Fundamentals of Image Processing for Integrating Science, Mathematics, and Technology

## Discovering Image Processing

Version 2.0

Center for Image Processing in Education

# Discovering Image Processing Version 2.0 Development Team 

## Editor

Wynne Brown

## Adaptation and Design

Larry Kendall

## Lesson Authors

LuAnn Dahlman
Carla McAuliffe
Larry Kendall

## CIPE Staff

Steven Moore, Executive Director
Wynne Brown, Director of Publications
Hedley Bond, Programmer
Mary Moore, Senior Media Specialist
Sam Jenkins, Senior Media Specialist
Kristine Rees, Associate Director
Sandy Stoll, Business Operations Associate
Jenny Brady, Director of Teacher Training

## Concept Development

LuAnn Dahlman
Larry Kendall
Carla McAuliffe
Deborah Alongi
Steven Moore
Don Adams

## Lesson Reviewers

Mike Slootmaker
Michelle Kinzel
James Stevenson
Yuhong Liu

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## Lesson authors

LuAnn Dahlman works as a project director at TERC, a nonprofit educational research and development organization based in Cambridge, Massachusetts. She is currently heading up the Eyes in the Sky, Earth Exploration Toolbook, and Digital Library for Earth System Education (DLESE) Data Services projects. LuAnn also served as curriculum director for the Exploring Earth project, which developed an extensive interactive Web site to augment the 2003 high school Earth Science textbook by Spaulding and Namowitz. LuAnn earned a B.S. in geology from Arizona State University and taught Earth science and chemistry for seven years at Dobson High School in Mesa, Arizona. In the 1990s, she wrote curriculum materials for the University of Arizona's Image Processing for Teaching program and taught professional development workshops for the Center for Image Processing in Education in Tucson, Arizona.

Carla McAuliffe works as a curriculum developer, staff development specialist, and internal evaluator for TERC. She holds a B.S. in cellular and developmental biology, an M.Ed. in secondary education and Ph.D. in learning and instructional technology. Carla's research focuses on the role of 3-D visualization in student learning. Carla has extensive teaching experience. She has taught middle school life and physical science, high school Earth science and biology, and various college-level instructional technology courses. She continues to provide preservice and in-service teacher training and has written a variety of curriculum materials using visualization technologies, including GIS and image analysis.

Larry Kendall teaches middle school science in Apple Valley, California and works as an independent educational consultant. He earned a B.S. in secondary education and an M.A. in Earth science, and holds secondary school teaching credentials in Arizona and California. Larry has extensive experience in designing, writing, and editing curriculum materials and providing in-service teacher training in imaging and spatial technologies.

## Field testers

The developers of Discovering Image Processing appreciate the assistance of these dedicated educators and other professionals in reviewing and/or field-testing the lessons in their classrooms:

## Lisa Alewine

Curriculum Specialist
Peoria, Arizona
Meg Davis
McKemy Middle School
Tempe, Arizona

## Phil Edwards

John Marshall High School
Rochester, Minnesota

## Joe Freeman

Field Elementary School
Mesa, Arizona

JoAnne Goodwin<br>Churchill Road Elementary School Herndon, Virginia<br>Peggy Kahlstorf<br>John Marshall High School<br>Rochester, Minnesota<br>Janet Kieber<br>Shakopee Senior High School<br>Shakopee, Minnesota<br>Gillian Lunn<br>Senior Industrial Engineer<br>Motorola<br>Tempe, Arizona

## Charles Murphy

Science \& Technology Consultant
St. Louis Public Schools
St. Louis, Missouri

## Rick Rutland

San Antonio USI
San Antonio, Texas
Dianne Schrick
IMPACT
Independence, Missouri

## The Center for Image Processing in Education

Founded in 1992, the Center for Image Processing in Education (CIPE) is a nonprofit organization whose mission is to promote the use of visualization technologies for innovative educational applications. CIPE offers professional development workshops on using state-of-the-art image processing and geographic information system (GIS) tools and techniques and publishes award-winning software products for the classroom. With support from the National Science Foundation, National Institutes of Health, and other funders, CIPE is bringing image processing and geospatial technology instructional materials to technological education at the high school and community college levels.

For more information on workshops and instructional materials available through the Center for Image Processing in Education, contact:
Steven Moore, PhD
Executive Director
Center for Image Processing in Education
PO Box 13750
Tucson, AZ 85732-3750
800/322-9884
stevem@evisual.org

## Opportunities for professional growth

The Center for Image Processing in Education offers a wide range of experiences for teachers who want to continue their growth in image processing, including:

- Workshops. We offer professional development workshops at your institution and at national education conferences.
- Lesson Development. We are looking for qualified developers, testers, and contributors for future instructional materials.
- Presentations. We make presentations at national, regional, state, and local science, mathematics, and technology conferences.
If you would like to know more about these and other exciting opportunities, call CIPE at:

800/322-9884
Or check our Web site:
http://www.evisual.org

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## Introduction

## About digital image processing

## What is a digital image?

A digital image is:

- a rectangular array of measurements (light, temperature, X-rays, etc.),
- sampled at regular intervals, and
- displayed according to a lookup table, which assigns a specific color to each different pixel value.
This concept is illustrated by the series of magnifications of Mona Lisa at right. Unlike graphics, where meaning comes only from the patterns formed by many pixels, each pixel in a digital image can contain meaningful information.


## Image processing and analysis

The brightness or color of each pixel often represents a measurement of some kind, such as elevations of points on the surface of Earth, or the intensity of infrared radiation across a scene. Pixels also represent distances and areas. A length of 20 pixels in an image may represent one micron in an electron micrograph or one hundred kilometers in a satellite image.

- Image processing involves manipulating the numeric data contained in a digital image to improve or alter its appearance for either aesthetic reasons or to make image features easier to measure. Contrast enhancement, noise reduction, and edge sharpening are examples of image processing functions.
- Image analysis involves collecting, measuring, and analyzing data from processed digital images.
- Image interpretation is the process of extracting meaning from the analyses in a larger scientific context.


## ImageJ—a powerful image processing tool

ImageJ is public domain image processing and analysis software written and maintained by Wayne Rasband of the National Institutes of Health and used by scientists in many disciplines around the world. The program can acquire, display, edit, enhance, analyze, print, and animate digital images.
ImageJ has tools for measuring distance, area, brightness, location, and angles on digital images. Measurement results can be printed or exported to other software packages for further analysis. Users can enhance images through false coloring, contrast enhancement, and digital filtering using both built-in and user-defined digital filters. ImageJ also provides tools for editing images, allowing the user to draw lines, shapes, and text. The program can flip, rotate, invert, and scale selections. It supports multiple windows and many levels of magnification, and most processes can be undone.

## What is Discovering Image Processing?

Discovering Image Processing is a creative and powerful introduction to the techniques and applications of digital imaging technology. These handson lessons use real-world images and ImageJ software to model the work that scientists and technicians do every day. Using this extraordinary tool, students learn fundamental concepts of digital image processing, analysis, and interpretation. In addition to science, mathematics, and technology, these lessons explore issues and problems from a variety of fields ranging from forensics to graphic arts.

Most of the images in Discovering Image Processing were not created for classroom use-they are data culled from research laboratories, government agencies, medical facilities, and the Internet.

## Exploration, problem solving, and creative thinking

Discovering Image Processing lessons are designed to engage students in exploration and directed discovery. Using ImageJ, students explore images from a variety of sources, including satellite photos, digital elevation models, and images from space probes, scanners, and digital cameras. Students make measurements, collect and analyze data, and develop their own solutions as they work through problems.

## Integrating science, mathematics, and technology

The lessons in Discovering Image Processing integrate basic concepts in science, mathematics, and technology by presenting lesson content in an applied context, such as using animation to visualize the motion and trajectory of a storm, or creating histograms of pixel values in an image to analyze and enhance the image for better visualization of details. As a result, students learn that digital imaging technology is a technical skill relevant to the workplace.

## Philosophy

We believe that technology should be presented in a way that encourages students to actively participate, utilize materials currently in use in the world outside the classroom, and apply learned skills to a variety of vocations. The lessons in Discovering Image Processing were developed with the following goals in mind:

- Teach fundamental concepts in technology through hands-on participation, guided discovery, and critical thinking.
- Explore real-world data with appropriate technology tools.
- Integrate different disciplines through the use of image processing technology.


## Teaching fundamental concepts in technology

With Discovering Image Processing, your students will learn fundamental but important concepts in digital technology by actively participating in guided discovery, image analysis, and problem solving. Using the tools of ImageJ, they will collect, organize, analyze, and interpret data from digital images, and can work independently or cooperatively in sharing and communicating their results with others.

What's included
Discovering Image
Processing CD-ROM

- ImageJ software and installer
- Lesson data
- Lesson text


## Discovering Image

 Processing guidebook- Introductory materials
- ImageJ Skill Sheets
- Teaching notes
- Classroom lessons
- Student data sheets

The images in Discovering Image Processing come from:

- students and teachers using scanners, video, and digital cameras
- research laboratories
- medical facilities
- government agencies
- industry

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## Exploring real-world data

Discovering Image Processing is designed to provide your students with opportunities to experience technology through the analysis of digital images collected from the world around us. This approach offers them a context for the relevancy of the technology skills they will acquire. They will use image processing to explore and analyze the data in the images with more precision and detail than can typically be conveyed with traditional classroom techniques.

## Integrating disciplines

The lessons in Discovering Image Processing integrate concepts not only from mathematics, science, and technology, but also from language arts and social studies. Image processing tools and techniques are taught in applied contexts ranging from motion analysis of weather to travel planning to the investigation of crime scene evidence. Lesson materials also lend themselves to further discovery about numerous professional fields that currently utilize image processing, ranging from the arts to aerospace.
The lessons emphasize many of the workplace competencies and foundational skills recommended by the Skills and Tasks for Jobs: A SCANS Report for America 2000. These abilities include the productive use of resources, information, systems, and technology, as well as teamwork and thinking skills. In particular, using computers to process information and applying technology to tasks are two competencies fulfilled by all Discovering Image Processing lessons. When the lessons are employed in a collaborative learning environment, interpersonal competencies may also be attained. Thinking skills, such as problem solving, decision making, reasoning, and creative thinking are utilized throughout Discovering Image Processing and are a key component of the guided discovery approach.
"A major question about this technical world is, 'Who will develop and control the technologies so that they can best serve all citizens?' In the broadest sense, the answer has to be, for a democratic society, a technically literate citizenry."

American Association for the Advancement of Science
Technology: A Project 2061
Panel Report

## Image processing in the classroom

Each lesson in Discovering Image Processing uses one or more image processing skills to manipulate, analyze, and interpret digital images. Lesson instructions guide the student through the required techniques. The following table shows which skills are used in each lesson.

| Lesson |  |  | $\begin{aligned} & \frac{\pi}{0} \\ & \frac{0}{0} \\ & \frac{0}{2} \\ & \frac{2}{2} \\ & 0 \\ & 0 \end{aligned}$ |  | $\begin{aligned} & \text { 읗 } \\ & \text { 든 } \end{aligned}$ |  | Thresholding | Image enhancement |  |  | $\frac{\cdot \frac{n}{n}}{\frac{\lambda}{\frac{0}{4}}}$ | N U $\sim$ $\sim$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Pixelated Pictures | - |  | - |  | - |  | - |  |  |  |  |  |
| Lights, Camera, Action! | $\bullet$ | - |  | $\bullet$ | $\bullet$ |  |  |  | - |  |  | - |
| Digital Detective | $\bullet$ | - | - |  | - | $\bullet$ | $\bullet$ | $\bullet$ | - |  | - | - |
| More Than Numbers | $\bullet$ |  |  |  | - |  | - |  |  |  | - | - |
| North American Tour |  |  |  | $\bullet$ |  | $\bullet$ | - | $\bullet$ | $\bullet$ | - | - | $\bullet$ |
| Digitizing Details | $\bullet$ | - | - |  | $\bullet$ |  | $\bullet$ |  |  |  |  | - |
| Picture Yourself | - | - |  | - | - | $\bullet$ |  |  | - | - |  |  |
| Color by Number | $\bullet$ | $\bullet$ | - |  | $\bullet$ |  |  |  |  |  |  | - |
| Photo Effects | - | - | - |  | - |  |  |  |  |  |  | - |
| Images for Science | - | - | - | - | $\bullet$ |  |  | - | - | $\bullet$ | $\bullet$ | - |

## Cross-curriculum integration

The lessons in Discovering Image Processing integrate well with other subject areas:

## Mathematics

Digital image processing is an inherently mathematical process. Working with scaling factors in "Picture Yourself" and analyzing histograms in "Photo Effects" provides practice with the mathematics skills of using ratios and interpreting graphs. Furthermore, measurements from digital images can be exported as data to a spreadsheet program for detailed analyses. Conversely, digital images can be created by entering formulas into a spreadsheet, allowing patterns to be readily detected and analyzed.

## Science

Many of the images used in Discovering Image Processing are from scientific research. Parts of "Digital Detective" can serve as a lead-in to a unit on forensic science. Digital Elevation Models (DEMs) can be used to study landforms and topographic features. Animation can be used to visualize dynamic processes in science.

## Social studies

The geography in "North American Tour" easily ties into social studies. Students can identify the states in the U.S. as well as locate cities and other points of interest. They might choose to investigate the population densities of different regions.
"The study of technological systems should be used as a basis for providing integrated and holistic learning."

National Science Board
Educating Americans
for the 21st Century

## Language and media arts

All of the above subject areas can be tied together by language and media arts. Students can compile what they have learned into a report using a variety of formats and media. If they have used an integrated software package containing word processing, spreadsheet, and graphics capabilities, everything they have done can be brought together using the computer.

## Discovering Image Processing lesson length

The lessons in Discovering Image Processing are designed to cover all of the major image processing and analysis capabilities of ImageJ. Each lesson is centered around one or more major image processing or analysis concepts or themes. As a result, some of the lessons are longer than many school class periods. To facilitate the implementation of these lessons, we have subdivided them into sections.

## Curriculum planning grid

Use the grid below to help you integrate science, math, social studies, language arts, and media arts into your technology program.

| Lesson |  | $$ |  | $\begin{aligned} & \tilde{0} \\ & \stackrel{H}{0} \\ & \stackrel{1}{0} \\ & \stackrel{N}{N} \\ & \sum \end{aligned}$ |  | $\begin{aligned} & \text { n } \\ & 0 \\ & 0 \\ & 0 \\ & 0 \\ & 0 \\ & 0 \\ & \hline \end{aligned}$ |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Pixelated Pictures |  | - | - | - |  |  |  |
| Lights, Camera, Action! | - | - | - |  |  |  | - |
| Digital Detective |  |  | - | - |  | - |  |
| More Than Numbers |  |  | - | - | - |  |  |
| North American Tour |  |  | - | - | - | - |  |
| Digitizing Details | - |  |  | - |  |  | - |
| Picture Yourself |  |  | - |  |  | - |  |
| Color by Number | - |  |  | - |  |  | - |
| Photo Effects |  | - |  | - |  | - | - |
| Images for Science | - | - | - | - |  |  | - |

## A platform for independent investigation

ImageJ is an image analysis research tool used by scientists around the world. The program is simple but powerful, capable of sophisticated image processing techniques but easy to learn and understand. As such, its application to the classroom is a natural. Used on its own, ImageJ is completely open-ended. Student researchers must conceive, design, and create original images through such procedures as videomicroscopy or photography and flatbed scanning. They must then develop a process for gathering data from the images, such as image enhancement, filtering, thresholding, and measuring. In these endeavors, they mirror the processes of research scientists.

## Instructional materials: A launchpad to discovery

Computer and imaging technology are increasingly prevalent in the workplace. Good computer and image interpretation skills will be invaluable to information professionals of the 21st century. However, many students lack experience and confidence with both computers and advanced imaging technologies. As a result, independent use of ImageJ as a learning tool becomes a formidable and even frightening prospect to the uninitiated.

CIPE instructional materials are designed to familiarize both the instructor and the student with the tools and techniques of digital image processing as well as to model science process skills. Through these guided-discovery lessons, students learn content while building confidence and experience with image processing as an investigative tool. After completing a few lessons, students may be ready to capture images to create their own digital data sets for independent analysis and exploration.

## Creating your own images

The Discovering Image Processing CD-ROM provides all the images needed for the activities. However, we encourage teachers and students to apply the image processing techniques they learn from Discovering Image Processing to images that they have produced themselves. Students can analyze images created by microscopes, telescopes, still and video cameras, and medical and industrial equipment.

