



microScience Imaging

September
2006
No 2



Analyzing Stardust Mission Samples

By Matthew Batchelor, *Meyer Instruments Applications Engineer*
and Lon Nelson, *Leica Marketing Manager*

On January 2, 2004, the Stardust spacecraft flew through the coma of Comet Wild-2 within 140 miles of the nucleus and collected dust samples that emanate from the comet's surface. The Stardust mission is important because the common theory is that comets represent pristine, ancient materials that are left over from the formation of our solar system over 4.5 billion years ago.

The Stardust spacecraft returned to Earth on January 15, 2006 at the Utah Test & Training Range (UTTR) in northern Utah. Following the recovery of the spacecraft, its precious cargo was flown back to Houston to be opened in the newly constructed Class 100 Stardust Laboratory cleanroom located at the Johnson Space Center (JSC). "The collection of cometary particles has exceeded our expectations," says Dr. Donald Brownlee, Stardust principal investigator from the University of Washington, Seattle. "We were absolutely thrilled to see thousands of impacts on the aerogel." (Aerogel is a substance used to collect the particles)

The Stardust Science Team at the JSC uses a system to scan aerogel collectors at high resolution for the locations of cometary particle penetrations and impacts. The overall Stardust sample collector is slightly larger than the head of a tennis racket and is divided into 132 individual cells, each holding one 2x4x3cm aerogel tile. The scanning system consists of a Leica MZ16 A stereomicroscope and high-precision Texonics, Inc. positioning devices, both controlled by a Dell™ computer system. The Stardust team is analyzing the particle capture cells and removing individual grains of comet and interstellar dust, which are being sent to select investigators worldwide.

The positioning system, Leica microscope, and Image-Pro Plus software by Media Cybernetics are controlled by Aardvark Software, Inc., a custom application developed by Leica Microsystems dealer, Meyer Instruments, in Houston, Texas. The Windows™-based application allows the Stardust Science Team to set up small or large areas of interest for investigation and automatically images these areas at any magnification that the microscope system can handle. The system automatically acquires and stores the images, and then moves onto the next frame to repeat the process until the defined area has been completely imaged.

After the overview imaging of the aerogel has provided sufficient information about the specific tiles, single particulates are dissected out and further analyzed. The Houston-based team, in collaboration with researchers at the University of California–Berkeley, has utilized the Leica DM6000 M compound research microscope for more detailed analysis of Wild-2's dust.

For more information about the Stardust mission, visit <http://stardust.jpl.nasa.gov>



Aerogel tracks



Composite Stardust image of Comet Wild 2

Photos courtesy of APSA/JPL/UW

Contents

Stardust Mission.....	page 1
Technology Fast Track.....	page 2
The Private Eye.....	page 3
Your Educational Resource.....	page 3
Open Forum.....	page 4
The Objective.....	page 4
For You.....	page 4
Upcoming Events.....	page 5
Your Local Leica Team.....	page 5

Technology Fast Track



Mt. Isarog, Camarines Sur Province, Philippines

Leica Microsystems, Inc. Aids Research in the Philippines

by Lon Nelson, *Leica Marketing Manager*

Far from the bustling cities of the Philippines rises Mt. Isarog, a dormant volcano blanketed in pristine rain forest. This jewel of mega-diversity rises 1,966 meters from sea level and is a prime location for entomologists to collect, identify, catalog, and preserve new and known varieties of ants.

Unfortunately, this process is very difficult due to the lack of significant research funding in the Philippines. Without funding forces like NIH, NSF, and USDA in the United States, researchers in the Philippines are forced to use sub-standard stereomicroscopes that provide cloudy images at best and offer limited opportunities for digital image capture and archiving.

Enter Dr. Gary Alpert, Entomologist, from Harvard University (Environmental Health and Safety Department) and Leica Microsystems. Dr. Alpert has teamed up with Dr. Dave General of the Institute for Environmental Conservation and Research (INECAR), Ateneo de Naga University (Naga City, Philippines) to assist in identifying, cataloging, and digitally archiving Philippine ants. The overriding quest of this project is to reduce disease, increase food production, stop destructive species, protect endangered ones, and enjoy rather than struggle with nature.

