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LLNL-TR-739115

Sperm Scoring Using Multi-Spectral Flow Imaging and FISH-IS Final Report CRADA No. TC02088.0

F. Marchetti, P. J. Morrissey

September 28, 2017

Disclaimer

This work performed under the auspices of the U.S. Department of Energy by Lawrence Livermore National Laboratory under Contract DE-AC52-07NA27344.

Sperm Scoring Using Multi-Spectral Flow Imaging and FISH-IS

Final Report
CRADA No. TC02088.0
Date Technical Work Ended: March 24, 2006

Date: April 28, 2006 Revision: 1

A. Parties

This project was a relationship between Lawrence Livermore National Laboratory (LLNL) and Amnis Corporation.

The Regents of the University of California
Lawrence Livermore National Laboratory
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Livermore, CA 94550
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Amnis Corporation
2505 Third Avenue, Suite 210
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B. Project Scope

This was to be a collaborative effort between The Regents of the University of California, Lawrence Livermore National Laboratory (LLNL) and Amnis Corporation, to develop an automated system for scoring sperm interphase cells for the presence of chromosomal abnormalities using fluorescence in situ hybridization and the Amnis ImageStream technology platform.

The project consisted of six (6) main tasks and the following major deliverables:

Deliverable 1
Protocol for hybridizing probes for chromosomes X, Y and 8 to murine sperm in suspension. (Months 1-2) (Amnis)

Deliverable 2
Truth file of imagery generated by the IS100. (Months 1-8) (Amnis & LLNL)

Deliverable 3
FISH spot counting classifier integrated into IDEAS software program. (Months 6-8) (Amnis & LLNL)

Deliverable 4
Results of analysis of normal murine sperm using imagery obtained with the IS100 and analyzed with the spot counting classifier in the IDEAS software package and by standard FISH scoring. (Months 8-9) (Amnis and LLNL)

Deliverable 5
Validated FISH spot classifier using frozen aneuploid murine sperm samples previously scored by conventional FISH scoring. (Months 10-12) (Amnis & LLNL)

Deliverable 6
Protocol for hybridizing chromosomal probes to human sperm and results of analysis of aneuploidy using imagery from the IS100 and analyzed with the spot counting classifier in the IDEAS software package. (Months 9-12) (Amnis)

Deliverable 7
Final Report and Abstract due within thirty (30) days of completion or termination of the project, as required under Article XI of the CRADA.

This CRADA was executed on February 3, 2006 and was originally designated as a twelve (12) month project. Due to changes in personnel and a shift in program priorities at LLNL, the CRADA was terminated effective April 24, 2006.

C. Technical Accomplishments

The following tasks were completed by LLNL under this CRADA prior to its termination.

1) Prepared mouse DNA probe for chromosome Y to be used by Amnis for hybridization in suspension.
2) Teleconference for discussing the functionality of Amnis image analysis software. Installed Amnis image analysis software and performed preliminary analysis on image file provided by Amnis.
3) Reviewed images of sperm that had been hybridized with the chromosomal probes and imaged with the Amnis ImageStream technology and provided feedback on scoring these images to Amnis.

This project work was terminated on March 24, 2006 due to changes in personnel and a shift in program priorities at LLNL.

D. Expected Economic Impact

This CRADA would have benefited US taxpayers by developing a high throughput technique to provide information for environmental protection and drug developments that would lead to lower abnormal pregnancy and birth defects. The substantial increase in efficiency provided by automating the sperm aneuploidy assay would result in a significant decrease in manpower currently required to perform the assay manually.

D.1 Specific Benefits

Benefits to DOE
This CRADA would have benefited DOE by contributing to accomplish DOE missions in environmental protection and in enhancing the health of the general public.

Benefits to Industry
This CRADA would have benefited industry and the Industrial Participant by developing a technology to assess aneuploidy rates in sperm to identify genetic, physiological and environmental risk factors with a greatly increased sample throughput and automated analysis.

E. Partner Contribution

Protocol for hybridizing probes for chromosomes X, Y and 8 to murine sperm in suspension was developed and provided by Amnis (Deliverable 1).

Amnis Corp provided image analysis software and an image file containing images of murine sperm hybridized to probes for chromosomes 8, X and Y.

No subject inventions were created during the CRADA project.

F. Documents/Reference List

Reports
None

Copyright Activity
None

Subject Inventions
None

Background Intellectual Property

LLNL disclosed the following Background Intellectual Property (BIP) for this project:

U.S. Patent Application No. 10/738947 (LLNL Docket IL-11097) - *Multilabeling Mouse FISH Assay for Detecting Structural and Numerical Chromosomal Abnormalties*; Inventors: Andrew J. Wyrobek, Francesca S. Pearson (formerly Hill), Francesco O. Marchetti

This CRADA was terminated shortly after execution, and Amnis Corporation has not expressed an interest in licensing this BIP.

Amnis Corporation disclosed 22 issued patents and 14 patent applications as Background Intellectual property for this project.

G. Acknowledgement

Industrial Participant's signature of the final report indicates the following:

1) The Participant has reviewed the final report and concurs with the statements made therein.
2) The Participant agrees that any modifications or changes from the initial proposal were discussed and agreed to during the term of the project.
3) The Participant certifies that all reports either completed or in process are listed and all subject inventions and the associated intellectual property protection measures generated by his/her respective company and attributable to the project have been disclosed and included in Section E or are included on a list attached to this report.
4) The Participant certifies that if tangible personal property was exchanged during the agreement, all has either been returned to the initial custodian or transferred permanently.
5) The Participant certifies that proprietary information has been returned or destroyed by LLNL.

Philip J. Morrissey — May 8, 2006

Philip J. Morrissey, Ph.D., Vice President/Biology — Date
Amnis Corporation

5/22/06

Francesco Marchetti, LLNL Principal Investigator — Date
Lawrence Livermore National Laboratory

Norma E Dunycal — June 2, 2006

Karena D. McKinley, IPAC Director — Date
Lawrence Livermore National Laboratory

Industrial Partnerships and Commercialization

Mail Station L-795

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Fax 3-8988

May 30, 2006

MEMORANDUM

To: Distribution

From: Karena McKinley

Subject: Signature Authority

In my absence May 30 through June 2, I hereby delegate signature authority to Norma Dunipace for all IPAC-related documents.

Director, Industrial Partnerships
and Commercialization

Distribution:
Norma Dunipace
Kathy Kaufman
IPAC Staff

University of California
**LAWRENCE LIVERMORE
NATIONAL LABORATORY**

Sperm Scoring Using Multi-Spectral Flow Imaging and FISH-IS

Final Abstract (Attachment I)
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B. Purpose and Description

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This CRADA was executed on February 3, 2006 and was originally designated as a twelve (12) month project. Due to changes in personnel and a shift in program priorities at LLNL, the CRADA was terminated effective April 24, 2006. Task 1 and Deliverable 1 were successfully completed prior to termination of the CRADA.

C. Benefit to Industry

This CRADA would have benefited industry and the Industrial Participant by developing a technology to assess aneuploidy rates in sperm to identify genetic, physiological and environmental risk factors with a greatly increased sample throughput and automated analysis.

D. Benefit to DOE/LLNL

This CRADA would have benefited DOE by contributing to accomplish DOE missions in environmental protection and in enhancing the health of the general public.

E. Project Dates

Planned project dates: February 3, 2006 through February 3, 2007
Actual project dates: February 3, 2006 through March 24, 2006