2254 Extension for response within fourth month	865.00
2051 Surcharge-Late oath or declaration	65.00

SUBMITTED BY					
Signature		Registration No. (Attorney/Agent)	53,624	Telephone	(617) 517-5543
Name (Print/Type)	Jonathan M. Sparks, Ph.D.			Date	October 23, 2009

BOS2 761896.1

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OCT 23 2009

PTO/SB/22 (07-09)
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PETITION FOR EXTENSION OF TIME UNDER 37 CFR 1.136(a) FY 2009 *(Fees pursuant to the Consolidated Appropriations Act, 2005 (H.R. 4818).)*	Docket Number (Optional) 68324(71699)
Application Number 12/227,458-Conf. #2285	Filed November 17, 2008
For METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS	
Art Unit N/A	Examiner Not Yet Assigned

This is a request under the provisions of 37 CFR 1.136(a) to extend the period for filing a reply in the above identified application.

The requested extension and fee are as follows (check time period desired and enter the appropriate fee below):

		Fee	Small Entity Fee	
[ ]	One month (37 CFR 1.17(a)(1))	$130	$65	$
[ ]	Two months (37 CFR 1.17(a)(2))	$490	$245	$
[ ]	Three months (37 CFR 1.17(a)(3))	$1110	$555	$
[x]	Four months (37 CFR 1.17(a)(4))	$1730	$865	$ 865.00
[ ]	Five months (37 CFR 1.17(a)(5))	$2350	$1175	$

[x] Applicant claims small entity status. See 37 CFR 1.27.

[ ] A check in the amount of the fee is enclosed.

[ ] Payment by credit card. Form PTO-2038 is attached.

[x] The Director has already been authorized to charge fees in this application to a Deposit Account.

[x] The Director is hereby authorized to charge any fees which may be required, or credit any overpayment, to Deposit Account Number 04-1105

WARNING: Information on this form may become public. Credit card information should not be included on this form. Provide credit card information and authorization on PTO-2038.

I am the

[ ] applicant/inventor.

[ ] assignee of record of the entire interest. See 37 CFR 3.71.
Statement under 37 CFR 3.73(b) is enclosed. (Form PTO/SB/96).

[x] attorney or agent of record. Registration Number 53,624

[ ] attorney or agent under 37 CFR 1.34.
Registration number if acting under 37 CFR 1.34

Signature — October 23, 2009 (Date)

Jonathan M. Sparks, Ph.D. (Typed or printed name) — (617) 517-5543 (Telephone Number)

NOTE: Signatures of all the inventors or assignees of record of the entire interest or their representative(s) are required. Submit multiple forms if more than one signature is required, see below.

[ ] Total of 1 forms are submitted.

10/26/2009 LLANDGRA 00000074 041105 12227458

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UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
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Alexandria, Virginia 22313-1450
www.uspto.gov

U.S. APPLICATION NUMBER NO.	FIRST NAMED APPLICANT	ATTY. DOCKET NO.
12/227,458	Ian MCNIECE	68324(71699)

49383
EDWARDS ANGELL PALMER & DODGE LLP
P.O. BOX 55874
BOSTON, MA 02205

INTERNATIONAL APPLICATION NO.	
PCT/US2007/011921	
I.A. FILING DATE	PRIORITY DATE
05/18/2007	05/19/2006

CONFIRMATION NO. 2285
371 FORMALITIES LETTER

OC000000035873451

Date Mailed: 05/12/2009

NOTIFICATION OF MISSING REQUIREMENTS UNDER 35 U.S.C. 371 IN THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US)

The following items have been submitted by the applicant or the IB to the United States Patent and Trademark Office as a Designated Office (37 CFR 1.494):

- Indication of Small Entity Status
- Priority Document
- Copy of the International Application filed on 11/17/2008
- Copy of the International Search Report filed on 11/17/2008
- Preliminary Amendments filed on 11/17/2008
- Information Disclosure Statements filed on 11/17/2008
- U.S. Basic National Fees filed on 11/17/2008
- Priority Documents filed on 11/17/2008

The following items **MUST** be furnished within the period set forth below in order to complete the requirements for acceptance under 35 U.S.C. 371:

- Oath or declaration of the inventors, in compliance with 37 CFR 1.497(a) and (b), identifying the application by the International application number and international filing date.

ALL OF THE ITEMS SET FORTH ABOVE MUST BE SUBMITTED WITHIN TWO (2) MONTHS FROM THE DATE OF THIS NOTICE OR BY 32 MONTHS FROM THE PRIORITY DATE FOR THE APPLICATION, WHICHEVER IS LATER. FAILURE TO PROPERLY RESPOND WILL RESULT IN ABANDONMENT.

The time period set above may be extended by filing a petition and fee for extension of time under the provisions of 37 CFR 1.136(a).

Applicant is reminded that any communications to the United States Patent and Trademark Office must be mailed to the address given in the heading and include the U.S. application no. shown above (37 CFR 1.5)

Registered users of EFS-Web may alternatively submit their reply to this notice via EFS-Web.
https://sportal.uspto.gov/authenticate/AuthenticateUserLocalEPF.html

PAGE 5/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

For more information about EFS-Web please call the USPTO Electronic Business Center at **1-866-217-9197** or visit our website at http://www.uspto.gov/ebc.

If you are not using EFS-Web to submit your reply, you must include a copy of this notice.

SHAKEEL AHMED

Telephone: (703) 756-1423

FORM PCT/ISO/EC/005 (371 Formalities Notice)

PAGE 6/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

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OCT 2 3 2009

Docket No.: 68324(71699)
(PATENT)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Patent Application of:
Ian Mcniece

Application No.: 12/227,458

Filed: November 17, 2008

For: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Confirmation No.: 2285

Art Unit: N/A

Examiner: Not Yet Assigned

RESPONSE TO NOTIFICATION OF MISSING REQUIREMENTS

MS Missing Parts
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir/Madam:

In response to the Notification of Missing Requirements – Filing Date Granted mailed May 12, 2009, Applicant respectfully submits a Combined Declaration and Power of Attorney, a Petition for Extension of Time, and Part 2 Copy of Notice.

Please charge our Deposit Account No. 04-1105 in the amount of $930.00 covering the fees set forth in 37 CFR 1.17(a)(4) and 1.16(f). The Director is hereby authorized to charge any deficiency in the fees filed, asserted to be filed or which should

have been filed herewith (or with any paper hereafter filed in this application by this firm) to our Deposit Account No. 04-1105, under Order No. 68324(71699).

Dated: October 23, 2009

Respectfully submitted,

By _____

Jonathan M. Sparks, Ph.D.
Registration No.: 53,624
EDWARDS ANGELL PALMER & DODGE LLP
P.O. Box 55874
Boston, Massachusetts 02205
(617) 517-5543
Attorneys/Agents For Applicant

PAGE 8/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

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OCT 23 2009

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

DECLARATION FOR PATENT APPLICATION

As the below named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor of the subject matter which is claimed and for which a patent is sought on the invention entitled:

METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

the specification of which was filed on November 17, 2008 as Application No. 12/227,458.

In the event that the filing date and/or Application No. are not entered above at the time I execute this document, and if such information is deemed necessary, I hereby authorize and request my attorneys/agent(s) at **Edwards Angell Palmer & Dodge LLP**, P.O. Box 55874, Boston, Massachusetts 02205, to insert above the filing date and/or Application No. of said application.

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims, as amended by an amendment, if any, specifically referred to herein.

I acknowledge the duty to disclose all information known to me that is material to patentability as defined in 37 CFR 1.56.

FOREIGN PRIORITY CLAIM

I hereby claim foreign priority benefits under Title 35, United States Code § 119(a)-(d) of any foreign application(s) for patent or inventor's certificate listed below and have also identified below any foreign application for patent or inventor's certificate having a filing date before that of the application on which priority is claimed:

[x] no such foreign applications have been filed

[ ] such foreign application have been filed as follows:

PAGE 9/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

Attorney Docket No.: 68324(71699)

EARLIEST FOREIGN APPLICATION(S), IF ANY FILED WITHIN 12 MONTHS (6 MONTHS FOR DESIGN) PRIOR TO THIS U.S. APPLICATION

Application Number	Country	Date of Filing	Priority Claimed Under 35 USC 119

ALL FOREIGN APPLICATION(S), IF ANY FILED MORE THAN 12 MONTHS (6 MONTHS FOR DESIGN) PRIOR TO THIS U.S. APPLICATION

Application Number	Country	Date of Filing

CLAIM FOR BENEFIT OF EARLIER U.S. PROVISIONAL APPLICATIONS

I hereby claim priority benefits under Title 35, United States Code §119(e), of any United States provisional patent application(s) listed below:

[ ] no such U.S. provisional applications have been filed.

[X] such U.S. provisional application have been filed as follows:

Application Number	Date of Filing	Priority Claimed Under 35 USC 119
60/801,661	May 19, 2006	X Yes No
		X Yes No

CLAIM FOR BENEFIT OF EARLIER U.S./PCT APPLICATION(S)

I hereby claim the benefit under Title 35, United States Code, §120 of the United States application(s) listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States application in the manner provided by the first paragraph of Title 35, United States Code, §112, I acknowledge the duty to disclose all information that is material to patentability as defined in 37 CFR 1.56 which became available to me between the filing date of the prior application and the national or PCT international filing date of this application:

[ ] no such U.S./PCT applications have been filed.

PAGE 10/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

Attorney Docket No.: 68324(71699)

[x] such U.S./PCT application have been filed as follows:

Application Number	Relationship	Parent Application	Date of Filing
This Application	Continuation	PCT/US2007/011921	May 18, 2007

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

I hereby appoint:

All practitioners at Customer Number 49383

jointly, and each of them severally, my attorneys at law/patent agent(s), with full power of substitution, delegation and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, and to transact all business in the U.S. Patent and Trademark Office connected therewith.

The undersigned hereby authorizes the U.S. attorney or agent named herein to accept and follow instructions from The Johns Hopkins University as to any action to be taken in the United States Patent and Trademark Office regarding this application without direct communication between the U.S. attorney or agent and the undersigned. In the event of a change in the persons from whom instructions may be taken, the U.S. attorney or agent named herein will be so notified by the undersigned.

Please mail all correspondence to Peter F. Corless , whose address is:

Edwards Angell Palmer & Dodge LLP
P.O. Box 55874
Boston, Massachusetts 02205

Please direct telephone calls to: Peter F. Corless at (617) 517-5557.

Please direct facsimiles to: (888) 325-9132

PAGE 11/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

Attorney Docket No.: 68324(71699)

Full name of sole or first inventor Ian McNiece	
Sole or first inventor's signature	Date 10/9/09
Residence Coral Gables, Florida	
Citizenship Australia / USA	
Mailing Address 821 Majorca Ave. Coral Gables, FL 33134	

PAGE 12/12 * RCVD AT 10/23/2009 2:21:50 PM [Eastern Daylight Time] * SVR:USPTO-EFXRF-6/43 * DNIS:2738300 * CSID:6172274420 * DURATION (mm-ss):01-42

UNITED STATES PATENT AND TRADEMARK OFFICE

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United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
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Alexandria, Virginia 22313-1450
www.uspto.gov

U.S. APPLICATION NUMBER NO.	FIRST NAMED APPLICANT	ATTY. DOCKET NO.
12/227,458	Ian MCNIECE	68324(71699)

49383
EDWARDS ANGELL PALMER & DODGE LLP
P.O. BOX 55874
BOSTON, MA 02205

INTERNATIONAL APPLICATION NO.	
PCT/US2007/011921	
I.A. FILING DATE	PRIORITY DATE
05/18/2007	05/19/2006

CONFIRMATION NO. 2285
371 FORMALITIES LETTER

Date Mailed: 05/12/2009

NOTIFICATION OF MISSING REQUIREMENTS UNDER 35 U.S.C. 371 IN THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US)

The following items have been submitted by the applicant or the IB to the United States Patent and Trademark Office as a Designated Office (37 CFR 1.494):

- Indication of Small Entity Status
- Priority Document
- Copy of the International Application filed on 11/17/2008
- Copy of the International Search Report filed on 11/17/2008
- Preliminary Amendments filed on 11/17/2008
- Information Disclosure Statements filed on 11/17/2008
- U.S. Basic National Fees filed on 11/17/2008
- Priority Documents filed on 11/17/2008

The following items **MUST** be furnished within the period set forth below in order to complete the requirements for acceptance under 35 U.S.C. 371:

- Oath or declaration of the inventors, in compliance with 37 CFR 1.497(a) and (b), identifying the application by the International application number and international filing date.

ALL OF THE ITEMS SET FORTH ABOVE MUST BE SUBMITTED WITHIN TWO (2) MONTHS FROM THE DATE OF THIS NOTICE OR BY 32 MONTHS FROM THE PRIORITY DATE FOR THE APPLICATION, WHICHEVER IS LATER. FAILURE TO PROPERLY RESPOND WILL RESULT IN ABANDONMENT.

The time period set above may be extended by filing a petition and fee for extension of time under the provisions of 37 CFR 1.136(a).

Applicant is reminded that any communications to the United States Patent and Trademark Office must be mailed to the address given in the heading and include the U.S. application no. shown above (37 CFR 1.5)

Registered users of EFS-Web may alternatively submit their reply to this notice via EFS-Web.
https://sportal.uspto.gov/authenticate/AuthenticateUserLocalEPF.html

For more information about EFS-Web please call the USPTO Electronic Business Center at **1-866-217-9197** or visit our website at http://www.uspto.gov/ebc.

If you are not using EFS-Web to submit your reply, you must include a copy of this notice.

SHAKEEL AHMED

Telephone: (703) 756-1423

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
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APPLICATION NUMBER	PATENT NUMBER	GROUP ART UNIT	FILE WRAPPER LOCATION
12/227,458		1636	0540

Correspondence Address/Fee Address Change

The following fields have been set to Customer Number 49383 on 02/03/2009

- **Correspondence Address**
- **Maintenance Fee Address**
- **Power of Attorney Address**

The address of record for Customer Number 49383 is:

49383
EDWARDS ANGELL PALMER & DODGE LLP
P.O. BOX 55874
BOSTON, MA 02205

IAP16 Rec'd PCT/PTO 17 NOV 2008

PTO-1390 (Rev. 09-08)
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U. S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
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TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A SUBMISSION UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER	
68324(71699)	
U.S. APPLICATION NO.	12/227458

INTERNATIONAL APPLICATION NO.	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/US2007/011921	18 May 2007	19 May 2006

TITLE OF INVENTION
METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

APPLICANT(S) FOR DO/EO/US
Ian Mcniece

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. [x] This is a **FIRST** submission of items concerning a submission under 35 U.S.C. 371.
2. [ ] This is a **SECOND** or **SUBSEQUENT** submission of items concerning a submission under 35 U.S.C. 371.
3. [ ] This is an express request to begin national examination procedures (35 U.S.C. 371(f)). The submission must include items (5), (6), (9) and (21) indicated below.
4. [x] The US has been elected (Article 31).
5. [x] A copy of the International Application as filed (35 U.S.C. 371 (c)(2))
   a. [ ] is attached hereto (required only if not communicated by the International Bureau).
   b. [x] has been communicated by the International Bureau.
   c. [ ] is not required, as the application was filed in the United States Receiving Office (RO/US).
6. [ ] An English language translation of the International Application as filed (35 U.S.C. 371(c)(2)).
   a. [ ] is attached hereto.
   b. [ ] has been previously submitted under 35 U.S.C. 154(d)(4).
7. [x] Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))
   a. [ ] are attached hereto (required only if not communicated by the International Bureau).
   b. [ ] have been communicated by the International Bureau.
   c. [ ] have not been made; however, the time limit for making such amendments has NOT expired.
   d. [x] have not been made and will not be made.
8. [ ] An English language translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. [ ] An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. [ ] An English language translation of the annexes of the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11 to 20 below concern document(s) or information included:

11. [x] An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
12. [ ] An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. [x] A preliminary amendment.
14. [x] An Application Data Sheet under 37 CFR 1.76.
15. [ ] A substitute specification.
16. [ ] A power of attorney and/or change of address letter.
17. [ ] A computer-readable form of the sequence listing in accordance with PCT Rule 13*ter*.2 and 37 CFR 1.821 – 1.825.
18. [ ] A second copy of the published International Application under 35 U.S.C. 154(d)(4).
19. [ ] A second copy of the English language translation of the international application under 35 U.S.C. 154(d)(4).

IAPEC'd PCT 17 NOV 2008

PTO-1390 (Rev. 09-08)
Approved for use through 2/28/2010. OMB 0651-0021
U. S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
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U.S. APPLICATION NO. (if known, see 37 CFR 1.5)	INTERNATIONAL APPLICATION NO.	ATTORNEY'S DOCKET NUMBER
12/227458	PCT/US2007/011921	68324(71699)

20. Other items or information: Return Receipt Postcard; Copy of published PCT Application No. WO 2007/136760 A2; Copy of International Search Report.

The following fees have been submitted	CALCULATIONS	PTO USE ONLY
21. [x] Basic national fee (37 CFR 1.492(a)) $330	$ 330.00	
22. [x] Examination fee (37 CFR 1.492(c)) If the written opinion prepared by ISA/US or the international preliminary examination report prepared by IPEA/US indicates all claims satisfy provisions of PCT Article 33(1)-(4) ..... $0 All other situations $220	$ 220.00	
23. [x] Search fee (37 CFR 1.492(b)) If the written opinion of the ISA/US or the international preliminary examination report prepared by IPEA/US indicates all claims satisfy provisions of PCT Article 33(1)-(4) ..... $0 Search fee (37 CFR 1.445(a)(2)) has been paid on the international application to the USPTO as an International Searching Authority ..... $100 International Search Report prepared by an ISA other than the US and provided to the Office or previously communicated to the US by the IB ..... $430 All other situations $540	$ 100.00	
TOTAL OF 21, 22 and 23 =	650.00	
[ ] Additional fee for specification and drawings filed in paper over 100 sheets (excluding sequence listing in compliance with 37 CFR 1.821(c) or (e) or in an electronic medium or computer program listing in an electronic medium) (37 CFR 1.492(j)). The fee is **$270** for each additional 50 sheets of paper or fraction thereof.		

Total Sheets	Extra Sheets	Number of each additional 50 or fraction thereof (round up to a whole number)	RATE	
22 - 100 =	/50 =		x $270	$

Surcharge of **$130** for furnishing any of the search fee, examination fee, or the oath or declaration after the date of commencement of the national stage (37 CFR 1.492(h)).				$ 130.00
CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE	
Total claims	18 - 20 =	0	x $52	0.00
Independent claims	1 - 3 =	0	x $220	0.00
MULTIPLE DEPENDENT CLAIM(S) (if applicable)			+ $390	
TOTAL OF ABOVE CALCULATIONS =				$ 780.00
[x] Applicant claims small entity status. See 37 CFR 1.27. Fees above are reduced by ½.				390.00
SUBTOTAL =				$ 390.00
Processing fee of **$130.00** for furnishing the English translation later than 30 months from the earliest claimed priority date (37 CFR 1.492(i)). +				$
TOTAL NATIONAL FEE =				$ 390.00
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). **$40.00** per property +				$
				$
TOTAL FEES ENCLOSED =				$ 390.00
Amount to be refunded:				$
Amount to be charged				$

PTO-1390 (Rev. 09-08)
Approved for use through 2/28/2010. OMB 0651-0021
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NOTE: Where an appropriate time limit under 37 CFR 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the International Application to pending status.

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SIGNATURE

Peter F. Corless
NAME

CUSTOMER NUMBER: 49383

33,860
REGISTRATION NUMBER

Application No. (if known): Not Yet Assigned | Attorney Docket No.: 68324(71699)

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Application Data Sheet (2 pages)
Transmittal Letter to the United States Designated-Elected Office (3 pages)
First Preliminary Amendment (3 pages)
IDS (Citation) by Applicant (3 References) (2 pages)
Information Disclosure Statement (2 pages)
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PTO-1390 (Rev. 09-08)
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TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A SUBMISSION UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER	
68324(71699)	
U.S. APPLICATION NO.	12/227458

INTERNATIONAL APPLICATION NO.	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/US2007/011921	18 May 2007	19 May 2006

TITLE OF INVENTION
METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

APPLICANT(S) FOR DO/EO/US
Ian Mcniece

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. [x] This is a **FIRST** submission of items concerning a submission under 35 U.S.C. 371.
2. [ ] This is a **SECOND** or **SUBSEQUENT** submission of items concerning a submission under 35 U.S.C. 371.
3. [ ] This is an express request to begin national examination procedures (35 U.S.C. 371(f)). The submission must include items (5), (6), (9) and (21) indicated below.
4. [x] The US has been elected (Article 31).
5. [x] A copy of the International Application as filed (35 U.S.C. 371 (c)(2))
   a. [ ] is attached hereto (required only if not communicated by the International Bureau).
   b. [x] has been communicated by the International Bureau.
   c. [ ] is not required, as the application was filed in the United States Receiving Office (RO/US).
6. [ ] An English language translation of the International Application as filed (35 U.S.C. 371(c)(2)).
   a. [ ] is attached hereto.
   b. [ ] has been previously submitted under 35 U.S.C. 154(d)(4).
7. [x] Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))
   a. [ ] are attached hereto (required only if not communicated by the International Bureau).
   b. [ ] have been communicated by the International Bureau.
   c. [ ] have not been made; however, the time limit for making such amendments has NOT expired.
   d. [x] have not been made and will not be made.
8. [ ] An English language translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. [ ] An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. [ ] An English language translation of the annexes of the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11 to 20 below concern document(s) or information included:

11. [x] An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
12. [ ] An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. [x] A preliminary amendment.
14. [x] An Application Data Sheet under 37 CFR 1.76.
15. [ ] A substitute specification.
16. [ ] A power of attorney and/or change of address letter.
17. [ ] A computer-readable form of the sequence listing in accordance with PCT Rule 13*ter*.2 and 37 CFR 1.821 – 1.825.
18. [ ] A second copy of the published International Application under 35 U.S.C. 154(d)(4).
19. [ ] A second copy of the English language translation of the international application under 35 U.S.C. 154(d)(4).

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U.S. APPLICATION NO. (if known, see 37 CFR 1.5)	INTERNATIONAL APPLICATION NO.	ATTORNEY'S DOCKET NUMBER
12/227458	PCT/US2007/011921	68324(71699)

20. Other items or information: Return Receipt Postcard; Copy of published PCT Application No. WO 2007/136760 A2; Copy of International Search Report.

The following fees have been submitted	CALCULATIONS	PTO USE ONLY
21. [x] Basic national fee (37 CFR 1.492(a)) $330	$ 330.00	
22. [x] Examination fee (37 CFR 1.492(c)) If the written opinion prepared by ISA/US or the international preliminary examination report prepared by IPEA/US indicates all claims satisfy provisions of PCT Article 33(1)-(4) ..... $0 All other situations $220	$ 220.00	
23. [x] Search fee (37 CFR 1.492(b)) If the written opinion of the ISA/US or the international preliminary examination report prepared by IPEA/US indicates all claims satisfy provisions of PCT Article 33(1)-(4) ..... $0 Search fee (37 CFR 1.445(a)(2)) has been paid on the international application to the USPTO as an International Searching Authority ..... $100 International Search Report prepared by an ISA other than the US and provided to the Office or previously communicated to the US by the IB ..... $430 All other situations $540	$ 100.00	
TOTAL OF 21, 22 and 23 =	650.00	
[ ] Additional fee for specification and drawings filed in paper over 100 sheets (excluding sequence listing in compliance with 37 CFR 1.821(c) or (e) or in an electronic medium or computer program listing in an electronic medium) (37 CFR 1.492(j)). The fee is **$270** for each additional 50 sheets of paper or fraction thereof.		

