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(71) Applicant *(for all designated States except US)*: **MONSANTO TECHNOLOGY LLC** [US/US]; 800 North Lindbergh Boulevard, St. Louis, Missouri 63102 (US).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **DURACK, Gary** [US/US]; 2505 Appaloosa Lane, Mahomet, IL 61853 (US). **HATCHER, Jeremy, T.** [US/US]; 509 East Shurts Street, Urbana, IL 61801 (US). **WESTFALL, Lon, A.** [US/US]; 1007 Timber Drive, Mahomet, IL 61853 (US). **HELBLING, David, R.** [US/US]; 904 South Fawn Drive, Mahomet, IL 61853 (US). **WALLACE, Jeffrey, D.** [US/US]; 666 Autumn Fields Lane, Rantoul, IL 61866 (US). **VANDRE, Gary, P.** [US/US]; 105 Sharon Drive, Mahomet, IL 61853 (US). **DIDION, Bradley** [US/US]; 220 Hickory Hollow, Washington, MO 63090 (US). **NAYAK, Niraj, V.** [US/US]; 504 Avenue G, Apt. #24, Redondon Beach, CA 90277 (US). **ANZAR, Muhammad** [CA/US]; 14506 Tienda Drive, Chesterfield, MO 63017 (US). **LUDWIG, Cindy, L.** [US/US]; 1412 Dautel Lane, St. Louis, MO 63146 (US). **GRAHAM, Jeffrey, A.** [US/US]; 49 Picardy Hill Drive, Chesterfield, MO 63017 (US). **CROWLEY, Kathleen, S.** [US/US]; 315 Carmel Road, Webster Groves, MO 63119 (US).

(74) Agents: **GODAR, Michael, E.** et al.; Senniger, Powers, Leavitt & Roedel, #1 Metropolitan Square, 16th Floor, St. Louis, Missouri 63102 (US).

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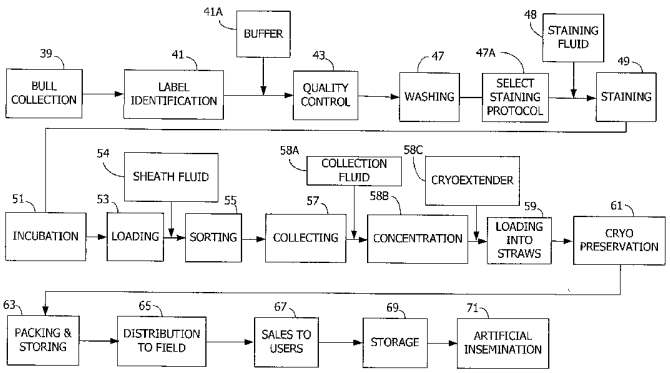
[Continued on next page]

(54) Title: APPARATUS AND METHODS FOR PROVIDING SEX-SORTED ANIMAL SPERM



(57) Abstract: Apparatus and methods for analyzing particles, including apparatus and methods for a sperm sorting process including: collecting sperm from an animal (30); selecting staining conditions (47A); staining the sperm with DNA selective fluorescent dye (48); sorting the sperm cells according to sex chromosome content (55); and cryopreserving a population of sorted sperm (61) until used for artificial insemination. One embodiment includes apparatus (1001) and methods for using a plurality of flow cytometry units (9) sharing an integrated platform to sort sperm cells. In one embodiment, flow cytometric sorting includes use of the following apparatus and methods: an orienting nozzle having a baffle (101); an epi-illumination optics system (109); slit scanning of localized DNA regions within cell nuclei (225); digital signal processing, including synchronous sampling of analog output signals (701), pulse waveform (497) feature extraction of an approximation of a first order derivative of a pulse waveform (497) at a point of the pulse, any of various sort strategies; and an automated sort calibration system (4201). In one embodiment, digital signal processing includes sampling analog output signals (701) at times relative to emission of pulses from an illumination laser. Other embodiments are substantially different from the foregoing, including embodiments directed to individual steps or systems that can be used for any of various applications involving particle analysis.