Leica Microsystems provided a Leica StereoZoom™ S6 D to Dr. General and his team of young researchers in the Philippines. Now armed with the proper imaging equipment, Dr. General's troops head out to the rain forest to collect specimens. Students collect ants by either plucking them from fallen trees and mounds of dirt or by catching them in large sheets as they fall from shaken vegetation. The ants are carefully transferred to vials containing alcohol and are transported back to the lab. Students learn to process, identify, and preserve rare ants and new species of ants under the direction of Dr. General. At present, no one else in the Philippines outside of this group studies ants to the species level.

Dr. General has attached a digital camera to the microscope and electronically sends clear images to Dr. Alpert at Harvard to consult with him about ant species identification. When a new species is discovered, Dr. General's group carefully mounts and packs the specimens for shipment to Harvard for higher-resolution imaging. Dr. Alpert has long been a Leica Microsystems customer and currently owns several Leica MZ16 and other M-series stereomicroscopes set up with AX carriers for on-axis image stacking (utilizing Syncrosopy's Auto Montage™ and Leica's LAS Montage software). Thus, Philippine biodiversity is documented and archived for future generations of scientists.

Dr. General also digitally preserves the ant collections of the Philippine National Museum and the Museum of Natural History (MNH) of the University of the Philippines at Los Baños, which are the two most important collections in the Philippines today. During this process, Dr. General and Dr. Alpert verify that previous species determinations are accurate, thereby ensuring the quality of the collections. Says Dr. General, "We are able to appreciate the historical value of the (existing) collections. The MNH collection contains specimens that may have been identified by the great Dr. John W. Chapman*



Dr. Dave General with the Leica S6 D.



A young researcher collects ant specimens from a decaying tree.



Students shake the vegetation and use blankets to catch ants that fall.



Lengthwise image stack of a Philippine ant (*Polyrhachis* species); single images taken with the Leica S6 D. **

continued on page 5

The Private Eye

Is NeuroLucida Right for My Research?

by Craig Wollschlager, *Leica Product Manager*

Application / Question:

I would like to analyze anatomical features of neurons and synaptic boutons in my knockout mice. What are my options and how much would they cost? Is NeuroLucida right for me?

Answer:

The answer is that it is all dependent upon how detailed your analysis needs to be. There are systems that provide anything from basic point-to-point measuring and 2-D imaging up to high-resolution confocal for real-time, 3-D imaging.

Basic:

Research light microscope with camera and basic software for capturing 2-D images.

Most commercially available systems provide image acquisition, documentation, and data analysis. This solution provides you with the ability to perform basic point-to-point measurements of neurons. You can then annotate or draw directly on the 2-D image for documentation or publication.

Leica solution: DM2500 B microscope with DFC300 FX camera and LAS software

Cost: \$30K to \$50K

Intermediate:

Automated research light microscope with camera and deconvolution software for 3-D image construction.

This solution provides everything listed in the basic package above plus image overlay, 3-D visualization with deconvolution, enhancement, and data analysis. This would allow you to view the neurons and boutons in 3-dimensions for more accurate analysis of dendritic branching.

Leica solution: DM5500 B microscope with DFC350 FX camera and LAS AF6000 software

Cost: \$60K to \$90K

Advanced:

High-resolution spectral scanning confocal and software.

This solution is a highly integrated system providing optimal resolution and 3-D visualization without the necessity of time-consuming, image deconvolution. Confocal systems give you the ability to do everything listed in the intermediate package above plus acquire real-time 3-D images for even more detailed neuronal analysis.

Leica solution: TCS SPE Personal Spectral Confocal

Cost: \$100K+

For a more quantitative, morphological analysis of dendritic branching, NeuroLucida (MicroBrightField, Inc.) software may be combined with any of the SE solutions. NeuroLucida is advanced scientific software for performing brain mapping, neuron tracing, anatomical mapping and branching, image analysis, and morphometry. Designed specifically for neuroscience applications, NeuroLucida can also be used in any situation where 3D reconstruction, volumetric analysis, or cell counting is required. NeuroLucida is definitely the pinnacle of neuronal analysis packages and prices for this solution vary widely depending upon the detail of your analysis.