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CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE	
Total claims	18 - 20 =	0	x $52	0.00
Independent claims	1 - 3 =	0	x $220	0.00
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TOTAL OF ABOVE CALCULATIONS =				$ 780.00
[x] Applicant claims small entity status. See 37 CFR 1.27. Fees above are reduced by ½.				390.00
SUBTOTAL =				$ 390.00
Processing fee of **$130.00** for furnishing the English translation later than 30 months from the earliest claimed priority date (37 CFR 1.492(i)). +				$
TOTAL NATIONAL FEE =				$ 390.00
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). **$40.00** per property +				$
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TOTAL FEES ENCLOSED =				$ 390.00
Amount to be refunded:				$
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Peter F. Corless
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Application Data Sheet

Application Information

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Subject Matter::	Utility
Suggested Group Art Unit::	N/A
CD-ROM or CD-R?::	None
Sequence submission?::	None
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Title::	METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS
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Request for Early Publication?::	No
Request for Non-Publication?::	No
Total Drawing Sheets::	6
Small Entity?::	Yes
Petition included?::	No
Secrecy Order in Parent Appl.?::	No

Applicant Information

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State or Province of mailing address:: MD

Postal or Zip Code of mailing address:: 21093

Correspondence Information

Correspondence Customer Number:: 49383

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Domestic Priority Information

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This Application	Continuation of	PCT/US2007/01192 1	05/18/07
PCT/US2007/01192 1	An application claiming the benefit under 35 USC 119(e)	60/801,661	05/19/06

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60/801,661 19 May 2006 (19.05.2006) US

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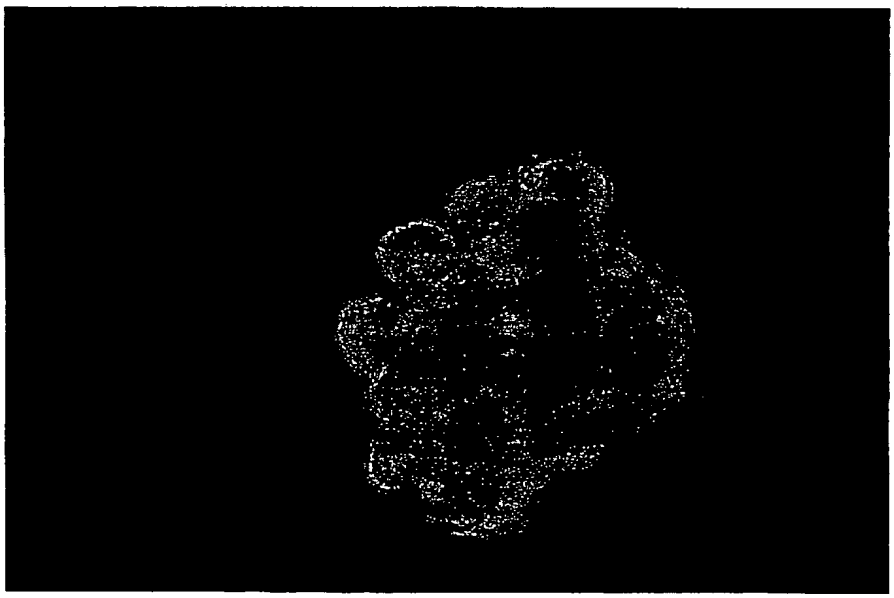
For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS



(57) Abstract: The invention provides methods for expanding mesenchymal stem cells (MSCs) in non-adherent cultures. The methods include the propagation of MSCs in or on non-adherent matrices. The invention further provides administration and the use of cells propagated by the method of the invention for administration and preparation of a therapeutic agent. The invention further provides kits including cells propagated by the methods of the inventions.

WO 2007/136760 A2

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12/227458

METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Related Applications

This application claims priority to Provisional Patent Application Serial No. 60/801,661, filing date May 19, 2006, which is incorporated herein by reference in its entirety.

Government Support

The invention was made under Grant Number CA088878 from the National Institutes of Health of the United States Government. The Government may have certain rights in relation to the application.

Field Of The Invention

This invention relates to methods of growth of mesenchymal cells under non-adherent conditions. The method allows for expansion of mesenchymal cells in suspension for research or therapeutic uses.

Background Of The Invention

Mesenchymal stem cells are the formative pluripotential blast cells found *inter alia* in bone marrow, blood, dermis, and periosteum that are capable of differentiating into any of the specific types of mesenchymal or connective tissues (i.e. the tissues of the body that support the specialized elements; particularly adipose, osseous, cartilaginous, elastic, and fibrous connective tissues) depending upon various influences from bioactive factors, such as cytokines. In contrast to their hematopoietic counterparts, MSCs are adherent and can be expanded in culture. A number of U.S. Patents, e.g., U.S. Patent Nos. 5,486,359; 5,591,625; 5,736,396; 5,811,094; 5,827,740; 5,837,539; 5,908,782; 5,908,784; 5,942,225; 5,965,436; 6,010,696; 6,022,540; 6,087,113; 5,858,390; 5,804,446; 5,846,796; 5,654,186; 6,054,121; 5,827,735; and 5,906,934 (all of which are incorporated herein by

reference) disclose mesenchymal stem cells (MSC), which can be differentiated into several progenitor cells, for example muscle progenitor cells, connective tissue cell progenitor cells, or hepatic oval cells. Muscle progenitor cells differentiate further into cardiac, skeletal, and smooth muscle cells, whereas the connective tissue cell progenitor may differentiate into bone. The patents above further teach transgenic MSCs that carry a transgene, methods to promote differentiation of MSCs along specific paths, and therapeutic methods including the use of MSCs.

Human MSC (hMSC) can be identified by the presence or absence of specific cell surface markers (Pittenger and Martin, *Circ. Res.* 95:9-20, 2004, incorporated herein by reference). Typically, hMSC can be identified by the presence of surface markers CD13, CD29, CD44, CD49a, b, c, e, f, CD51, CD54, CD58, CD71, CD73, CD90, CD102, CD105, CD106, CDw11, CD120a, CD120b, CD123, CD124, CD126, CDC127, CD140a, CD166, P75, TGFb1R, TGFbIIR, HLA-A, B, C, SSEA-3, SSEA-4, D7; and the absence of surface markers CD3, CD4, CD6, CD9, CD10, CD11a, CD14, CD15, CD18, CD21, CD25, CD31, CD34, CD36, CD38, CD45, CD49d, CD50, CD62E, L, S, CD80, CD86, CD95, CD117, CD133, SSEA-1. Monoclonal antibodies specific to MSCs have also been identified (e.g., US Patents 5,486,359 and 5,811,094). However, most surface markers have been found inadequate as a means to identify stem cells because putative marker(s) may also be found on nonstem cells, or a particular marker may only be expressed on a stem cell at a certain stage or under certain conditions, such as CD34 on hematopoietic stem cells. Nevertheless, surface markers and other attributes are useful in characterizing a stem cell as isolated or cultured, to detect changes in cells in culture over time, and as a means to begin to understand its potential interactions with neighboring cells and the cell environment (Pittenger and Martin, *Circ. Res.* 95:9-20, 2004).

Mesenchymal stem cells can be isolated from a number of cells and tissues including bone marrow, embryonic yolk sac, placenta, umbilical cord, fetal and adolescent skin, and blood, and propagated in culture. Friedenstein et al. (*Exp. Hematol.* 4:267-274, 1976, incorporated herein by reference) initially isolated MSCs

by their adherence to tissue culture surfaces. Similar methods for isolation of MSCs are still commonly used.

Plating studies indicate that MSCs are present at as a rare population of cells in bone marrow, representing about 0.001-0.01% of nucleated cells. However, MSCs can be readily expanded when grown at a very low plating density. Cotler et al. (*Proc. Natl. Acad. Sci. USA.* 97:3213-3218) noted that the number of colonies formed per 100 cells plated remained constant when the density of plating was varied from 0.5 to 12 cells per cm^2. However, the size of the colonies decreased markedly when the cells were plated at higher densities. Colonies of maximal size were obtained when cells were plated at 1.5 to 3.0 cells per cm^2. Plating at such low densities requires the use of large amount of tissue culture dishes, reagents, and space. Methods for culturing of MSCs in a less resource intensive manner is desirable.

Adult bone marrow-derived MSCs engraft in numerous organs and differentiate along tissue-specific lineages when transplanted into animals. They migrate into areas of muscle degeneration to undergo myogenic differentiation in immunodeficient mice. Injection of MSCs directly into infracted swine heart has been shown to induce myocardial regeneration and improved cardiac function (Shake et al., *Ann. Thorac. Surg.* 73:1919-1925, 2002). In addition, MSCs implantation has been demonstrated to induce therapeutic angiogenesis in a rat model of hindlimb ischemia through vascular endothelial growth factor (VEGF) production by MSCs (Al-Khaldi et al., *Gene Ther.* 10:621-629, 2003). In humans, bone marrow-derived MSCs have been used to regenerate the marrow microenvironment after myeloablative therapy. When introduced into the infracted heart, MSCs prevent deleterious modeling and improve recovery. Interestingly, implanted cells do not appear to expand after implantation when engrafted to tissue other than bone. Experiments using MSCs labeled with membrane dyes that would be diluted out after about 3 cell divisions were found months later even in repairing tissue (Pittenger and Martin, *Circ. Res.* 95:9-20, 2004).

Clinical trials have been initiated in several countries to test cell-based therapies for the treatment of the injured heart. However, no studies have

demonstrated incorporation of MSCs into regenerating tissue. It has been suggested that the MSCs exert a therapeutic effect by paracrine actions exerted by the cells through the release of soluble factors (See e.g., Gnecchi et al., *FASEB J.* 20:661-669, 2006; and Nagaya et al., *Circulation.* 112:1128-1135, 2005). This theory is supported by data therein demonstrating that conditioned media from transgenic MSCs overexpressing the prosurvival gene Akt limits hypoxia-induced apoptosis and triggers vigorous spontaneous contraction of adult rat cardiomyocytes in culture. Moreover, injection of concentrated conditioned media from the Akt transgenic MSCs into infracted rat hearts significantly limited infarct size and improved ventricular function relative to controls (Gnecchi et al., 2006).

Studies have demonstrated that upon transplantation of cells into cardiac tissue (e.g., by injection) less than 3% of injected MSCs persist after 2 weeks (Mazhari & Hare, Nature Clinical Practice Cardiovascular Medicine 4: suppl 1; S21-S26, 2007). This may be due to the adherent culture methods used to culture the MSCs. MSCs in bone marrow are able to adhere to bone to allow for proliferation. No comparable surface is present in muscle or many other tissues in which MSCs have been demonstrated to be beneficial. Current culture methods select for cells that are able to adhere to culture dishes through repeated rounds of trypsinization. When transplanted into cardiac tissue for example, MSCs may fail to proliferate due to their inability to adhere to a cardiac tissue surface, minimizing the contribution of MSCs to regenerating tissue.

Methods of culture of MSCs that do not include adherence to a surface and/or reduce the need for multiple rounds of trypsinization for propagation of cells may improve the effects of MSC at sites of injury, for example, by providing cells that are more able to proliferate at the site of injury.

Summary Of The Invention

The invention provides methods for the propagation of mesenchymal stem cells (MSCs) in non-adherent culture, eliminating the need for trypsinization in propagation of MSCs.

Accordingly, an aspect of the invention features a method for culturing MSCs under non-adherent conditions in or on a non-adherent matrix to obtain an expanded population of MSCs. The methods include formation of MSC spheres (MSCS) in or on several different non-adherent matrices, including incorporation of cells into biocompatible matrices such as Hydrogel and Matrigel™; culture of cells on or between layers of agarose; and culture of cells in Teflon® bags. After isolation of MSCs from a sample, the cells are propagated without treatment with trypsin after initial cell selection. MSCS are optionally mechanically manipulated, collected by centrifugation, and resuspended in fresh media for continued propagation, or resuspended in an appropriate buffer for administration to a subject.

An aspect of the invention features a method for therapeutic administration to a subject in need of treatment with MSCs comprising; i)obtaining MSCs, for example by isolating the cells from a sample, ii) culturing the cells in a non-adherent manner to generate an expanded population of cells, and iii) administering the cells to the subject. In an embodiment, the MSCs are administered to an individual having a condition or disease susceptible to treatment with MSCs

An aspect of the invention provides for the use of MSCs cultured under non-adherent conditions for use as a medicament for the treatment of a condition or disease susceptible to treatment with MSCs.

An aspect of the invention includes kits containing MSCs expanded under non-adherent conditions in appropriate packing material. In an embodiment, the kits further include reagents or materials for propagation of the cells under adherent and/or non-adherent conditions.

In some embodiments of the invention, the methods further include obtaining a sample that contains MSCs, and may further include isolating the MSCs to obtain a substantially purified sample of MSCs.

In some embodiments of the invention, culturing the MSCs increases the expansion of the cells by at least 2 fold, preferably at least 10 fold or 100 fold, more preferably 1000 fold, 10,000 fold, or 100,000 fold. In another embodiment of the

first or second aspects of the invention, the MSCs are maintained in non-adherent culture for at least one week, preferably at least two weeks, at least a month, or at least two months.

In some embodiments of the invention, the cultured MSCs are suitable for administration to a subject, preferably a human subject.

In some embodiments of the invention, the MSCs are allogenic or autologous to the subject to whom the cells are administered.

In an embodiment, the MSCs may express classic surface markers including CD105, CD73 and CD90 but lack expression of CD34 or CD45.

Definitions

By "administering", "therapeutic administration" and the like is meant providing to a human patient a pharmaceutical preparation containing the MSCs, optionally in the form of MSC spheres or foci, or their progeny or derivatives in a suitable formulation. The preferred method of administration can vary depending on various factors, e.g., the components of the pharmaceutical preparation, site of the potential or actual disease, and severity of disease.

By "allogenic" is meant involving, derived from, or being individuals of the same species that are sufficiently unlike genetically to interact antigenically.

By "animal" is meant to be preferably a mammal. A mammal can be human or non-human including, but not limited to laboratory and/or commercially important mammals, such as mouse, rat, rabbit, monkey, dog, cat, pig, cow, sheep, and goat.

By "autologous" is meant derived from the same individual or involving one individual as both donor and recipient.

By "cell culture" is meant grown outside of the body in a dish, flask, or other container in the presence of growth media. Cell culture can be performed with transformed or immortalized cell lines. Cell culture can also be performed with "primary cells" removed from an animal, such as a mammal, and are not transformed or immortalized. Primary cells can be dividing or non-dividing cells. For example, the cells can be bone marrow cells, umbilical cord blood cells, or mesenchymal stem cells.

By a "condition or disease susceptible to treatment with MSCs" is meant a malady that has been demonstrated to be treated using MSCs, for example muscle disease, neural disease, and vascular disease. Theses diseases have been demonstrated to be susceptible to treatment with MSCs. For example, demonstrated therapeutic effects include those shown in US Patents 5,811,094 to promote connective tissue regeneration; 5,858,930 for repair of skin and soft tissue defects; 6,387,369 for cardiac muscle regeneration; 6,875,430 for treatment of immune responses in transplantation; 7,029,666 for muscle and connective tissue repair; 7,097,832 for enhancing blood vessel formation; and 7,160,724 for repair of the brain and spinal cord.

By "effective amount" is an amount sufficient to effect beneficial or desired clinical or biochemical results. An effective amount can be administered one or more times. For purposes of this invention, an effective amount is the amount of MSCs to effect beneficial engraftment of the cells.

By "engraftment" is meant the implantation of cells in the body, and/or replacement of lost or damaged cells with injected cells. The engrafted cells persist in a particular location over time following transplantation of the cells into a mammal (e.g., a human).

By the term "expanded population" is meant a population of cells, e.g., MSCs isolated from bone marrow or other tissue, wherein at least 50% of the cells have divided at least once.

A molecule is a "marker" of a desired cell type if it is found on a sufficiently high percentage of cells of the desired cell type, and found on a sufficiently low percentage of cells of an undesired cell type, such that one can achieve a desired level of purification of the desired cell type from a population of cells comprising both desired and undesired cell types by selecting for cells in the population of cells that have the marker. A marker can be displayed on, for example, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 99% or more of the desired cell type, and can be displayed on fewer than 50%, 45%, 40%, 35%, 30%, 25%, 20%, 15%, 10%, 5%, 1% or fewer of an undesired cell type. It is preferred that a marker be displayed on 90% or more of a desired cell type, or on fewer than 10% of a desired cell type.

A desired cell type is negative for a cell surface-expressed marker or lacks expression of the marker if fewer than 50 marker molecules per cell are present on the cell surface of the desired cell type. Techniques for detecting cell surface-expressed marker molecules are well known in the art and include, e.g., flow cytometry. One skilled in the art can also use enzymatic amplification staining techniques in conjunction with flow cytometry to distinguish between cells expressing a low number of a marker molecule and cells that do not express the marker molecule (see, e.g., Kaplan, *Front. Biosci.* 7:c33-c43, 2002; Kaplan *et al.*, *Amer. J. Clin. Pathol.* 116:429-436, 2001; and Zola *et al.*, *J. Immunol. Methods* 135:247-255, 1990).

By "non-adherent matrix" is meant a material which cells can grow in, or on a material that prevents adhesion to a cell culture container surface. For example, growing cells in a non-adherent matrix (e.g., Hydrogel, BD Biosciences or Matrigel®, BD Biosciences) can prevent attachment to a cell culture container surface. MSCs may adopt their typical fibroblast-like shape on the matrices, but do not attach to the plastic culture surface. Alternatively a non-adherent matrix can be understood to be a matrix that the cells can grow on, but do not attach tightly to (e.g., agarose, or Teflon®). With such matrices, the MSCs retain a rounded, rather than fibroblast shape which they obtain when grown on plastic. In a preferred embodiment, the non-adherent matrix is preferably biocompatible such that it can be

administered to a subject for transplant without separation from the matrix. Alternatively, the matrix can be of a size, shape, and resiliency that readily allows for removal of the cells from the matrix (e.g., Teflon®) to allow the cells to be administered to a subject.

By "mesenchymal stem cell" (MSC) is meant an adherent stroma cell, for example from a biological sample such as bone marrow or umbilical cord blood, isolated by methods such as those provided herein and by US Patents 5,486,359; 5,654,186; 5,827,735; 5,858,390; 5,906,934; 5.908,784; 5,965,436; and 7,060,494. Such cells have been characterized by being multipotent stem cells that have the capacity to differentiate into osteoblasts, adipocytes and chondrocytes in vitro and express the surface antigens CD105, CD73 and CD90, but not CD45 or CD34 (Dominici et al, *Cytotherapy* 8:315-317, 2007)

By a "muscle cell" is meant a skeletal, smooth, or cardiac cell.

By "muscle disease" is meant a disease or disorder that affects or involves the musculature, e.g., cardiac, smooth, or skeletal muscles. Examples of muscle diseases include neuromuscular disease, e.g., muscular dystrophy (e.g., Duchenne muscular dystrophy (DMD), Becker muscular dystrophy (BMD), Limb-girdle muscular dystrophy, and congenital muscular dystrophy), congenital myopathy, and myasthenia gravis, cardiomyopathy, e.g., heart disease, aortic aneurysm (Marfan's disease), cardiac ischemia, congestive heart failure, heart valve disease, and arrhythmia, and metabolic muscle diseases.

By a "neural cell" is meant a neuron (e.g., a sensory neuron, a motor neuron, or an interneuron) or a support cell of the central or peripheral nervous system. Examples of neurons include pyramidal cells, Betz cells, stellate cells, horizontal cells, granule cells, Purkinje cells, spinal motor neurons, and ganglion cells. Examples of support cells include glial cells, oligodendroglial cells, astrocytes, satellite cells, microglial cells, and Schwann cells.

By "neural disease" is meant a disease or disorder that affects or involves the central or peripheral nervous system. Examples of neural diseases include multi-

infarct dementia (MID), vascular dementia, cerebrovascular injury, Alzheimer's disease (AD), neurofibromatosis, Huntington's disease, amyotrophic lateral sclerosis, multiple sclerosis, stroke, Parkinson's disease (PD), pathologies of the developing nervous system, pathologies of the aging nervous system, and trauma, e.g., head trauma. Other examples of neural diseases are those that affect tissues of the eye, e.g., the optic stalk, retinal layer, and lens of the eye, and the inner ear. In certain embodiments, the patient may have suffered a neurodegenerative disease, a traumatic injury, a neurotoxic injury, ischemia, a developmental disorder, a disorder affecting vision, an injury or disease of the spinal cord, or a demyelinating disease.

By "non-adherent culture" is meant herein as a method of propagation of cells *in vitro* as in a container in the presence of growth media in a manner in which the cells do not attach to the surface of the container such that a substantial portion of the cells can be removed from the surface of the container by mechanical manipulations that do not cause significant damage to the cells. It is understood that the cells can still be retained in or on a non-adherent matrix (e.g., on Hydrogel spheres) and be removed from the surface of the container. Such manipulations include, for example, gentle agitation, massage, or manual manipulation of the container, or rinsing the container with growth media. As used herein, a substantial portion of the cells to be removed is at least 70%, preferably at least 75%, 80% or 85%, more preferably at least 90% or 95%. Manipulations that cause damage to the cells can be identified by determining the viability of the cells before and after manipulation, for example by trypan blue staining. Mechanical manipulations should cause damage to less than 20%, preferably less than 15%, or 10%, more preferably less than 5%, 2%, or 1% of the cells.

By "obtaining" as in "obtaining an agent" or "obtaining a cell" refers to purchasing, synthesizing, or otherwise procuring an agent or cell. Cells can be obtained, for example, from an animal including human and non-human animals. Cells can also be obtained from cell and tissue repositories.

By "prevent," "preventing," "prevention," "prophylactic treatment" and the like is meant reducing the probability of developing a disorder or condition in a subject, who does not have, but is at risk of or susceptible to developing a disorder

or condition. Prevention or prophylactic treatment can require administration of more than one dose of the compositions of the invention.

By "propagate", "passage", and the like is meant increasing the volume of a cell culture and/or decreasing the amount of cells in a specific culture volume by diluting cells in at least some fresh growth media to allow for maintenance and/or expansion of the cell population.

By "sample" or "biological sample" is meant any biological sample obtained from an individual, body fluid, cell line, tissue culture, or other source.

By "stem cell" or "pluripotent stem cell," which can be used interchangeably, is meant a cell having the ability to give rise to two or more cell types of an organism.

By "subject" is meant a vertebrate, preferably a mammal, more preferably a human.

By "substantially purified" is meant that the desired cells (e.g., MSCs) are enriched by at least 30%, more preferably by at least 50%, even more preferably by at least 75%, and most preferably by at least 90% or even 95%.

By "transgene" is meant any piece of a nucleic acid molecule (for example, DNA) that is inserted by artifice into a cell transiently or permanently, and becomes part of the organism if integrated into the genome or maintained extrachromosomally. Such a transgene may include a gene that is partly or entirely heterologous (foreign) to the transgenic organism, or may represent a gene homologous to an endogenous gene of the organism. The transgene may be introduced into the organism from which the MSCs are isolated. Alternatively, the transgene may be introduced using viral vectors, such as retroviral vectors (See, e.g., Gnecchi et al., 2006).

By "transgenic cell" is meant a cell containing a transgene. For example, a cell transformed with an expression vector operably linked to a heterologous nucleic acid molecule can be used to produce a population of cells having altered phenotypic

characteristics. A cell derived from a transgenic organism is also a transgenic cell so long as the cell contain the transgene.

By "transplant" or "transplanting" is meant administering one or more cells (or parts thereof), cell products, tissue, or cell culture products derived from cells that are grafted into a human host. For example, a transplant can include an MSC transplant.

By "treatment" is meant an approach for obtaining beneficial or desired clinical results. For the purposes of this invention, beneficial or desired clinical results include, but are not limited to, alleviation of symptoms, diminishment of extent of disease, stabilization (i.e., not worsening) of a state of disease, delay or slowing of disease progression, amelioration or palliation of the disease state, and remission (whether partial or total), whether detectable or undetectable. "Treatment" can also mean prolonging survival as compared to expected survival if not receiving treatment. "Treatment" refers to both therapeutic treatment and prophylactic or preventative measures. Those in need of treatment include those already with the disorder as well as those in which the disorder is to be prevented. "Palliating" a disease means that the extent and/or undesirable clinical manifestations of a disease state are lessened and/or the time course of the progression is slowed or lengthened, as compared to a situation without treatment. Typically, the "treatment" entails administering an effective dose of MSCs to the patient to regenerate tissue.

By a "vascular cell" is meant an endothelial cell. Endothelial cells line the blood and lymph vessels and are present in and play a key role in the development of organs, such as the brain, heart, liver, pancreas, lungs, spleen, stomach, intestines, and kidneys.