Digital still and video cameras and inexpensive flatbed scanners produce images that can easily be imported into the computer and analyzed using ImageJ. Analog images from videotapes, laserdiscs, and traditional film cameras can also be converted to digital format using appropriate hardware and software. You can even create images directly from grids of measurement data. With digital imaging, you are limited only by your imagination!

## Image resources

## Parents, neighbors, friends, and relatives of students

A rich source of support and great connections to other sources.

## Physicians and hospitals

X-rays; CT, MRI, and PET scans; EKG tracings; sonograms, etc.

## Veterinarians, animal clinics, and zoos

X-rays of domestic and exotic animals, fur, feathers, and other structures for analysis.

## Colleges and universities

Medical schools, nursing colleges, medical technician programs, biology departments, educational outreach programs, etc.

## City/county government

Departments of transportation, engineering, and law enforcement; also libraries, zoos, museums, etc.

## Fair use guidelines

The guidelines for fair use of copyrighted works for educational purposes have recently been updated and clarified to deal with the use of intellectual property in digital technologies. These fair use guidelines were adopted on September 27, 1996, as a nonlegislative report by the Subcommittee on Courts and Intellectual Property, Committee of the Judiciary, U.S. House of Representatives.
To get a copy of the guidelines, visit their Web site:
http://www.libraries.psu.edu/ avs/fairuse/default.html

## Important reminders

- Exercise caution in downloading works from the Internet. Some works may be copyrighted and may not be reused without permission.
- When using an image under fair use, credit the source and display the copyright notice and ownership if shown in the original work.
- Include a notice that certain materials in your presentation are included under the fair use exemption of U.S. Copyright Law and are restricted from further use.
- If you make alterations to copyrighted works to achieve specific instructional objectives, note that alterations have been made.


## State government

Departments of health, environmental quality, water resources, land, fish and game, and agriculture. Cooperative extension, geological survey, mines, parks, law enforcement, historical society, museums, etc.

## Federal government

Federal government sources of image data include the USDA Forest Service, Department of Energy (DOE), National Park Service, Drug Enforcement Agency (DEA), Department of Defense (military bases), National Oceanographic and Atmospheric Administration (NOAA), as well as the Food and Drug Administration (FDA), Federal Bureau of Investigation (FBI), Environmental Protection Agency (EPA), National Aeronautics and Space Administration (NASA), National Institutes of Health (NIH), etc. Many federal agencies publish image data on inexpensive CD-ROMs or via the Internet.

## The Internet

ImageJ reads most of the popular image file formats found on the Internet. Larger images may be compressed for faster download, requiring the user to decompress or expand image files before they can be opened. CIPE's Web site at http://www.evisual.org provides links to many images and resources.

## Student assessment

In Discovering Image Processing, students organize information, apply previously learned information to new situations, make conjectures based on data they have gathered, and explain and justify their analyses. Teachers may use a variety of tools and methods for assessing how well students have accomplished these tasks as they complete the lessons in Discovering Image Processing. These tools are described below:

## Image printouts and Data Sheets

The most direct method is to print out final images and use the Data Sheet included with each lesson. The Data Sheets provide students with a structured format to respond to the questions in the lessons. Answer keys to these questions are included in the Teaching Notes section of the lessons.

## Direct observation

The lessons in Discovering Image Processing present open-ended problems, giving students the chance to work cooperatively, think critically, and form conclusions. Spot-checking students as they work at the computer, asking questions that probe for comprehension, and allowing the students to ask for instructor feedback are all effective ways to informally assess student understanding.

## A few image resources on the Web

- American Memory (Library of Congress)
http://memory.loc.gov
- Astronaut Photography of Earth http://eol.jsc.nasa.gov
- Earth Observatory http://earthobservatory. nasa.gov
- NASA ESA Landsat Photography http://landsat.gsfc.nasa.gov
- NASA Planetary Photojournal http://photojournal.jpl.nasa. gov
- NOAA Image Library
http://www.photolib.noaa. gov
- National Space Science Data Center (NSSDC) Image Library
http://nssdc.gsfc.nasa.gov/ photo_gallery/
- Terraserver
http://terraserver.microsoft. com
- USGS EROS Data Center http://edc.usgs.gov
- USGS Seamless Map Server http://seamless.usgs.gov
- Visible Earth
http://visibleearth.nasa.gov
- Visible Human Project
http://www.nIm.nih.gov/
research/visible


## Image Processing Skills Checklist

A sample checklist for assessing basic image processing skills is provided on the next page. The list can be used to assess student skill mastery or for student self-evaluation. Modify it to meet your specific needs.

The list includes real-world, marketable computer skills that your students can learn through the Discovering Image Processing lessons. Students can also apply these skills to conduct authentic scientific research using ImageJ. For more about each skill, see the ImageJ Skill Sheets on pages 25-40.

## Student collaboration

Many of the lessons in Discovering Image Processing are particularly suited to collaborative learning environments. When all groups have finished a lesson, they can come back together as a class and share their results.

## Student journals

Many teachers assess students' written work by reading daily journal entries. For teachers who already include this tool in their technology curriculum, students can record journal entries for each lesson. They might be asked to reflect on things that they have learned on their own or from a classmate while completing the lesson or to tell something about their favorite image from a lesson.

## Student portfolios

For teachers who use student portfolios in their assessment repertoire, images can be printed and saved as hard copies along with other samples of student work. Students can include printouts showing images before and after they completed the assigned task. If the portfolio is in an electronic format, students' work may be saved to disk as well.

## Student presentations

A very effective way to assess student understanding of these lessons is to have them present what they have learned to the class or another group. These presentations demonstrate both the students' understanding of the underlying technology concepts taught in the lesson, as well as their ability to apply image processing to these concepts. Students could be encouraged to use the computer as part of their demonstration.

## Web sites

Many educators have students construct imaging-based Web sites using simple WYSIWYG (what-you-see-is-what-you-get) HTML editors.

## HTML

HTML stands for HyperText Markup Language. It is a set of instructions, called tags, that describe the layout and format of text, images, and other elements on Web pages.
$\qquad$ Class $\qquad$

## Image Processing Skills Checklist

## Window management

Activating windows
Organizing windows $\qquad$
Changing window size and location

## Image modes

Changing image modes $\qquad$
Selecting appropriate image mode

## Lookup tables

Applying new lookup tables to images
Adjusting image brightness and contrast
$\qquad$

Adjusting lookup table windowing for 16- and 32-bit images

## Selecting

Making selections $\qquad$
Modifying selections
Moving selections

## Editing

Cutting and copying selections
Pasting selections/using the Paste Control
Cropping images
Rotating and flipping images $\qquad$
Resizing images

## Drawing \& Text

Drawing lines and shapes $\qquad$
Painting with selection tools
Changing foreground/background color
Changing line width $\qquad$
Drawing text

## Thresholding

Adjusting the threshold range
Measuring with thresholding
Using the auto-selection (wand) tool

## Using the skills checklist

These skills are not all taught in every lesson. The matrix on page 4 identifies the major image processing skills that are utilized in each lesson.
Use the following scale to assess skill level. You may want to establish criteria for each level depending upon your students' abilities and experiences.

1 Novice (just beginning to learn, needs much help)

2 Beginner (needs a little help)
3 Able (can accomplish by referring to instructions)

4 Accomplished (can help others)
5 Expert (highly skilled)
The list can be also used for student self-assessment.

## Image enhancement

Enhancing image contrast
Applying image filters

## Calibration

Spatial calibration $\qquad$
Density calibration $\qquad$
Removing image calibration

## Measurement

Setting measurement options
Measuring distance and area
Measuring angles

## Analysis

Making and interpreting histograms
Making and interpreting profile plots
Making and interpreting surface plots

## Stacks

Creating stacks
Flipping through slices in stacks
Animating stacks
Editing stacks
Making montages
Split color images/merge RGB channels
Create Z-axis profiles of stacks

## Installation

## Windows installation

## System requirements

- 200 MHz Pentium II or faster computer with color monitor
- Windows 98, ME, 2000, or XP
- Minimum 64 MB available RAM (256 MB or more preferred)
- The ImageJ application requires 21 MB available hard drive space. Approximately 200 MB is required for installing the lesson data.


## Install ImageJ

- Insert the Discovering IP 2 CD-ROM into your computer's CD drive.
- Click the Start button on your computer's desktop, choose My Computer, then double-click the CD drive icon to view the CD contents.
- Open the Software folder on the CD (in Windows, a folder is also called a directory), then open the Windows folder.
- Double-click the ImageJ.msi application icon to start the ImageJ installer.
- Follow the on-screen instructions in the ImageJ Setup Wizard to install ImageJ.
- On the Ready to Install screen, review your installation settings, then click Install to complete the installation.

- An ImageJ application shortcut should now be available on your desktop. Double-click $\frac{\substack{\text { s. } \\ m \text { mos }}}{}$ to launch the ImageJ application.


## Need more help?

Additional installation instructions and troubleshooting notes are available via the Internet at
http://rsb.info.nih.gov/ij/ download.html

When you insert the CD-ROM into your computer's CD drive, you'll see a dialog box that reads: "What do you want Windows to do?"
Just click Cancel.

When you open the Windows folder inside the Software folder, you'll see the ImageJ Installer.msi icon. (Many Windows computers are set to hide the three-letter filename extensions, so you may see "ImageJ" instead of"ImageJ. msi," as shown below.).


## Install lesson data

Lesson images can be accessed directly from the Discovering IP 2 CDROM. However, for best performance, it is recommended that you copy the lesson data to your computer's hard drive.

- Open the Lesson Data folder on the Discovering IP 2 CD-ROM.
- Copy the desired lesson folders to your computer's hard drive. You may copy them into the ImageJ folder or to any convenient location. (Refer to your computer's documentation for instructions on copying files.)


## Test your installation

- If you have not already launched ImageJ, double-click $\frac{\underset{\sim}{~ d ~}}{\frac{d}{\text { masea }}}$ (on your desktop) to open it, or choose Start > All Programs > ImageJ.
- Choose File > Open..., navigate to a lesson data folder on the CD or your hard drive, and open an image file.


## Displaying file extensions

In this guide, computer filenames end in three-character file extensions such as .txt, .tif, .jpg, and .gif. Unfortunately, many Windows computers are set to hide these extensions. To force file extensions to display:

1. Choose Start > My Computer.
2. Choose Tools > Folder Options... (Windows XP, NT, 2000, or ME) or View > Folder Options... (Windows 98).

3. On the View tab, uncheck the box next to Hide extensions for known file types.
4. Click Apply, then OK to close the Folder Options window.
5. Close the My Computer window.

## Uninstalling ImageJ

The MSI installer used to install ImageJ on your PC creates an entry in the Windows registry, a database of installed software. To remove ImageJ:

- Click Start in the lower left of your screen, and then click Control Panel.
- In the Control Panel, double-click Add or Remove Programs. Choose ImageJ from the list and click Remove.
Both ImageJ and the quick launch icon on the desktop will then be removed from your computer.



## Re-installing ImageJ

You can install ImageJ again from the CD if you wish or from the ImageJ Web site (http://rsb.info. nih.gov/ij). However, if you install a newer version from the Web site, you will have to separately install the several plugins used in the lessons.

## Macintosh installation

## System requirements

- Power Macintosh Computer with color monitor
- MacOS 8.x or newer or Mac OS X.x
- Minimum 64 MB RAM ( 256 MB or more preferred)
- The ImageJ application requires 3 MB available hard drive space. Approximately 200 MB is required for installing the lesson data, and additional space is needed for the Images to Explore folder.


## Install ImageJ

- Insert the Discovering IP 2 CD-ROM into your computer's CD drive.
- Double-click the Discovering IP 2 CD icon on your desktop to view the CD contents.
- For OS X, double-click the Software folder, then the Mac OS X folder. Drag and drop the entire ImageJ folder to the Applications folder on your hard drive.
- For OS 8/9, double-click the Software folder, then the Macintosh folder. Drag and drop the entire ImageJ $\mathbf{1 . 3 4}$ folder to the Applications (Mac OS 9) folder or a convenient location on your hard drive.
- To run ImageJ, double-click on the ImageJ icon.


## Create an ImageJ alias

- Open the ImageJ folder you installed, single-click to select ImageJ, and choose File > Make Alias.
- Drag the alias you created to the desktop or to the OS X dock. You may move or rename the alias at any time, as long as you do not move the original application.


## Install the lesson data

Lesson images can be accessed directly from the Discovering IP 2 CDROM. However, for best performance, it is recommended that you copy the lesson data from the CD-ROM to your computer's hard drive.

- Open the Discovering IP 2 CD-ROM.
- Copy the entire Lesson Data folder, or open the Lesson Data folder and copy selected lesson folders to your computer's hard drive. You may copy the lesson data folders into the ImageJ folder you installed earlier.


## Test your installation

- Double-click the ImageJ alias icon on your desktop.
- Choose File > Open... from the ImageJ menu bar, navigate to a lesson data folder on the CD or your hard drive, and open an image file.


## Uninstalling ImageJ

- For OS X, drag and drop the entire ImageJ folder from the Applications folder on your hard drive to the Trash. In the Finder, choose Finder > Empty trash ..., and click OK.
- For OS 8/9, drag and drop the entire ImageJ $\mathbf{1 . 3 4}$ folder from the Applications (Mac OS 9) folder (or other location where you stored it on your hard drive) to the Trash. From the Special menu, choose Empty trash.


## Locking images to prevent changes

The files on the Discovering IP 2 CD-ROM cannot be modified or overwritten. However, after copying image files to your hard drive, they can be changed. To lock these files to prevent them from accidentally being altered, follow the instructions below. If a file is accidentally changed or deleted, replace it with a fresh copy from the CD.

## Macintosh

- In the Finder, select the icon of the image file you want to lock
- Choose File > Get Info to open the file's Info window.
- Check the Locked checkbox, and close the Info window.


## Windows

- In My Computer, right-click the icon of the image file you want to lock.
- Choose Properties from the menu that appears, and select the General tab.
- Check the Read-only attribute checkbox, and click OK.


## Re-installing ImageJ

You can install ImageJ again from the CD if you wish or from the ImageJ Web site (http://rsb.info. nih.gov/ij). However, if you install a newer version from the Web site, you will have to separately install the several plugins used in the lessons.

## Locking files



## Troubleshooting

## Updating ImageJ (Mac \& Win)

ImageJ is a work in progress, and is continuously being updated and improved by its author, Wayne Rasband of the National Institutes of Health.
Problem: "How do I update ImageJ?"
Solution: Download the latest ij.jar file from http://rsb.info.nih.gov/ij/ upgrade. To install the file:

- Windows and MacOS 8-9: Open the ImageJ folder, and replace the old ij.jar file with the new version.
- Mac OS X: Open the ImageJ folder, control-click the ImageJ icon, and choose Show Package Contents from the contextual menu. Replace the old ij.jar file in the Contents $\backslash$ Resources $\backslash$ Java folder with the new version.


## Increasing ImageJ memory (Mac \& Win)

By default, ImageJ is set to reserve 128 MB of RAM for storing and processing images. If you need to work with more or larger images, you may wish to make more memory available to ImageJ.
Problem: "How do I increase the amount of memory available to ImageJ?"
Solution: To increase the memory allocation for ImageJ:

- Launch the ImageJ application.
- Choose Edit > Options > Memory..., enter a new Maximum Memory size, and click OK.
- Quit and re-launch ImageJ.


## Missing file extensions (Win)

Problem: "I don't see the .txt file extension at the end of the file names."
Solution: In Windows, open the Files and Folders control panel, and uncheck the Hide known file extensions option.

## ImageJ won't let me do anything (Mac \& Win)

Problem: "ImageJ seems to be frozen and won't let me do anything. I can activate and move windows but can't modify the images in any way."
Solution: There may be a hidden window or dialog box waiting for input that must be closed before you can proceed. Move windows around, close or hide other applications, and look for this window or dialog box.

## The ImageJ menus disappear (Mac)

Problem: "When I switch between windows, the ImageJ menu headings sometimes disappear."

## Update with care!

Warning: Program features and the user interface of future versions of ImageJ may not match the instructions given in the Discovering Image Processing lessons. The instructions are based on ImageJ Version 1.34 s provided on the Discovering IP 2 CD.

## Don't overdo it!

Entering more than 75 percent of your installed RAM is not advised. The maximum amount of memory that can be allocated to ImageJ regardless of the amount of installed RAM is about 1.7 GB .

Solution: Press the Enter key on your keyboard to move the ImageJ window to the front. This should update the menu.

## ImageJ is labeling and drawing in the wrong colors (Mac \& Win)

Problem: "When I draw labels, shapes, text, etc. on an image, they are not drawn in the foreground color, but in another color or a shade of gray."

