(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
13 December 2012 (13.12.2012)

WIPO | PCT

(10) International Publication Number
WO 2012/170490 A3

(51) International Patent Classification:
A01N 1/02 (2006.01) *C12N 5/02* (2006.01)
C12N 5/071 (2010.01)

(21) International Application Number:
PCT/US2012/041048

(22) International Filing Date:
6 June 2012 (06.06.2012)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
61/493,695 6 June 2011 (06.06.2011) US

(71) Applicant *(for all designated States except US)*: **INVIVOSCIENCES, LLC** [US/US]; 6102 Canyon Parkway, McFarland, WI 53558 (US).

(72) Inventor; and

(75) Inventor/Applicant *(for US only)*: **WAKATSUKI, Tetsuro** [US/US]; 6102 Canyon Parkway, McFarland, WI 53558 (US).

(74) Agents: **VANHEYNINGEN, Tambryn, K.** et al.; Andrus, Sceales, Starke & Sawall, LLP, 100 East Wisconsin Avenue, Suite 1100, Milwaukee, WI 53202 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: 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, 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.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (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,

[Continued on next page]

(54) Title: ENGINEERED CARDIAC TISSUES AND METHODS OF USING THEM

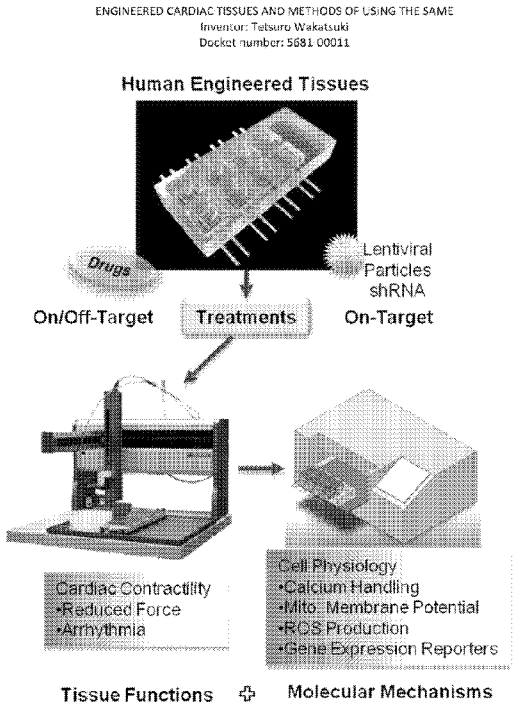

Fig. 1

(57) Abstract: Disclosed are engineered cardiac tissues. The tissues include cardiomyocyte cells derived from a pluripotent cell, fibroblast cells and extracellular matrix components. The growth ability of the fibroblast cells may be limited. The tissue contracts coherently or synchronously with electrical pacing by an external electrical stimulator. Methods of using the tissues described herein are also provided.

WO 2012/170490 A3

TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

(88) Date of publication of the international search report: 8 May 2014

Published:

— *with international search report (Art. 21(3))*

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 12/41048

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - A01N 1/02; C12N 5/071; C12N 5/02 (2012.01)
USPC - 435/1.1; 435/366; 435/377
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC(8) - A01N 1/02; C12N 5/071; C12N 5/02; C12N 15/00 (2012.01)
USPC - 435/1.1; 435/366; 435/377; 435/325; 435/375; 435/455

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
IPC(8) - A01N 1/02; C12N 5/071; C12N 5/02; C12N 15/00 (2012.01) - see keyword below
USPC - 435/1.1; 435/366; 435/377; 435/325; 435/375; 435/455 - see keyword below

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
PubWEST(USPT,PGPB,EPAB,JPAB); Medline, Google: engineered, cardiac tissue, ECT, cardiomyocyte, cardiac myocyte, pluripotent, cell, fibroblast, extracellular matrix, ECM, contract, coherent, synchronous, heart, EHT, 3D tissue, ROCK kinase inhibitor, stem cell, SC, hSC, ES, iPSC, electric, pacing, paced, stimulate, Hz, human, stimulating, inhibit

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- Y	GUO et al. Creation of Engineered Cardiac Tissue In Vitro From Mouse Embryonic Stem Cells. Circulation. 2006, Vol. 113(18), 2229-37. Abstract; pg 2230, col 2, para 3; pg 2231, col 1, para 1, and col 2, para 1 and 2; and pg 2235, col 2, up para	3, 5/(3) ---------- 1-2, 4, 5/(1-2,4)
Y	BOATENG et al. Inhibition of fibroblast proliferation in cardiac myocyte cultures by surface microtopography. Am J Physiol Cell Physiol. 2003, Vol. 285(1), C171-82. Abstract; pg C171, col 1, lower para, and col 2, top para and last para; pg C172, col 1, top para ; pg C177, col 2, para 1; and pg C180, col 2, middle para	1-2, 4, 5/(1-2,4)
A	PARKER et al. Extracellular matrix, mechanotransduction and structural hierarchies in heart tissue engineering. Philos Trans R Soc Lond B Biol Sci. 2007, Vol. 362(1484), p. 1267-79. pg 1267, col 2, lower para	1-5
A	US 2009/0202977 A1 (OTT et al.) 13 August 2009 (13.08.2009), para [0006], [0016], [0108], and [0109]	1-5
A	GERECHT-NIR et al. Biophysical regulation during cardiac development and application to tissue engineering. Int J Dev Biol. 2006, Vol. 50(2-3), 233-43. Entire documentation, especially Abstract; pg 234, col 2, middle para; and pg 237, Table 1	1-5
A	ZIMMERMANN et al. Engineered heart tissue for regeneration of diseased hearts. Biomaterials. 2004, Vol. 25(9), p.1639-47. Abstract; pg 1640, col 2; pg 1642, col 1, para 1; and pg 1643, col 1, last para and Fig 5	1-5
A	US 2009/0169521 A1 (LEVENBERG et al.) 02 July 2009 (02.07.2009), Abstract, para [0008], [0009], [0039], [005], [0057], [0060], [0111], [0115], [0165], [0181], [0193], and [0196]	1-5

☐ 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	Date of mailing of the international search report
25 July 2012 (25.07.2012)	16 OCT 2012
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) (July 2009)