By "vascular disease" is meant a disease or disorder that affects or involves the vasculature. Examples of vascular disease include peripheral vascular disease, peripheral arterial disease, venous disease (e.g., deep vein thrombosis), ischemia, cardiovascular disease, tissue organ engraftment rejection, or sequelae of ischemic reperfusion injury. In still another embodiment, the peripheral vascular disease is

atherosclerosis, thromboembolic disease, or Buerger's disease (thromboangiitis obliterans). In a further embodiment, the cardiovascular disease is myocardial infarction, heart disease, or coronary artery disease.

As used herein, "a", "an", and "the" are understood to be either singular or plural unless otherwise obvious from context.

As used herein, "or" is meant to be inclusive unless otherwise obvious from context.

As used herein, ranges are understood to include all values within the range. For example, 1 to 50 is understood to mean 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, and 50. A series of values are understood to represent a range, and thereby all of the values within the range unless otherwise obvious from context.

Brief Description of the Drawings

FIGURES 1A-1B are images of MSC harvested from plastic adherent culture of MSC by trypsinization and then cultured for 1 week in (A) plastic tissue culture dish, (magnification 100x), or (B) grown in a double layer agarose culture (magnification 100x), or cultured for 2 weeks (C) in liquid culture above a single layer of agarose to prevent adherence to plastic (magnification 100x)

FIGURE 2A-2B is an image of MSC spheres generated in culture of MSCs in Teflon® bags (A) grown in culture for 2 weeks (mag 100x) and (B) for 6 weeks (mag 100x).

FIGURE 3 is an image of proliferating MSCs in hydrogel for 2 weeks (mag 100x)

FIGURE 4 is an image of MSCs in a tissue culture flask after seven passages in Teflon® bags and then transferred to a plastic culture flask. The MSC spheres adhered to the surface of the flask within 2 to 3 days and obtained a morphology essentially identical to that observed in cells passaged in adherent cultures (mag 100x).

Detailed Description

Mesenchymal stem cells have been demonstrated to be useful in the therapeutic methods for the repair and regeneration of tissue, especially muscle tissue, including cardiac tissue. This is somewhat surprising as MSCs have been demonstrated to be quiescent after injection, have low engraftment into tissue other than bone, and to have a very low persistence after injection.

Mesenchymal stem cells are adherent cells, and can be selected for growth in culture by their ability to adhere to tissue culture containers (i.e., plastic). In culture, cells are propagated by repeated rounds of trypsinization and replating, effectively selecting for cells that are adherent. The observed low level of engraftment and cell division *in vivo* may be due to the *in vitro* methods of propagation of the MSCs in adherent cultures, as no comparable surfaces are available *in vivo*, for example in muscle, vascular, and neural cells.

The invention provides methods for mesenchymal stem cells (MSCs) growth in non-adherent culture, eliminating the need for trypsinization in propagation of MSCs. The non-adherent culture methods of the invention allow for the propagation of MSCs that may more readily engraft into recipient tissue and be more viable for longer periods after transplant as they do not require a surface to which they can adhere to divide.

The non-adherent culture methods of the invention also allow for propagation of cells in a less resource intensive manner by allowing the cells to be grown in larger numbers in the same culture container area as the cells do not need to all grow in the same plane of the culture container as with adherent cells.

The invention provides culture methods that enable the generation of MSC in non-adherent foci in various support matricies. MSCs grown under these conditions can be passaged without trypsinization. Methods include growth of cells encapsulated in matrices such as Hydrogel and Matrigel®, on or between layers of agarose, or in Teflon® bags. Cells can grow in contact with the non-adherent matrices, but do not adhere to plastic culture containers. The lack of adherence to a

surface is notable in the MSCs grown on agarose or in Teflon® bags as can be determined by the maintenance of their rounded shape. MSCs grown in adherent cultures on plastic adopt an elongated, fibroblastic shape (see, e.g., compare Figure 1A with Figures 1B-1C and 2A-B).

Mesenchymal stem cells have been cultured for up to 10 passages and can be subcultured without the need of treatment with trypsin. The non-adherent cells express similar surface markers as cells grown under adherent conditions (e.g., CD105), and they maintain their ability to differentiate into multiple cell types. Optimal growth of the cells is stimulated by basic fibroblast growth factor (bFGF) and other growth factors including stem cell factor (SCF) and vascular endothelial growth factor (VEGF).

Growth of non-adherent MSCs in Teflon® bags provides an additional advantage for translation into therapeutic applications as the MSCs can be cultured by massaging the bag to detach the cells from the surface. When the MSCs are detached the can be maintained as MSC spheres by regular massaging of the bag and inversion of the bag for continued incubation. Performance of this manipulation about twice daily allows for the MSC spheres to increase in size, and for the MSCs to continue to proliferate and expand. The cells can readily be removed from the culture media by centrifugation and resuspension into an appropriate buffer for injection (e.g., phosphate buffered saline (PBS), physiological saline solution) without the need to remove the cells from a less sturdy non-adherent surface (e.g., Matrigel® or agarose) and without the use of trypsin which would need to be removed from the cells prior to administration.

It is understood that the initial source of and method of isolation of the MSCs to be grown by the culture methods of the invention is not a limitation of the invention. A number of methods of isolation of MSCs are known to those skilled in the art including, but not limited to, those set forth in US Patents 5,486,359; 5,654,186; 5,827,735; 5,858,390; 5,906,934; 5.908,784; 5,965,436; and 7,060,494.

It is further understood that the methods provided herein can be used to culture both wild-type and transgenic MSCs such as those taught, for example in US

Patent 5,591,625 or in Gnecchi et al. (both incorporated herein by reference). Transgenic MSCs can be isolated from transgenic animals or can be transduced using vectors, including viral vectors, for the insertion of expression constructs into the cells.

Mesenchymal stem cells cultured by the methods of the invention can be used for any of a number of research or therapeutic purposes. For example, a number of therapeutic methods using MSCs are known, such as those taught in US Patents 5,811,094 for connective tissue regeneration; 5,858,930 for repair of skin and soft tissue defects; 6,387,369 for cardiac muscle regeneration; 6,875,430 for treatment of immune responses in transplantation; 7,029,666 for muscle and connective tissue repair; 7,097,832 for enhancing blood vessel formation; and 7,160,724 for repair of the brain and spinal cord (all of which are incorporated herein by reference).

Mesenchymal stem cells cultured by the methods of the invention can be used for the generation of cultured media to promote the growth of cells, for therapeutic uses, or for research purposes to identify secreted growth factors that may be responsible for the beneficial therapeutic effects provided by MSCs.

Mesenchymal stem cells cultured by methods of the invention can be incorporated into a kit including the cells in a container with appropriate packing material. The kit can further contain reagents and/or materials for culturing MSCs in adherent and/or non-adherent manner(s).

EXAMPLE 1- Isolation of MSCs from human bone marrow

Human bone marrow cells were obtained from normal donors following informed consent under an Institutional Review Board approved protocol. The mononuclear cell fraction of the bone marrow was isolated on a Ficoll gradient and plated in a T150 Corning (Acton, MA) tissue culture flask at 1-5 x 10^6 cells/ml in α-MEM media containing 20% fetal calf serum (FCS). The cells were incubated in a humidified environment at 5% CO_2 at 37°C. The media was changed weekly. Adherent cells were grown in culture and passaged using trypsin when confluent.

EXAMPLE 2- Culture of MSCs in Hydrogel

MSCs were isolated and propagated as set forth above. MSCs were collected from adherent, confluent cultures using trypsin and encapsulated in Hydrogel (Becton Dickson) following the manufacturer's instructions. The encapsulated MSCs were cultured in α-MEM + 20% FCS in T75 culture flasks. At regular intervals, the non-adherent cells were passaged by removing the supernatant, centrifuging the Hydrogel/MCS mixture, and resuspending the cells in growth media. As shown in Figure 3, MSCs encapsulated in the Hydrogel proliferated and maintained a fibroblast-like morphology. Cells encapsulated in Matrigel™ gave comparable results.

EXAMPLE 3- Culture of MSCs in agarose

Single layer agarose cultures were established in 100 mm culture dishes on preformed layers of 0.5% agarose for double layer, and 1% agarose for single layer agarose in α-MEM + 30% FCS. MSCs were harvested from confluent cell cultures by trypsinization and resuspended in α-MEM + 20% FCS. The MSCs were added in 10 ml of α-MEM + 20% FCS above the agarose layer. The non-adherent cells were passaged by removing the supernatant from the agarose underlay. The cells were centrifuged and the supernatant discarded. The cells were resuspended in fresh media and replated over the agarose underlay. Double layer agarose cultures were generated by incorporating the cells into a top agarose layer (0.66%).

Figure 1B shows cells cultured in a double layer agarose culture in the top agarose layer. The MSCs could be visualized as single, round cells. No proliferation was observed. However, when the cells were plated in a liquid phase in α-MEM + 20% FCS on a lower layer of 1% agarose to prevent adherence, the MSC formed spheres and proliferated as shown in Figure 1C. Cells were passaged multiple times.

EXAMPLE 4- Culture of MSCs in Teflon® bags

MSCs were harvested from confluent cell cultures by trypsinization and resuspended in 50 ml of α-MEM + 20% FCS. The cells were placed in 100 ml

Teflon® bags (American Fluoroceal Corp, Gaithersburg, MD) and cultured. At weekly intervals the bags were harvested, the cells were centrifuged, resuspended in fresh media and placed into new Teflon® bags.

MSCs can be cultured by massaging the bag to detach the cells from the surface. When the MSCs are detached the can be maintained as MSC spheres by regular massaging of the bag and inversion of the bag for continued incubation. Performance of this manipulation twice daily allows for the MSC spheres to increase in size, and for the MSCs to continue to proliferate and expand (see, Figure 2).

The culture methods have been replicated beginning with bone marrow harvested from pig. Non-adherent cultures of pig MSC have now been generated for animal studies. One hundred million non-adherent pig MSCs were generated after 3 weeks of culture in Teflon® bags.

EXAMPLE 5- Adherence of cells to plastic after culture under non-adherent conditions

Culturing of cells under non-adherent conditions does not alter the ability of the MSCs to adhere when provided with an appropriate substrate. Figure 4 shows cells grown in a tissue culture flasks after seven passages in Teflon® bags. The morphology of the cells appears to be identical to that of MSCs propagated continuously in adherent culture.

EXAMPLE 6- Stimulation of MSCs by growth factors

The effect of several growth factors on MSC sphere proliferation were evaluated, including macrophage colony stimulating factor (M-CSF), vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF), stem cell factor (SCF), and media conditioned by 5637 cells (a human bladder carcinoma cell line that constitutively secretes functional cytokines). Optimal growth factors for of MSC spheres was addition of 10% 5637 conditioned media to the cells. However, as this source of growth factors is limited for clinical applications, the effects of recombinant growth factors were also analyzed. A combination of recombinant

human bFGF (50 ng/ml) and recombinant human SCF (100 ng/ml) resulted in maximal proliferation of MSCs and sphere formation.

EXAMPLE 7- Transplantation of non-adherent MSCs for the treatment of cardiac infarction

MSCs are isolated from rat bone marrow by standard Ficoll gradient followed by adherent culture methods. After expansion of the cells, the culture is split. A portion of the cells are maintained in adherent culture, and a portion of the cells are transferred to Teflon® bags for propagation. Cells in Teflon® bags are manipulated twice daily to promote growth of MSC spheres, and media is changed as needed. Adherent cells are propagated using trypsin as needed. Cells can include a marker such as GFP or beta-galactosidase to facilitate identification of the transplanted cells at the end of the experiment. Cells are collected and resuspended in an appropriate buffer for administration (e.g., normal saline).

Age and sex matched laboratory rats of a single type are divided into four groups, sham myocardial infarction (MI), adherent MSC treated, non-adherent MSC treated, and normal saline. In all but the sham MI group, ligation of the left coronary artery is performed using well known methods (see, e.g., Gnecchi et al). Briefly, animals are anesthetized and a left thoracotamy is performed under artificial respiration. The heart is accessed through the intercostal space, the pericardial sac is cut, and the heart is exteriorized through the space. The left coronary artery is legated with a silk suture about midway between the left atrium and the apex of the heart and EKG is recorded to confirm the presence of infarction. In sham operated animals, the artery is not legated. One hour after infarction, an equal number of adherent or non-adherent MSCs are injected into a total of five sites per infarct area. Normal saline is injected into the infarct area in the control animals.

Cardiac function is analyzed at regular intervals after the surgery and administration of the cells, for example by EKG. Either throughout the course of the experiment, or at the end of the experiment, rats are euthanized and hearts are excised. Analysis is performed to determine any of a number of outcomes including, but not limited to, infarct area, engraftment of MSCs into the infarct area,

angiogenesis in the infarct area, and/or mRNA or protein expression. Methods for performing such analyses are known to those skilled in the art. The therapeutic effect of the cells grown in adherent culture and non-adherent culture are compared to each other and to control animals.

It is understood that comparable experiments can be performed using different animals including, for example, pigs.

The invention may be embodied in other specific forms without departing from the spirit or essential characteristics thereof. The foregoing embodiments are therefore to be considered in all respects illustrative rather than limiting on the invention described herein. Scope of the invention is thus indicated by the appended claims rather than by the foregoing description, and all changes which come within the meaning and range of equivalency of the claims are therefore intended to be embraced therein.

All references, patents, patent applications, and GenBank numbers cited are incorporated herein by reference in their entirety.

CLAIMS

1. A method for propagation of a non-adherent culture of mesenchymal stem cells (MSCs) comprising expanding MSCs in or on a non-adherent matrix.

2. The method of claim 1, comprising encapsulation of MSCs in Matrigel™ or Hydrogel.

3. The method of claim 1, comprising the cells propagated on agarose or on Teflon®.

4. The methods of any of claims 1 to 3, wherein the cells are propagated in the non-adherent culture without the use of trypsin.

5. The methods of any of claims 1 to 4, comprising mechanical manipulation of the MSCs.

6. The method of any of claims 1 to 5, further comprising a biological sample containing MSCs.

7. The method of claim 6, further comprising isolating the MSCs from the biological sample containing the MSCs.

8. The method of claim 7, wherein the isolated MSCs are substantially purified.

9. The method of any of claims 1-8, wherein the MSCs are expanded at least 2-fold, 10-fold, 100-fold, 1000-fold, 10,000-fold, or 100,000 fold.

10. The method of any of claims 1-9, wherein the MSCs are suitable for administration to a subject.

11. The method of claim 10, wherein the subject is a human subject.

12. The method of any of claims 1-11 wherein the MSCs are propagated in non-adherent culture for at least a week, at least 2 weeks, at least a month, or at least 2 months.

13. A method for treatment of a subject having a disease or condition susceptible to treatment with MSCs comprising administration of MSCs grown in a non-adherent culture of any of the methods of claims 1 to 12.

14. The method of claim 13, wherein the disease or condition susceptible to treatment with MSCs is selected from the group consisting of muscle disease, neural disease, and vascular disease.

15. The method of claim 13 or 14, wherein the MSCs are allogenic or autologous to the subject.

16. The method of any of claims 13 to 15, wherein the subject is human.

17. The use of a MSC propagated by any of the methods of claims 1 to 12 for use as a therapeutic agent for the treatment of a disease or condition susceptible to treatment with MSCs.

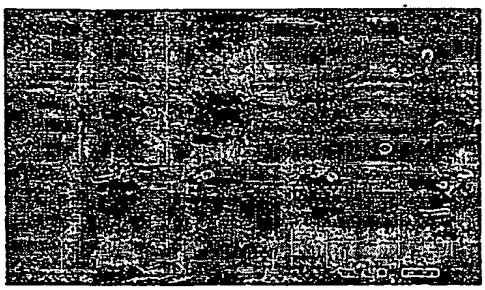
18. The use of claim 17, wherein the disease or condition susceptible to treatment with MSCs is selected from the group consisting of muscle disease, neural disease, and vascular disease.

19. A kit comprising an MSC of any of claims 1 to 12 and appropriate packing material.

20. The kit of claim 19, further comprising reagents or supplies for propagation of MSCs under adherent or non-adherent conditions or both.

FIGURE 1

A



B

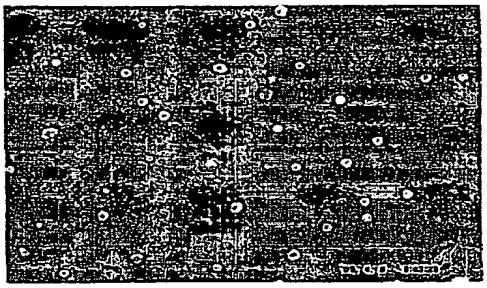


FIGURE 1C

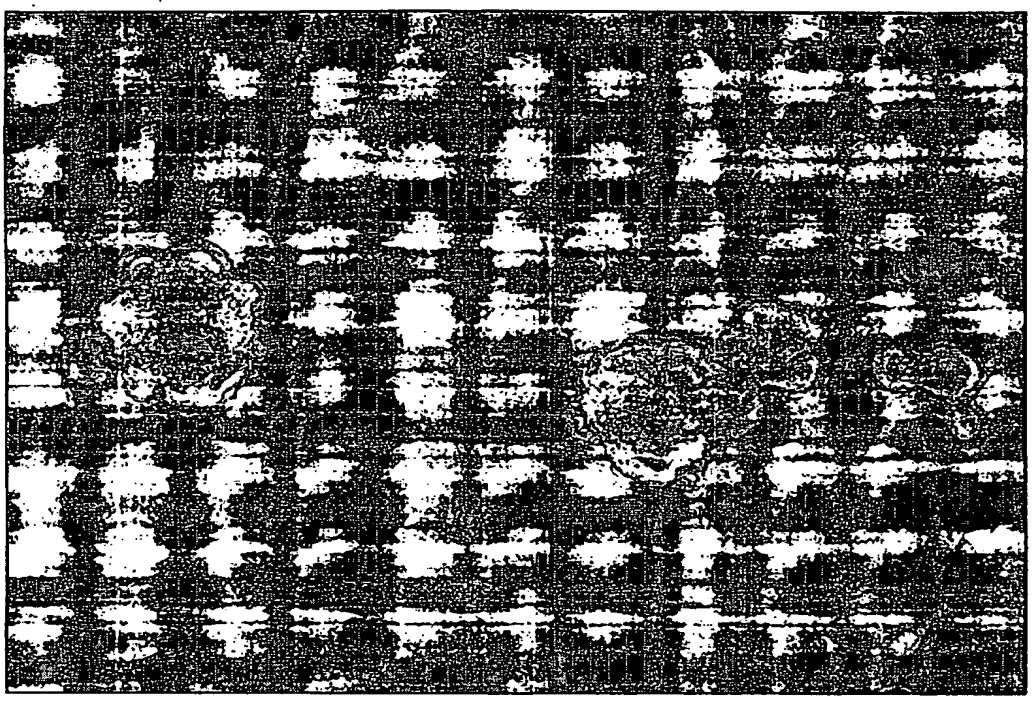


FIGURE 2A

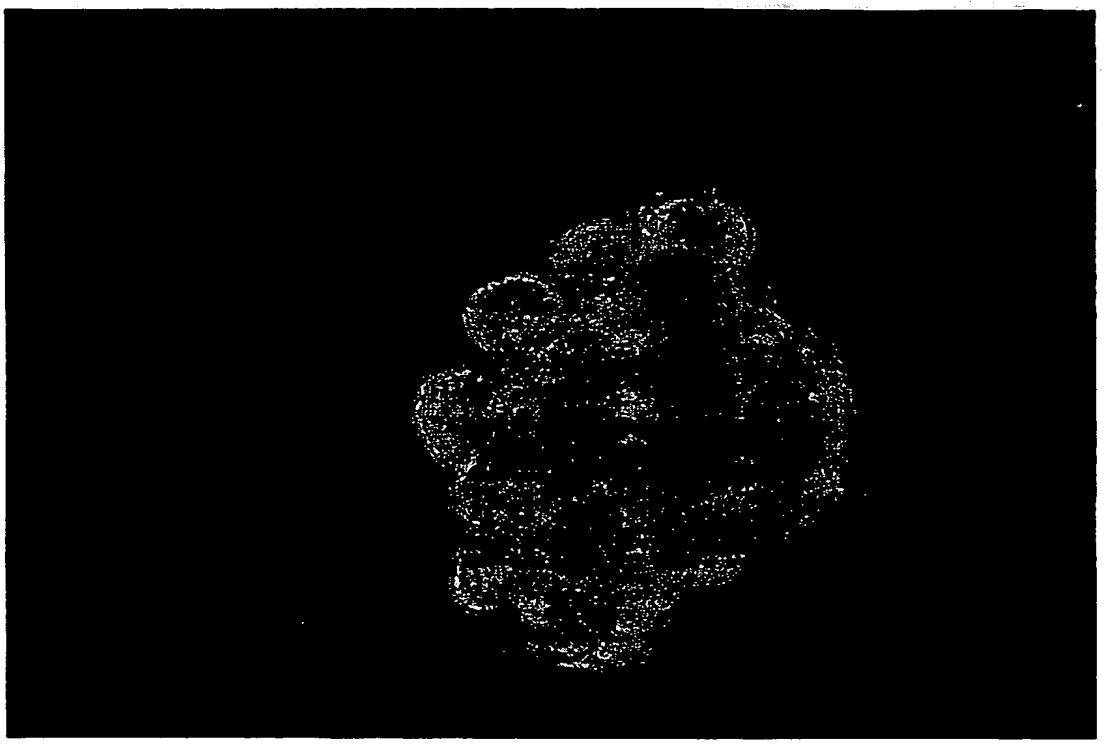


FIGURE 2B

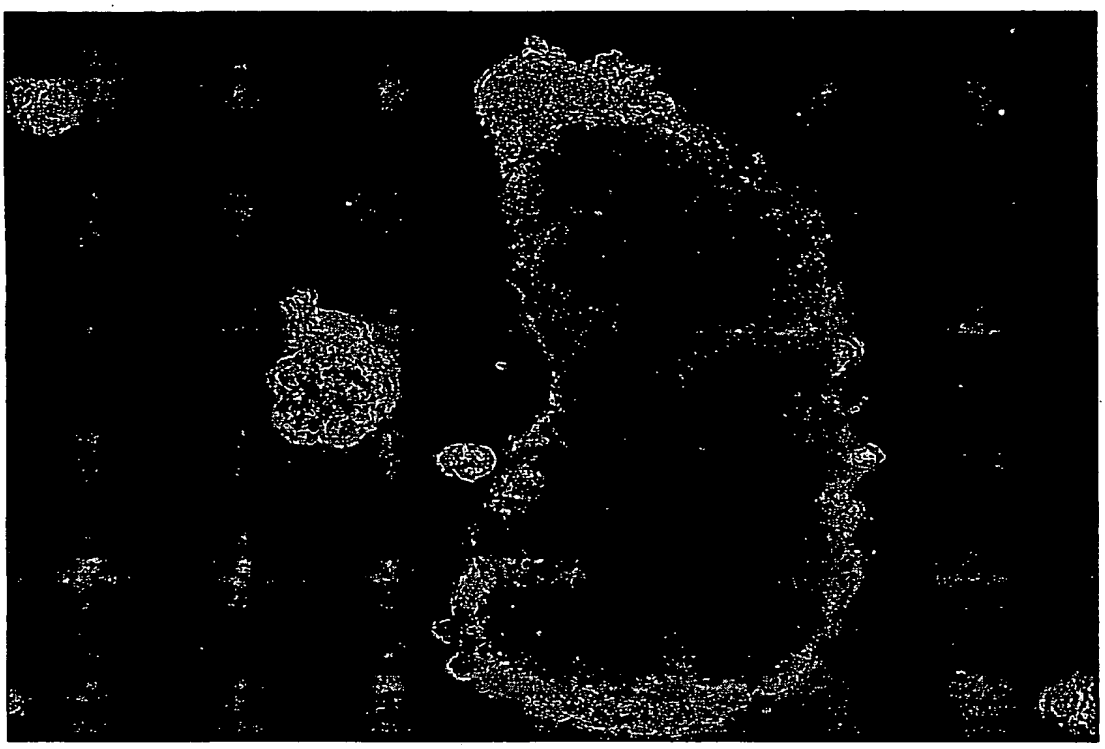
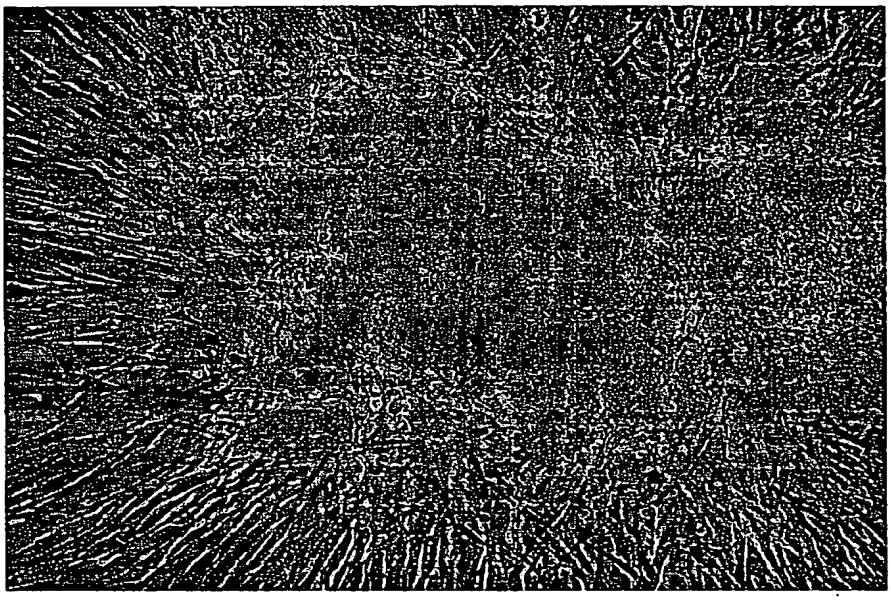