Solution: Choose Image > Type > RGB Color to convert the image (even grayscale images) to an RGB color image.

## I can't copy and paste between ImageJ and other applications (Mac \& Win)

Problem: "I selected and copied (Edit > Copy) an image in ImageJ, but when I try to paste it into my word processor (or other application), nothing is there to paste."
Solution: Use the System Clipboard plugin to copy and paste between ImageJ and other applications. Choose Plugins > System Clipboard to display the System Clipboard window. Click the Copy button to copy the current ImageJ selection to the system clipboard, and click the Paste button to paste the contents of the system clipboard into the current ImageJ window. (Note: The System Clipboard plugin does not work with Mac OS 8 or OS 9.)

## I don't see the <br> $\qquad$ plugin. Where is it? (Mac \& Win)

Problem: "I am doing a lesson that calls for a specific plugin, but I don't see it listed at the bottom of the Plugins menu."
Solution: Copy the TurboReg and Stacks folders from the Plugins folder on the Discovering IP 2 CD to the Plugins folder of your ImageJ location.

## My Y-coordinates do not match those in the answer key (Mac \& Win)

Problem: "My answers are different from those in the answer key. The Xcoordinates agree, but all of my Y-coordinates are different."
Solution: Choose Analyze > Set Measurements... and turn on (check) the Invert Y Coordinates option.

## ImageJ dialog boxes, menus, or tools don't match those shown in the book (Mac \& Win)

Problem: "The dialog boxes, menus, or tools in ImageJ look or work differently than the ones shown in this book."
Solution: You are using a version of ImageJ that is older or newer than the one provided with this book ( 1.34 n ). Do so at your own risk.

## ImageJ quits for no apparent reason (Mac)

Problem: "At random times, ImageJ quits and gives the message 'The application JavaApplicationStub has unexpectedly quit.'"

## More info...

At the time of this writing, this issue has been mostly fixed, but still occurs occasionally. It is simply a menu update issue that can be forced by changing windows.

## More info...

The colors available for drawing on an image are limited by the type of image. Grayscale images can only be drawn on with grays.

## More info...

Most applications hold copied data in a section of memory called the system clipboard, which is managed by the operating system. ImageJ uses its own clipboard memory for these operations, and this clipboard is not available to other applications.

## More info...

Plugins for ImageJ can be downloaded from the ImageJ Web site. Choose Help > ImageJ Web Site..., and click the Plugins link on the Web page. Read and follow all installation instructions carefully.

## More info...

ImageJ displays images starting at the upper left corner of the window, so the default origin ( 0,0 ) is located there. The Invert Y Coordinates option is a convenience for those preferring the origin in the lower left corner, to match the Cartesian coordinate system.

## More info...

If you want to use a newer (or an older) version of ImageJ, you can install more than one version on your computer. Just give each a name that helps you tell the versions apart.

Solution: This is a known issue when running ImageJ under Mac OS X, but there is no definitive solution. While not a solution, it may be helpful to increase the memory allocated to ImageJ. Choose Edit > Options... Memory... and enter a higher value. It's probably wise to keep your memory allocation to about half of the installed RAM. (For example, on a

Would you like to submit a bug report to Apple? Submit Report... Cancel computer with 1 GB of installed RAM, set the memory allocation to 512 MB.) Fortunately, the crash does not affect other applications, so re-launch ImageJ, re-open the images you were using, and pick up where you left off. If you are performing complex processes on images, be sure to save your work often.

## Problems opening files by drag-and-drop (Mac)

Problem: "When I drag a group of files and drop them onto the ImageJ application or alias icon, some of the files open, some open multiple times (identical copies), and some don't open at all."
Solution: This is another known issue with ImageJ. Instead of dropping the files onto the ImageJ icon, you can drop them on the ImageJ window and they will open properly. However, it is usually best to use the File > Open... menu command within ImageJ to open files. Type either the letter O or Control-O as a shortcut to quickly open the Open dialog box.

# ImageJ Quick Reference 

## The ImageJ user interface

The ImageJ user interface is nearly identical for Windows and Macintosh operating systems, except for the location of the menu bar.

## Windows

Under Windows, the menu bar is at the top of the ImageJ window.


## Macintosh

On the Macintosh, the menu bar is at the top of the screen, separate from the ImageJ window.


## The ImageJ toolbar



## ImageJ updates

ImageJ is constantly being updated and improved. Updating your copy of ImageJ from the version included with this package may result in changes to the user interface. The capabilities of the program may also change significantly, and lesson instructions and results may need to be modified.

You can have access to the latest tools and functions while retaining compatibility with these materials by installing multiple copies (versions) of ImageJ on your computer. Each copy can be launched and run independently.

## Need to know more?

For additional information about ImageJ's tools and menus, refer to the online user's guide at:
http://rsb.info.nih.gov/ij/docs/
This guide can be opened from within ImageJ by choosing Help > Online Docs...

## Drawing tools

The blank buttons will be replaced by additional tools in future versions of ImageJ.

## Tool descriptions

## Area selection tools

The area selection tools allow you to select an area, or region of interest (ROI), of the image using a rectangle, oval, polygon, or freehand shape, or some combination of these shapes. To move a selection, click and drag within the selection outline, or use the arrow keys to nudge the selection one pixel at a time in any direction. The selected areas can be edited, enhanced, analyzed, outlined, filled, or copied using menu commands and keyboard shortcuts. As you make a selection, the location and dimensions of the selection are displayed in the ImageJ status bar.
To measure the selected area, press $\mathbf{M}$ (measure); to outline the selected area using the foreground color, press $\mathbf{D}$ (draw); to fill the selected region with the foreground color, type $\mathbf{F}$ (fill); and to fill the area with the background color, press the Backspace key (Delete key on Macs).
To subtract from an existing selection using any of these tools, hold down the Alt (Win) or Option (Mac) key as you make the selection. To add to an existing selection, hold down the Shift key as you make additional selections. Note: When adding selections with the rectangle and oval tools, you can avoid constraining your selection to a square or circle by releasing the Shift key before you release the mouse button.

## Rectangle

When creating rectangular selections, click on one corner of the area and drag to the opposite corner. Hold the Shift key down to constrain the selection to a square. You can modify the selection by dragging any of the eight square handles around the edges of the selection. In addition, you can use the Alt (Win) or Option (Mac) key combined with the Arrow keys to change the height and width of the selection by one pixel at a time.

## Oval

Creates elliptical selections that can be moved and resized using the same methods as rectangular selections.

## Polygon

Creates irregularly shaped selections composed of a series of line segments. Click at the end of each segment, then click in the small box at the starting point to complete the selection. Alternatively, you can double-click at the end of the next-to-last segment, and ImageJ will automatically complete the last segment. To modify the polygon, drag the square handles at each vertex. To delete a vertex, hold down the Alt (Win) or Option (Mac) key and click on the vertex. To add a vertex, hold down Shift and click on a segment.

## Freehand area

The freehand area tool lets you drag the cursor to create irregularly shaped selections. The area will automatically close when you release the mouse button. To edit a freehand area, choose Edit > Selection > Fit Spline. The resulting selection can then be edited using the same methods as the polygon selection tool.

## Changing the selection outline color

By default, ImageJ uses a yellow line to outline selection. This may not always be appropriate, such as when selecting against a light (or even yellow) background. To change the selection outline color, choose Edit > Options > Colors... and choose a different color.

## Setting line width

To set the drawing line width for all of the selection tools, double-click any of the line selection tools and set the desired width in pixels.


## Editing selections with handles



To modify a selection with editing handles (the small squares along the selection line), drag a handle to the desired location.
With polygon selections, you can Alt-(Win) or Option-(Mac) click on a handle to delete it, and Shift-click on a handle to add a new handle.

## Line selection tools

The line selection tools create straight, segmented, or freehand line selections. Drag or use the arrow keys to move line selections, and drag handles to edit the selection. To measure the selected line, press $\mathbf{M}$ (measure); to draw the selected line using the foreground color, press D (draw) or $\mathbf{F}$ (fill); and to draw the line with the background color, press the Backspace key.

Caution: Regardless of the shape selected, measurements made with the line selection tools include only the pixels under or behind the line. Using the segmented or freehand line selection tools to select and measure areas will produce incorrect results.

## Straight line

Click, drag, and release the mouse button to define the endpoints of a line selection. Hold down the Shift key while dragging to constrain the line to vertical or horizontal. While dragging, the length and angle of the line is displayed in the status bar.

## Segmented line

Use the segmented line tool to select lines made of straight line segments. Click once at the end of each segment, and double-click at the final endpoint. If double-clicking is difficult, you can also click in the box at the beginning of the line. (Unlike the polygon area tool, the segmented line tool cannot "close in" an area.)

## Freehand line

Use the freehand line tool to create irregular line selections by dragging on the image. To convert the freehand line to an editable curved line, choose Edit > Selection > Fit Spline.

## Angle tool

The angle tool selects angles on images using three clicks-the first defines one leg, the second the vertex, and the third the other leg. After the first click, the status bar displays the angle of the cursor, measured $\pm 180^{\circ}$ relative to the 3 o'clock position. After the second click, the status bar displays the interior angle ( $\leq 180^{\circ}$ ) of the angle defined by the first leg and the current cursor position.
To edit an angle, drag any of the three handles. As you drag a handle, the changing angle measurement is displayed in the status bar.

## Point selection tool

Click on the image with the point selection tool to select, mark, and record locations on the image. To select multiple points, hold down the Shift key while clicking on each location. Drag a point to move it, and Alt-click (Win) or Option-click (Mac) a point to delete it.
In Auto Measure mode, each click on the image records the coordinates and brightness value of the pixel in the Results window. Double-click the point selection tool button to turn on the auto-measure mode option or set the mark width. If a mark width greater than zero pixels is selected, a point will automatically be drawn each time you click on the image. To draw points without measuring, turn off Auto Measure, select a point, and press the $\mathbf{D}$ (draw) key.

## Line tool areas

Although it appears possible to select an area using the freehand line tool, the area reported when you measure the selection is not the area enclosed by the selection. Rather, it is the area of the pixels crossed by the selection line, shown here in black. Always use the area selection tools to select and measure areas.


## Using the Angle tool

Selecting with the angle tool requires three clicks: (1) the first leg, (2) the vertex, and (3) the second leg.


Point tool options


Note: with RGB color images, pixel values are reported on the status bar as separate red, green, and blue channel values. The value recorded in the Results window is the average of the $R, G$, and $B$ values (not very useful!).

## Wand (tracing) tool

The wand tool automatically finds the edge of an object and traces its shape. It works best with high contrast images. Place the wand to the left of an edge, and click to have ImageJ trace along the edge of the object until it returns to the starting point.

An object is defined as a cluster of pixels having the same color or value. ImageJ does not currently have the ability to define an object based on a range of pixel values.

## A. Text tool

The text tool is used to add text labels to images. Adding text is a simple four-part process:

- Click at the text's desired location.
- Type the text. This creates a selection shaped like the desired text, which expands as you type. Type Return (Enter on some keyboards) to create multi-line text. Use the backspace key to edit the text.
- Drag the text selection, or use the arrow keys to move the selection to the proper location.
- Draw the text using Ctrl+D or by choosing Edit > Draw.

Double-click the text tool button to select the text font, size, and style. Text draws in the foreground color defined in the color picker tool button.

Note: Drawing text on an image destroys data by replacing the original pixel values with the foreground color. Text cannot be edited after it is drawn, but it can be deleted by immediately choosing Edit > Undo. After this, the only way to delete text is to choose File > Revert, which will undo all changes made to the image since it was last saved.

## Magnifying glass (zoom) tool

To zoom in or magnify the image up to $32 x$, click on the image or press the + key repeatedly. To zoom out or reduce an image down to $1 / 32 \times$, you may right-click, Option-click (Mac), Alt-click (Win), or press the - key repeatedly. Double-clicking $Q_{Q}$ returns the image to full size.

## Scrolling tool

This tool allows you to move the image within its window if it is larger than the window. While using any other tool except the text tool, you can quickly toggle on the scrolling tool by holding down the space bar.

## Color picker tool

This tool sets the foreground drawing color or text color by "picking up" colors from images with the cursor. Colors may also be picked up from the $\mathbf{C P}$ (color picker) window by double-clicking the color picker button. Alt-click (or Option-click) in the $\mathbf{C P}$ window to change the background color. The eyedropper in the color picker tool button shows the current foreground color while the frame around it shows the background color.

## Selecting areas using the wand tool



Image of leaf selected using the wand (tracing) tool. To isolate the background, the contrast had to be increased using the Brightness and Contrast control panel, and the contrast change permanently applied to the image.

## Text tip

To add color labels to grayscale or 8bit color images, convert the image to RGB color first, using Image > Type > RGB.

## Foreground \& background colors

The current foreground and background colors are shown on the color picker tool button. Doubleclick this button to set these colors.


Caution: Even though colors appear in the color picker tool, accurate colors are only available when working with RGB color images. On 8 -bit color images, drawing colors will not match the foreground and background colors; on grayscale images, actual drawing colors will be shades of gray.

## Drawing tools

In addition to drawing and filling shapes using the selection tools, ImageJ provides familiar drawing tools like those found in most graphic applications. The actual tools are provided through a macro file that ImageJ loads automatically at startup. If these tool buttons do not appear on the toolbar, you may need to install or replace this file.
Remember that the drawing tools permanently modify pixel values, and that scientific analysis may not be valid if the image has been modified in any way.

## Pencil tool

The pencil tool draws points and lines in the foreground color. Doubleclick the tool button to set the diameter of the pencil tool.

回 Paint brush tool
The paint brush tool draws with a circular brush, using the foreground color. Double-click the tool button to set the diameter of the paintbrush tool.

## Eraser tool

The eraser tool draws with a circular shape, using the background color. Double-click the tool button to set the diameter of the eraser tool.
( Q $_{\text {a }}$ Flood fill tool
The flood fill tool fills the pixel you click on and any adjacent pixels having the same value with the foreground color. Double-click the tool button to set the foreground color.

## ImageJ menus

| File Edit | Image |
| :--- | ---: |
| New．．． | Proce |
| Open．．． | HO |
| Open Samples |  |
| Open Recent |  |
| Import | HW |
| Close | HS |
| Save | HR |
| Save As |  |
| Revert | HP |
| Page Setup．．． |  |
| Print．．． |  |
| Quit |  |


| Image Process | Analyze |
| :---: | :---: |
| Type | － |
| Adjust | － |
| Show Info．．． | \％1 |
| Properties．．． |  |
| Color | － |
| Stacks | － |
| Crop |  |
| Duplicate．．． | 仑भD |
| Rename．．． |  |
| Scale．．． | HE |
| Rotate | － |
| Zoom | － |
| Lookup Tables | － |


| Analyze Plugins | Windo |
| :--- | :--- |
| Measure | $\mathscr{M}$ |
| Analyze Particles．．． |  |
| Summarize |  |
| Label |  |
| Clear Results |  |
| Set Measurements．．． |  |
| Set Scale．．． |  |
| Calibrate．．． |  |
| Histogram | $\mathscr{}$ Plot Profile |
| Surface Plot．．． |  |
| Gels |  |
| Tools |  |

## File menu

The File menu allows you to create new image windows，open existing image files，as well as print and save image files．

## Edit menu

The Edit menu provides standard editing functions such as undo，cut， copy，and paste．In addition，it offers the powerful Paste Control feature， drawing commands，and selection and program options．

| Edit Image | Proces |
| :---: | :---: |
| Undo | HZ |
| Cut | HX |
| Copy | HC |
| Paste | \＆V |
| Paste Control．．． |  |
| Clear |  |
|  |  |
| Fill | HF |
| Draw | HD |
| Invert | 仓み！ |
| Selection | － |
| Options | － |

## Need to know more？

For a more detailed description of ImageJ＇s tools and menus，refer to the online user＇s guide at：

## http：／／rsb．info．nih．gov／ij／docs

This guide can be opened from within ImageJ by choosing Help＞ Online Docs．．．

| Window Help |
| :--- |
| ImageJ［enter］ |
| Put Behind［tab］ |
| Cascade |
| Tile |
| $\checkmark$ Image＿1 4216K |
| Image＿2 4216K |

## Window menu

The Window menu allows the user to control the placement of image and program windows in ImageJ．

| Help menu | Help |
| :---: | :---: |
|  | About Plugins |
| The Help menu links to the ImageJ Web site and online documentation． | ImageJ Web Site．．． Online Docs．．． About ImageJ．．． |

## Image menu

The Image menu provides functions for manipulating the display of images．

## Process menu

The Process menu includes powerful data filtering and manipulation tools．

| Process Analyze | Plugins |
| :---: | :---: |
| Smooth | vfs |
| Sharpen |  |
| Find Edges | へみF |
| Enhance Contrast |  |
| Noise | $\checkmark$ |
| Shadows | $\checkmark$ |
| Binary | － |
| Math | － |
| FFT | $\stackrel{\rightharpoonup}{*}$ |
| Filters | － |
| Image Calculator．．． |  |
| Subtract Background．．． |  |
| Repeat Command | 饣HR |

## Analyze menu

The Analyze menu offers a variety of tools and functions for measuring and representing image data．


## Lesson features

Right-hand column gives menu commands, helpful hints, definitions, and references to "ImageJ Skill Sheets" for further instruction.