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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, US, UZ, VC, VN, YU, ZA, ZM, ZW.

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International Application No
PCT/US2004/009646

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 G01N33/487 G01N15/14 G01N33/50 C12N5/06 C12Q1/04

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 7 G01N C12N C12Q

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

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

EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	JOHNSON L A ET AL: "SEX PRESELECTION: HIGH-SPEED FLOW CYTOMETRIC SORTING OF X AND Y SPERM FOR MAXIMUM EFFICIENCY" THERIOGENOLOGY, LOS ALTOS, CA, US, vol. 52, no. 8, 1999, pages 1323-1341, XP001023997 ISSN: 0093-691X	1-15
X	the whole document	16,19, 118,145

Y	US 6 149 867 A (SCHENK JOHN ET AL) 21 November 2000 (2000-11-21)	136,137, 140-144, 149
X	the whole document	16,19, 22,30, 37,39, 41,43,45

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
2 June 2005	10. 06. 2005
Name and mailing address of the ISA European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31–70) 340–2040, Tx. 31 651 epo nl, Fax: (+31–70) 340–3016	Authorized officer Diez Schlereth, D

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Form PCT/ISA/210 (second sheet) (January 2004)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 00/06193 A (MCCUE PATRICK M ; SEIDEL GEORGE E (US); XY INC (US); COLORADO STATE UN) 10 February 2000 (2000-02-10)	1-15
X	the whole document	16,19, 22,30, 37,39, 41,43,45
A	WO 01/85913 A (EVANS KENNETH M ; XY INC (US); MUNSTER ERIK B VAN (US)) 15 November 2001 (2001-11-15)	1-15
X	the whole document	16,19, 22,30, 37,39, 41,43,45
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X	the whole document	16,19, 46,55, 63,65, 67,71,77
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Y	the whole document	98-117
Y	US 6 263 745 B1 (HERICKHOFF LISA A ET AL) 24 July 2001 (2001-07-24)	127-131
X	the whole document	118,120, 124-126, 132,145
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Y	claims 1-22; figures 2-12	13, 98-117
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Y	the whole document & DATABASE WPI Derwent Publications Ltd., London, GB; AN 1987-169389 abstract	98-117

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INTERNATIONAL SEARCH REPORT

International Application No
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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 6 042 025 A (CRAMPTON ET AL) 28 March 2000 (2000-03-28) the whole document	127-131
Y	WO 02/052244 A (AMERSHAM BIOSCIENCES AB; NORLING, BORJE; PERTOFT, HAKAN) 4 July 2002 (2002-07-04) the whole document	136,137
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Y	FR 2 813 182 A (MOAZZEZI SAADOLLAH) 1 March 2002 (2002-03-01) the whole document	149

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2004/009646

Box 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. [X] Claims Nos.: 148
because they relate to subject matter not required to be searched by this Authority, namely:

6 fees paid for inventions i-vi

2. [ ] Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box 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:

see additional sheet

1. [ ] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [X] 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.:

1-147,149

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.

[X] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-15

   Multichannel system comprising a plurality of flow cytometry units and methods for classifying/sorting particles using said system

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2. claims: 16-45,138

   Flow cytometer apparatus/system for sorting particles having a control responsive to information for varying sorting strategy or for controlling fluid delivery system and sorting method using said apparatus

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3. claims: 46-80,139

   Flow cytometer system having a A/D converter and a digital signal processor and methods of using it.

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4. claims: 81-117

   Flow cytometer for sorting particles comprising epi-illumination optics and sorting methods using said apparatus

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5. claims: 118-135,145-147

   Nozzle for flow cytometer, flow cytometer comprising said nozzle and methods for orienting and sorting particles using said nozzle

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6. claims: 136- 137,140-144,149

   methods for processing sperm cells

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7. claim: 148

   method for evaluation of a set of conditions for staining a population of cells

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/US2004/009646

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