([http:// www.mbfbioscience.com](http://www.mbfbioscience.com))



Your Educational Resource

Upcoming Science Courses in 2006:

Cold Spring Harbor:

- X-ray Methods in Structural Biology (**October 16-31**)
- Immunocytochemistry, In Situ Hybridization & Live Cell Imaging (**October 23-November 5**)

<http://meetings.cshl.edu/courses.html>

The Jackson Laboratory

- Workshop on Surgical Techniques in the laboratory Mouse (**November 5-10**)
- Colony Management: Principles and Practices (**December 3-7**)

<http://www.jax.org/courses/events>

Mount Desert Island Biological Laboratory:

- Health and Colony Management of Laboratory Fish (**September 18-22**)

<http://www.mdibl.org/course>

Open Forum

How Do I Reduce Autofluorescence When Imaging Fixed Cells?

Autofluorescence can be a frustrating problem that obscures your signal of interest when imaging fixed cells. Typically, the shorter wavelengths are affected, which can make imaging reporter proteins such as CFP and GFP difficult.

Although there are inherent cellular substances that autofluoresce, there are also treatments to reduce some of these problems. For example, autofluorescence from lipofuscin, a substance that accumulates with age, can be reduced with sudan black or CuSO₄. Autofluorescing structural components like elastin or collagen can be masked with pontamine sky blue. Also, commonly used aldehyde fixatives; i.e., formaldehyde and glutaraldehyde, can generate fluorescence, so alternative fixatives such as carnoys or methacarn may be helpful.

One should also verify that the spectral characteristics of the fluorescence filter set are appropriate for the desired fluorophore. New narrow band, high-efficiency filters help to pull out your desired signals and block those that are extraenous. These fluorescence filters are now available as standard for all Leica compound and stereo microscopes. Hopefully these suggestions will provide solid starting points to isolate the source of autofluorescence and eliminate or reduce it from your images. — Chris Vega, *Leica Product Manager*

Special thanks to NFS Graduate Research Fellow Teresa Murray, Center for BioOptical Nanotechnology, Biodesign Institute at Arizona State University, for submitting her question on autofluorescence. Teresa will receive a FREE Leica Lens Care Kit and Teresa's Lab will receive \$500 toward their next Leica purchase.

We welcome your questions! Submit a question to the Open Forum section and receive a FREE Leica Lens Care Kit! If your question is chosen for use in a future issue, you also will receive a **\$500 Leica Product Voucher** on your next purchase!



For You

Special product offers currently available from Leica Microsystems:

Upgrade to LMD Promotion

Trade in any existing Laser capture MicroDissection (LMD) system and receive up to a 40% discount on a new Leica LMD6000!

New Investigator Program

Starting up a new lab? Beginning a new research project? Contact your local Leica Microsystems Sales Executive to see if you qualify for thousands in discount!

FREE Fluorescence Illuminator Promotion

Purchase a completely configured upright or inverted research microscope with fluorescence (DM5000 B or DM14000 B models) and receive a FREE Leica EL6000 fluorescence illuminator (>\$5,000 value!). Get 50% off of the same illuminator when you purchase a Leica MZ16 F stereo-fluorescence microscope.

Demonstration Equipment Available at Deep Discounts

Contact your local Leica Sales Executive to find out about certain microscope accessories that are available from our demonstration stock; with some you receive up to a 60% discount.

Call 800-248-0123 and press 2 for more information.