## Lights, Camera, Action! Creating and using image stacks

Have you ever wanted to make a movie? Well, here's your chancel With your imagination and a little help from image processing technology you can easily create digital animations using stacks of images. You can even save thern as QuickTime ${ }^{\text {II }}$ and AVI movies for use with other applications.

Image file name

Computer icons indicate actions
to carry out on the computer.

Numbered items are questions to answer on the
Data Sheet.

## At the movies

First, let's observe a ready-made animation.

- Open Galloping Horse.tif
- Drag the scroll bar at the bottom of the stack window left and right to see the frames, or slices of the stack.

This series of images was created in 1878 by Eadward Muybridge, an early pioneer in the use of photography to study animal locomotion.


Microscope icons indicate ImageJ menu commands, tools, or
detailed
instructions.

- Press the (backslash) key while holding down Control (Windows) or Command (Macintosh) to start the animation. Click on the image to stop the animation, or use the $\backslash$ key alone.
- Experiment with changing the speed of the animation. To change the speed, choose Image > Stacks > Animation Options....

1. What are the fastest and slowest animation speeds in frames per second?

## What is a stack?

A set of images displayed within a single window is called a stack. A stack is similar to a pad of paper or a deck of cards, except that each "card" is a digital image. These images can show many things-a sequence of events, parts of a larger image, or slices through an object at different depths. In fact, ImageJ refers to the individual, "still" images within the stack as slices.
This lesson will focus on the use of stacks for animating a series of images. Perhaps you have created a simple flip-book animation using a stack of paper. When the images are shown quickly, the eye "sees" motion. This is the basic principle that is used for making movies. Films and videos are really just series of still pictures shown in a rapid sequence. By arranging a series of images in a stack and flipping through them, you can create your own animation.

- Use the > and < keys or the scroll bar to move forward and backward through the stack as you view the individual slices.
- Below the title bar of the Galloping Horse.tif window is a row of
 numbers. The first number refers to the slice of the stack that you are currently viewing, and the second number refers to the total number of slices in the stack.

2. How many slices are there in Galloping Horse.tif?


## ImageJ Skill Sheets

## Using the ImageJ Skill Sheets

There is no right or best way to use these resources. Here are two suggestions for using them in your classroom.

- Copy and laminate the Skill Sheets, and post them near the computers.
- Bind copies of the Skill Sheets, along with other useful reference materials, into handbooks. Give a copy to each student or workgroup, or place one at each computer.


## If you need to know more

For more extensive information on image processing techniques using ImageJ, consult the online ImageJ user manual at:
http://rsb.info.nih.gov/ij/docs/index.html
The online documentation can also be accessed directly from ImageJ by choosing Help > Online Docs. Additional information is provided in the Troubleshooting section of this guidebook.

## Lookup Tables

## What is a lookup table?

Digital images are produced by matching numerical values to colored pixels using a lookup table (LUT). The diagram below shows this relationship.

Numerical values

| 0 | 0 | 0 | 0 | 3 | 8 | 9 | 5 | 5 | 5 | 8 | 3 | 0 | 0 | 0 | 0 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 0 | 0 | 0 | 7 | 9 | 12 | 6 | 4 | 4 | 5 | 6 | 11 | 13 | 3 | 0 | 0 |
| 0 | 0 | 8 | 6 | 4 | 10 | 5 | 4 | 10 | 10 | 11 | 12 | 12 | 11 | 7 | 0 |
| 0 | 8 | 12 | 6 | 5 | 4 | 4 | 6 | 12 | 12 | 11 | 15 | 12 | 11 | 6 | 3 |
| 2 | 11 | 12 | 10 | 11 | 4 | 4 | 10 | 12 | 11 | 11 | 11 | 12 | 10 | 5 | 4 |
| 7 | 6 | 12 | 12 | 10 | 1 | 4 | 13 | 10 | 14 | 14 | 14 | 14 | 6 | 4 | 4 |
| 4 | 6 | 15 | 12 | 4 | 1 | 8 | 13 | 8 | 14 | 14 | 14 | 14 | 8 | 4 | 4 |
| 5 | 5 | 11 | 5 | 1 | 3 | 9 | 14 | 14 | 14 | 13 | 10 | 13 | 1 | 1 | 4 |
| 5 | 4 | 6 | 5 | 1 | 3 | 13 | 14 | 14 | 14 | 7 | 7 | 8 | 1 | 1 | 4 |
| 4 | 4 | 4 | 6 | 10 | 3 | 7 | 13 | 14 | 13 | 2 | 3 | 9 | 7 | 4 | 4 |
| 7 | 5 | 4 | 9 | 12 | 9 | 2 | 7 | 14 | 9 | 2 | 3 | 7 | 4 | 9 | 4 |
| 2 | 5 | 4 | 5 | 14 | 9 | 3 | 7 | 14 | 9 | 3 | 2 | 3 | 13 | 10 | 7 |
| 0 | 7 | 5 | 4 | 10 | 9 | 3 | 3 | 10 | 9 | 3 | 3 | 8 | 10 | 13 | 2 |
| 0 | 0 | 7 | 4 | 10 | 4 | 1 | 3 | 4 | 3 | 3 | 1 | 4 | 8 | 3 | 0 |
| 0 | 0 | 0 | 7 | 6 | 7 | 3 | 3 | 3 | 3 | 1 | 7 | 7 | 2 | 0 | 0 |
| 0 | 0 | 0 | 0 | 3 | 7 | 7 | 7 | 7 | 7 | 7 | 3 | 0 | 0 | 0 | 0 |

Lookup table


## Manipulating the lookup table using the Brightness and Contrast (B\&C) window

Choose Image > Adjust > Brightness/Contrast to open the $\mathbf{B} \& \mathbf{C}$ window. The $\mathbf{B} \& \mathbf{C}$ window shows a histogram of the pixel values in the image and provides sliders to control how ImageJ maps pixel values to the lookup table. The following example shows how to enhance the contrast of an image using these controls.


In the original low-contrast image, the pixel values cluster near the center of the histogram. This corresponds to the middle shades of gray in the lookup table.


Adjust the Minimum and
Maximum slider controls to match the lookup table to the range of pixel values.


To make the contrast change permanent, click the Apply button in the B\&C window. Note: this changes the original pixel values! the transfer function equation to convert the old pixel values to new pixel values.

## Transfer function

The diagonal line on the histogram in the $\mathbf{B \& C}$ window represents the transfer function (equation) that mathematically relates pixel values to the lookup table.

Clicking the Apply button uses
pixels $=$ picture elements, the small colored squares or rectangles that make up a digital image.
histogram = graph showing the number of pixels in an image with each different pixel value. Also called a frequency distribution chart.



## Lookup tables and image types

Computers store and process information in the form of binary digits or bits. The number of bits that are used to represent each pixel in an image determines the number of possible values a pixel can have. ImageJ can work with 8 -bit, 16 -bit, and 32 -bit grayscale images, as well as 8 -bit and

24-bit (RGB) color images. 8-bit images can have up to 256 different values, 16 -bit images can have over 65 thousand values, and 32-bit images can have billions of possible values.

In ImageJ, all lookup tables contain a maximum of 256 colors. In 8-bit grayscale and color images, each value corresponds to one color of the lookup table. Since the human eye cannot distinguish billions, or even thousands of different colors, it would be pointless to use 16- or 32-bit lookup tables. Therefore, these images are displayed using an 8-bit lookup table, with each color representing a range of adjacent pixel values, rather than a single value.
The process of displaying 16- and 32-bit data using a 256-color lookup table is called windowing. In ImageJ, windowing is controlled using the Window and Level (W\&L) panel. To open the $\mathbf{W} \& L$ panel, choose Image > Adjust > Window/Level. The Window control determines the number of adjacent pixel values that the 256-color lookup table is applied to. The Level control sets the center pixel value of the range defined by the Window control. Pixel values above this range appear white, and values below the range appear black. The example below, using a 16-bit image of a galaxy, shows the use of the Window and Level controls.


The original 16-bit image contains more information than a 256 -color lookup table can display effectively. As a result, only the bright center of the galaxy and a nearby star can be seen.


By narrowing the window and adjusting the level, additional detail can be seen in the central region of the galaxy.


Narrowing the window further shows more detail in the faint, outer parts of the galaxy. The inner part of the galaxy is now outside the range of the lookup table, and appears "overexposed."

## Using a different lookup table

ImageJ includes several built-in lookup tables and can open saved ImageJ and NIH Image lookup tables. Lookup table files have a .lut file extension.

- Choose Image > Lookup Tables to use a built-in lookup table, and pick a lookup table from the list.
- Use File > Open... to open lookup table (.lut) files.


## Combining images with different lookup tables

To copy and paste between two images, or to combine multiple images into a montage or a stack, they must have the same lookup table. If they have different lookup tables, change the image type of all images involved in the operation to 8-bit grayscale (Image > Type > 8-bit) or RGB color (Image > Type > RGB Color).

## $\boldsymbol{b i t}=$ "binary digit"

## Bits and colors

4 bits $=2^{4}=16$ colors
8 bits $=2^{8}=256$ colors
16 bits $=2^{16}=65,536$ colors
32 bits $=2^{32}=4.3$ billion colors

## RGB Color and Lookup Tables

RGB Color images do not use lookup tables. Rather, they define the color of each pixel using three values, one each for the Red, Green, and Blue brightness of the pixel. Since each pixel can have any of 256 possible values for red, green, and blue, there are a total of $256 \times 256 \times 256$ or 16.7 million color combinations!

## 8-bit color

8-bit color images are like "paint-bynumber" pictures. Each image has a unique palette of up to 256 colors. In the same way that the colors from one picture may not work well for painting a different scene, the lookup tables from 8-bit color images are not interchangeable. Thus, cutting and pasting between 8 -bit color images usually does not work well. To combine 8 -bit color images, you must first convert each image to RGB color using Image > Type > RGB Color.


Combining the two 8 -bit color images above does not produce the desired result (below left). Converting the images to RGB color before combining them produces the appropriate result (lower right).


## Selecting

Selecting tells the computer which pixels you are interested in. This is done using the selection tools in the ImageJ window. Selected pixels are defined by a colored line or outline. The selected pixels can be copied, filled, erased, outlined, filtered, or measured. The ImageJ status bar displays relevant information about selections as you make them.


## Area selection tools

Use these tools to create area selections.
$\square$ Rectangle - Creates rectangular or square selections. Holding the Shift key down forces the selection to be a square. Use the arrow keys with the Alt (Win) or Option (Mac) key pressed to change the width or height one pixel at a time.
O Oval - Creates elliptical selections. Holding the Shift key down forces the selection to be circular. Use the arrow keys with the Alt (Win) or Option (Mac) key pressed to change the width or height by one pixel.
Polygon - Creates irregularly shaped selections defined by a series of line segments. Click at each vertex of the polygon, and either doubleclick or click in the box at the starting point to complete the polygon.
$\bigcirc$ Freehand - Creates irregularly shaped selections by clicking and dragging with the mouse. When you release the mouse button, the area automatically completes.
W Wand Tool - Creates a selection by tracing objects of uniform color or thresholded objects. To trace an object, either click inside near the right edge, or outside to the left of the object.

## Line selection tools

Use these tools to create line selections.
Straight Line - Creates straight line selections. Holding the Shift key down forces the line to be horizontal or vertical.
Z Segmented Line - Creates segmented line selections by repeatedly clicking with the mouse. Each click defines a vertex. Double-click when finished.
$\boxed{\text { Freehand Line - Creates freehand line selections by clicking and }}$ dragging with the mouse.
$\triangle$ Angle - Creates angle selections using three mouse clicks. After the second click defines the angle vertex, the status bar displays the angle until you click the third time.

## Reshaping selections

To resize or reshape selections, drag the small handles on the selection outline. To create editable handles on freehand selections, choose Edit > Selections > Fit Spline.


## Creating complex selections

Area selections can be added to and subtracted from to create complex geometric and freehand selections. To add to an existing selection, hold down the Shift key while you select using any of the area selection tools. (When using the rectangle or oval selection tools, release the Shift key after you begin making the new selection unless you want to constrain your selection to a square or circle.) An example of a complex selection is shown at right.

## Moving selections

Position the cursor inside the selection and drag it to a new location. You can nudge a selection one pixel at a time in any direction by pressing the arrow keys on your keyboard.

## Selecting the entire image

Choose Edit > Selection > Select All, or press the A key.

## Cancelling selections

Click anywhere on the image outside the selection boundary, choose Edit > Selection > Select None, or press Shift-A.

## Restoring cancelled selections

Choose Edit > Selection > Restore Selection or press Shift-E.

## Drawing selection boundaries

Choose Edit > Draw or press D. Set the drawing color using the Color Picker tool by choosing Image > Color > Color Picker, or press Shift-K. Set the line width by choosing Edit > Options > Line Width... or by doubleclicking one of the line selection tools.

## Filling selections

Choose Edit > Fill or press F. Set the drawing color (also called the foreground color) with the Color Picker tool by choosing Image > Color > Color Picker, by pressing Shift-K, or by double-clicking the Color Picker tool itself. (Note: if the image type is 8 -, 16 -, or 32 -bit, the drawing color will be the shade of gray closest to the chosen foreground color. See Image types and drawing colors at right.)

## Erasing selections

Press the Backspace key to erase the selection to the background color.

## Measuring selections

After making a selection, choose Analyze > Measure (or press M). To measure in spatially calibrated units, first set the image scale by choosing Analyze > Set Scale.... To spatially calibrate an image, create a line selection corresponding to a known distance (e.g. 10 mm ), then enter that distance in the Analyze > Set Scale dialog box. (See the Measuring Skill Sheet for more information on setting scale and measuring.)


A complex selection created using the rectangle, oval, and polygon area selection tools by adding to a simple rectangular selection.

## Saving selections for later use

There may be times when you need to use the same selection over and over on a series of images. To save a selection, choose File > Save As... > Selection... and name the selection (do not change the .roi file extension).
To select the same region on another image or at another time, choose File > Open... and locate and open the saved selection file.

## Image types and drawing colors

"Help! The foreground color is set to red, but outlines are drawn in gray!"
This "problem" is normal ImageJ behavior. Colors are only available when working with color images. If the image type is 8 -, 16 -, or 32 -bit, lines and fills will be made in shades of gray rather than the selected foreground color.
If you need to draw on a grayscale image in color, you must first convert the image to RGB color (Image > Type > RGB Color).

# Drawing \& Text 

## Drawing tools

ImageJ includes four drawing tools that work the same as similar tools found in most graphic applications.

The Pencil tool draws in the foreground color. The line width can be set by double-clicking the pencil tool button.
The Paintbrush tool draws in the foreground color. The brush diameter is set by double-clicking the paintbrush tool button.
(G) The Eraser tool "erases" to the background color. The eraser diameter is set by double-clicking the eraser tool button.

Th The Flood fill tool fills groups of adjacent pixels that have the same value with the foreground color.

In addition to the drawing tools, you can use any selection as a paintbrush. Create an area or line selection, and drag it slowly while holding down the F (fill) key to paint in the foreground color, or the Backspace key to paint with the background color.

## Drawing lines and shapes

Use one or more selection tools to make a selection on the image. (See the Selecting ImageJ Skill Sheet for more information on making selections.) To draw the selection boundary, choose Edit > Draw or press the $\mathbf{D}$ key. Choose Edit > Fill or press the $\mathbf{F}$ key to fill the shape with the foreground color. Choose Edit > Clear or press the Backspace key to fill the selection with the background color.
To set the width of the outline, choose Edit > Options > Line Width... or double-click any of the line selection tools.

## Adding text to images

To add text to an image:

- Choose Edit > Options > Fonts... or double-click the text tool A. to set the font, size, and style of text.
- Click on the image using the text tool A, and enter the desired text (backspace to delete and edit the text).
- Drag the text frame to reposition the text, if necessary.
- Press Ctrl-D (Win) or Cmd-D (Mac) to draw the text in the foreground color. (To delete text, choose Edit > Undo or press Ctrl-Z immediately.)