FIGURE 3

FIGURE 4



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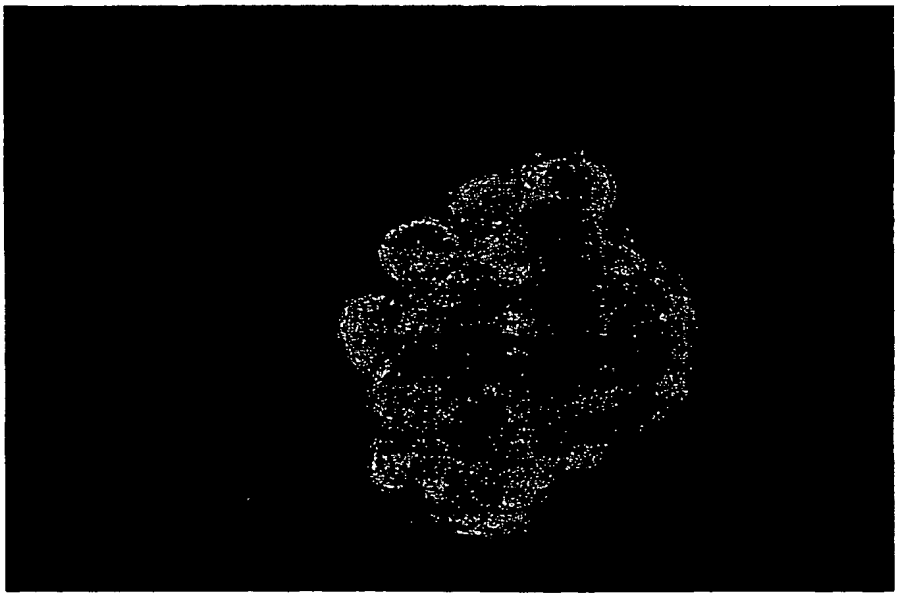
(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*
— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

(88) Date of publication of the international search report:
14 February 2008

(54) Title: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS



(57) Abstract: The invention provides methods for expanding mesenchymal stem cells (MSCs) in non-adherent cultures. The methods include the propagation of MSCs in or on non-adherent matrices. The invention further provides administration and the use of cells propagated by the method of the invention for administration and preparation of a therapeutic agent. The invention further provides kits including cells propagated by the methods of the inventions.

WO 2007/136760 A3

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 07/11921

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - C12N 5/08 (2007.01)
USPC - 435/366
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)
USPC: 435/366

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
USPC: 435/7.21, 435/372, 435/440, 424/93.7 (text search-see terms below)

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
PubWEST(USPT,PGPB, USOC, EPAB,JPAB); Google Scholar- propagat$, expand$, grow$, non-adherent, culture, non-adherent culture, mesenchymal, mesenchymal stem cell, stem cells, matrigel, hydrogel, matrix, monlece ian, without trypsin, agarose, teflon

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 2005/0265980 A1 (CHEN et al.) 01 December 2005 (01.12.2005) para [0007], [0047], [0088]	1 ------ 2-4
Y	US 2004/0092011 A1 (WILKISON et al.) 13 May 2004 (13.05.2004), para [0109], [0142]	2 and 4
Y	US 2005/0013804 A1 (KATO et al.) 20 January 2005 (20.01.2005), para [0028]	3

☐ 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
[08 November 2007 (08.11.2007)]

Date of mailing of the international search report
13 DEC 2007

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:
Lee W. Young

PCT Helpdesk: 571-272-4300
PCT OSP: 571-272-7774

Form PCT/ISA/210 (second sheet) (April 2007)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 07/11921

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.: 5-20
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:

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.:

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.

IAP20Rec'd PCT 17 NOV 2008

Docket No.: 68324(71699)
(PATENT)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Patent Application of:
Ian McNiece

Application No.: Continuation of PCT/US2007/011921

Confirmation No.: N/A

Filed: Herewith

Art Unit: N/A

For: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Examiner: Not Yet Assigned

FIRST PRELIMINARY AMENDMENT

MS PCT
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

INTRODUCTORY COMMENTS

Prior to examination on the merits, please amend the above-identified U.S. patent application as follows:

Amendments to the Claims are reflected in the listing of claims which begins on page 2 of this paper.

Remarks begin on page 4 of this paper.

AMENDMENTS TO THE CLAIMS

1. (original) A method for propagation of a non-adherent culture of mesenchymal stem cells (MSCs) comprising expanding MSCs in or on a non-adherent matrix.

2. (original) The method of claim 1, comprising encapsulation of MSCs in Matrigel™ or Hydrogel.

3. (original) The method of claim 1, comprising the cells propagated on agarose or on Teflon®.

4. (currently amended) The methods of claim 1~~any of claims 1 to 3~~, wherein the cells are propagated in the non-adherent culture without the use of trypsin.

5. (currently amended) The methods of claim 1 ~~any of claims 1 to 4~~, comprising mechanical manipulation of the MSCs.

6. (currently amended) The method of claim 1 ~~any of claims 1 to 5~~, further comprising a biological sample containing MSCs.

7. (original) The method of claim 6, further comprising isolating the MSCs from the biological sample containing the MSCs.

8. (original) The method of claim 7, wherein the isolated MSCs are substantially purified.

9. (currently amended) The method of claim 1 ~~any of claims 1-8~~, wherein the MSCs are expanded at least 2-fold, 10-fold, 100-fold, 1000-fold, 10,000-fold, or 100,000 fold.

10. (currently amended) The method of claim 1 ~~any of claims 1-9~~, wherein the MSCs are suitable for administration to a subject.

11. (original) The method of claim 10, wherein the subject is a human subject.

12. (currently amended) The method of claim 1 ~~any of claims 1-11~~ wherein the MSCs are propagated in non-adherent culture for at least a week, at least 2 weeks, at least a month, or at least 2 months.

13. (currently amended) A method for treatment of a subject having a disease or condition susceptible to treatment with MSCs comprising administration of MSCs grown in a non-adherent culture of claim 1 ~~any of the methods of claims 1 to 12~~.

14. (original) The method of claim 13, wherein the disease or condition susceptible to treatment with MSCs is selected from the group consisting of muscle disease, neural disease, and vascular disease.

15. (currently amended) The method of claim 13 ~~or 14~~, wherein the MSCs are allogenic or autologous to the subject.

16. (currently amended) The method of claim 13 ~~any of claims 13 to 15~~, wherein the subject is human.

17-18. (cancelled)

19. (currently amended) A kit comprising an MSC of claim 1 ~~any of claims 1 to 12~~ and appropriate packing material.

20. (original) The kit of claim 19, further comprising reagents or supplies for propagation of MSCs under adherent or non-adherent conditions or both.

REMARKS

Claims 4-6, 9, 10, 12, 13, 15, 16 and 19 have been amended to remove multiple claim dependency, and claims 17 and 18 have been cancelled without prejudice. The amendments are non-substantive.

Early consideration and allowance of the application are earnestly solicited.

Dated: November 17, 2008

Respectfully submitted,

By _____

Peter F. Corless
Registration No.: 33,860
EDWARDS ANGELL PALMER & DODGE LLP
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Boston, Massachusetts 02205
(617) 517-5557
Attorneys/Agents For Applicant

PTO/SB/08a (09-08)
Approved for use through 10/31/2008. OMB 0651-0031
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Substitute for form 1449A/PTO

INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Use as many sheets as necessary)

Sheet	1	of	2

Complete if Known	
Application Number	Not Yet Assigned
Filing Date	Herewith
First Named Inventor	Ian Mcniece
Art Unit	N/A
Examiner Name	Not Yet Assigned
Attorney Docket Number	68324(71699)

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document	Pages, Columns, Lines, Where Relevant Passages or Relevant Figures Appear
	AA*	US-20050265980	12-01-2005	Chen et al.	
	AB*	US-20040092011	05-13-2004	Wilkison et al.	
	AC*	US-20050013804	01-20-2005	Kato et al.	

FOREIGN PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Foreign Patent Document Country Code[3]-Number[4]-Kind Code[5] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document	Pages, Columns, Lines, Where Relevant Passages Or Relevant Figures Appear	T[6]

Examiner Signature		Date Considered	

*EXAMINER: Initial if reference considered, whether or not citation is in conformance with MPEP 609. Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. * CITE NO.: Those application(s) which are marked with an single asterisk (*) next to the Cite No. are not supplied (under 37 CFR 1.98(a)(2)(iii)) because that application was filed after June 30, 2003 or is available in the IFW. [1] Applicant's unique citation designation number (optional). [2] See Kinds Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English language Translation is attached.

IAP Rec'd PCT 17 NOV 2008

PTO/SB/08b (09-06)
Approved for use through 10/31/2008. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
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INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Use as many sheets as necessary)

Sheet	2	of	2

Complete if Known	
Application Number	Not Yet Assigned
Filing Date	Herewith
First Named Inventor	Ian Mcniece
Art Unit	N/A
Examiner Name	Not Yet Assigned
Attorney Docket Number	68324(71699)

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.), date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]

Examiner Signature		Date Considered	

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[1]Applicant's unique citation designation number (optional). [2]Applicant is to place a check mark here if English language Translation is attached.

DocCode - SCORE

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PATENT APPLICATION SERIAL NO._____

U.S. DEPARTMENT OF COMMERCE
PATENT AND TRADEMARK OFFICE
FEE RECORD SHEET

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01 FC:2631 165.00 DA
02 FC:2633 110.00 DA
03 FC:2641 50.00 DA
04 FC:2617 65.00 DA

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(5/87)

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IAP Rec'd PCT 17 NOV 2008

12/227458

Docket No.: 68324(71699)
(PATENT)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Patent Application of:
Ian Mcniece

Application No.: Continuation of PCT/US2007/011921

Confirmation No.: N/A

Filed: Herewith

Art Unit: N/A

For: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Examiner: Not Yet Assigned

INFORMATION DISCLOSURE STATEMENT (IDS)

MS PCT
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

Pursuant to 37 CFR 1.56, 1.97 and 1.98, the attention of the Patent and Trademark Office is hereby directed to the documents listed on the attached PTO/SB/08. It is respectfully requested that the information be expressly considered during the prosecution of this application, and that the documents be made of record therein and appear among the "Documents Cited" on any patent to issue therefrom.

This Information Disclosure Statement accompanies the new patent application submitted herewith.

In accordance with 37 CFR 1.98(a)(2)(ii), Applicant has not submitted copies of U.S. patents and U.S. patent applications. Applicant submits herewith copies of foreign patents and non-patent literature in accordance with 37 CFR 1.98(a)(2).

In accordance with 37 CFR 1.97(g), the filing of this Information Disclosure Statement shall not be construed to mean that a search has been made or that no other

material information as defined in 37 CFR 1.56(a) exists. In accordance with 37 CFR 1.97(h), the filing of this Information Disclosure Statement shall not be construed to be an admission that any patent, publication or other information referred to therein is "prior art" for this invention unless specifically designated as such.

It is submitted that the Information Disclosure Statement is in compliance with 37 CFR 1.98 and the Examiner is respectfully requested to consider the listed documents.

The Director is hereby authorized to charge any deficiency in the fees filed, asserted to be filed or which should have been filed herewith (or with any paper hereafter filed in this application by this firm) to our Deposit Account No. 04-1105, under Order No. 68324(71699).

Dated: November 17, 2008

Respectfully submitted,

By _____

Peter F. Corless
Registration No.: 33,860
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(617) 517-5557
Attorneys/Agents For Applicant

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

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PCT/US2007/011921

(22) International Filing Date: 18 May 2007 (18.05.2007)

(25) Filing Language: English

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(30) Priority Data:
60/801,661 19 May 2006 (19.05.2006) US

(71) Applicant *(for all designated States except US)*: **THE JOHNS HOPKINS UNIVERSITY** [US/US]; 3400 N. Charles Street, Baltimore, MD 21218 (US).

(72) Inventor; and
(75) Inventor/Applicant *(for US only)*: **MCNIECE, Ian** [AU/US]; 1609 Pot Spring Road, Lutherville, MD 21093 (US).

(74) Agents: **CORLESS, Peter, F.** et al.; Edwards Angell Palmer & Dodge Llp, P. O. Box 55874, Boston, MA 02205 (US).

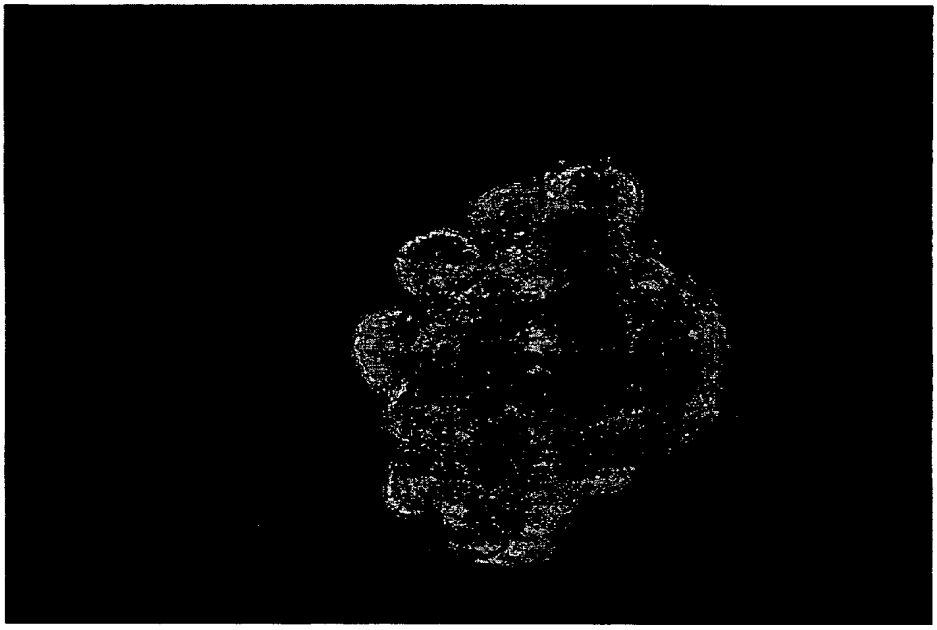
(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *without international search report and to be republished upon receipt of that report*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS



(57) Abstract: The invention provides methods for expanding mesenchymal stem cells (MSCs) in non-adherent cultures. The methods include the propagation of MSCs in or on non-adherent matrices. The invention further provides administration and the use of cells propagated by the method of the invention for administration and preparation of a therapeutic agent. The invention further provides kits including cells propagated by the methods of the inventions.

WO 2007/136760 A2

METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS

Related Applications

This application claims priority to Provisional Patent Application Serial No. 60/801,661, filing date May 19, 2006, which is incorporated herein by reference in its entirety.

Government Support

The invention was made under Grant Number CA088878 from the National Institutes of Health of the United States Government. The Government may have certain rights in relation to the application.

Field Of The Invention

This invention relates to methods of growth of mesenchymal cells under non-adherent conditions. The method allows for expansion of mesenchymal cells in suspension for research or therapeutic uses.

Background Of The Invention

Mesenchymal stem cells are the formative pluripotential blast cells found *inter alia* in bone marrow, blood, dermis, and periosteum that are capable of differentiating into any of the specific types of mesenchymal or connective tissues (i.e. the tissues of the body that support the specialized elements; particularly adipose, osseous, cartilaginous, elastic, and fibrous connective tissues) depending upon various influences from bioactive factors, such as cytokines. In contrast to their hematopoietic counterparts, MSCs are adherent and can be expanded in culture. A number of U.S. Patents, e.g., U.S. Patent Nos. 5,486,359; 5,591,625; 5,736,396; 5,811,094; 5,827,740; 5,837,539; 5,908,782; 5,908,784; 5,942,225; 5,965,436; 6,010,696; 6,022,540; 6,087,113; 5,858,390; 5,804,446; 5,846,796; 5,654,186; 6,054,121; 5,827,735; and 5,906,934 (all of which are incorporated herein by

reference) disclose mesenchymal stem cells (MSC), which can be differentiated into several progenitor cells, for example muscle progenitor cells, connective tissue cell progenitor cells, or hepatic oval cells. Muscle progenitor cells differentiate further into cardiac, skeletal, and smooth muscle cells, whereas the connective tissue cell progenitor may differentiate into bone. The patents above further teach transgenic MSCs that carry a transgene, methods to promote differentiation of MSCs along specific paths, and therapeutic methods including the use of MSCs.

Human MSC (hMSC) can be identified by the presence or absence of specific cell surface markers (Pittenger and Martin, *Circ. Res.* 95:9-20, 2004, incorporated herein by reference). Typically, hMSC can be identified by the presence of surface markers CD13, CD29, CD44, CD49a, b, c, e, f, CD51, CD54, CD58, CD71, CD73, CD90, CD102, CD105, CD106, CDw11, CD120a, CD120b, CD123, CD124, CD126, CDC127, CD140a, CD166, P75, TGFb1R, TGFbIIR, HLA-A, B, C, SSEA-3, SSEA-4, D7; and the absence of surface markers CD3, CD4, CD6, CD9, CD10, CD11a, CD14, CD15, CD18, CD21, CD25, CD31, CD34, CD36, CD38, CD45, CD49d, CD50, CD62E, L, S, CD80, CD86, CD95, CD117, CD133, SSEA-1. Monoclonal antibodies specific to MSCs have also been identified (e.g., US Patents 5,486,359 and 5,811,094). However, most surface markers have been found inadequate as a means to identify stem cells because putative marker(s) may also be found on nonstem cells, or a particular marker may only be expressed on a stem cell at a certain stage or under certain conditions, such as CD34 on hematopoietic stem cells. Nevertheless, surface markers and other attributes are useful in characterizing a stem cell as isolated or cultured, to detect changes in cells in culture over time, and as a means to begin to understand its potential interactions with neighboring cells and the cell environment (Pittenger and Martin, *Circ. Res.* 95:9-20, 2004).

Mesenchymal stem cells can be isolated from a number of cells and tissues including bone marrow, embryonic yolk sac, placenta, umbilical cord, fetal and adolescent skin, and blood, and propagated in culture. Friedenstein et al. (*Exp. Hematol.* 4:267-274, 1976, incorporated herein by reference) initially isolated MSCs

by their adherence to tissue culture surfaces. Similar methods for isolation of MSCs are still commonly used.

Plating studies indicate that MSCs are present at as a rare population of cells in bone marrow, representing about 0.001-0.01% of nucleated cells. However, MSCs can be readily expanded when grown at a very low plating density. Cotler et al. (*Proc. Natl. Acad. Sci. USA.* 97:3213-3218) noted that the number of colonies formed per 100 cells plated remained constant when the density of plating was varied from 0.5 to 12 cells per cm^2. However, the size of the colonies decreased markedly when the cells were plated at higher densities. Colonies of maximal size were obtained when cells were plated at 1.5 to 3.0 cells per cm^2. Plating at such low densities requires the use of large amount of tissue culture dishes, reagents, and space. Methods for culturing of MSCs in a less resource intensive manner is desirable.

Adult bone marrow-derived MSCs engraft in numerous organs and differentiate along tissue-specific lineages when transplanted into animals. They migrate into areas of muscle degeneration to undergo myogenic differentiation in immunodeficient mice. Injection of MSCs directly into infracted swine heart has been shown to induce myocardial regeneration and improved cardiac function (Shake et al., *Ann. Thorac. Surg.* 73:1919-1925, 2002). In addition, MSCs implantation has been demonstrated to induce therapeutic angiogenesis in a rat model of hindlimb ischemia through vascular endothelial growth factor (VEGF) production by MSCs (Al-Khaldi et al., *Gene Ther.* 10:621-629, 2003). In humans, bone marrow-derived MSCs have been used to regenerate the marrow microenvironment after myeloablative therapy. When introduced into the infracted heart, MSCs prevent deleterious modeling and improve recovery. Interestingly, implanted cells do not appear to expand after implantation when engrafted to tissue other than bone. Experiments using MSCs labeled with membrane dyes that would be diluted out after about 3 cell divisions were found months later even in repairing tissue (Pittenger and Martin, *Circ. Res.* 95:9-20, 2004).

Clinical trials have been initiated in several countries to test cell-based therapies for the treatment of the injured heart. However, no studies have

demonstrated incorporation of MSCs into regenerating tissue. It has been suggested that the MSCs exert a therapeutic effect by paracrine actions exerted by the cells through the release of soluble factors (See e.g., Gnecchi et al., *FASEB J.* 20:661-669, 2006; and Nagaya et al., *Circulation.* 112:1128-1135, 2005). This theory is supported by data therein demonstrating that conditioned media from transgenic MSCs overexpressing the prosurvival gene Akt limits hypoxia-induced apoptosis and triggers vigorous spontaneous contraction of adult rat cardiomyocytes in culture. Moreover, injection of concentrated conditioned media from the Akt transgenic MSCs into infracted rat hearts significantly limited infarct size and improved ventricular function relative to controls (Gnecchi et al., 2006).

Studies have demonstrated that upon transplantation of cells into cardiac tissue (e.g., by injection) less than 3% of injected MSCs persist after 2 weeks (Mazhari & Hare, Nature Clinical Practice Cardiovascular Medicine 4: suppl 1; S21-S26, 2007). This may be due to the adherent culture methods used to culture the MSCs. MSCs in bone marrow are able to adhere to bone to allow for proliferation. No comparable surface is present in muscle or many other tissues in which MSCs have been demonstrated to be beneficial. Current culture methods select for cells that are able to adhere to culture dishes through repeated rounds of trypsinization. When transplanted into cardiac tissue for example, MSCs may fail to proliferate due to their inability to adhere to a cardiac tissue surface, minimizing the contribution of MSCs to regenerating tissue.

Methods of culture of MSCs that do not include adherence to a surface and/or reduce the need for multiple rounds of trypsinization for propagation of cells may improve the effects of MSC at sites of injury, for example, by providing cells that are more able to proliferate at the site of injury.

Summary Of The Invention

The invention provides methods for the propagation of mesenchymal stem cells (MSCs) in non-adherent culture, eliminating the need for trypsinization in propagation of MSCs.

Accordingly, an aspect of the invention features a method for culturing MSCs under non-adherent conditions in or on a non-adherent matrix to obtain an expanded population of MSCs. The methods include formation of MSC spheres (MSCS) in or on several different non-adherent matrices, including incorporation of cells into biocompatible matrices such as Hydrogel and Matrigel™; culture of cells on or between layers of agarose; and culture of cells in Teflon® bags. After isolation of MSCs from a sample, the cells are propagated without treatment with trypsin after initial cell selection. MSCS are optionally mechanically manipulated, collected by centrifugation, and resuspended in fresh media for continued propagation, or resuspended in an appropriate buffer for administration to a subject.