The Objective

Embryonic Stem Cell Research Shows Promise for Degenerative Diseases

by Nicola Bettsworth,
Leica Product Manager

Stem cell research and treatment may yield extraordinary results for heart disease, Parkinson's disease, cancer, diabetes, as well as paralysis. The U.S. news program *60 Minutes* ran a segment concerning this subject on February 26, 2006. The program featured an interview by reporter Ed Bradley with Leica customer Dr. Robert Robbins, Chair of Cardiothoracic Surgery at Stanford University. "The cells that make up the heart muscle cells," Dr. Robbins explains, "started out as cells from embryos with the potential to develop into any type of cell." Dr. Robbins hopes to one day inject the cells, which actually beat like a heart, into someone whose heart has suffered damage. In theory, those cells would replace the damaged part of the heart.

Dr. Robbins and his team injected cells like those directly into the hearts of mice with severe cardiac disease. Researchers then tracked the cells and determined that they had stayed in the heart and were all beating in unison. After six weeks, the new heart cells had replaced the damaged ones, and heart function was restored to near normal. "There's a lot of enthusiasm for this area of research," says Dr. Robbins. "There's great hope for patients that suffer from heart failure that may be too old or can't get a heart transplant."

Stem cell research is a life science application that Leica Microsystems addresses with a comprehensive, application-oriented product solution that is built on a microscope/micromanipulation platform. The full solution can include anti-vibration stages, climate chambers, digital video systems,

continued on page 5

Leica Microsystems, Inc. Aids Research in the Philippines *(cont.)*

himself, when he was working in the Philippines.” In fact, Dr. General is now able to digitally compare the handwritten labels of Chapman locally with those in the Harvard collection to verify their authenticity.

According to Dr. Alpert, “Discover Life (www.discoverlife.org) supports our efforts to put the Philippine Ant images onto the web.” There now exists a comprehensive list of the ants of the world on the Discover Life website. Discover Life’s website was created and is authored by John Pickering from the University of Georgia at Athens, and the site’s mission is to “assemble and share knowledge about nature in order to improve education, health, agriculture, economic development, and conservation throughout the world”.

* *Well-known myrmecologist Dr. John W. Chapman was captured by the Japanese during World War II. He buried his ant collection in the mountains and lived to dig them up and send them on to Harvard University. Chapman escaped the Japanese for 17 months while he continued to collect ants in the mountains.*

** *The ant photo is a product of software assembly of multiple images, each with in-focus information from different horizontal planes. Recently, Leica Microsystems signed an agreement with the designers of this software, Syncrosopy (of Cambridge, UK), to license it for use with all Leica microscopes and digital cameras. This software package is called Leica LAS Montage and is currently available.*



Embryonic Stem Cell Research Shows Promise for Degenerative Diseases *(cont.)*

and other accessories that allow a researcher to perform stem cell research.

Since embryonic stem cells are capable of becoming any type of cell in the body, and can be grown in infinite numbers, there are many potential treatment applications such as diabetes, Alzheimer’s, and heart disease. It is hoped that stem cell procedures will replace many procedures that currently require surgery.

For more information, go to:

<http://www.cbsnews.com/stories/2006/02/23/60minutes/main1341635.shtml>



Your Local Leica Team



To find your local Leica Sales and Service Team, visit: www.leica-microsystems.us

Click on “Find Your Local Sales Representative”



Upcoming Events

Visit Leica Microsystems at the following exhibitions:

- National Association of Biology Teachers, Booth #201, Albuquerque, NM (Oct. 11)
- Society for Neuroscience Annual Meeting, Booth #1804, Atlanta, GA (Oct 14)
- NIH Tent Show (Oct 19)
- American Society for Cell Biology, Booth #600 (Dec 9)
- Entomological Society of America (Dec 11)

For more events: <http://www.leica-microsystems.us>
(click on Company, then Events)



Editorial Staff

Editors-in-Chief:	Molly Lundberg
Managing Editors:	Pam Jandura, Lon Nelson
Graphic Design:	M.N. Kennedy
Contributors:	Nicola Bettesworth, Chris Vega Craig Wollschlager, Micah Harrel
Special Thanks:	Gary Alpert, Dave General, Teresa Murray, Matthew Batchelor, NASA

Note: We are interested in your comments and thoughts about the newsletter. Please feel free to email your comments to: microscience.imaging@leica-microsystems.com