## Antialiased Text ON

## Antialiased Text OFF

If you want your text to look less jagged, choose Edit > Options > Misc... and turn on the Antialiased Text option. For crisper (but jaggier) text, turn the antialiased text option off.

## Image types and drawing colors

If the image type is 8-, 16-, or 32-bit, lines and fills will be made in shades of gray rather than the selected foreground color.
To draw on a grayscale image in color, convert the image to RGB color (Image > Type / RGB Color).

## Foreground and background colors?

The foreground and background colors appear in the Color Picker tool on the ImageJ tool bar.


Background Color
To set the foreground color, click on a pixel in an image or in the Color Picker
 window using the Color Picker tool
Alt-click (Win), or click the Color Picker's toggle button (Mac) to set the background color. Double-click the Color Picker tool to open the Color Picker window.

## Precision color

Double-click anywhere in the Color Picker window to open a window with sliders for adjusting the red, green, and blue components of the foreground color.


## Cutting \& Pasting

## Basic cutting, copying, and pasting

1. Make a selection (see the Selecting Skill sheet for more about making selections).
2. Choose Edit > Cut or Edit > Copy to copy the selected pixels to the ImageJ clipboard.
3. Choose Edit > Paste to put the copied selection back in the image window as a floating layer that can be moved around with the cursor.
4. Drag the pasted selection to the desired location.
5. Click outside the colored selection boundary to finish the paste operation. The original pixels are replaced with the pasted pixels.

## Cutting and pasting between different image types

Different image types handle lookup tables differently. When you cut or copy a selection from an 8-bit grayscale image and paste it into an 8-bit color image, the results usually look awful. For best results, the images you are cutting or copying and pasting between should be of the same type.

- If you need a grayscale result, convert all images to 8-bit grayscale.
- If you need a color result, convert all images to RGB color.


## The Paste Control

Normally, when you paste a selection, the original pixel values of the image are replaced by the pixel values of the floating layer. The Paste Control allows you to perform operations between the pixel values of the two layers, producing interesting transparency and blending effects.
To show the Paste Control window, choose Edit > Paste Control. The Paste Control only works while the pasted layer is still "floating" over the original image, and has no effect after the paste operation has been finished. Click the Close box in the Paste Control window to close the window.

## What is the Transfer Mode?



The Transfer Mode determines how the pixel values in the original image and the floating layer interact to produce new pixel values. Different Transfer Modes cause the floating layer to appear solid, transparent, or blend with the original image.

## Changing the Transfer Mode

Pull down the Transfer Mode pop-up menu and choose a transfer mode.

## Cut or Copy?

What is the difference between the Cut and the Copy commands? Both commands copy the selected pixels to the clipboard, but Cut replaces the selected pixels with the background color, while
Copy leaves the selected pixels unchanged.

## Copying and pasting between ImageJ and other applications

ImageJ currently does not utilize the system clipboard that is used to move data between different applications.
You may, however, be able to copy to or paste from the system clipboard through the System Clipboard plugin. Choose Plugins > System Clipboard and click the appropriate button. (Note: The System Clipboard plugin is not available for Mac OS 8.x or 9.x.)


## What does each Transfer Mode do?

To learn what each transfer mode does, refer to the ImageJ online documentation at:
http://rsb.info.nih.gov/ij/docs

## Thresholding

## ImageJ

 Skill
## What is thresholding?

Thresholding allows you to highlight the pixels throughout an image whose values are in a specific range. For example, if pixel values represent elevation, you can select all of the pixels with values between 10 and 20 meters to represent a specific flooding region. Once thresholded, you can perform many functions on the highlighted pixels, such as measuring the area they cover.

## Turning Thresholding on and off

- To turn Thresholding on, choose Image > Adjust > Threshold....
- To turn Thresholding off, click the Reset button in the Threshold window.


## Adjusting the range of thresholded values

Drag the Lower and Upper Level slider controls in the Threshold window or click the Set button and enter the Upper and Lower Threshold levels manually.


## Measuring only the thresholded pixels

- Choose Analyze > Set Measurements... and check the Limit to Threshold checkbox.
- Make measurements as described in the Measuring Skill Sheet.
- To measure all of the pixels in the image, either turn off Thresholding mode or choose Analyze > Set Measurements... and uncheck the Limit to Threshold checkbox.


## Threshold display options

Thresholding offers three different display options:

- Red - pixels with values in the specified range of values are highlighted red.

- Black \& White - pixels within the specified range appear black, and pixels outside the limits appear white.

- Over/Under - pixels with values below the specified range are highlighted blue, and pixels above the range are highlighted green. Pixels within the specified range are not highlighted.



## Calibrating Images

Calibrating an image allows you to measure distance, area, and pixel value in meaningful units.

## Spatial calibration

Spatial calibration defines horizontal and vertical dimensions of an image in real-world units such as millimeters, centimeters, miles, etc.

## When there is a known distance in the image

When the image contains a grid, ruler, or object of known size:

1. Use the straight line selection tool to select a known distance on the image.
2. Choose Analyze > Set Scale...
3. Enter the Known Distance.
4. Enter the Unit of Length.
5. Click OK.

## When you know the scale (e.g., 1 pixel $=0.025 \mathrm{~mm}$ )

1. Choose Analyze > Set Scale....
2. Enter the Distance in Pixels (1).
3. Enter the Known Distance (0.025).
4. Enter the Unit of Length (mm)
5. Click OK.

## Uncalibrating an image (removing the scale)

1. Choose Analyze > Set Scale...
2. Enter 0 for the Known Distance, or delete the Unit of Length.
3. Click OK.

## Density calibration

Pixel values often represent measurements-temperature, elevation, brightness, etc.-recorded at regular intervals in a grid pattern. Sometimes the pixel values are the actual measurements, but often they are only proportional to the actual measured values. Density calibration is the process of scaling the pixel values to approximate the original measured quantities.

The information needed to density calibrate an image can come from either of two sources - measurements made on the image itself (such as from a calibration bar), or from image metadata (information about the image, often provided in a separate text file, printed manual, or Web page).


ImageJ Skill

## Setting scale




## What is the Pixel Aspect Ratio?

ImageJ always displays pixels as squares, but some imaging devices use rectangular pixels. The Pixel Aspect Ratio setting tells ImageJ how to adjust for non-square pixels, so that length and area measurements are accurate.
You can determine the pixel aspect ratio of a camera or scanner by creating an image of a circular object and carefully measuring the height and width of the object in the image. Divide the measured width by the height (both in pixels) to get the pixel aspect ratio.
For example, the width of this scanned nickel was 255 px and the height was 250 px, a pixel aspect ratio of 1.02. Unless otherwise noted, leave the pixel aspect ratio set to 1 (square pixels).


## Density calibration from image measurements

If an image includes a density scale or regions of known value, you can calibrate it by taking measurements directly from the image. In this example, the image represents sea surface temperature near the East coast of the United States. The calibration bar relates pixel brightness to the temperature in degrees Celsius.

1. Choose Analyze > Clear Results to delete any previous measurements.
2. Use a selection tool to select a small portion of the calibration bar on the image, and press $\mathbf{M}$ to measure the average pixel value of the selection.
 Repeat this for one or more other regions of known temperature.

3. Choose Analyze > Calibrate... to open the Calibrate window.
4. The average pixel values you measured should be listed in the left-hand column of the Calibrate window. Enter the known value (Temperature in this case) in the right-hand column for each measured value.
5. Enter the appropriate units in the Unit box.
6. Select Straight Line from the Function popup menu.
7. Click OK. A plot of the calibration function and a list of pixel values and their corresponding calibrated values are displayed. Close these windows.


As you move the cursor around the image, the status bar will now display both the calibrated and uncalibrated pixel values. Pixel measurements (mean, minimum, maximum, etc.) will also be made using calibrated values.


## The Calibrate window



## Calibration functions

Not all image calibrations use a straight line function. In general, you should choose the simplest function that "fits" the calibration data you entered.

The correlation coefficient, labeled $\mathrm{R}^{2}$ on the calibration plot, tells how well the function you chose fits the data. The closer this value is to 1 , the better the fit. (A correlation coefficient of 1 represents a"perfect fit." However, don't get too excited if your $R^{2}=1$. With only two points, you will always get a perfect fit, but that doesn't mean that your calibration is perfect!)

## Density calibration from image metadata

In this example, you are working with an image of the surface of Mars. The pixel values represent elevation (dark = low, bright $=$ high $)$.


The image is accompanied by a label (.lbl) file containing image metadata (information about the image data). From this file, you learn that the elevation values in the image range from -8068 meters to 21134 meters. The minimum and maximum values in the image range from 0 to 255.


1. Choose Analyze > Calibrate... to open the Calibrate window.
2. Enter the known minimum and maximum pixel values in the left-hand column and the corresponding minimum and maximum elevation values in the right-hand column.
3. Enter the appropriate units in the Unit box.
4. Select Straight Line from the Function popup menu.
5. Click OK. A plot of the calibration function and a list of the pixel values and their corresponding calibrated values are displayed. Close these windows. The ImageJ status bar and Results window will now display calibrated pixel values.

## How to tell if an image is calibrated

Spatially calibrated images show X and Y in both calibrated units and pixels in the ImageJ status bar. The image window status bar also shows the dimensions of the image in calibrated distance units.


Density calibrated images show pixel values in both calibrated units and raw values in the ImageJ status bar.

## Removing density calibration

To remove the density calibration from an image:

- Choose Analyze > Calibrate...
- Set the calibration function to None.
- Click OK.


## Be an image detective

Spatial and density calibration data can come from many sources. It may be found in separate label (.lbl) or header (.hdr) files, or it may be embedded at the beginning of the image file itself or in the image documentation. You may even be able to get the information from the person or agency that provided the images.

## The Calibrate window

To enter or change values in the Calibrate window, click in a column and type the number. Press the Return key to enter the next value.


## Measuring

Before you begin measuring images, you may need to:

1. Calibrate the image (see the Image Calibration Skill Sheet.)
2. Choose (set) the measurements you want to make.

## Setting measurements

1. Choose Analyze > Set Measurements....
2. Check the measurements you wish to make and set the number of decimal places for your measurements. See opposing page for descriptions of each measurement.
3. Click OK.

## Measuring

1. You can measure the entire image or select a portion of the image using the selection tools. See the Selecting Skill Sheet for more information about making selections. If nothing is selected, ImageJ measures the entire image.
2. Choose Analyze > Measure, or press $\mathbf{M}$ to measure the selection.
3. After the first measurement, the Results window appears. The Results window is updated after each measurement.

## The Results window

Measurement results are displayed in the Results window. Measurements can be saved to a text file, cut, or copied for use in other programs such as spreadsheets or graphing applications using the options available from the Results window menu bar. On Windows computers, the results menu bar is part of the Results window; on Macintosh computers, the results menus appear at the top of the screen when the Results window is active.


## Deleting all measurements

To delete all measurements, choose Analyze > Clear Results on the ImageJ menu bar or Edit > Clear Results on the Results window menu bar.

## Summarizing measurements

To calculate basic statistics (mean, standard deviation, minimum, and maximum) for each measurement field, choose Analyze > Summarize or Edit > Summarize in the Results window menu bar.

## Changing Results column widths

If the columns of numbers in the Results window overlap or are difficult to read, resize the columns by dragging the line separating column headings.

| 00 | O Results |  |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  | Label | Area | +1ean | 5 StD 4 |
| 1 | leaf | 1715 | 74.53 | 16.3: 7 |
| 2 | leaf | 2255 | 98.82 | 33.8 |
| 3 | leaf | 820 | 78.59 | 10.6: |
| 4 | leaf | 1273 | 84.31 | 19.0 |
| 5 | leaf | 1407 | 149.50 | 42.4i |
| 6 | leaf | 1043 | 163.93 | 44.4 |
| Mean <br> SD <br> Min <br> Max |  | 1418.83108 .28 |  | 27.8 |
|  |  | $511.50 \quad 38.68$ |  | 14.3 |
|  |  | 820 | 74.53 | 10.6 |
|  |  | 2255 | 163.93 | 44.4 |
| $\square \square$ |  |  |  |  |
|  |  |  |  |  |

Measurement summaries appear at the bottom of the Results window.

## Measuring angles

Use the angle tool $\Delta$ to measure angles with three clicks as shown below. After the second click, the interior angle formed by the two rays is displayed in the ImageJ status bar as you move the cursor.

To record angle measurements, choose Analyze > Measure, or press $\mathbf{M}$. When measuring with the angle tool, the angle measurement option is automatically enabled.


## Redirecting measurements

There are times when it is necessary to use the features of one image to select areas to measure in another image.

The image at right shows elevation data for the area around Mt. Denali, Alaska. The lower image shows vegetation in the same area. To measure the vegetation within a specific elevation range:

- Open the source image, and select the region you want to measure.
- Open the target image containing the data you want to measure.
- Choose Analyze > Set Measurements... and set Redirect To: to the target image.
- With the source image active, press the $\mathbf{M}$ key to measure. The measurements will be made using the pixel values of the target image.

This technique is also used for measuring changes in images created at different times, or images that have to be highly processed to see the features you want to measure, where the processing changes the original pixel values.


## Measurement options

Area - Area in pixels, if uncalibrated, square units if calibrated.
Mean Gray Value - Average gray value within the selection.

Standard Deviation - Standard deviation of the values used to generate the mean gray value.

Modal Gray Value - Most frequently occurring gray value within the selection.

Min \& Max Gray Level - Minimum and maximum gray values within the selection.

Centroid - The center point of the selection. This is the average of the $x$ and $y$ coordinates of all of the pixels in the selection.

Center of Mass - The brightnessweighted average of the $x$ and $y$ coordinates of all pixels in the selection

Perimeter - The length of the outside boundary of the selection.
Bounding Rectangle - Defines the smallest rectangle enclosing the selection.

Fit Ellipse - Fits an ellipse to the selection.

Circularity - A value of 1.0 indicates a perfect circle.
Feret's Diameter - The longest distance between any two points along the selection boundary.
Integrated Density - Sum of pixel values in selection (number of pixels $x$ mean pixel value).
Median - The "middle" value of the distribution.

Skewness - Statistical measure of the symmetry of the distribution.
Kurtosis - Statistical measure of the "sharpness" or "flatness" of the distribution.

Limit to Threshold - Only thresholded pixels are included in measurement calculations.

Display Label - Lists the image name and slice number (for stacks) in the first column of the results table.

Invert Y Coordinates - If checked, the XY origin is assumed to be the lower left corner of the image window instead of the upper left corner.

Redirect To - Allows you to outline a structure on one image and measure the corresponding region in another image.

Decimal Places - This is the number of digits to the right of the decimal point in real numbers displayed in the results table.

## Stacks \& Animation

## What is a stack?

A stack is a series of two or more images in a single window. These images, called slices, can represent motion, time, space, or wavelength. Slices can be displayed in sequence either manually or automatically, and can be used to create two- and three-dimensional views of a scene or object. The features of a stack window are shown below.


## Moving through a stack

- Manually-Use the > and < keys or the scroll bar (slice slider) to move forward and backward through the slices in a stack. (Hold the > and < keys down to move quickly through many slices.)
- Animation-Choose Image > Stacks > Animate or press the backslash (I) key to start and stop the animation. (You can also click on the image to stop the animation.)


## Making a stack

To make a stack, all images must be the same type and have the same dimensions (height, width, and bit depth).

From a series of open images-Choose Image > Stacks > Convert Images to Stack. The new stack window is named Stack. (The original image names are shown to the right of the slice counter, but are lost when you save the stack.)
From a series of saved files-Choose File > Import > Image Sequence, and specify the sequence options for the slices you want to import.

## Order of slices

When you make a stack, the slices are arranged in the order in which they were created or opened, which is shown at the bottom of the Windows menu. When you import an image sequence, images are opened in alphanumeric order by image name.

## The slice counter

The stack window status bar shows the current slice number and the total number of slices.

## Setting the animation speed and direction

Choose Image > Stacks > Animation Options..., and enter the desired speed in frames per second ( $0.5=$ one frame every two seconds).


Normally, the animation plays in 1-2-3-1-2-3 order. By checking the Loop Back and Forth option, the animation will play in 1-2-3-2-1 order.

Sequence options

| Sequence Options |  |
| ---: | ---: |
| Number of Images: $\sqrt{27}$ |  |
| Starting Image: $\sqrt{1}$ |  |
| Increment: $\sqrt{1}$ |  |
| File Name Contains: | $\square$ |
| Scale Images | $\boxed{100} \%$ |
| T Convert to 8-bit Grayscale |  |
| $320 \times 240 \times 27(2.0 \mathrm{MB})$ |  |
|  | OK |

## Unstacking a stack

To separate a stack into separate windows, choose Image > Stacks > Convert Stack to Images. The windows are numbered 001, 002, 003, etc.