An aspect of the invention features a method for therapeutic administration to a subject in need of treatment with MSCs comprising; i)obtaining MSCs, for example by isolating the cells from a sample, ii) culturing the cells in a non-adherent manner to generate an expanded population of cells, and iii) administering the cells to the subject. In an embodiment, the MSCs are administered to an individual having a condition or disease susceptible to treatment with MSCs

An aspect of the invention provides for the use of MSCs cultured under non-adherent conditions for use as a medicament for the treatment of a condition or disease susceptible to treatment with MSCs.

An aspect of the invention includes kits containing MSCs expanded under non-adherent conditions in appropriate packing material. In an embodiment, the kits further include reagents or materials for propagation of the cells under adherent and/or non-adherent conditions.

In some embodiments of the invention, the methods further include obtaining a sample that contains MSCs, and may further include isolating the MSCs to obtain a substantially purified sample of MSCs.

In some embodiments of the invention, culturing the MSCs increases the expansion of the cells by at least 2 fold, preferably at least 10 fold or 100 fold, more preferably 1000 fold, 10,000 fold, or 100,000 fold. In another embodiment of the

first or second aspects of the invention, the MSCs are maintained in non-adherent culture for at least one week, preferably at least two weeks, at least a month, or at least two months.

In some embodiments of the invention, the cultured MSCs are suitable for administration to a subject, preferably a human subject.

In some embodiments of the invention, the MSCs are allogenic or autologous to the subject to whom the cells are administered.

In an embodiment, the MSCs may express classic surface markers including CD105, CD73 and CD90 but lack expression of CD34 or CD45.

Definitions

By "administering", "therapeutic administration" and the like is meant providing to a human patient a pharmaceutical preparation containing the MSCs, optionally in the form of MSC spheres or foci, or their progeny or derivatives in a suitable formulation. The preferred method of administration can vary depending on various factors, e.g., the components of the pharmaceutical preparation, site of the potential or actual disease, and severity of disease.

By "allogenic" is meant involving, derived from, or being individuals of the same species that are sufficiently unlike genetically to interact antigenically.

By "animal" is meant to be preferably a mammal. A mammal can be human or non-human including, but not limited to laboratory and/or commercially important mammals, such as mouse, rat, rabbit, monkey, dog, cat, pig, cow, sheep, and goat.

By "autologous" is meant derived from the same individual or involving one individual as both donor and recipient.

By "cell culture" is meant grown outside of the body in a dish, flask, or other container in the presence of growth media. Cell culture can be performed with transformed or immortalized cell lines. Cell culture can also be performed with "primary cells" removed from an animal, such as a mammal, and are not transformed or immortalized. Primary cells can be dividing or non-dividing cells. For example, the cells can be bone marrow cells, umbilical cord blood cells, or mesenchymal stem cells.

By a "condition or disease susceptible to treatment with MSCs" is meant a malady that has been demonstrated to be treated using MSCs, for example muscle disease, neural disease, and vascular disease. Theses diseases have been demonstrated to be susceptible to treatment with MSCs. For example, demonstrated therapeutic effects include those shown in US Patents 5,811,094 to promote connective tissue regeneration; 5,858,930 for repair of skin and soft tissue defects; 6,387,369 for cardiac muscle regeneration; 6,875,430 for treatment of immune responses in transplantation; 7,029,666 for muscle and connective tissue repair; 7,097,832 for enhancing blood vessel formation; and 7,160,724 for repair of the brain and spinal cord.

By "effective amount" is an amount sufficient to effect beneficial or desired clinical or biochemical results. An effective amount can be administered one or more times. For purposes of this invention, an effective amount is the amount of MSCs to effect beneficial engraftment of the cells.

By "engraftment" is meant the implantation of cells in the body, and/or replacement of lost or damaged cells with injected cells. The engrafted cells persist in a particular location over time following transplantation of the cells into a mammal (e.g., a human).

By the term "expanded population" is meant a population of cells, e.g., MSCs isolated from bone marrow or other tissue, wherein at least 50% of the cells have divided at least once.

A molecule is a "marker" of a desired cell type if it is found on a sufficiently high percentage of cells of the desired cell type, and found on a sufficiently low percentage of cells of an undesired cell type, such that one can achieve a desired level of purification of the desired cell type from a population of cells comprising both desired and undesired cell types by selecting for cells in the population of cells that have the marker. A marker can be displayed on, for example, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 99% or more of the desired cell type, and can be displayed on fewer than 50%, 45%, 40%, 35%, 30%, 25%, 20%, 15%, 10%, 5%, 1% or fewer of an undesired cell type. It is preferred that a marker be displayed on 90% or more of a desired cell type, or on fewer than 10% of a desired cell type.

A desired cell type is negative for a cell surface-expressed marker or lacks expression of the marker if fewer than 50 marker molecules per cell are present on the cell surface of the desired cell type. Techniques for detecting cell surface-expressed marker molecules are well known in the art and include, e.g., flow cytometry. One skilled in the art can also use enzymatic amplification staining techniques in conjunction with flow cytometry to distinguish between cells expressing a low number of a marker molecule and cells that do not express the marker molecule (see, e.g., Kaplan, *Front. Biosci.* 7:c33-c43, 2002; Kaplan *et al.*, *Amer. J. Clin. Pathol.* 116:429-436, 2001; and Zola *et al.*, *J. Immunol. Methods* 135:247-255, 1990).

By "non-adherent matrix" is meant a material which cells can grow in, or on a material that prevents adhesion to a cell culture container surface. For example, growing cells in a non-adherent matrix (e.g., Hydrogel, BD Biosciences or Matrigel®, BD Biosciences) can prevent attachment to a cell culture container surface. MSCs may adopt their typical fibroblast-like shape on the matrices, but do not attach to the plastic culture surface. Alternatively a non-adherent matrix can be understood to be a matrix that the cells can grow on, but do not attach tightly to (e.g., agarose, or Teflon®). With such matrices, the MSCs retain a rounded, rather than fibroblast shape which they obtain when grown on plastic. In a preferred embodiment, the non-adherent matrix is preferably biocompatible such that it can be

administered to a subject for transplant without separation from the matrix. Alternatively, the matrix can be of a size, shape, and resiliency that readily allows for removal of the cells from the matrix (e.g., Teflon®) to allow the cells to be administered to a subject.

By "mesenchymal stem cell" (MSC) is meant an adherent stroma cell, for example from a biological sample such as bone marrow or umbilical cord blood, isolated by methods such as those provided herein and by US Patents 5,486,359; 5,654,186; 5,827,735; 5,858,390; 5,906,934; 5.908,784; 5,965,436; and 7,060,494. Such cells have been characterized by being multipotent stem cells that have the capacity to differentiate into osteoblasts, adipocytes and chondrocytes in vitro and express the surface antigens CD105, CD73 and CD90, but not CD45 or CD34 (Dominici et al, *Cytotherapy* 8:315-317, 2007)

By a "muscle cell" is meant a skeletal, smooth, or cardiac cell.

By "muscle disease" is meant a disease or disorder that affects or involves the musculature, e.g., cardiac, smooth, or skeletal muscles. Examples of muscle diseases include neuromuscular disease, e.g., muscular dystrophy (e.g., Duchenne muscular dystrophy (DMD), Becker muscular dystrophy (BMD), Limb-girdle muscular dystrophy, and congenital muscular dystrophy), congenital myopathy, and myasthenia gravis, cardiomyopathy, e.g., heart disease, aortic aneurysm (Marfan's disease), cardiac ischemia, congestive heart failure, heart valve disease, and arrhythmia, and metabolic muscle diseases.

By a "neural cell" is meant a neuron (e.g., a sensory neuron, a motor neuron, or an interneuron) or a support cell of the central or peripheral nervous system. Examples of neurons include pyramidal cells, Betz cells, stellate cells, horizontal cells, granule cells, Purkinje cells, spinal motor neurons, and ganglion cells. Examples of support cells include glial cells, oligodendroglial cells, astrocytes, satellite cells, microglial cells, and Schwann cells.

By "neural disease" is meant a disease or disorder that affects or involves the central or peripheral nervous system. Examples of neural diseases include multi-

infarct dementia (MID), vascular dementia, cerebrovascular injury, Alzheimer's disease (AD), neurofibromatosis, Huntington's disease, amyotrophic lateral sclerosis, multiple sclerosis, stroke, Parkinson's disease (PD), pathologies of the developing nervous system, pathologies of the aging nervous system, and trauma, e.g., head trauma. Other examples of neural diseases are those that affect tissues of the eye, e.g., the optic stalk, retinal layer, and lens of the eye, and the inner ear. In certain embodiments, the patient may have suffered a neurodegenerative disease, a traumatic injury, a neurotoxic injury, ischemia, a developmental disorder, a disorder affecting vision, an injury or disease of the spinal cord, or a demyelinating disease.

By "non-adherent culture" is meant herein as a method of propagation of cells *in vitro* as in a container in the presence of growth media in a manner in which the cells do not attach to the surface of the container such that a substantial portion of the cells can be removed from the surface of the container by mechanical manipulations that do not cause significant damage to the cells. It is understood that the cells can still be retained in or on a non-adherent matrix (e.g., on Hydrogel spheres) and be removed from the surface of the container. Such manipulations include, for example, gentle agitation, massage, or manual manipulation of the container, or rinsing the container with growth media. As used herein, a substantial portion of the cells to be removed is at least 70%, preferably at least 75%, 80% or 85%, more preferably at least 90% or 95%. Manipulations that cause damage to the cells can be identified by determining the viability of the cells before and after manipulation, for example by trypan blue staining. Mechanical manipulations should cause damage to less than 20%, preferably less than 15%, or 10%, more preferably less than 5%, 2%, or 1% of the cells.

By "obtaining" as in "obtaining an agent" or "obtaining a cell" refers to purchasing, synthesizing, or otherwise procuring an agent or cell. Cells can be obtained, for example, from an animal including human and non-human animals. Cells can also be obtained from cell and tissue repositories.

By "prevent," "preventing," "prevention," "prophylactic treatment" and the like is meant reducing the probability of developing a disorder or condition in a subject, who does not have, but is at risk of or susceptible to developing a disorder

or condition. Prevention or prophylactic treatment can require administration of more than one dose of the compositions of the invention.

By "propagate", "passage", and the like is meant increasing the volume of a cell culture and/or decreasing the amount of cells in a specific culture volume by diluting cells in at least some fresh growth media to allow for maintenance and/or expansion of the cell population.

By "sample" or "biological sample" is meant any biological sample obtained from an individual, body fluid, cell line, tissue culture, or other source.

By "stem cell" or "pluripotent stem cell," which can be used interchangeably, is meant a cell having the ability to give rise to two or more cell types of an organism.

By "subject" is meant a vertebrate, preferably a mammal, more preferably a human.

By "substantially purified" is meant that the desired cells (e.g., MSCs) are enriched by at least 30%, more preferably by at least 50%, even more preferably by at least 75%, and most preferably by at least 90% or even 95%.

By "transgene" is meant any piece of a nucleic acid molecule (for example, DNA) that is inserted by artifice into a cell transiently or permanently, and becomes part of the organism if integrated into the genome or maintained extrachromosomally. Such a transgene may include a gene that is partly or entirely heterologous (foreign) to the transgenic organism, or may represent a gene homologous to an endogenous gene of the organism. The transgene may be introduced into the organism from which the MSCs are isolated. Alternatively, the transgene may be introduced using viral vectors, such as retroviral vectors (See, e.g., Gnecchi et al., 2006).

By "transgenic cell" is meant a cell containing a transgene. For example, a cell transformed with an expression vector operably linked to a heterologous nucleic acid molecule can be used to produce a population of cells having altered phenotypic

characteristics. A cell derived from a transgenic organism is also a transgenic cell so long as the cell contain the transgene.

By "transplant" or "transplanting" is meant administering one or more cells (or parts thereof), cell products, tissue, or cell culture products derived from cells that are grafted into a human host. For example, a transplant can include an MSC transplant.

By "treatment" is meant an approach for obtaining beneficial or desired clinical results. For the purposes of this invention, beneficial or desired clinical results include, but are not limited to, alleviation of symptoms, diminishment of extent of disease, stabilization (i.e., not worsening) of a state of disease, delay or slowing of disease progression, amelioration or palliation of the disease state, and remission (whether partial or total), whether detectable or undetectable. "Treatment" can also mean prolonging survival as compared to expected survival if not receiving treatment. "Treatment" refers to both therapeutic treatment and prophylactic or preventative measures. Those in need of treatment include those already with the disorder as well as those in which the disorder is to be prevented. "Palliating" a disease means that the extent and/or undesirable clinical manifestations of a disease state are lessened and/or the time course of the progression is slowed or lengthened, as compared to a situation without treatment. Typically, the "treatment" entails administering an effective dose of MSCs to the patient to regenerate tissue.

By a "vascular cell" is meant an endothelial cell. Endothelial cells line the blood and lymph vessels and are present in and play a key role in the development of organs, such as the brain, heart, liver, pancreas, lungs, spleen, stomach, intestines, and kidneys.

By "vascular disease" is meant a disease or disorder that affects or involves the vasculature. Examples of vascular disease include peripheral vascular disease, peripheral arterial disease, venous disease (e.g., deep vein thrombosis), ischemia, cardiovascular disease, tissue organ engraftment rejection, or sequelae of ischemic reperfusion injury. In still another embodiment, the peripheral vascular disease is

atherosclerosis, thromboembolic disease, or Buerger's disease (thromboangiitis obliterans). In a further embodiment, the cardiovascular disease is myocardial infarction, heart disease, or coronary artery disease.

As used herein, "a", "an", and "the" are understood to be either singular or plural unless otherwise obvious from context.

As used herein, "or" is meant to be inclusive unless otherwise obvious from context.

As used herein, ranges are understood to include all values within the range. For example, 1 to 50 is understood to mean 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, and 50. A series of values are understood to represent a range, and thereby all of the values within the range unless otherwise obvious from context.

Brief Description of the Drawings

FIGURES 1A-1B are images of MSC harvested from plastic adherent culture of MSC by trypsinization and then cultured for 1 week in (A) plastic tissue culture dish, (magnification 100x), or (B) grown in a double layer agarose culture (magnification 100x), or cultured for 2 weeks (C) in liquid culture above a single layer of agarose to prevent adherence to plastic (magnification 100x)

FIGURE 2A-2B is an image of MSC spheres generated in culture of MSCs in Teflon® bags (A) grown in culture for 2 weeks (mag 100x) and (B) for 6 weeks (mag 100x).

FIGURE 3 is an image of proliferating MSCs in hydrogel for 2 weeks (mag 100x)

FIGURE 4 is an image of MSCs in a tissue culture flask after seven passages in Teflon® bags and then transferred to a plastic culture flask. The MSC spheres adhered to the surface of the flask within 2 to 3 days and obtained a morphology essentially identical to that observed in cells passaged in adherent cultures (mag 100x).

Detailed Description

Mesenchymal stem cells have been demonstrated to be useful in the therapeutic methods for the repair and regeneration of tissue, especially muscle tissue, including cardiac tissue. This is somewhat surprising as MSCs have been demonstrated to be quiescent after injection, have low engraftment into tissue other than bone, and to have a very low persistence after injection.

Mesenchymal stem cells are adherent cells, and can be selected for growth in culture by their ability to adhere to tissue culture containers (i.e., plastic). In culture, cells are propagated by repeated rounds of trypsinization and replating, effectively selecting for cells that are adherent. The observed low level of engraftment and cell division *in vivo* may be due to the *in vitro* methods of propagation of the MSCs in adherent cultures, as no comparable surfaces are available *in vivo*, for example in muscle, vascular, and neural cells.

The invention provides methods for mesenchymal stem cells (MSCs) growth in non-adherent culture, eliminating the need for trypsinization in propagation of MSCs. The non-adherent culture methods of the invention allow for the propagation of MSCs that may more readily engraft into recipient tissue and be more viable for longer periods after transplant as they do not require a surface to which they can adhere to divide.

The non-adherent culture methods of the invention also allow for propagation of cells in a less resource intensive manner by allowing the cells to be grown in larger numbers in the same culture container area as the cells do not need to all grow in the same plane of the culture container as with adherent cells.

The invention provides culture methods that enable the generation of MSC in non-adherent foci in various support matricies. MSCs grown under these conditions can be passaged without trypsinization. Methods include growth of cells encapsulated in matrices such as Hydrogel and Matrigel®, on or between layers of agarose, or in Teflon® bags. Cells can grow in contact with the non-adherent matrices, but do not adhere to plastic culture containers. The lack of adherence to a

surface is notable in the MSCs grown on agarose or in Teflon® bags as can be determined by the maintenance of their rounded shape. MSCs grown in adherent cultures on plastic adopt an elongated, fibroblastic shape (see, e.g., compare Figure 1A with Figures 1B-1C and 2A-B).

Mesenchymal stem cells have been cultured for up to 10 passages and can be subcultured without the need of treatment with trypsin. The non-adherent cells express similar surface markers as cells grown under adherent conditions (e.g., CD105), and they maintain their ability to differentiate into multiple cell types. Optimal growth of the cells is stimulated by basic fibroblast growth factor (bFGF) and other growth factors including stem cell factor (SCF) and vascular endothelial growth factor (VEGF).

Growth of non-adherent MSCs in Teflon® bags provides an additional advantage for translation into therapeutic applications as the MSCs can be cultured by massaging the bag to detach the cells from the surface. When the MSCs are detached the can be maintained as MSC spheres by regular massaging of the bag and inversion of the bag for continued incubation. Performance of this manipulation about twice daily allows for the MSC spheres to increase in size, and for the MSCs to continue to proliferate and expand. The cells can readily be removed from the culture media by centrifugation and resuspension into an appropriate buffer for injection (e.g., phosphate buffered saline (PBS), physiological saline solution) without the need to remove the cells from a less sturdy non-adherent surface (e.g., Matrigel® or agarose) and without the use of trypsin which would need to be removed from the cells prior to administration.

It is understood that the initial source of and method of isolation of the MSCs to be grown by the culture methods of the invention is not a limitation of the invention. A number of methods of isolation of MSCs are known to those skilled in the art including, but not limited to, those set forth in US Patents 5,486,359; 5,654,186; 5,827,735; 5,858,390; 5,906,934; 5.908,784; 5,965,436; and 7,060,494.

It is further understood that the methods provided herein can be used to culture both wild-type and transgenic MSCs such as those taught, for example in US

Patent 5,591,625 or in Gnecchi et al. (both incorporated herein by reference). Transgenic MSCs can be isolated from transgenic animals or can be transduced using vectors, including viral vectors, for the insertion of expression constructs into the cells.

Mesenchymal stem cells cultured by the methods of the invention can be used for any of a number of research or therapeutic purposes. For example, a number of therapeutic methods using MSCs are known, such as those taught in US Patents 5,811,094 for connective tissue regeneration; 5,858,930 for repair of skin and soft tissue defects; 6,387,369 for cardiac muscle regeneration; 6,875,430 for treatment of immune responses in transplantation; 7,029,666 for muscle and connective tissue repair; 7,097,832 for enhancing blood vessel formation; and 7,160,724 for repair of the brain and spinal cord (all of which are incorporated herein by reference).

Mesenchymal stem cells cultured by the methods of the invention can be used for the generation of cultured media to promote the growth of cells, for therapeutic uses, or for research purposes to identify secreted growth factors that may be responsible for the beneficial therapeutic effects provided by MSCs.

Mesenchymal stem cells cultured by methods of the invention can be incorporated into a kit including the cells in a container with appropriate packing material. The kit can further contain reagents and/or materials for culturing MSCs in adherent and/or non-adherent manner(s).

EXAMPLE 1- Isolation of MSCs from human bone marrow

Human bone marrow cells were obtained from normal donors following informed consent under an Institutional Review Board approved protocol. The mononuclear cell fraction of the bone marrow was isolated on a Ficoll gradient and plated in a T150 Corning (Acton, MA) tissue culture flask at 1-5 x 10^6 cells/ml in α-MEM media containing 20% fetal calf serum (FCS). The cells were incubated in a humidified environment at 5% CO_2 at 37°C. The media was changed weekly. Adherent cells were grown in culture and passaged using trypsin when confluent.

EXAMPLE 2- Culture of MSCs in Hydrogel

MSCs were isolated and propagated as set forth above. MSCs were collected from adherent, confluent cultures using trypsin and encapsulated in Hydrogel (Becton Dickson) following the manufacturer's instructions. The encapsulated MSCs were cultured in α-MEM + 20% FCS in T75 culture flasks. At regular intervals, the non-adherent cells were passaged by removing the supernatant, centrifuging the Hydrogel/MCS mixture, and resuspending the cells in growth media. As shown in Figure 3, MSCs encapsulated in the Hydrogel proliferated and maintained a fibroblast-like morphology. Cells encapsulated in Matrigel™ gave comparable results.

EXAMPLE 3- Culture of MSCs in agarose

Single layer agarose cultures were established in 100 mm culture dishes on preformed layers of 0.5% agarose for double layer, and 1% agarose for single layer agarose in α-MEM + 30% FCS. MSCs were harvested from confluent cell cultures by trypsinization and resuspended in α-MEM + 20% FCS. The MSCs were added in 10 ml of α-MEM + 20% FCS above the agarose layer. The non-adherent cells were passaged by removing the supernatant from the agarose underlay. The cells were centrifuged and the supernatant discarded. The cells were resuspended in fresh media and replated over the agarose underlay. Double layer agarose cultures were generated by incorporating the cells into a top agarose layer (0.66%).

Figure 1B shows cells cultured in a double layer agarose culture in the top agarose layer. The MSCs could be visualized as single, round cells. No proliferation was observed. However, when the cells were plated in a liquid phase in α-MEM + 20% FCS on a lower layer of 1% agarose to prevent adherence, the MSC formed spheres and proliferated as shown in Figure 1C. Cells were passaged multiple times.

EXAMPLE 4- Culture of MSCs in Teflon® bags

MSCs were harvested from confluent cell cultures by trypsinization and resuspended in 50 ml of α-MEM + 20% FCS. The cells were placed in 100 ml

Teflon® bags (American Fluoroceal Corp, Gaithersburg, MD) and cultured. At weekly intervals the bags were harvested, the cells were centrifuged, resuspended in fresh media and placed into new Teflon® bags.

MSCs can be cultured by massaging the bag to detach the cells from the surface. When the MSCs are detached the can be maintained as MSC spheres by regular massaging of the bag and inversion of the bag for continued incubation. Performance of this manipulation twice daily allows for the MSC spheres to increase in size, and for the MSCs to continue to proliferate and expand (see, Figure 2).

The culture methods have been replicated beginning with bone marrow harvested from pig. Non-adherent cultures of pig MSC have now been generated for animal studies. One hundred million non-adherent pig MSCs were generated after 3 weeks of culture in Teflon® bags.

EXAMPLE 5- Adherence of cells to plastic after culture under non-adherent conditions

Culturing of cells under non-adherent conditions does not alter the ability of the MSCs to adhere when provided with an appropriate substrate. Figure 4 shows cells grown in a tissue culture flasks after seven passages in Teflon® bags. The morphology of the cells appears to be identical to that of MSCs propagated continuously in adherent culture.

EXAMPLE 6- Stimulation of MSCs by growth factors

The effect of several growth factors on MSC sphere proliferation were evaluated, including macrophage colony stimulating factor (M-CSF), vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF), stem cell factor (SCF), and media conditioned by 5637 cells (a human bladder carcinoma cell line that constitutively secretes functional cytokines). Optimal growth factors for of MSC spheres was addition of 10% 5637 conditioned media to the cells. However, as this source of growth factors is limited for clinical applications, the effects of recombinant growth factors were also analyzed. A combination of recombinant

human bFGF (50 ng/ml) and recombinant human SCF (100 ng/ml) resulted in maximal proliferation of MSCs and sphere formation.

EXAMPLE 7- Transplantation of non-adherent MSCs for the treatment of cardiac infarction

MSCs are isolated from rat bone marrow by standard Ficoll gradient followed by adherent culture methods. After expansion of the cells, the culture is split. A portion of the cells are maintained in adherent culture, and a portion of the cells are transferred to Teflon® bags for propagation. Cells in Teflon® bags are manipulated twice daily to promote growth of MSC spheres, and media is changed as needed. Adherent cells are propagated using trypsin as needed. Cells can include a marker such as GFP or beta-galactosidase to facilitate identification of the transplanted cells at the end of the experiment. Cells are collected and resuspended in an appropriate buffer for administration (e.g., normal saline).

Age and sex matched laboratory rats of a single type are divided into four groups, sham myocardial infarction (MI), adherent MSC treated, non-adherent MSC treated, and normal saline. In all but the sham MI group, ligation of the left coronary artery is performed using well known methods (see, e.g., Gnecchi et al). Briefly, animals are anesthetized and a left thoracotamy is performed under artificial respiration. The heart is accessed through the intercostal space, the pericardial sac is cut, and the heart is exteriorized through the space. The left coronary artery is legated with a silk suture about midway between the left atrium and the apex of the heart and EKG is recorded to confirm the presence of infarction. In sham operated animals, the artery is not legated. One hour after infarction, an equal number of adherent or non-adherent MSCs are injected into a total of five sites per infarct area. Normal saline is injected into the infarct area in the control animals.

Cardiac function is analyzed at regular intervals after the surgery and administration of the cells, for example by EKG. Either throughout the course of the experiment, or at the end of the experiment, rats are euthanized and hearts are excised. Analysis is performed to determine any of a number of outcomes including, but not limited to, infarct area, engraftment of MSCs into the infarct area,

angiogenesis in the infarct area, and/or mRNA or protein expression. Methods for performing such analyses are known to those skilled in the art. The therapeutic effect of the cells grown in adherent culture and non-adherent culture are compared to each other and to control animals.

It is understood that comparable experiments can be performed using different animals including, for example, pigs.

The invention may be embodied in other specific forms without departing from the spirit or essential characteristics thereof. The foregoing embodiments are therefore to be considered in all respects illustrative rather than limiting on the invention described herein. Scope of the invention is thus indicated by the appended claims rather than by the foregoing description, and all changes which come within the meaning and range of equivalency of the claims are therefore intended to be embraced therein.

All references, patents, patent applications, and GenBank numbers cited are incorporated herein by reference in their entirety.

CLAIMS

1. A method for propagation of a non-adherent culture of mesenchymal stem cells (MSCs) comprising expanding MSCs in or on a non-adherent matrix.

2. The method of claim 1, comprising encapsulation of MSCs in Matrigel™ or Hydrogel.

3. The method of claim 1, comprising the cells propagated on agarose or on Teflon®.

4. The methods of any of claims 1 to 3, wherein the cells are propagated in the non-adherent culture without the use of trypsin.

5. The methods of any of claims 1 to 4, comprising mechanical manipulation of the MSCs.

6. The method of any of claims 1 to 5, further comprising a biological sample containing MSCs.

7. The method of claim 6, further comprising isolating the MSCs from the biological sample containing the MSCs.

8. The method of claim 7, wherein the isolated MSCs are substantially purified.

9. The method of any of claims 1-8, wherein the MSCs are expanded at least 2-fold, 10-fold, 100-fold, 1000-fold, 10,000-fold, or 100,000 fold.

10. The method of any of claims 1-9, wherein the MSCs are suitable for administration to a subject.

11. The method of claim 10, wherein the subject is a human subject.

12. The method of any of claims 1-11 wherein the MSCs are propagated in non-adherent culture for at least a week, at least 2 weeks, at least a month, or at least 2 months.

13. A method for treatment of a subject having a disease or condition susceptible to treatment with MSCs comprising administration of MSCs grown in a non-adherent culture of any of the methods of claims 1 to 12.

14. The method of claim 13, wherein the disease or condition susceptible to treatment with MSCs is selected from the group consisting of muscle disease, neural disease, and vascular disease.

15. The method of claim 13 or 14, wherein the MSCs are allogenic or autologous to the subject.

16. The method of any of claims 13 to 15, wherein the subject is human.

17. The use of a MSC propagated by any of the methods of claims 1 to 12 for use as a therapeutic agent for the treatment of a disease or condition susceptible to treatment with MSCs.

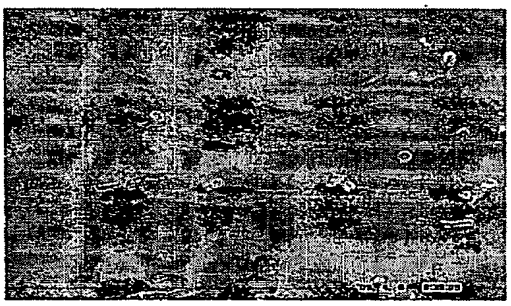
18. The use of claim 17, wherein the disease or condition susceptible to treatment with MSCs is selected from the group consisting of muscle disease, neural disease, and vascular disease.

19. A kit comprising an MSC of any of claims 1 to 12 and appropriate packing material.

20. The kit of claim 19, further comprising reagents or supplies for propagation of MSCs under adherent or non-adherent conditions or both.

FIGURE 1

A



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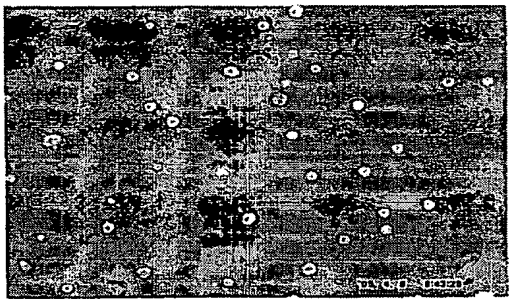


FIGURE 1C

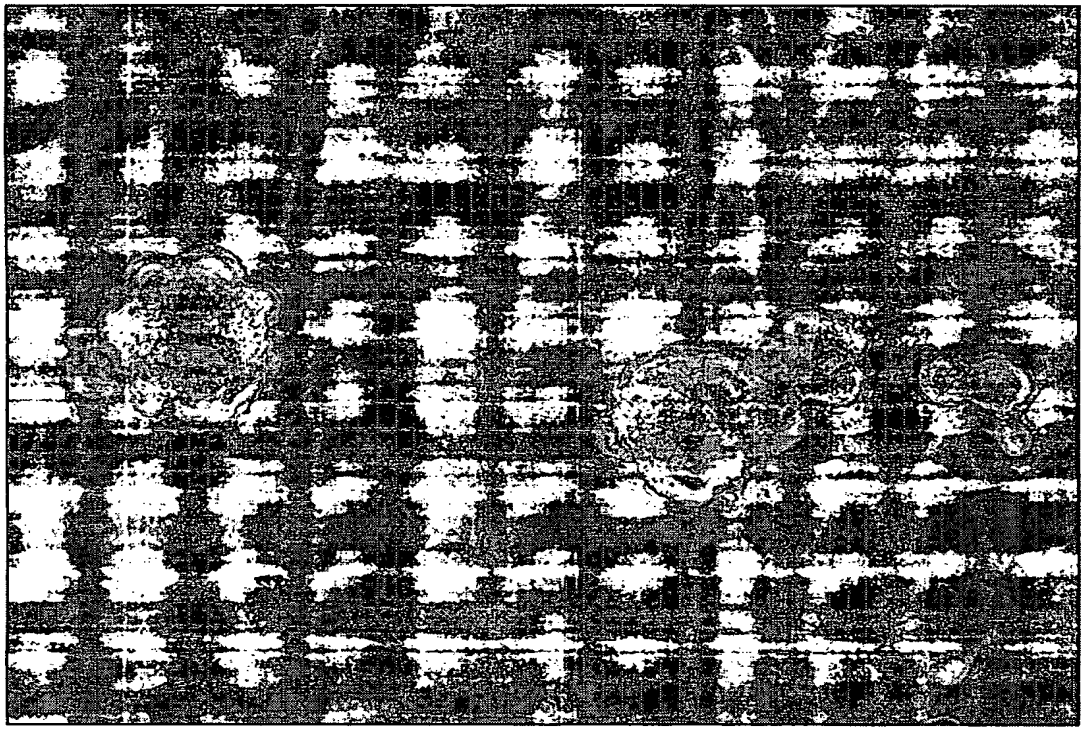


FIGURE 2A

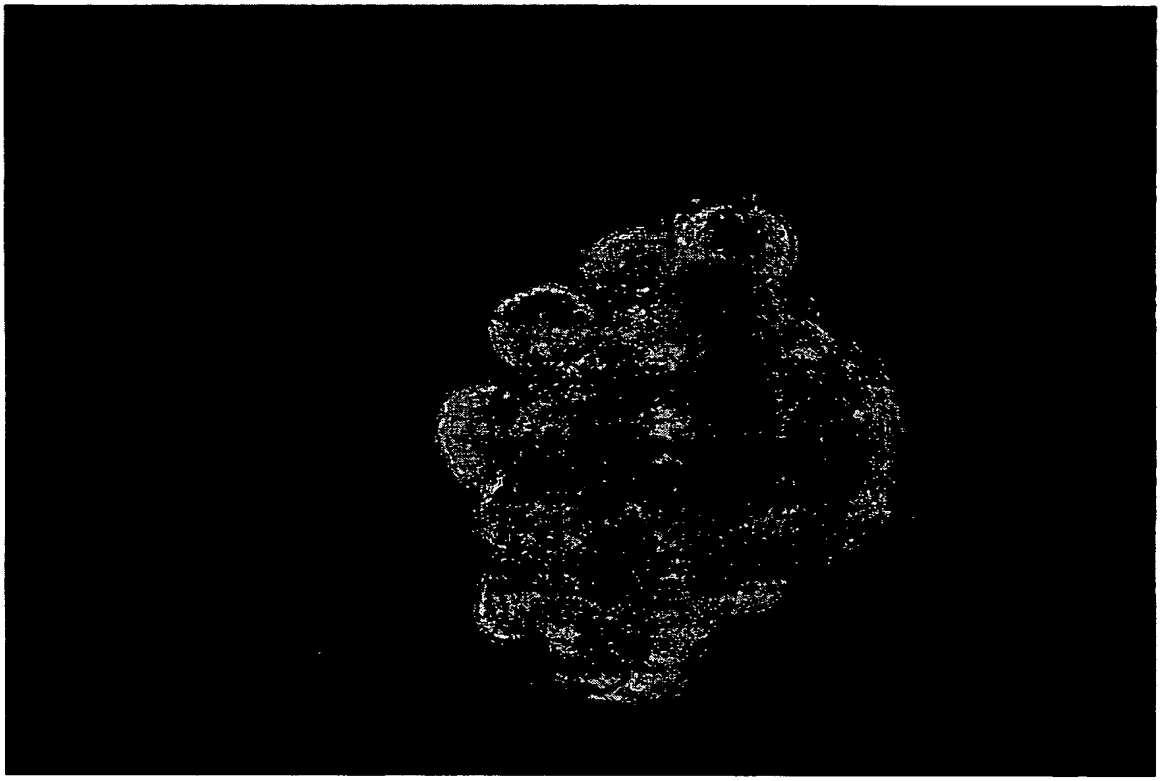


FIGURE 2B

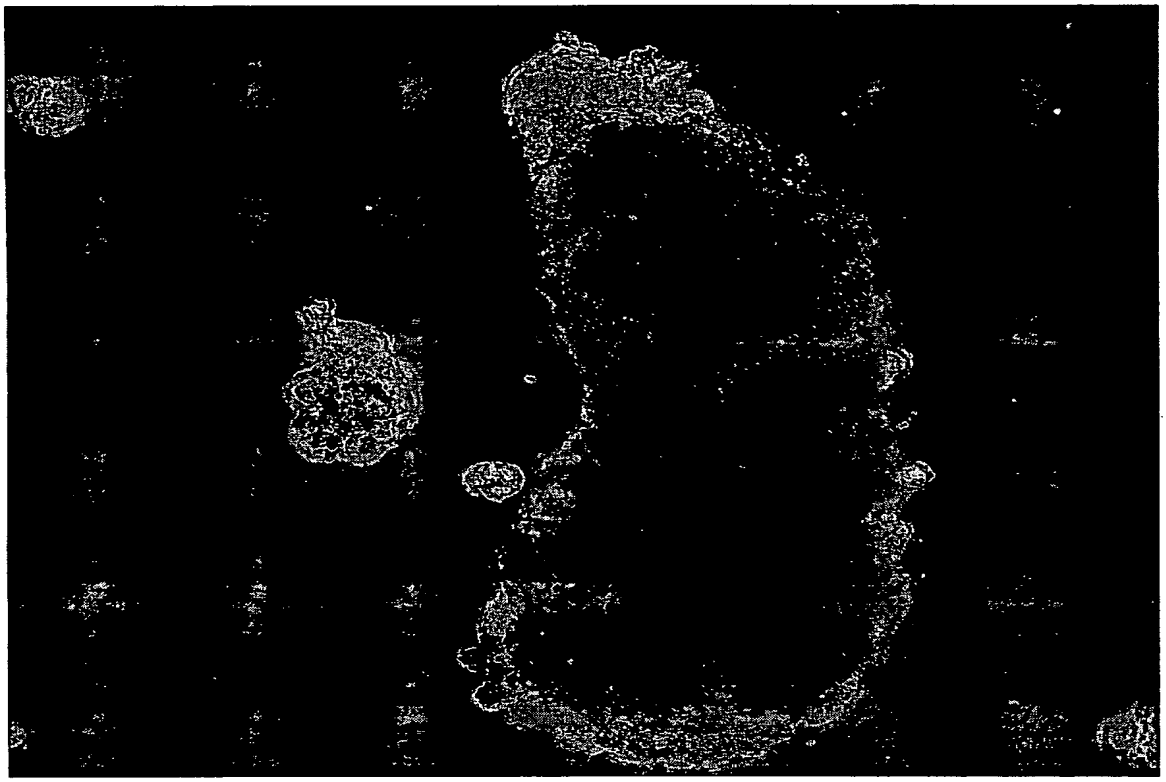


FIGURE 3

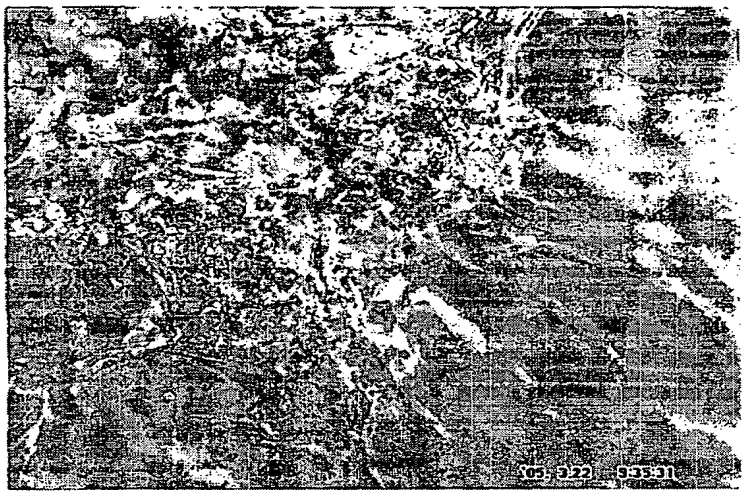
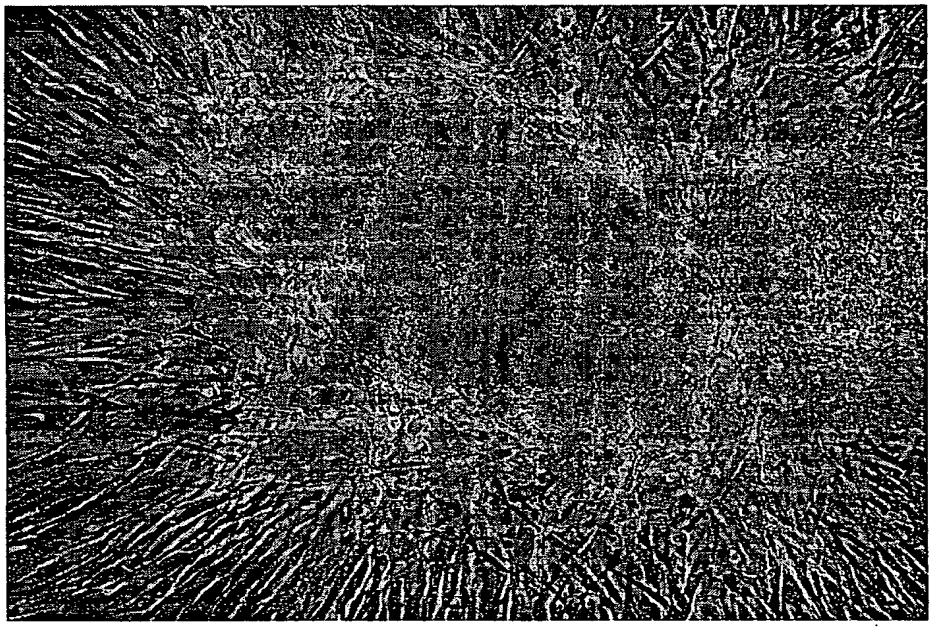


FIGURE 4



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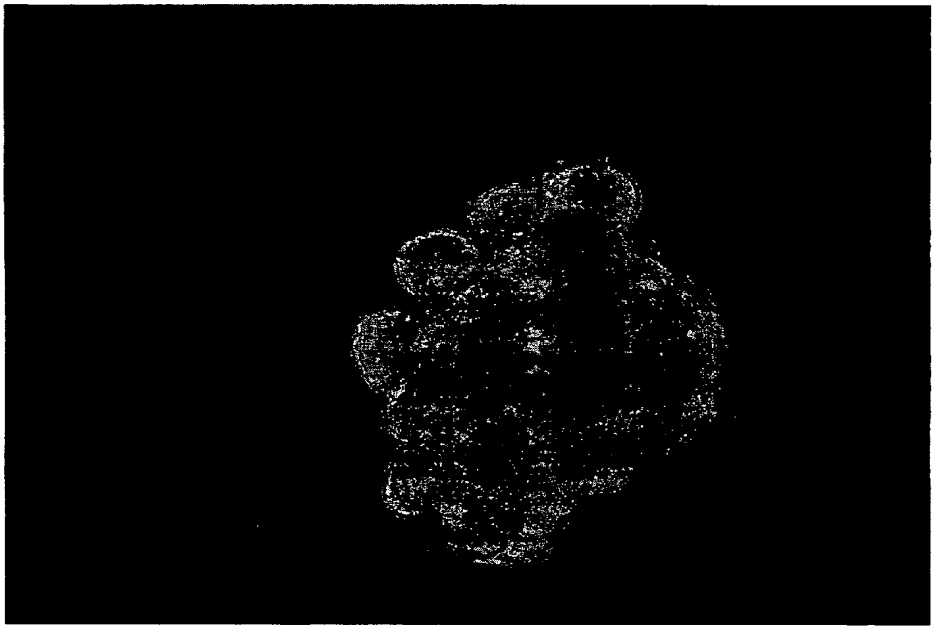
(54) Title: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS



(57) Abstract: The invention provides methods for expanding mesenchymal stem cells (MSCs) in non-adherent cultures. The methods include the propagation of MSCs in or on non-adherent matrices. The invention further provides administration and the use of cells propagated by the method of the invention for administration and preparation of a therapeutic agent. The invention further provides kits including cells propagated by the methods of the inventions.

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FILING DATE: *May 19, 2006*
RELATED PCT APPLICATION NUMBER: *PCT/US07/11921*

THE COUNTRY CODE AND NUMBER OF YOUR PRIORITY APPLICATION, TO BE USED FOR FILING ABROAD UNDER THE PARIS CONVENTION, IS *US60/801,661*

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PROVISIONAL APPLICATION FOR PATENT COVER SHEET – Page 1 of 2

This is a request for filing a PROVISIONAL APPLICATION FOR PATENT under 37 CFR 1.53(c).

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110264 U.S. PTO 60/801661 051906

INVENTOR(S)		
Given Name (first and middle [if any])	Family Name or Surname	Residence (City and either State or Foreign Country)
Ian	Mc Niece	Lutherville, MD

Additional inventors are being named on the — *separately numbered sheets attached hereto*

TITLE OF THE INVENTION (500 characters max):

Growth of Mesenchymal Stem Cells Under Non Adherent Conditions

Direct all correspondence to: **CORRESPONDENCE ADDRESS**

☐ The address corresponding to Customer Number:

OR

☑ Firm or Individual Name The Johns Hopkins University

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5th Floor

City Baltimore	State MD	Zip 21201
Country USA	Telephone 410-516-8300	Email techlicense@jhmi.edu

ENCLOSED APPLICATION PARTS *(check all that apply)*

☐ Application Data Sheet. See 37 CFR 1.76

☑ Specification *Number of Pages* 18

☐ Drawing(s) *Number of Sheets* _____

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U.S. Provisional Patent Application

JHU Ref. No.: JHU-4683

Growth of Mesenchymal Stem Cells under Non Adherent Conditions

By: Ian McNiece

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INVENTION INFORMATION

Title of Invention: [Title should be sufficiently descriptive to identify the invention yet not reveal unique unpublished details.]
Growth of mesenchymal stem cells under non adherent conditions for clinical applications

Name of Lead Inventor:	McNiece	Ian	Keith	PhD
	Last	First	Middle	Degree

Lead Inventor Information: [The Lead Inventor should be a full time JHU faculty member. He/She will be the primary contact person for LTD on all matters associated with this Report of Invention, including processing, patent prosecution and licensing. For reasons of administrative efficiency, it is the responsibility of the Lead Inventor to keep all other JHU inventors named on this Report of Invention informed of the status of such matters.]

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School: Medicine	Department: Oncology
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Interdepartmental address: CRB287	
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Are you a Kennedy Krieger Institute employee or investigator? ☐ Yes X No
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Additional inventors: ☐ Yes X No If yes, please complete Additional Inventors section for each inventor.

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INVENTION DESCRIPTION

Describe the invention completely, using the outline given below. Please provide an **electronic copy** of the invention disclosure document, references, and abstracts in Windows format on CD-ROM or floppy disk if possible

1. **Marketing Summary** [Please provide a non-confidential summary of the invention that can be used for marketing purposes. Unique details that are published may also be included.]

A novel culture methodology is described for the generation of mesenchymal stem cells (MSC) under non adherent conditions. MSC generated under these conditions form MSC foci (MSCF) that can be passaged without trypsinization and cultured long term. Upon culture on plastic the MSCF generate adherent MSC. The continued growth of MSC in the MSCF provide a product capable of seeding damaged tissue in organs where bone surfaces or other solid surfaces are not available for adherence and growth of MSC..

SOFTWARE –Does this disclosure include a software element or software is implemented in the invention ☐ Yes X No

If yes, please complete the Software Information Form which can be found at: _____

BIOLOGICAL MATERIAL – Does this disclosure include biological material, ☐ Yes X No

If yes, please attach a list of materials for reference. A Tangible Property Report of Invention form may be completed if the disclosure is biological materials only. You can find this form at: http://www.ltd.jhu.edu/For Hopkins Inventors/reporting.html

2. Problem Solved [Describe the problem solved by this invention]

This invention provides the methods to optimally generate a MSC product for cellular regeneration. This product enables the transplantation of viable MSC to tissues and enables the continued growth of the MSC in these tissues.

3. Novelty [Identify those elements of the invention that are new when compared to the current state of the art]

The novely of this invention relates to generation of MSCF under non adherent conditions to generate a product capable of continued growth in tissues in vivo. The invention also teaches how to propagate and maintain the growth of MSCF in vivtro.

4. Potential Commercial Use – [What products can be produced with this invention.]

This invention would provide the methods to routinely provide cells for regenerative therapies that are capabale of enhancing in vivo engraftment.

5. Commercialization - List any companies that you feel may be interested in this technology or are doing similar research. Indicate how the invention complements the company's existing technology. If known, provide the names of any companies (and a contact person) that have contacted you regarding your research related to the invention.

X No company interest known at this time.

Keywords – Please circle the categories and keywords that accurately describe the present invention.