## Editing a stack

To add a slice after the current slice-choose Image > Stacks > Add Slice.

To add a slice before the current slice-hold down the Alt (Win) or Option (Mac) key, then choose Image > Stacks > Add Slice.

To delete a slice-choose Image > Stacks > Delete Slice.

## Creating a montage

Since a stack cannot be printed for a report or poster, ImageJ provides a way to arrange the slices of a stack in rows and columns as a single image called a montage.

To make a montage, choose Image > Stacks > Montage..., specify the number of rows and columns, and click OK.


## Converting a stack to an .avi movie

To convert a stack into an .avi movie that can be played on many freeware movie player applications such as QuickTime Player, choose File > Save As... > AVI... and save the file. Optional plugins are available from the ImageJ download site that may allow users to save stacks in other movie formats.


## Processing stacks

When you conduct processing operations on a stack, you are given the option to process the current slice only (No), all slices (Yes), or cancel the process.


## Montage options

In addition to the number of rows and columns, you can resize the images, include every Nth image, and other options.


## Managing Windows

## Parts of a window



## To activate a window

Click on part of the window, or choose the window you want from the
Window menu. The active window has a shaded title bar or lines along it.

## To move a window

Drag the window by its title bar to a new location.

## To show the ImageJ window

If the ImageJ window is hidden, press the Enter key (Return on Macs) to move it to the front.

## To put an image window behind other windows

Sometimes, one image completely covers another. To move an image behind other images, choose Window > Put Behind.

## Organizing windows

The Window menu provides several ways to organize image windows on the screen.

1. Cascade-Piles and fans out the windows, in order, so that you can see the title bars.
2. Tile-Lays out the image windows on the screen so they do not overlap.

## Lesson Introduction

## What is image processing technology?

Image processing technology allows users to gather and analyze an incredible amount and range of information from digital images. Digital images are composed of rows and columns of picture elements or pixels, each representing a number. Like a paint-by-number picture, each number corresponds to a specific color or shade of gray. Digital images contain information that may not be visible at first. Image processing can help you "see" this information.
The lessons in this guide will teach you how to use image processing tools to view, analyze, and evaluate images for various purposes. For example, you may be asked to do forensic detective work on images of fingerprints or plan a cross-country rock-and-roll tour by measuring distances on a satellite image. Once you've mastered some of these techniques, the possibilities are endless. You can eventually capture and analyze your own images. Image processing allows you to use your math and science skills, as well as your creativity, so have fun and enjoy!

## Lesson Descriptions

Pixelated Pictures
Use a "stadium picture" to discover what a digital image is and how it can be manipulated. Explore different uses of digital images in fields ranging from medicine to art to geography.
Lights, Camera, Action! ..... 55

Learn how to create and manipulate image stacks. Make your own
animated cartoon, track a hurricane, see a lightning strike in slow
motion, and slice a brain to see inside it.

Digital Detective ........................................................ 67
Process low-contrast images to reveal the picture of a suspected drug dealer, compare fingerprints from a crime scene with those of the suspects, and look for evidence of a forger using a new alias.
More Than Numbers ..... 83

Construct two- and three-dimensional graphs to help you "see" the numbers in digital maps, photographs, and other images.
North American Tour ..... 99

Plan your band's cross-country concert tour using image processing! Locate cities and other features on a satellite image and measure distances as you search for the most cost-effective route.
image, $n$. An optically formed representation of an object.
process, v. In computer science, to perform operations on data.
technology, n . The application of scientific knowledge, especially in industry or business.
Digitizing Details ..... 107Learn about image resolution, bit depth, and file formats, andhow to use them to get the best possible images for a variety ofsituations and uses.
Picture Yourself ..... 125Have you ever dreamed of yourself as president or a famoussports or entertainment figure? Ever wanted to see your face ona $\$ 100$ bill? Using digital images, measurement, scaling, copyingand pasting, you can picture yourself doing what you've alwaysdreamed of!
Color by Number ..... 133
Take color images apart and put them back together to learnthe principles of color imaging. Make both true- and false-colorsatellite images, and use color to create cool "three-dimensional"images!
Photo Effects ..... 149
Edit and enhance images to create zany, as well as useful, photographic effects. Put yourself on the moon, become fish bait, turn into an animal, design an ad for a product, or bring an old photo back to life!
Images for Science 167
Download a set of raw images from the Internet (alternative images are provided if Internet access is unavailable) and prepare them for scientific analysis and display.

## Pixelated Pictures Understanding Digital Images

## Overview

Digital image processing is used by professionals in many fields, from biomedical imaging to graphic arts. This lesson introduces students to digital images and a few basic image processing techniques.
In Part 1, students examine the pixels (picture elements) that make up digital images. They are introduced to the concept of a digital image as a series of numbers that are displayed according to a color lookup table. With image processing technology, students can change the appearance of images for aesthetic or scientific purposes by changing the numerical values of pixels using drawing tools, mathematical operations, or by modifying the lookup table.


In Part 2 of the lesson, students compare digital images to the large pictures formed by sports fans holding up colored cards in a stadium. By developing this analogy, students better understand how digital information is displayed and manipulated. The X,Y coordinates used as pixel "addresses" in a digital image are shown to be analogous to seat and row numbers in a stadium. Students also compare this coordinate system to the Cartesian coordinate system used for drawing mathematical graphs. Students explore the relationship between pixel values and the lookup table by changing color tables and reading pixel values. They use the stadium picture analogy to build an understanding of image histograms and the use of thresholding to highlight specific ranges of pixel values.

In the final section, students explore images that demonstrate how image processing is used in the workplace. Students are introduced to a few simple imaging techniques as they interact with the images.


Teaching Notes

## Goal

Students use the tools of ImageJ to discover the characteristics of digital images and explore how they are used.

## IP Skills

- Changing image modes
- Changing color tables
- Interpreting histograms


## ImageJ Skill Sheets

- Lookup Tables
- Thresholding
- Managing Windows


## Vocabulary

- pixel
- digital image
- image processing
- lookup table


## Objectives

Students will:

- explain the key properties of pixels, their coordinates, and their values;
- explain relationships between pixel values, lookup tables, and images;
- use lookup table manipulation and filtering to process digital images, and
- discover how digital images are used in a different careers.


## Related Activities

- See "Digital Detective" for an introduction to image processing in the field of forensics.


## Career Links

Many government and public agencies use digital images to archive information. For example, space agencies, weather service bureaus, and hospitals either create digital images or scan images and documents for storage onto CD-ROM, DVD, and other media. Image processing is also common in fields such as graphic arts and city planning.

## Important Setup Note

The "normal" origin $(0,0)$ for ImageJ is in the upper left corner of the image, with Y values increasing toward the bottom. Before your students do this lesson, you should modify this behavior so that ImageJ matches the familiar graphing space of Quadrant I of the Cartesian coordinate system.

- Choose Analyze > Set Measurements.
- Turn on (check) the Invert Y Coordinates option.

This will move the origin $(0,0)$ to the lower left corner. This setting is saved with other ImageJ preferences, and will remain in effect until it is turned off. (It is not saved with the image.)

## Acknowledgments

Lesson developed by Carla McAuliffe and LuAnn Dahlman, Center for Image Processing in Education, Tucson, Arizona. Updated for use with ImageJ by Larry Kendall.

Parts of this lesson are based on the lesson "Stadium Pictures," developed by LuAnn Dahlman and Larry Kendall, Image Processing for Teaching project, University of Arizona, Tucson, Arizona.

Slice one of Stadium Picture © Herb Weitman/NFL Photos, slice two of Stadium Picture © Baron Wolman/NFL Photos, and slice three of Stadium Picture © Paul Spinelli/NFL Photos. Used with permission.
House Drawing is courtesy of Patti Parsons, Phoenix, Arizona. Winter and First Eagle are courtesy of Nelson Begay, Tuba City, Arizona.
Other images courtesy of the National Institutes of Health, National Aeronautics and Space Administration (NASA), and the authors.

## Inverting Y Coordinates

| $\pm$ Set Measurements |  | x |
| :---: | :---: | :---: |
| $\checkmark$ Area | F Mean Gray Value |  |
| V Standard Deviation | - Modal Gray Value |  |
| V Min \& Max Gray Value | $\Gamma$ Centroid |  |
| $\Gamma$ Center of Mass | $\Gamma$ Perimeter |  |
| $\Gamma$ Bounding Rectangle | 「 Fit Ellipse |  |
| $\Gamma$ circularity | $\Gamma$ Feret's Diameter |  |
| V Limit to Threshold | $\checkmark$ Display Label |  |
| F invert Y Coordinates |  |  |
| Redirect To: None | $\square$ |  |
| Decimal Places: ${ }^{2}$ | OK | Cancel |

## Answers

1. Small squares or pixels are visible.
2. Numbers in the status bar (the $X$ and $Y$ coordinates and the pixel value) change as the cursor is moved.
3. Answers will vary. Dragging the sliders to the right:

Minimum-increases contrast and decreases brightness; pixels with lower values are rendered lighter.
Maximum—increases contrast and brightness; pixels with lower values are rendered lighter.
Brightness-increases brightness.
Contrast-increases contrast.
4. If fans traded cards randomly, they would not form a recognizable or meaningful picture.
5. The value reported for X changes one unit for every one pixel moved in the horizontal direction.
6. The value of Y shows a pixel's vertical, or up-and-down location.
7. Answers will vary. X is equivalent to the seat number, and Y is equivalent to the row number in a stadium.
8. The $X, Y$ coordinate system of this digital image is the same as mathematical graphs, X increases to the right and Y increases toward the top.
9. The origin $(0,0)$ is in the lower left corner of the image.
10. To find the total number of pixels, go to the upper right (don't forget to add 1 to the X and Y coordinates, since the row and seat numbers begin with zero). Multiply the number of rows $(\mathrm{Y})$ by the number of seats or columns (X).
11. $415 \times 640=265,600$ total pixels.
12. Answers will vary. The values range from 0 to 255.
13. Answers will vary. Complete answers will show numbers for $\mathrm{X}, \mathrm{Y}$, and Value.
14. No. The test pixel's value does not change when a new lookup table is used.
15. The pixel value does not change. Changing lookup tables only changes the color assigned to each value, not the pixel values.
16. The lookup table determines the colors currently assigned to each of the 256 possible pixel values in the image. Each pixel in the image is shown as the color or shade of gray assigned to it in the lookup table. Changing the color assignments by applying a new color (i.e. lookup) table can dramatically change the appearance of the image, although the pixel values are not changed.
17. Answers will vary since changing one value changes the other. Setting the Level to around 900 and the Window to 1700 looks pretty good.
18. The pixel value of 40 appears most frequently in the Mona Lisa image.

Note: questions 8 and 9 assume that you have set the origin $(0,0)$ of the images from the default position in the upper-left corner to the lowerleft corner of the image window. See the Troubleshooting section of this lesson for instructions on changing the location of the origin.
19. Pixels with a value of 227 are mostly located in Mona Lisa's face and neck.
20. The noisy pixels disappeared. This makes the actual surface features easier to see. You may notice that remaining objects in the image are more blurry or less sharp.

# Pixelated Pictures Understanding Digital Images 



## What is a digital image?

Open X-ray.tif.

You are looking at an X-ray image of major arteries in the neck and chest. The patient is facing you, although his head is turned to the side. Notice how the fillings in the patient's teeth are visible. The large loop at the bottom of the image is the aorta. When pictures like this are displayed on a computer screen, they are digital images. But, what exactly are digital images? How are they used, and what characteristics do they possess?

呾 Use the Magnifying Glass tool on the image four or five times to zoom in. (You can click 8 while holding down Control [Win] or Option [Mac] to restore the image.)

1. What do you see?

The small squares that make up a digital image are called picture elements or pixels.

Watch the status bar as you move the $\mathrm{Q}_{\text {tool across the pixels. }}$
2. What do you notice about the status bar? (Be specific!)

You should see numbers changing. The pixels in a digital image represent a grid or table of numerical values. Each pixel value is a single measurement recorded at a specific time and place. By assigning a specific color to each different pixel value, a picture or digital image is formed. An X-ray is an image made up of X-ray "brightness" measurements. Where the X-rays are blocked, by bone for example, the brightness is low. Where the X-rays pass through, in substances such as air or soft tissue, the brightness is high. Scanners and digital cameras collect these brightness measurements and display them as digital images.
With image processing technology you can analyze and change features of pixels such as their brightness, contrast, or numerical value.
[ Choose Image > Adjust > Brightness/Contrast to open the $\mathbf{B} \& \mathbf{C}$ (Brightness \& Contrast) window. Adjust each of the four slider controls, and watch how each slider affects the image appearance.
3. Describe what happens to the appearance of the image when you move each of the four sliders in the $\mathbf{B} \& \mathbf{C}$ window.
You should have noticed that changing the brightness and contrast changes the appearance of different features in the image. Changing the appearance of an image to make features easier to see is called image enhancement. Another simple way to enhance images is to change the colors used to display the pixel values.

Choose Image > Lookup Tables > Spectrum, and observe the details now visible in the image. (Feel free to "try on" some of the other color tables.)

## Gray is a color too!

Most people don't think of black, white, and gray as colors, but we'll call them colors and we encourage

## Enhancing science

Image enhancement is a powerful tool. For example, being able to better see the details of a person's heart and vessels can help a doctor to make a more accurate diagnosis.
$\frac{1}{2}$ File > Close

[^1]you to do it too!
Lile > Open

## Big pictures from little squares

Open Stadium Picture.tif. This image is really three images, each in a separate layer, or slice.

Press the > and < keys to move through the slices of Stadium Picture.tif. Or use the arrows on the slider bar at the base of the image.

Have you ever seen fans in a stadium form a big picture as part of a halftime show? Each seat in the "card section" is assigned one or more large cards, each a solid color. On command, each fan holds up the appropriate card. Viewed from across the field or from high above the stadium, the individual cards blend together to form a large picture.

Close up, all you can see are the individual colored cards. The pixels in a digital image are similar to the cards held by each fan.
4. How would the stadium picture look if the fans randomly traded cards with each other?

The location of each colored card is extremely important for displaying the picture. Stadium sections have "addresses" for every seat-locations are given by seat and row numbers. Digital images have a similar system for describing the location of each pixel.

## Close Stadium Picture.tif.

## Discovering pixel "addresses"

品 Open Mona Lisa.tif.
品 Click $\mathrm{Q}_{\mathrm{a}}$, then click the image several times until you can see individual pixels.
Move your cursor horizontally across a row of pixels, and watch the changes in the status bar.
5. Which number in the status bar changes one unit for every one pixel you move in the horizontal direction?

Move the cursor vertically (up or down) in a column of pixels, and again watch the changes in the status bar.
6. Which item in the status bar shows a pixel's vertical location?
7. Describe how seat and row numbers in a stadium compare to the coordinate system used to locate specific pixels in a digital image.
8. How is the $\mathrm{X}, \mathrm{Y}$ coordinate system in this digital image similar to mathematical graphs you have used?
Double-click 0 to view the entire image again.
몰 Based on what you observed, move your cursor to the origin of the image (the pixel with the lowest $\mathrm{X}, \mathrm{Y}$ coordinates).
9. Where is the origin (row 0 , seat 0 ) of the image?
10. How could you find out the total number of pixels in this image?
11. How many pixels are in this image?

## Mona Lisa's pixel locations

The Mona Lisa image is moderately large and is not displayed at full size by ImageJ. As a result, pixel locations will not change uniformly as you move the cursor over the image.
To display it at full size, select Edit
> Options > Image . . . and click
Open Images at $\mathbf{1 0 0 \%}$. Then reopen Mona Lisa.tif.
Make sure you reset this option when you have finished this part of the lesson (click Open Images at 100\%).

## Image width and height

Remember that in ImageJ, the "row" and "seat" numbers start with 0,0 not 1,1. That means that you have to add 1 to the maximum $X$ and $Y$ coordinates to get the width and height of the image.

## The value of a pixel

Besides the specific address given by X and Y coordinates, each pixel in a digital image has another number associated with it.

ㅁ. Use the tool to zoom in several times until you can see individual pixels.

를 Place your cursor on the lightest pixel in your field of view and read its Value in the status bar. Do the same for the darkest pixel you see.
12. What are the highest and lowest pixel values you can find in this image?

## Assigning colors to values

In a stadium picture, each fan is given a card with a specific color. Likewise, in a digital image each pixel value is represented by a specific color. In digital images, a color is assigned to every pixel value using a lookup table. A simple lookup table for an image with up to 16 different values is shown at the right.
ImageJ uses 8-bit lookup tables containing $2^{8}$, or 256, different colors. Normally, pixels with a value of 0 are shown as white and pixels with a value of 255 are shown as black. (If that seems backward to you, ImageJ can also be set to assign 0 to black and 255 to white—both ways are "correct.") Two hundred fifty-four colors or shades of gray are used to show the values between white and black.
13. Zoom in on any pixel in your image. Write down its location and value.

D Choose Image > Lookup Tables, and pick a new lookup table from the available choices. The new lookup table assigns new colors to each pixel value.
四 Move your cursor back to your test pixel from question 13.
14. Has the pixel value changed?

ㄴ. Try other lookup tables, and check the value of your test pixel.
15. What happens to the pixel values when you apply a new lookup table to an image?
16. Describe how the lookup table affects the appearance of a digital image.
$\square$ Change the lookup table back to grayscale when you are finished by choosing File > Revert.

## Bit depth

The number of possible pixel values in an image is often called the bit depth of the image. Bit depth is determined by the number of binary digits, or bits, that are used to represent the value of each pixel. ImageJ can work with 8-, 16-, and 32-bit images. The table below shows the number of different values that are possible at each bit depth.

| Bit Depth | Possible Pixel Values |
| :---: | :---: |
| 8 -bit | $2^{8}=256$ |
| 16 -bit | $2^{16}=65,536$ |
| 32 -bit | $2^{32}=4,294,967,296$ |

Wow！A pixel in a 32－bit image can represent any one of more than 4 billion possible values！Unfortunately，ImageJ uses lookup tables with only 256 colors．When displaying 16 －and 32 －bit images，ImageJ divides the range of possible pixel values into 256 groups，called bins，and assigns a single color to all of the pixels in each bin．That means that in a 16 －bit image，a single color can represent a range of up to 256 different pixel values，and in 32－bit images，each color of the lookup table can represent nearly 16.8 million different values！The trick to working with 16－and 32－bit images is learning how to assign the 256 available colors to the range of pixel values you＇re most interested in．This process is called windowing．

Open the Galaxy．tif image．（Do not close Mona Lisa．tif．）
This is a 16 －bit image of the Andromeda Galaxy taken with a digital camera mounted on a telescope on Kitt Peak in Arizona．The image doesn＇t contain a full 65，000 different pixel values，but it does use far more than 256 different values to record faint details of the galaxy．At first，it doesn＇t look like much，but looks can be deceiving．

## Choose Image＞Lookup Tables＞Fire．

Our eyes are more sensitive to different colors，so you should see a little more detail in the image using the Fire lookup table．

旺 Choose Image＞Adjust＞Window／Level．
The Window／Level（W\＆L）panel contains two sliders．One controls the window width－the range of pixel values in the image that the lookup table is applied to．Pixel values above this range are displayed as black，and values below it are displayed as white．The other slider controls the Level－ the pixel value where the window（and the lookup table）is centered．These adjustments will make more sense after you experiment with them a bit．

虽 Adjust the Window and Level controls in the W\＆L panel until you think the maximum detail is visible in the image．
17．What Window and Level settings produce the best range of detail in the image？

Close the Galaxy．tif image and the W\＆L window．

## Back to the stadium

Imagine that you want to create the Mona Lisa as a stadium picture．To produce the individual cards to give to the fans，you will need to know how many cards of each shade to paint．

曹 Choose Analyze＞Histogram to see a histogram of the pixels used to create Mona Lisa．

Think of a histogram as a way of organizing your stadium cards．If you sort them by pixel value along a number line from 0 to 255 ，the resulting piles of cards would resemble a histogram．The most－used colors（values）would have the highest piles while the least－used colors would have smaller piles． The piles show the amount（the number of cards）of each color（value） needed to produce the picture．
18．What is the most frequently used pixel value（color）in Mona Lisa？
믄 Close the Histogram window．

## 32－bit floating point

The 32－bit image format used by ImageJ represents values using 32－bit floating point notation．This allows pixels to represent values containing a decimal point．It is especially useful for scientific imaging，because each pixel value can be an actual measurement value，such as $31.625 \mathrm{~W} / \mathrm{m} 2$ ．（The units，of course，are not stored with the pixel value．）These＂32－bit float＂ values can represent numbers ranging from about $1.7 \times 10^{38}$ to $5.9 \times 10^{-39}$ ．The possibilities are nearly endless！

## More practice

If you want to practice working with more 16－and 32－bit images，you will find them in the $\mathbf{1 6}$－bit Images and 32－bit Images folders．

## Sorting the cards



## Mona a la mode

Another name for the most frequently used value is the Mode．

## Highlighting pixels in the image

Sometimes it is useful to identify the pixels in an image whose values are within a certain range. For example, let's pretend for a moment that the pixel values in Mona Lisa represent temperature instead of brightness. As part of a diagnostic procedure or research study, you might want to know which parts of the image are between $30^{\circ} \mathrm{C}$ and $45^{\circ} \mathrm{C}$.

If the back of each "card" used to create the image was colored bright red, you could highlight where specific pixel values occur in the image. Imagine flashing this message on the scoreboard while the card section is showing the Mona Lisa image:

## IF THE RUGBBER ON YOUR CRRD <br> 15 between 30 RחO 45, 5HOW <br> THE RED SIDE OF YOUR CRRD!

You can do something similar in ImageJ. It is a process called Thresholding.
岛 Choose Image > Adjust > Threshold to turn on Thresholding mode.
In Thresholding mode, the pixels whose values are within a range of values are highlighted.

모․ Highlight all of the pixels with values between 30 and 45 by adjusting the Threshold range using the two slider controls in the Threshold panel.

모 You can also set the upper and lower Threshold limits precisely by clicking the Set button and entering exact values. Use this method to set the Threshold range to 49-73.
回 With Thresholding on, create a new histogram to see which pixels are within the Threshold limits.

You can highlight a single pixel value by setting the upper and lower limits of the Threshold to the same value.

Set the Threshold to highlight pixels with a value of 227 in the Mona Lisa image.
19. Where in the image are most of the pixels with a value of 227 located?

Close Mona Lisa.tif and the Threshold window.

## Careers using digital images

Many careers use digital images. Government and public agencies as well as private businesses use digital images to display, interpret, and archive information. For example, space agencies, weather bureaus, and hospitals make digital images or scan images and documents for storage onto CDROMs and other media. Today, image processing is used in a variety of fields ranging from graphic arts to medical technology to city planning. In this section, you will explore a few workplace applications of digital imaging.

## Open Medical Image.tif.

This is a magnetic resonance image (MRI) of a patient's head. In the field of medicine, image processing techniques can allow a doctor to visualize anatomy without surgery. Health-care technicians use a wide variety of specialized equipment for collecting images like this.

## Thresholding

The Threshold dialog box lets you choose from three different methods of highlighting a range of pixel values.


## MRI = NMR

Magnetic resonance imaging is sometimes called nuclear magnetic resonance imaging (NMR) because it measures the response of atomic nuclei to strong magnetic fields.
Essentially, MRI maps the locations of different kinds of atoms along a plane or slice of an object, usually some part of a person's body.

Since an MRI is in a digital format, technicians can easily isolate and select specific structures and types of tissue for further analysis. In this image, the dark areas behind the eyes and nose are the sinus cavities. A doctor may want to study these to identify or diagnose breathing disorders.

Turn on Thresholding, and adjust the range to highlight the sinuses.
回 Close Medical Image.tif and the Threshold window.
When an image is in a digital format, technicians can easily isolate and enhance particular portions of the image, whether it's the image of a heart or the surface of another planet.

## Open Mercury Surface.tif.

In 1974-75, the Mariner 10 spacecraft made three passes by the planet Mercury. This image of Mercury's surface is one of the photos taken by Mariner 10. Interference from radio waves and the sun produced static, or noise, that gives the image a salt-and-pepper appearance. Noise like this can be removed by processing the image with a digital filter that identifies and replaces the noisy pixels with new, more realistic values. Planetary geologists and astronomers frequently use such filtering techniques to enable them to more clearly see features they might not otherwise notice or be able to detect.
$\square$ Use the Rectangular selection tool $\square$ to select part of the image.
Choose Process > Noise > Despeckle to process the image.
20. What happened to the part of the image inside the area you selected?

Close Mercury Surface.tif.
Open Math Image.tif.
Any grid of numbers can be displayed as a digital image. This image was created using a spreadsheet program and shows the equations $\mathrm{xy}=1$ and $x y=-1$. Some mathematical functions show intricate and attractive patterns when displayed as digital images. Artists, scientists, and engineers create interesting and beautiful images using mathematical functions to generate pixel values. The colors in the image depend on the lookup table used.

㩆 Apply various lookup tables to the image, and observe how its appearance changes.
Close Math Image.tif.
Open Painting.tif.
This image is a drawing created by artist Patti Parsons. She selectively changed pixel values from white to a new color using the selection tools as drawing tools. Patti uses the computer as a sketchbook to try out different color combinations before painting her works on canvas. Graphic artists rely heavily on digital imagery to create print and electronic art, advertisements, and video special effects.

## Close Painting.tif.

## Sinuses?

The sinuses are the large, air-filled cavities between the forehead, eyes, nose, and the brain. The air outside the patient is black, so black areas inside the head are likely filled with air.

## Filtering noise, Part II

Another way to filter noise is to choose Process > Filters > Median. Try this method on another part of the Mercury image, and compare the results to the first method you used. Experiment with the Median filter's radius setting to find the radius that gives the best results.

## On your own

- Look at a newspaper photo with a magnifying glass. Describe how the picture is similar to and different from a digital image.
- Design and create your own stadium picture. Use a piece of graph paper to create your design.
- Visit someone in a career that uses digital images. Describe how they use digital images in their job.
$\qquad$


## Pixelated Pictures



Data
Sheet

1. What do you see?
2. What do you notice about the status bar? (Be specific!)
3. Describe what happens to the image when you move each of the four sliders in the $\mathbf{B} \& \mathbf{C}$ Window.
a. Minimum
b. Maximum
c. Brightness
d. Contrast
4. How would the stadium picture look if the fans randomly traded cards with each other?
5. Which number in the status bar changes one unit for every one pixel you move in the horizontal direction?
6. Which item in the status bar shows a pixel's vertical location?
7. Describe how seat and row numbers in a stadium compare to the coordinate system used to locate specific pixels in a digital image.
8. How is the $\mathrm{X}, \mathrm{Y}$ coordinate system in this digital image similar to mathematical graphs you have used?
9. Where is the origin (row 0 , seat 0 ) of the digital image?
10. How could you find out the total number of pixels in this image?
11. How many pixels are in this image?
12. What are the highest and lowest pixel values you can find in this image?
a. highest = $\qquad$
b. lowest = $\qquad$
13. Zoom in on any pixel in your image. Write down its location and value.
a. location: $\mathrm{X}=$ $\qquad$ $\mathrm{Y}=$ $\qquad$
b. value $=$ $\qquad$
14. Has the pixel value changed?
15. What happens to the pixel values when you apply a new lookup table to an image?
16. Describe how the lookup table affects the appearance of a digital image.
17. What Window and Level settings produce the best range of detail in the image?
a. Window $=$ $\qquad$
b. Level = $\qquad$
18. What is the most frequently used pixel value (color) in Mona Lisa?
19. Where in the image are most of the pixels with a value of 227 located?
20. What happened to the part of the image inside the area you selected?

## Lights, Camera, Action! Creating and using image stacks

## Overview

From special effects in movies to weather forecasting to interpreting structures within the brain, digital animation is a technique used in a wide variety of fields. Graphic artists, meteorologists, sports physiologists, and biomedical engineers all use animation in some facet of their work. In this lesson, students learn to create animations using image stacks. They begin by exploring and interacting with a ready-made cartoon animation. They unstack and edit the animation to learn the basics of working with stacks. Next, students construct their own animation.


In the second half of the lesson, students discover that animation provides a means for analyzing motion and visualizing three-dimensional structures. They observe how a hurricane travels and how lightning strikes as they use animation to speed up and slow down processes. Last, they reslice and project magnetic resonance images (MRIs) of the brain to reveal anatomical structures. More advanced stack techniques can be found in the On your own section at the end of the lesson.

## Objectives

Students will:

- create and manipulate digital animations
- use digital animation to analyze motion
- visualize three-dimensional stacked data


## Related Activities

See the "Digitizing Details" lesson for information on how to capture and make movies using scanners and digital video and still cameras.

## Career Links

Graphic arts, meteorology, sports physiology, medical imaging, anatomy, and biomechanics are some of the fields that employ digital animation.


Teaching Notes

## Goal

Students create digital animations, analyze motion, and visualize three-dimensional data.

## IP Skills

- Animation
- Editing stacks
- Creating stacks
- Reslicing and projecting stacks


## ImageJ Skill Sheets

- Stacks \& Animation
- Cutting \& Pasting
- Stacks
- Drawing \& Text
- Managing Windows


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Dog Stack courtesy of Kevin Bowman, a graduate of Wenatchee High School, and his former teacher, John Barnes, chairman of the Wenatchee High School Art Department.
Galloping Horse from Horse in Motion (1878), by Eadward Muybridge, public domain.

AZ Storm courtesy of Mike Charles, Eastern Michigan University, and Charles Hemann, University of Arkansas.

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- See the About... files in the More Stacks folder for acknowledgment of additional contributors.


## Answers

1. Answers will vary. The maximum animation speed depends on the computer. It may be 100 fps or more. The minimum speed is 0.1 fps (one frame every ten seconds).

## 2. There are nine slices in Galloping Horse.tif.

3. Galloping Horse.tif is 292K.
4. Answers will vary. Nine individual images are listed in place of the one stack. The names of all of the open image windows and the memory occupied by each appear at the bottom of the Window menu. The active window is indicated with a checkmark.
5. Each window is 32 K , or roughly $1 / 9$ the size of Galloping Horse.tif.
6. Added slices are inserted before the currently active slice.
7. Only the About Andrew WV.tif image opens and a Log window appears with the following messages:

8. In order to calculate the speed, students would need to measure the distance the hurricane travels over a known time period.
9. Answers will vary. Examples may include photo finishes of horse, car, and foot races or analysis of figure skating.
10. Answers will vary but may include features like the ventricles, sinuses, and cerebellum.

## Lights, Camera, Action! Creating and using image stacks

Have you ever wanted to make a movie? Well, here's your chance! With your imagination and a little help from image processing technology you can easily create digital animations using stacks of images. You can even save them as QuickTime ${ }^{\circledR}$ and AVI movies for use with other applications.

## At the movies

First, let's observe a ready-made animation.
员 Open Galloping Horse.tif.
D Drag the scroll bar at the bottom of the stack window left and right to see the frames, or slices, of the stack.

This series of images was created in 1878 by Eadward Muybridge, an early pioneer in the use of photography to study animal locomotion.

Press the backslash (1) key while holding down Control (Win) or Command (Macs) to start the animation. Click on the image to stop the animation, or use the $\backslash$ key alone.
몽 Experiment with changing the speed of the animation. To change the speed, choose Image > Stacks > Animation Options....

1. What are the fastest and slowest animation speeds in frames per second?

## What is a stack?

A set of images displayed within a single window is called a stack. A stack is similar to a pad of paper or a deck of cards, except that each "card" is a digital image. These images can show many things-a sequence of events, parts of a larger image, or slices through an object at different depths. In fact, ImageJ refers to the individual, "still" images within the stack as slices.

This lesson will focus on the use of stacks for animating a series of images. Perhaps you have created a simple flip-book animation using a stack of paper. When the images are shown quickly, the eye "sees" motion. This is the basic principle that is used for making movies. Films and videos are really just series of still pictures shown in a rapid sequence. By arranging a series of images in a stack and flipping through them, you can create your own animation.

Use the > and < keys or the scroll bar to move forward and backward through the stack as you view the individual slices.

Below the title bar of the Galloping Horse.tif window is a row of numbers. The first number refers to the slice of the stack that you are currently viewing, and the second number refers to the total number of slices in the stack.

## 2. How many slices are there in Galloping Horse.tif?

\& File > Open...

央 Image $>$ Stacks $>$ Animation Options...

## Stack Window



몽 The other numbers below the title bar refer to the image's dimensions (width $\times$ height), bit depth (number of possible colors), and size (amount of computer memory the stack occupies). The size of each open image is also listed next to the image name in the bottom section of the Window menu.
3. How large (in kilobytes, kb) is Galloping Horse.tif?