CHEMICAL
- ☐ Additives
- ☐ Alternative Energy
- ☐ Antioxidants
- ☐ Batteries
- ☐ Catalyst
- ☐ Coal Conversion
- ☐ Coatings
- ☐ Effluent Treatment
- ☐ Elastimers
- ☐ Electrochemistry
- ☐ Exhaust Treatment
- ☐ Foams
- ☐ Food Chemistry
- ☐ Fuel Cells
- ☐ Gas Conversion
- ☐ Gels
- ☐ Monomers
- ☐ Oxidation
- ☐ Petroleum
- ☐ Photochemistry
- ☐ Polymers
- ☐ Remediation
- ☐ Solvents

DIAGNOSTIC
- ☐ Antibody
- ☐ Assay
- ☐ Biochip
- ☐ Contrast Agent
- ☐ Detection
- ☐ DNA Probe
- ☐ Elisa
- ☐ Imaging
- ☐ Immunoassay
- ☐ In Situ
- ☐ Marker
- ☐ Measurement
- ☐ MRI
- ☐ Point of Use
- ☐ Radioisotope
- ☐ Transgenic
- ☐ Ultrasound

GENOMICS
- ☐ Allele
- ☐ Bioinformatic
- ☐ cDNA
- ☐ Epidemiology
- ☐ EST
- ☐ Gene
- ☐ Homologue
- ☐ Isogene
- ☐ Library
- ☐ Mutation
- ☐ Pharmacogenomics
- ☐ Polymorphism
- ☐ Positional Cloning
- ☐ Proteomics
- ☐ Receptor
- ☐ RNA
- ☐ Target Validation

MEDICAL DEVICE
- ☐ Delivery
- ☐ Diagnosis
- ☐ Imaging
- ☐ Measurement
- ☐ Optical
- ☐ Safety
- ☐ Surgical
- ☐ Treatment

RESEARCH TOOL
- ☐ Animal Model
- ☐ Antibody
- ☐ Cell Line
- ☐ Culture
- ☐ Directed Evolution
- ☐ DNA Probe
- ☐ DNA/RNA Sequencing
- ☐ DNA/RNA Synthesis
- ☐ Electrophoresis
- ☐ Elisa
- ☐ Enzyme
- ☐ Equipment
- ☐ Expression System
- ☐ Immunoassay
- ☐ Label
- ☐ PCR
- ☐ Protein Sequencing
- ☐ Protein Synthesis
- ☐ Reagent
- ☐ Spectroscopy
- ☐ Tissue Culture
- ☐ Vector

SCREENING
- ☐ Assay
- ☐ Biochip
- ☐ Combinatorial Biology
- ☐ Combinatorial Chemistry
- ☐ Detection
- ☐ HTS
- ☐ Phage Display
- ☐ Screen
- ☐ Target

THERAPEUTIC
- ☐ Analgesic
- ☐ Anesthetic
- ☐ Angiogenesis
- ☐ Antibiotic
- ☐ Antibody
- ☐ Antifungal
- ☐ Antiinflammatory
- ☐ Antisense
- ☐ Antiviral
- ☐ Apoptosis
- ☐ Cell Signaling
- X Cell Therapy
- ☐ Disease Model
- ☐ Drug Delivery
- ☐ Drug Design
- ☐ Fertility
- ☐ Gene Therapy
- ☐ Hormone
- ☐ Immunotherapy
- ☐ Natural Product
- ☐ Peptides
- ☐ Pro-drug
- ☐ Proteins
- ☐ Small Molecule
- X Tissue Engineering
- X Transplant
- ☐ Vaccine
- ☐ Virus
- ☐ Wound Healing

DISEASES
- ☐ Aging
- ☐ Blood
- X Cancer
- ☐ Cardiovascular
- ☐ Dermatologic
- ☐ Endocrine
- ☐ Gastrointestinal
- ☐ Genitourinary
- ☐ Hepatic
- ☐ Immune
- ☐ Infectious
- ☐ Metabolic
- X Musculoskeletal
- XNeurological
- ☐ ObGyn
- ☐ Ophthalmological
- ☐ Oral
- ☐ Pediatric
- ☐ Psychiatric
- ☐ Respiratory

ADDITIONAL KEY WORDS:

_____

_____

_____

STAGE OF DEVELOPMENT
- ☐ Unspecified
- ☐ Discovery
- X Preclinical
- ☐ Prototype
- ☐ Phase I
- ☐ Phase II
- ☐ Phase III
- ☐ NCE

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7. Detailed Description of the invention - On a separate page(s), attach a detailed description of how to make and use the invention. The description must contain sufficient detail so that one skilled in the same discipline could reproduce the invention. Include the following as necessary:

1- data pertaining to the invention;
2- drawings or photographs illustrating the invention;
3- structural formulae if a chemical;
4- procedural steps if a process;
5- a description of any prototype or working model;

In general, a manuscript that has been prepared for submission to a journal will satisfy this requirement.

8. Workable Extent/Scope [Describe the future course of related work, and possible variations of the present invention in terms of the broadest scope expected to be operable; if a ***compound,*** describe substitutions, breadth of substituents, derivatives, salts etc., if ***DNA or other biological material,*** describe modifications that are expected to be operable, if a ***machine or device,*** describe operational parameters of the device or a component thereof, including alternative structures for performing the various functions of the machine or device]

Future work is planned to evaluate the use of different matrices for generation of MSCF. Pre clinical studies are also planned to evaluate the in vivo potential of the MSCF in animal models.

9. References [Please cite relevant journal citations, patents, general knowledge or other public information related to the invention and distinguish between references that (A) contain a description of the current invention from those that (B) contains background information.]

X No references available at this time.

7. Detailed Description of the Invention

The use of bone marrow derived mesenchymal stem cells (MSC) has been proposed for a number of regenerative therapies including repair of myocardial tissue. The data to date has suggested improved functional outcomes but have failed to demonstrate incorporation of MSC into the tissue. These studies have utilized MSC grown as adherent cells in plastic tissue culture flasks and trypsinized for harvest and infusion.

We have developed culture methodologies that enable the generation of MSC in non adherent foci in various support matrices. MSC grown under these conditions can be passaged without trypsinization. The foci of MSC (MSCF) can be harvested for infusion as shown in Figure #1. For example purposes, hydrogel was used as a supportive matrix to generate MSCF. The matrix was prepared following the manufacturers recommended procedure and MSC were trypsinized and added to the hydrogel in alpha MEM plus 20% FCS. Over the next several days adherent MSC were observed on the surface of the wells and the supernatant from the well was collected and transferred to a secondary well without addition of any further MSC. The MSC could be visualized in the hydrogel and the wells were passaged several times over the next 2 weeks.

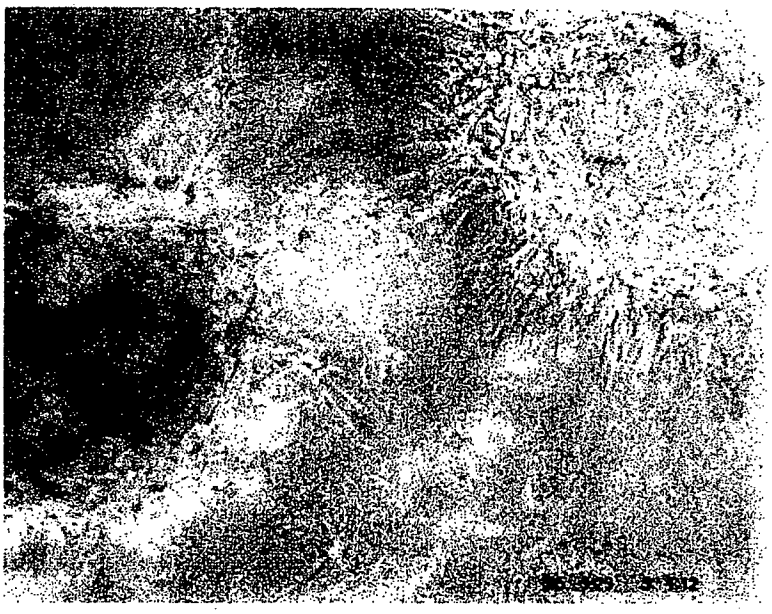

Figure 1: MSC foci generated from bone marrow derived MSC

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Culture of MSC Under Non-Adherent Conditions For Tissue Repair.

Ian McNiece, Adeline Chia*, Heming Wei.

Division of Biomedical Sciences Johns Hopkins in Singapore, Singapore.

INTRODUCTION

Human mesenchymal stem cells (MSC) are multipotent cells, which are present in adult marrow and can differentiate to lineages of mesenchymal tissues, including bone, cartilage, fat, tendon, muscle, and marrow stroma. MSC can be isolated and expanded in culture with the potential of up to 40 doublings. Recent studies have explored the potential use of human MSC (hMSC) to improve cardiac function in patients after irreversible ischemic injury. A number of reports have demonstrated that MSC can be injected into infracted cardiac tissue and improved cardiac function demonstrated. However, the extent of proliferation and integration of MSC into the regenerative tissue has been limited. Some experiments have demonstrated that less than 3% of injected MSCs persist after 2 weeks.

MSC are typically cultured as adherent cells in culture flasks and require trypsinization for passage. When transplanted into cardiac tissue it is likely that MSC fail to proliferate due to their inability to adhere to a surface and this minimizes the contribution of MSC to regenerating tissue. We hypothesized that if MSC could be cultured without adhering to a solid surface, it may be possible to generate MSC that are capable of in vivo proliferation in tissue and that these non adherent type MSC would contribute to tissue regeneration.

METHODS

Generation of Mesenchymal Stem Cells

Human BM cells were obtained from normal donors following informed consent under a Johns Hopkins University (Baltimore, MD) Institutional Board Review approved protocol. The mononuclear fraction of the BM was isolated on a Ficol gradient and placed in T150 Corning (Acton, MA) tissue culture flasks at 1 – 5 x 10^6 cell/ml in alpha MEM media containing 20% FCS (a-MEM+20%FCS). The cells were incubated at 5% CO_2, 37^{0}C and the media changed weekly. Adherent cells grew in the cultures and were passaged using trypsin when confluent.

Culture of MSC Under Non Adherent Conditions

1. **Hydrogel:** MSC were harvested using trypsin from confluent cultures and encapsulated in Hydrogel (Becton Dickinson, Franklin Lakes, NJ) following the manufacturers instructions. The MSC were mixed with activated Hydrogel and cultured in T75 tissue culture flasks. At regular intervals the non adherent cells were passaged by removing the supernatant, centrifuging the hydrogel/MSC mix and resuspending the cells in a-MEM + 20%FCS.

2. **Agarose Culture:** Preformed layers of 0.5% agarose in a-MEM + 30% FCS were established in 100 mm culture dishes and allowed to gel. MSC were harvested from confluent cultures by trypsinization and resuspended in a-MEM + 20%FCS. The MSC were added in 10 mls of a-MEM + 20%FCS above the agarose layer. The non adherent cells were passaged by removing the supernatant from the agarose underlay. The cells were centrifuged and the supernatant discarded and the cells were resuspended in fresh media and replated over agarose underlays.

3. **Culture in Teflon Bags:** MSC were harvested from confluent cultures by trypsinization and resuspended in 50 ml of a-MEM + 20% FCS. The cells were then placed in 100 ml Teflon bags (American Fluoroceal Corp, Gaithersburg, MD) and cultured. At weekly intervals the bags were harvested, the cells were centrifuged, resuspended in fresh media and placed into new Teflon bags.

RESULTS AND DISCUSSION

Hydrogel: Encapsulation of MSC resulted in proliferation of MSC and incorporation into the Hydrogel matrix. As shown in Figure 1,

Agarose Culture

When MSC were trypsinized and cultured in a double layer agarose culture in the top agarose layer (0.66%) the MSC could be visualized as single round cells (Figure 2). However, when the MSC were plated in a liquid phase in alpha MEM + 20%FCS on a lower layer of 1% agarose to prevent adherence, the MSC formed spheres and proliferated as shown in Figure 3.

Culture in Teflon Bags:

We have utilized Teflon bags for cell culture experiments based upon minimal adherence of cells to the Teflon surface. Therefore we evaluated the potential of the Teflon bags for generation of non adherent MSC. As shown in Figure 4, the MSC proliferated as non adherent spheres of cells.

The generation of non adherent MSC was achieved with all three culture conditions, namely, growth in Hydrogel, coculture on agarose and culture in teflon bags. In each case foci of MSC formed that continued to proliferate and the foci could be subcultured by removing the cells and transferring to secondary cultures. We have concentrated on the growth of MSC in Teflon bags as this approach can be scaled up to produce large numbers of MSC spheres. Also the culture in Teflon bags requires no substrates or scaffolds that require GMP manufacture.

The MSC spheres could be cultured for extended periods in Teflon bags and the cells continue to proliferate (Figure 5) and the size of the spheres increased continually. As shown in Figure 4, individual MSC can be visualized as large round cells on the surface of the spheres. When the spheres are placed into standard tissue culture flasks they can adhere to the plastic surface and appear identical to the original MSC generated in culture flasks (Figure 6). A number of MSC adhere

to the Teflon surface of the bags and can be eliminated by weekly transfer to new bags. Alternatively, by massaging the bags daily the adherent MSC can be dislodged and remain viable as non adherent cells leading to proliferation and sphere formation.

Stimulation of MSC by Growth Factors

We evaluated the effect of several growth factors on MSC sphere proliferation including macrophage colony stimulating factor (M-CSF or CSF-1), vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF), stem cell factor (SCF) and media conditioned by 5637 cells (5637CM). The optimal growth factors for proliferation of MSC spheres was addition of 10% 5637CM. Howver as the clinical application of this source of growth factors is limited we concentrated on recombinant growth factors. The combination of rh bFGF (50 ng.ml) and rhSCF (100 ng/ml) resulted in maximal proliferation of MSC and sphere formation.

We propose that the MSC spheres have the capacity to proliferate in tissues, such as cardiac tissue, and will remain viable. In addition, we propose that the MSC spheres will integrate into the tissue and be stimulated by local cytokines to differentiate into tissue specific cells, such as cardio myocytes. The cellular grafts have the potential to regenerate damaged tissue.

Figure 1: Growth of MSC in Hydrogel

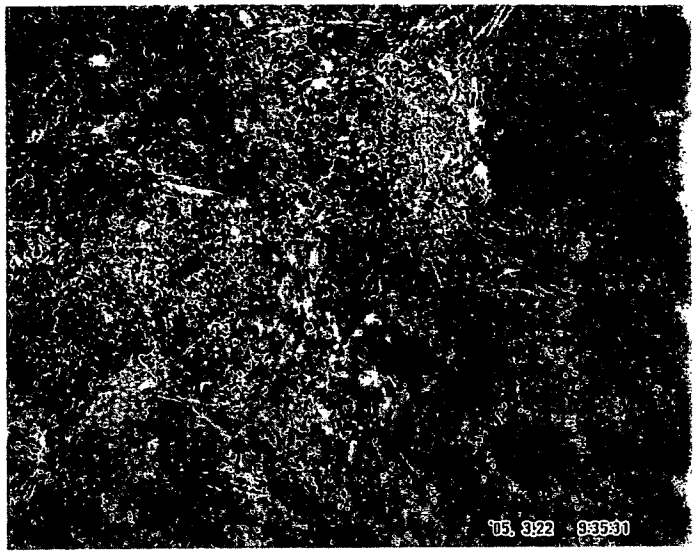

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Figure 2: MSC grown generated as adherent cells on plastic (top panel) were trypsinized and cultured in double layer agarose culture (bottom panel).

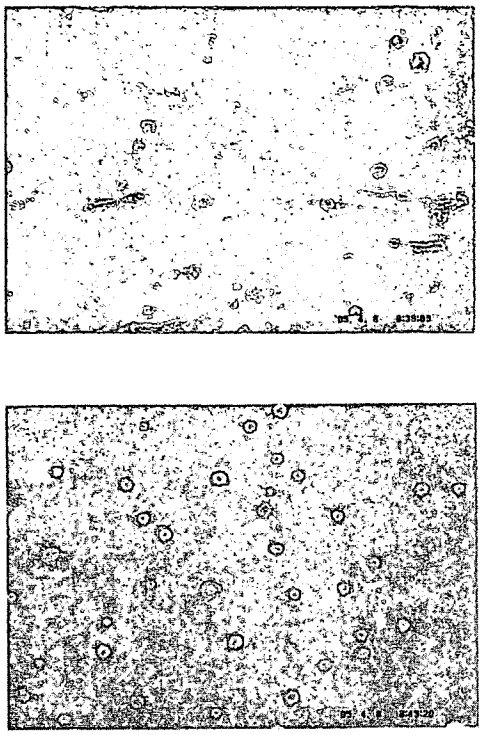

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Figure 3: Growth of MSC in double layer agarose culture

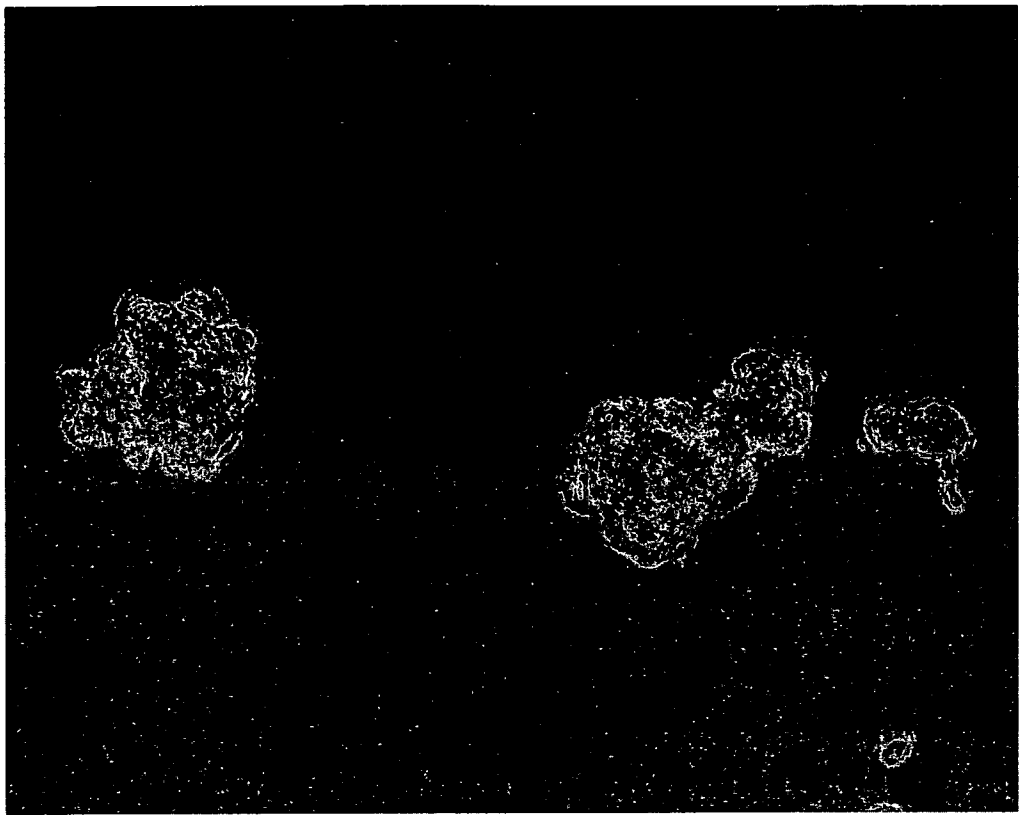

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Figure 4: MSC spheres generated in culture of MSC in Teflon bags.

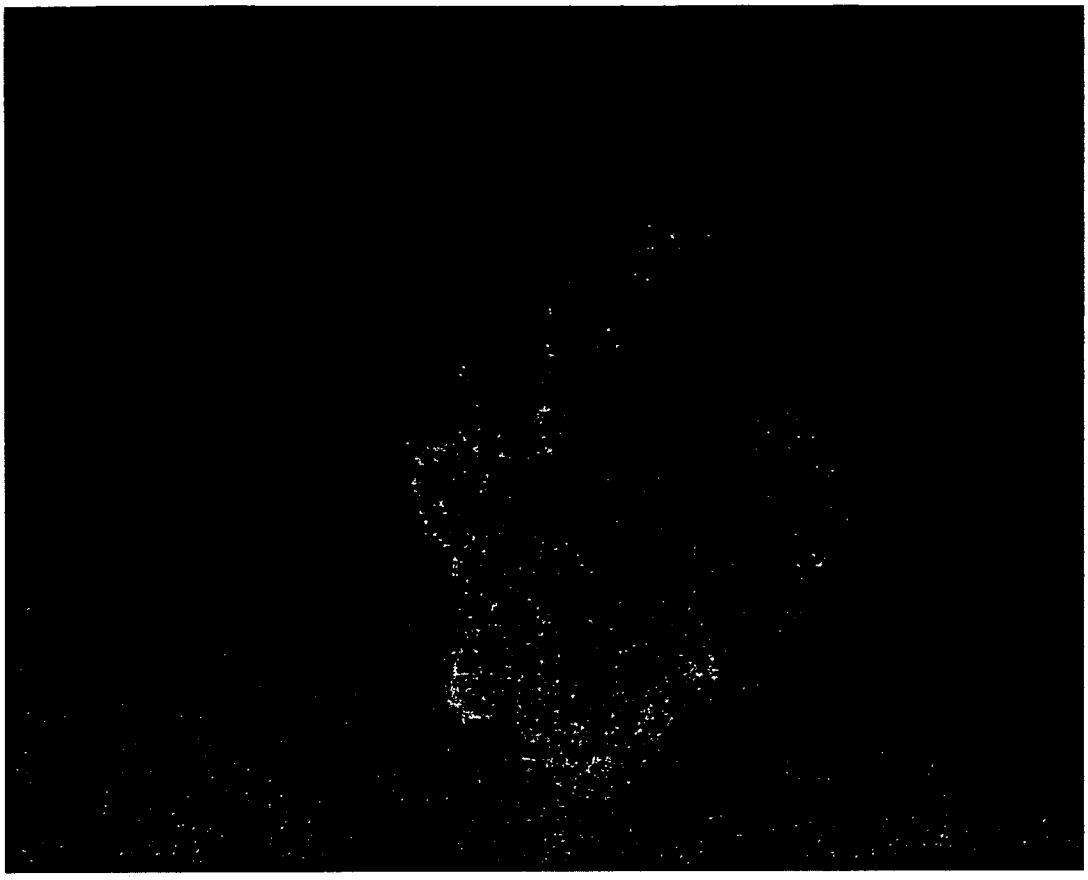

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Figure 5: MSC Spheres after culture for 6 weeks in Teflon bags.

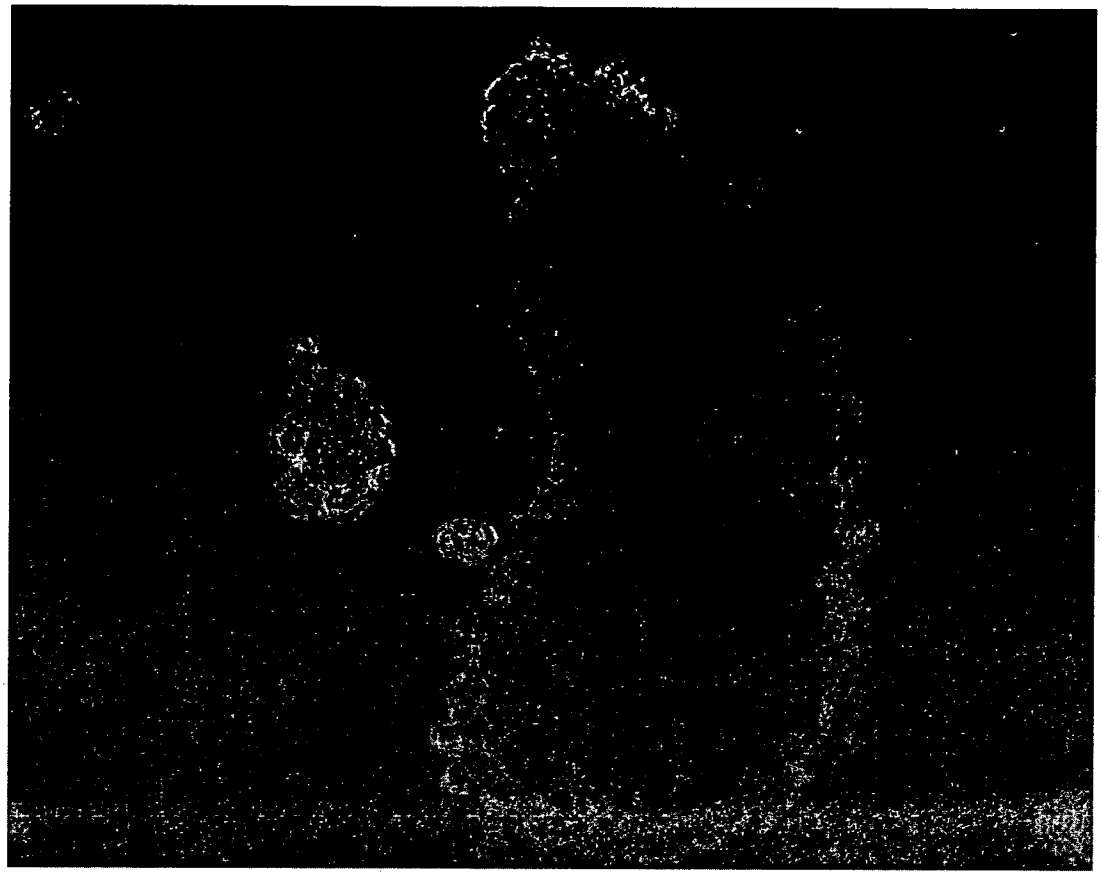

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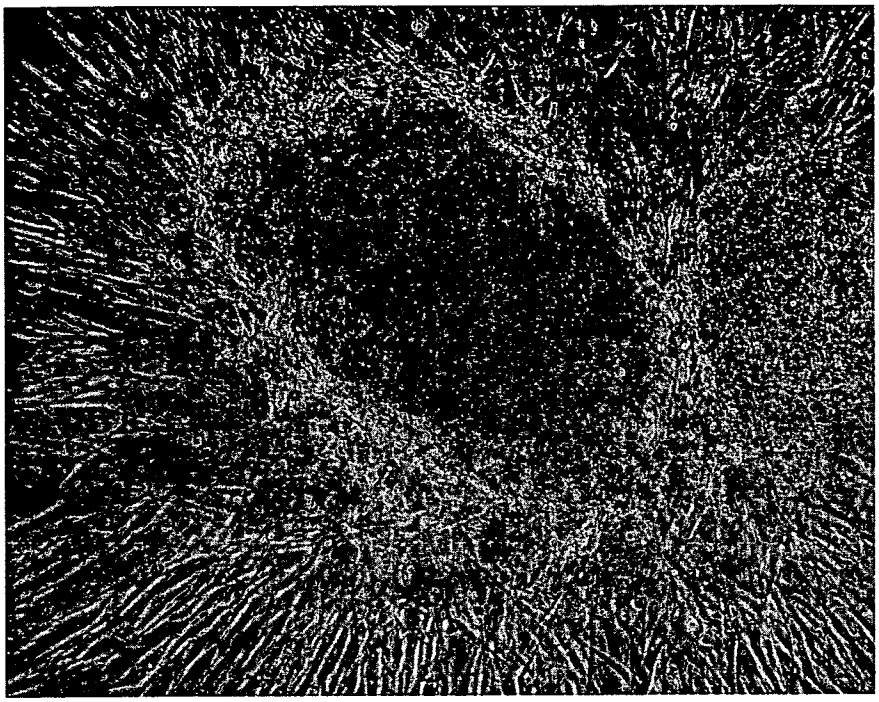
Figure 6: MSC spheres were culture for 7 passages in Teflon bags and then transferred to tissue culture flasks. The MSC spheres formed adherent MSC identical to the original MSC generated in culture flasks.



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All publications, patents and patent applications disclosed herein are incorporated into this application by reference in their entirety.

For example: "Sambrook *et al.*, Molecular Cloning, A Laboratory Manual (volumes I-III) 1989, Cold Spring Harbor Laboratory Press, USA", "Harlowe and Lane, Antibodies a Laboratory Manual 1988 and 1998, Cold Spring Harbor Laboratory Press, USA" and "Ausubel *et al.*, Current Protocols 2001, John Wiley and sons, Inc." provide sections describing methodology for antibody generation and purification, diagnostic platforms, cloning procedures, etc. that may be used in the practice of the instant invention.

Abstract

The use of bone marrow derived mesenchymal stem cells (MSC) has been proposed for a number of regenerative therapies including repair of myocardial tissue. The data to date has suggested improved functional outcomes but have failed to demonstrate incorporation of MSC into the regenerating tissue. These studies have utilized MSC grown as adherent cells in plastic tissue culture flasks and trypsinized for harvest and infusion. Upon trypsinization the MSC round up and fail to proliferate unless they become adherent to a solid surface. In the BM environment MSC proliferate as adherent cells on the bone surface, however in tissues such as the heart, there is no substrate for the MSC to attach and proliferate. We have developed culture methodologies that enable the generation of MSC under non adherent conditions resulting in the formation of non adherent spheres of MSC. The MSC speheres (MSCS) have been generated using several different culture techniques including; incorporation into matrices such as Hydrogel and Matrigel; ii) coculture on preformed layers of 0.5% agarose; and iii) culture in Teflon bags. MSCS have been cultured for up to 10 passages under these conditions and can be sub cultured without the need for treatment with trypsin. When cultured in flasks directly on plastic the MSCS adhere to plastic surface and grow as typical adherent MSC. The MSCS express similar surface markers as MSC grown under adherent condition, eg CD105, and they maintain their potential to differentiate into multiple cell types. Optimal growth of the MSCS is stimulated by basic fibroblast growth factor (bFGF) and other growth factors including SCF and VEGF can augment growth. For translation to clinical applications we are currently establishing MSCS in Teflon bags in alpha MEM plus 20% FCS plus 50 ng/ml of rhbFGF plus 100 ng/ml rhSCF. The MSC can be cultured as adherent layers in these bags and passaged by simple mechanical massaging of the bags to detach the MSC from the bag. When the MSC are detached they can be maintained as MSCS by regular massaging of the bag and invertion of the bag for continued incubation. This manipulation is performed twice daily and the MSCS increase in size as the MSC continue to proliferate. Current studies are generating MSCS using this approach for studies in animal models of cardiac ischemia.

BEFORE THE OFFICE OF ENROLLMENT AND DISCIPLINE
UNITED STATES PATENT AND TRADEMARK OFFICE

LIMITED RECOGNITION UNDER 37 CFR § 11.9(b)

Mr. Martin Phillip Devenport is hereby given limited recognition under 37 CFR §11.9(b) as an employee of the Johns Hopkins University Medical Institutions to prepare and prosecute patent applications wherein the assignee of record of the entire interest is the Johns Hopkins University Medical Institutions. This limited recognition shall expire on the date appearing below, or when whichever of the following events first occurs prior to the date appearing below: (i) Mr. Martin Phillip Devenport ceases to lawfully reside in the United States, (ii) Mr. Martin Phillip Devenport's employment with the Johns Hopkins University Medical Institutions ceases or is terminated, or (iii) Mr. Martin Phillip Devenport ceases to remain or reside in the United States on an H-1B visa.

This document constitutes proof of such recognition. The original of this document is on file in the Office of Enrollment and Discipline of the U.S. Patent and Trademark Office.

Limited Recognition No. **L0235**
Expires: January 12, 2007

Harry I. Moatz
Director of Enrollment and Discipline

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PATENT COOPERATION TREATY

PCT

12/227458

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY
(Chapter I of the Patent Cooperation Treaty)

(PCT Rule 44*bis*)

Applicant's or agent's file reference 68324WO (71699)	**FOR FURTHER ACTION**	See item 4 below
International application No. PCT/US2007/011921	International filing date *(day/month/year)* 18 May 2007 (18.05.2007)	Priority date *(day/month/year)* 19 May 2006 (19.05.2006)
International Patent Classification (8th edition unless older edition indicated) See relevant information in Form PCT/ISA/237		
Applicant THE JOHNS HOPKINS UNIVERSITY		

1. This international preliminary report on patentability (Chapter I) is issued by the International Bureau on behalf of the International Searching Authority under Rule 44 *bis*.1(a).

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

   In the attached sheets, any reference to the written opinion of the International Searching Authority should be read as a reference to the international preliminary report on patentability (Chapter I) instead.

3. This report contains indications relating to the following items:

☒	Box No. I	Basis of the report
☐	Box No. II	Priority
☒	Box No. III	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
☐	Box No. IV	Lack of unity of invention
☒	Box No. V	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
☐	Box No. VI	Certain documents cited
☐	Box No. VII	Certain defects in the international application
☐	Box No. VIII	Certain observations on the international application

4. The International Bureau will communicate this report to designated Offices in accordance with Rules 44*bis*.3(c) and 93*bis*.1 but not, except where the applicant makes an express request under Article 23(2), before the expiration of 30 months from the priority date (Rule 44*bis* .2).

	Date of issuance of this report 21 November 2008 (21.11.2008)
The International Bureau of WIPO 34, chemin des Colombettes 1211 Geneva 20, Switzerland Facsimile No. +41 22 338 82 70	Authorized officer Beate Giffo-Schmitt e-mail: pt03.pct@wipo.int

Form PCT/IB/373 (January 2004)

PATENT COOPERATION TREATY

From the
INTERNATIONAL SEARCHING AUTHORITY

To: Peter F. Corless
Edwards Angell Palmer & Dodge LLP
P.O. Box 55874
Boston, Massachusetts 02205

PCT

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

(PCT Rule 43*bis*.1)

Date of mailing *(day/month/year)*	13 DEC 2007
Applicant's or agent's file reference 68324WO(71699)	FOR FURTHER ACTION See paragraph 2 below

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US 07/11921	18 May 2007 (18.05.2007)	19 May 2006 (19.05.2006)

International Patent Classification (IPC) or both national classification and IPC
IPC(8) - C12N 5/08 (2007.01)
USPC - 435/366

Applicant The Johns Hopkins University

1. This opinion contains indications relating to the following items:

- ☒ Box No. I Basis of the opinion
- ☐ Box No. II Priority
- ☒ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☐ Box No. VII Certain defects in the international application
- ☐ Box No. VIII Certain observations on the international application

2. FURTHER ACTION

If a demand for international preliminary examination is made, this opinion will be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA") except that this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notified the International Bureau under Rule 66.1*bis*(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of 3 months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

3. For further details, see notes to Form PCT/ISA/220.

Name and mailing address of the ISA/US	Date of completion of this opinion	Authorized officer
Mail Stop PCT, Attn: ISA/US Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-3201	08 November 2007 (08.11.2007)	Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US 07/11921

Box No. I Basis of this opinion

1. With regard to the **language**, this opinion has been established on the basis of:

   ☒ the international application in the language in which it was filed.

   ☐ a translation of the international application into _____ which is the language of a translation furnished for the purposes of international search (Rules 12.3(a) and 23.1(b)).

2. ☐ This opinion has been established taking into account the **rectification of an obvious mistake** authorized by or notified to this Authority under Rule 91 (Rule 43*bis*.1(a))

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, this opinion has been established on the basis of:

   a. type of material

      ☐ a sequence listing

      ☐ table(s) related to the sequence listing

   b. format of material

      ☐ on paper

      ☐ in electronic form

   c. time of filing/furnishing

      ☐ contained in the international application as filed

      ☐ filed together with the international application in electronic form

      ☐ furnished subsequently to this Authority for the purposes of search

4. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

5. Additional comments:

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US 07/11921

Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non obvious), or to be industrially applicable have not been examined in respect of

☐ the entire international application

☒ claims Nos. 5-20

because:

☐ the said international application, or the said claims Nos. _____ relate to the following subject matter which does not require an international search *(specify)*:

☒ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. 5-20 are so unclear that no meaningful opinion could be formed *(specify)*:
Claims 5-20 are improper multiple dependent claims because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

☐ the claims, or said claims Nos. _____ are so inadequately supported by the description that no meaningful opinion could be formed *(specify)*:

☒ no international search report has been established for said claims Nos. 5-20

☐ a meaningful opinion could not be formed without the sequence listing; the applicant did not, within the prescribed time limit:

- ☐ furnish a sequence listing on paper complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Searching Authority in a form and manner acceptable to it.
- ☐ furnish a sequence listing in electronic form complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Searching Authority in a form and manner acceptable to it.
- ☐ pay the required late furnishing fee for the furnishing of a sequence listing in response to an invitation under Rule 13*ter*.1(a) or (b).

☐ a meaningful opinion could not be formed without the tables related to the sequence listings; the applicant did not, within the prescribed time limit, furnish such tables in electronic form complying with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions, and such tables were not available to the International Searching Authority in a form and manner acceptable to it.

☐ the tables related to the nucleotide and/or amino acid sequence listing, if in electronic form only, do not comply with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions.

☐ See Supplemental Box for further details.

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US 07/11921

Box No. V **Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

1. Statement

Novelty (N)	Claims	2-4	YES
	Claims	1	NO
Inventive step (IS)	Claims	None	YES
	Claims	1-4	NO
Industrial applicability (IA)	Claims	1-4	YES
	Claims	None	NO

2. Citations and explanations:

Claim 1 lacks novelty under PCT Article 33(2) as being anticipated by US 2005/0265980 A1 to Chen et al. (hereinafter 'Chen').

Regarding claim 1, Chen discloses a method for propagation of a non-adherent culture (in hydrogels) of mesenchymal stem cells (MSCs) comprising expanding MSCs in or on a non-adherent matrix (hydrogels) (para [0007], [0047], [0088]).

Claims 2 and 4 lack an inventive step under PCT Article 33(3) as being obvious over Chen in view of US 2004/0092011 A1 to Wilkison et al. (hereinafter 'Wilkison').

Regarding claim 2, claim 1 is obvious over Chen, as described above. Chen does not expressly disclose encapsulation of MSCs in Hydrogel. In a similar invention, Wilkison discloses encapsulation of MSCs (adipose-derived stem cells) in Hydrogel (para [0109]). It would have been obvious to one of ordinary skill in the art to combine the teaching of Chen with that of Wilkison to practice the claim as described since both are directed to methods for propagation of stem cells.

Regarding claim 4, claim 1 is obvious over Chen, as described above. Chen does not expressly disclose that the cells are propagated in the non-adherent culture without the use of trypsin. In a similar invention, Wilkison discloses cells propagated in a culture without the use of trypsin (para [0142]). It would have been obvious to one of ordinary skill in the art to combine the teaching of Chen with that of Wilkison to practice the claim as described since both are directed to methods for propagation of stem cells.

Claim 3 lacks an inventive step under PCT Article 33(3) as being obvious over Chen in view of US 2005/0013804 A1 to Kato et al. (hereinafter 'Kato').

Regarding claim 3, claim 1 is obvious over Chen, as described above. Chen does not expressly disclose that the cells are propagated on agarose or on Teflon. In a similar invention, Kato discloses cells propagated on agarose (cultured mesenchymal stem cells transferred to an agarose culture system) (para [0028]). It would have been obvious to one of ordinary skill in the art to combine the teaching of Chen with that of Kato to practice the claim as described since both are directed to methods for propagation of mesenchymal stem cells.

Claims 1-4 have industrial applicability as defined by PCT Article 33(4) because the subject matter can be made or used in industry.

24/c

U.S. Appl. No. 12/227458 Internatio Appl No. US07/011921

Application filed by: ☐ 20 months ☑ 30 months

INTERNATIONAL APPLICATION PAPERS IN THE APPLICATION FILE:

- ☑ International application (RECORD COPY)
- ☐ Article 19 amendments
- ☐ PCT/IB/331
- → ☐ PCT/IPEA/409 IPER (PCT/IPEA/416 on front) 237
- ☐ Annexes to 409
- ☑ Priority document(s) No. 1
- ☐ INTERNATIONAL APPLICATION ON DOUBLE SIDED PAPER (COPIES MADE)
- ☐ Request form PCT/RO/101
- ☐ PCT/IB/302
- ☑ PCT/ISA/210-Search Report
- ☐ Search Report references
- ☐ Other _____

RECEIPTS FROM THE APPLICANT: (other than checked above)

- ☑ Basic National Fee (paid or authorized to charge)
- Translation of international application as filed:
  - ☑ Description
  - ☑ Claims
  - ☑ Words in the drawing figure(s)
  - ☐ Article 19 amendments
  - ☐ Annexes to 409
- ☐ Oath / Declaration
- ☐ DNA diskette
- ☑ Preliminary amendment(s) filed _____
- ☑ Information Disclosure Statement
- ☐ Assignment document
- ☐ Power of attorney/Change of address
- ☐ Substitute specification
- ☐ Verified small status claim
- ☐ Other _____

Notes: Use IA from IB

35 U.S.C. 371 - Receipt of Request (PTO-1390)	17 Nov '08
Date acceptable oath / declaration received	
Date complete 35 U.S.C 371 requirements met	
102(e) Date	
Date of completion of DO/EO 906 - Notification of Missing 102(e) Requirements	
Date of completion of DO/EO 907 - Notification of Acceptance for 102(e) date	
Date of completion of DO/EO 911 - Application accepted under 35 U.S.C. 1.11	
Date of completion of DO/EO 905 - Notification of Missing Requirements	
Date of completion of DO/EO 916 - Notification of Defective Response	
Date of completion of DO/EO 903 - Notification of Acceptance	
Date of completion of DO/EO 909 - Notification of Abandonment	

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Publication No.
WO07/136760A2

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29 Nov '07

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(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
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PCT

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(30) Priority Data:
60/801,661 19 May 2006 (19.05.2006) US

(71) Applicant *(for all designated States except US)*: **THE JOHNS HOPKINS UNIVERSITY** [US/US]; 3400 N. Charles Street, Baltimore, MD 21218 (US).

(72) Inventor; and
(75) Inventor/Applicant *(for US only)*: **MCNIECE, Ian** [AU/US]; 1609 Pot Spring Road, Lutherville, MD 21093 (US).

(74) Agents: **CORLESS, Peter, F.** et al.; Edwards Angell Palmer & Dodge Llp, P. O. Box 55874, Boston, MA 02205 (US).

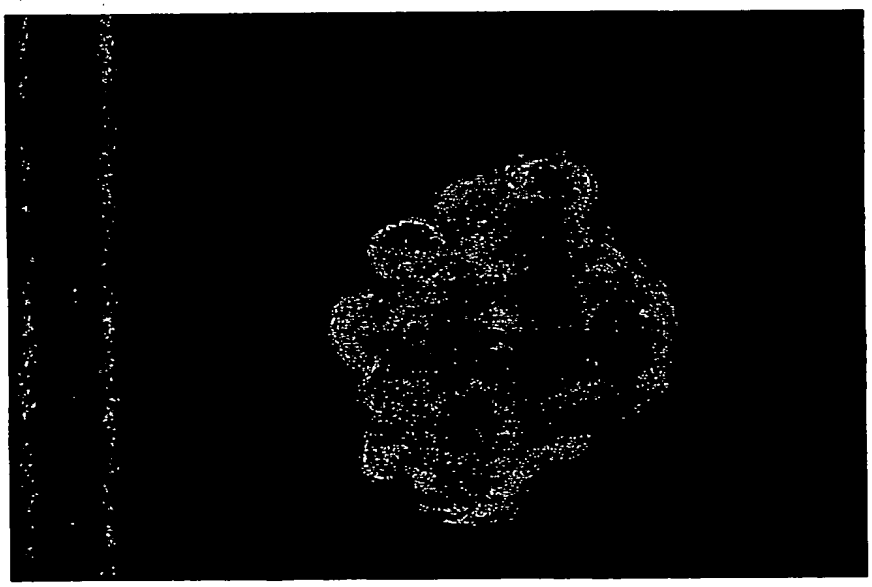
(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *without international search report and to be republished upon receipt of that report*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: METHOD OF GROWTH OF MESENCHYMAL CELLS UNDER NON-ADHERENT CONDITIONS FOR CLINICAL APPLICATIONS



(57) Abstract: The invention provides methods for expanding mesenchymal stem cells (MSCs) in non-adherent cultures. The methods include the propagation of MSCs in or on non-adherent matrices. The invention further provides administration and the use of cells propagated by the method of the invention for administration and preparation of a therapeutic agent. The invention further provides kits including cells propagated by the methods of the inventions.

WO 2007/136760 A2

PATENT APPLICATION FEE DETERMINATION RECORD

Effective October 2, 2008

Application or Docket Number: 12/227458

CLAIMS AS FILED - PART I

	(Column 1)		(Column 2)
U.S. NATIONAL STAGE FEES			
BASIC FEE			
EXAMINATION FEE			
SEARCH FEE			
FEE FOR EXTRA SPEC. PGS.		minus 100 =	/ 50 =
TOTAL CHARGEABLE CLAIMS	18	minus 20 =	*
INDEPENDENT CLAIMS	1	minus 3 =	*
MULTIPLE DEPENDENT CLAIM PRESENT			☐

* If the difference in column 1 is less than zero, enter "0" in column 2

SMALL ENTITY TYPE ☐	RATE	FEE	OR	OTHER THAN SMALL ENTITY	RATE	FEE
	BASIC FEE	165	OR		BASIC FEE	330
	EXAM. FEE	110			EXAM. FEE	220
	SEARCH FEE	~~245~~ 350			SEARCH FEE	430
	X $ 135 =				X $ 270 =	
	X $ 26 =		OR		X $ 52 =	
	X $ 110 =		OR		X $ 220 =	
	+ $ 195 =		OR		+ $ 340 =	
	TOTAL	325 / 390	OR		TOTAL	

CLAIMS AS AMENDED - PART II

AMENDMENT A

	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA
Total	*	Minus	**	=
Independent	*	Minus	***	=
FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM				☐

SMALL ENTITY	RATE	ADDITIONAL FEE	OR	OTHER THAN SMALL ENTITY	RATE	ADDITIONAL FEE
	X $ 25 =		OR		X $ 50 =	
	X $ 100 =		OR		X $ 200 =	
	+ $ 180 =		OR		+ $ 360 =	
	TOTAL ADDIT. FEE		OR		TOTAL ADDIT. FEE	

AMENDMENT B

	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA
Total	*	Minus	**	=
Independent	*	Minus	***	=
FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM				☐

	RATE	ADDITIONAL FEE	OR		RATE	ADDITIONAL FEE
	X $ 25 =		OR		X $ 50 =	
	X $ 100 =		OR		X $ 200 =	
	+ $ 180 =		OR		+ $ 360 =	
	TOTAL ADDIT. FEE		OR		TOTAL ADDIT. FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than '20', enter "20".
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than '3', enter "3".
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.

MULTIPLE DEPENDENT CLAIM FEE CALCULATION SHEET

(FOR USE WITH FORM PTO-875)

SERIAL NO. 12/227458

FILING DATE

APPLICANT(S)

CLAIMS

	AS FILED		AFTER 1st AMENDMENT		AFTER 2nd AMENDMENT	
	IND.	DEP.	IND.	DEP.	IND.	DEP.
1	1		1			
2		1		1		
3		1		1		
4		3		1		
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TOTAL IND.	1	⬇	1	⬇		⬇
TOTAL DEP.	21	⬅	17	⬅		⬅
TOTAL CLAIMS	23		18			

	AS FILED		AFTER 1st AMENDMENT		AFTER 2nd AMENDMENT	
	IND.	DEP.	IND.	DEP.	IND.	DEP.
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100						
TOTAL IND.		⬇		⬇		⬇
TOTAL DEP.		⬅		⬅		⬅
TOTAL CLAIMS						

PTO/SB/06 (07-06)
Approved for use through 1/31/2007. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
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PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 12/227,458	Filing Date 10/23/2009	☐ To be Mailed

APPLICATION AS FILED – PART I

	(Column 1)	(Column 2)	SMALL ENTITY ☒		OR	OTHER THAN SMALL ENTITY	
FOR	NUMBER FILED	NUMBER EXTRA	RATE ($)	FEE ($)		RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A	N/A			N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A	N/A			N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A	N/A			N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*	X $ =		OR	X $ =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*	X $ =			X $ =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $250 ($125 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).						
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))							
* If the difference in column 1 is less than zero, enter "0" in column 2.			TOTAL			TOTAL	

APPLICATION AS AMENDED – PART II

AMENDMENT	11/17/2008	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	SMALL ENTITY RATE ($)	ADDITIONAL FEE ($)	OR	OTHER THAN SMALL ENTITY RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 18	Minus	** 20	= 0	X $26 =	0	OR	X $ =	
	Independent (37 CFR 1.16(h))	* 1	Minus	***3	= 0	X $110 =	0	OR	X $ =	
	☐ Application Size Fee (37 CFR 1.16(s))									
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							OR		
						TOTAL ADD'L FEE	0	OR	TOTAL ADD'L FEE	

AMENDMENT		(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=	X $ =		OR	X $ =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=	X $ =		OR	X $ =	
	☐ Application Size Fee (37 CFR 1.16(s))									
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							OR		
						TOTAL ADD'L FEE		OR	TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.

** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".

*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".

The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.

Legal Instrument Examiner:
/ROZENIA HARMON/

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

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