## Unstacking and restacking

The slices of a stack can be separated into individual windows using the Image > Stacks > Convert Stack to Images command. The images can be reassembled into a stack using the Image > Stacks > Convert Images to Stack command.

Practice unstacking and restacking Galloping Horse.tif.
4. Look at the bottom of the Window menu when Galloping Horse.tif is unstacked. How is it different from when the images are in a stack?
5. How do the sizes of the individual images compare to the size of the stack?

If necessary, reassemble the galloping horse images into a stack.

## Editing a stack

$\square$ To draw on an image, select a line or region by using one of the selection tools ( $\square, 0, \square, \Omega, \square, \Omega$, or $\Theta$ ). Hold down the $\mathbf{F}$ key (for "Fill") while you drag the selection around on the image to use the selection as a paintbrush. Use this drawing technique to make changes to the images in the stack.Try inserting and deleting individual slices in the stack using Image > Stacks > Add Slice and Delete Slice. Experiment with holding down the Alt (Win) or Control (Mac) key while inserting slices and notice where the slice is added.
6. When you add a slice with the Alt or Control key pressed, is the slice added before or after the current slice?

Close Galloping Horse.tif.

## Make your own movie

Try creating your own animation.
$\square$ Choose File > New to create a new blank stack named My Movie. To make a convincing animation, you will probably need at least 12 slices.
[0. On the first slice, draw a simple scene. Be sure to include an object that you want to move in the animation.

Use the > key to advance to the next slice, and draw the next frame of your animation. Use the scroll bar at the bottom of the stack window to move through the stack and test your animation. (See Drawing hints on the next page.)

## Can't see the image size?

If you can't see the size of the image in the window, try stretching the window out a bit.

Creating a new blank stack


Here are the basic techniques you will use:

| To do this... | ...follow these procedures |
| :--- | :--- |
| Make a selection | Use $\square, \square, \boxed{\square}, \boldsymbol{Q}$, or $\boldsymbol{*}$ to select a region of interest. |
| Copy a selection | Choose Edit > Copy. |
| Paste a selection | Choose Edit > Paste. |
| Draw or paint <br> on the image | Make a selection using any of the selection tools. Press <br> the $\mathbf{D}$ key to draw an outline, F to fill the shape, or <br> hold down F while dragging the selection to use the <br> selection shape as a "paintbrush." |
| Add text | Use A. to enter text, then choose Edit > Draw. |
| Change the text <br> or drawing color | Double-click <br> palette. and choose a new color from the |

When you are finished with your animation, press <br>(Control $\backslash$ for Windows and Command $\backslash$ for older Macs) to start the animation. If your movie runs too slow or too fast, choose Image > Stacks > Animation Options to change the speed. (Movies and television both animate at a rate of around 30 frames per second.)

A stack is great for looking at a series of images one after the other. However, if you want to include the stacked images in a printed report, you need a different way to display them. A montage allows you to see all of a stack's slices in a single image.

ㅁ. Choose Image > Stacks > Make Montage to create a montage of your stacked animation. Experiment with the settings in the montage dialog box until you are pleased with the results.

ㅁ If possible, print your montage.
品 Save the montage and/or the stack if you like. Choose the AVI format to save your stack as a movie file that will play on most computers.
모 Close all open images.

## "Seeing" with animation

In addition to its use in the film industry, digital animation is a valuable technique widely used in other fields. Processes that occur too quickly or too slowly for the eye to perceive can be "seen" using animation. Images captured over an extended period of time can be sped up to show changes taking place that might not otherwise be noticeable. For example, meteorologists use animation to follow the movement of clouds, hurricanes, and weather fronts.

國 Choose File > Import > Image Sequence. Navigate to the Hurricane Andrew folder, and open it. In the Open Image Sequence dialog box, select the first file, named About Andrew WV.tif, and click Open.

In the Sequence Options dialog box, accept the default settings, and click $\mathbf{O K}$.
7. Describe what happens.

Close all open image windows and the Log window.

## Drawing hints

You can either draw each frame of your animation from scratch, or modify a copy of the previous slice. To duplicate a slice:

- Choose Edit > Selection > Select All.
- Choose Edit > Copy.
- Use the slider at the bottom of the stack window to advance to the next slice.
- Choose Edit > Paste.


You have just learned an important rule for stacking a set of images-all of the images must be the same size (width and height) in order to stack them.

Choose File > Import > Image Sequence... again, and open
Hurricane Andrew. This time, in the Sequence Options dialog box, enter 2 for the starting image. ImageJ will now skip over the first image (the one that's a different size than the others) and should create a stack from the remaining images in the folder.

In 1992, Hurricane Andrew swept through the Caribbean, Florida, and Louisiana in 72 hours. Using animation to track the movement of hurricanes like Andrew can help provide early warnings for people seeking safety during dangerous storms.
$\square$ Animate the hurricane stack.
8. What measurements would you need to make to be able to calculate the speed of the hurricane?

Close the hurricane stack.
In a similar manner, animation can be used to slow down events that happen too fast for us to see. Sports trainers often use animation to analyze motion such as the swing of a golfer or tennis player. Physicists study the paths of moving objects and the forces that cause them to move using animation. Biomechanical engineers sometimes use animation to help them determine the points of greatest wear when designing running shoes.
$\square$ Import the sequence of images in the Lightning folder.
$\square$ Animate the images.
Carefully observe these images of lightning taken during a typical Arizona summer thunderstorm. This event lasted less than two seconds.
[ Use the animation slider or set the animation speed to move slowly through the stack and observe the behavior of the lightning.
9. Write down another phenomenon that occurs too quickly to "see" with our eyes. Describe how imaging could be used to slow down the event so that it can be analyzed.

Close the Lightning stack.
Animation makes it easier to observe, interpret, and predict patterns.

## Using stacks to show three dimensions

In the field of medicine, animation is often combined with other imaging techniques to provide noninvasive methods for visualizing anatomy.
$\square$ Import the sequence of Brain images in the Makes My Head Spin folder.

Magnetic Resonance Imaging (MRI) is a powerful way to examine the structure of a human body without cutting it open. This tool is especially useful when examining delicate structures like the human brain. An MRI is made by placing a patient in a strong magnetic field, exposing him or her to radio waves, and then recording and analyzing the way the patient's atoms respond. Unlike x-rays, which reveal bone and other hard tissue, MRI excels at creating images of soft tissue. For example, a brain tumor that cannot be detected with an x-ray might be easily identified in an MRI.

直 File > Import >
Image Sequence...

## Wrong image sequence?

When importing an image sequence, the images are added to the stack in alphabetical (or numerical) order.
If the images are not named in the order that they are to be animated, you will have to open them one at a time in the correct sequence, then choose Image > Stacks > Convert Images to Stack to create the animation stack.
Remember to close any windows that are not part of the animation before building the stack.

步 File > Import > Image Sequence...

L File > Import > Image Sequence...

Another advantage of MRI technology is that its images represent very thin slices through the subject, making the exact size and location of features easy to determine. The 27 images in the Makes My Head Spin stack show cross-sections of a human head (with no tumors, fortunately!). Images were made every 5 mm , starting from the middle of the nose and continuing up through the eyes.

## Animating the brain

## Animate the Brain stack.

When you animate the stack, you take a trip through the brain from the nose to the top of the head. Although each individual slice has only two dimensions, the stack of slices gives us a three dimensional view.

## Reslicing the brain

ImageJ can use data from the individual slices in a stack to create new views of an object. Imagine cutting straight down through the brain stack and looking at the cut from the side. This is called reslicing.Use - to make a straight line selection across the brain image in any direction.
[ Choose Image > Stacks > Reslice. Set the Input Z Spacing to 5, and click $\mathbf{O K}$. (For comparison, try setting the spacing to $\mathbf{3}$.)
[ Reslice the stack along other straight line selections drawn at various angles, and experiment with the options in the Reslice dialog box.
10. What structures can you see within the brain? Use the diagram at right or a textbook to help you identify and describe a few of them.

## Projecting the brain

Imagine that each slice of the brain stack was printed on a sheet of clear plastic and you held the stack of plastic sheets in your hand. By tilting and rotating the stack of images, you could view them from different angles. ImageJ can create a series of images that show how a stack would appear as it is rotated around any of three axes.

Close all of the resliced images, but leave the brain stack open.
ㅁ. Choose Edit > Selection > Select None to make sure there are no selections on the brain stack.

ㄷ. Choose Image > Stacks > 3D Project to project the stack. Fill in the 3D Projection dialog box as shown at right. Don't worry for now what all of these settings mean. They were chosen to allow you to look "through" the hard bone tissue of the skull at the softer, fainter brain tissue. Click OK.

ㅁ. To increase the contrast of the images, choose Image > Adjust > Brightness/Contrast, and click the Auto button.
$\square$ Click the Apply button, then click Yes when asked if you want to adjust the contrast of all of the slices in the stack.
므․ Close the $\mathbf{B} \& \mathbf{C}$ window.
品 Animate the completed projection stack.

## Upside-down?

Your resliced brain image will be upside-down with this procedure. To make it right-side up, check the Flip Vertically box.

## Major structures of the brain



## On your own

Use the images in the On Your Own folder to try out the following techniques and to explore stacks and animation techniques．

## Making a stack from a montage

Earlier in this lesson you turned a stack into a single image called a montage． You can also go the other direction，turning a montage into a stack．

㖘 Close all open images，and open 1 Running Horse．tif．
$\square$ Click $\square$ ，and select the first frame of the running horse．
［Choose Image＞Duplicate to duplicate the selection into its own window．Click OK．Drag the new window out of the way．
Choose 1 Running Horse．tif from the Windows menu to activate the source image again．
$\square$ Drag the selection outline you made to the second running horse frame．

回 Duplicate this selection．Repeat this process until you have made a separate image from each of the nine running horse frames．Close the Running Horse montage image．
Stack and animate the running horse frames．Close the stack．

## Registering slices in a stack

The Rocket Stack shows a model rocket being launched．Unfortunately， the camera moved，making it difficult to analyze the motion of the rocket． Philippe Thevenaz，a researcher of the Biomedical Imaging Group of the Swiss Federal Institute of Technology，created an amazing ImageJ add－on that automates the process of registering the images in a stack．

O Open the $\mathbf{2}$ Rocket Stack．tif image．
$\square$ Choose Image＞Duplicate to make a copy of the first slice of the stack（not the whole stack，though it will still work if you do．）

品 Choose Plugins＞TurboReg＞TurboReg．
国 Use the default settings，and select the Accurate option．


Click the Batch button．
The plugin takes over and registers the stack！
$\square$ Animate the registered rocket stack．To compare the stacks before and after the registration process，you can click on one of the original rocket stack copies and animate it．

Image registration－correcting for differences in position of features in two or more images due to camera motion and other effects．

Note to Mac OS 9 users－
TurboReg is only available for Windows and Mac OSX．
$\qquad$

## Lights, Camera, Action!



Data Sheet

1. What are the fastest and slowest animation speeds in frames per second?
a. fastest $=$ $\qquad$ fps (frames per second)
b. slowest = $\qquad$ fps (frames per second)
2. How many slices are there in Galloping Horse.tit?
3. How large (in kilobytes, kb) is Galloping Horse.tit?
4. Look at the bottom of the Window menu when Galloping Horse is unstacked. How is it different from when the images are in a stack?
5. How do the sizes of the individual images compare to the size of the stack?
6. When you add a slice with the Alt or Control key pressed, is the slice added or before or after the current slice?
7. Describe what happens.
8. What measurements would you need to make to be able to calculate the speed of the hurricane?
9. What do you notice about the path the electricity travels?
10. What structures can you see within the brain? Use a textbook to help you identify and describe a few of them.

# Digital Detective Investigating lookup tables 

## Overview

This lesson explores techniques of manipulating image lookup tables and introduces the use of digital images in forensic science. In Part 1 of the lesson, students explore methods for enhancing low-contrast images to reveal important details. In Part 2, they measure angles and calculate ratios to compare handwriting samples. In Part 3, students enhance, classify, and compare fingerprints in search of a burglary suspect. In Part 4, students enhance and measure images of hair and tire tracks to look for matches to crime scene evidence.

## Objectives

Students will:

- manipulate lookup tables to reveal hidden evidence in digital images
- measure angles and letter heights and calculate proportions to compare and identify signatures
- enhance, classify, and compare fingerprints
- enhance and measure images of trace evidence to solve crimes


## Related Activities

- See "Pixelated Pictures" for an introduction to digital image processing.


## Career Links

Digital images of fingerprints have been compiled into Automated Fingerprint Identification Systems (AFIS) in many states. Police can submit fingerprints from a crime scene to the AFIS, and it will search the database for a match. Banks also use digital images of signatures to aid forgery detection. Some banks are requiring thumbprints from patrons before cashing checks. More recently, some states have begun cataloging entire palm prints to help improve their ability to solve crimes.

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Images courtesy of the authors.


Teaching Notes

## Goal

Students process crime scene evidence to reveal hidden clues and solve crimes.

## IP Skills

- Manipulating lookup tables
- Pasting selections using Paste Control
- Measuring angles


## ImageJ Skill Sheets

- Lookup Tables
- Cutting \& Pasting
- Measuring
- Drawing \& Text


## Teaching tips

There are three additional handwriting analysis problems in the $\mathbf{0 7}$ More Signatures folder.
Challenge your more advanced students or those who finish their work quickly to see how many different ways they can find to reveal the hidden information in the images.

## References

Kaye, Brian H. (1995) Science and the Detective. VCH Publishers, Inc. New York, NY.

Deedrick, Douglas W. (2000) "Hairs, Fibers, Crime, and Evidence." Forensic Science Communications. July 2000 Volume 2 Number 3. Trace Evidence Unit, FBI. (http:>>www.fbi.gov>hq>lab>fsc>backissue> july2000>deedric1.htm)

## Answers

1. $\$ 215,000.00$ was deposited in account number 4490-60695-004.
2. Techniques that reveal the information include, but are not limited to:
a. Adjust Image > Adjust > Brightness/Contrast so that the Minimum value is 245 or above (see hint at right).
b. Choose Process > Enhance Contrast, and set Saturated Pixels to $3 \%$ or more.
c. Choose Image > Adjust > Window/Level, and set the Level to 251 and the Window to 15 or lower.
d. Choose Image > Adjust > Threshold, and set the Minimum to 0-3 and the Maximum to 250.
e. Choose Process > Sharpen several times.
3. Descriptions will vary. The suspect is a white male in his thirties, wearing jeans and a three-tone jacket. Techniques include, but are not limited to, those described in 2a through 2d above.
4. The license plate number is JKP 154. (Minimum = 0, Maximum = 0 in B\&C window.)
5. Techniques that reveal the information include but are not limited to:
a. Choose Image $>$ Adjust $>$ Brightness/Contrast, and set Minimum $=$ 0 and Maximum $=0$.
b. Choose Image > Adjust > Window/Level, and set Level = 23 and Window $=52$.
c. Choose Image > Adjust > Threshold, and set the Minimum = 1 and the Maximum = 70-255.
d. Choose Image > Lookup Tables.... Several of the available color tables faintly reveal the license plate.
6. The original date was April 13, 1985. Techniques used to reveal the date include but are not limited to:
a. Choose Image $>$ Adjust $>$ Brightness/Contrast and set Minimum $=$ 244 and Maximum $=244$.
b. Choose Image > Adjust > Window/Level, and set Level = 255 and Window $=25$.
c. Choose Image > Adjust > Threshold, and set the Minimum =0 and the Maximum = 244.
7. The Blend and And transfer modes make the pasted signature transparent.

Hint: In question 1, the receipt is readable when the brightness and contrast Minimum is set at 245 or higher, and the Maximum is set at 255.

8. The lowercase " a " and the uppercase " S " share similar shapes and styles between the signatures. The lowercase " $a$ " makes a stronger case for Sarah Black as an alias of Susan Jones since there are more of them to compare.
9. Baseline angle $\approx 258^{\circ}$ (or $179^{\circ}$ ).
10. Answers will vary, depending upon the letters chosen for measuring. The following angles were measured for the letters indicated in parentheses.

Table 1

| Letter <br> Number | Letter Angle |  |
| :---: | :---: | :---: |
|  | Sample <br> Signatures | Questioned <br> Signature |
| $\mathbf{1}$ | $68(\mathrm{a})$ | $90(\mathrm{a})$ |
| $\mathbf{2}$ | $84(\mathrm{a})$ | $98(\mathrm{r})$ |
| $\mathbf{3}$ | $65(\mathrm{a})$ | $85(\mathrm{~h})$ |
| $\mathbf{4}$ | $69(\mathrm{a})$ | $90(\mathrm{~B})$ |
| $\mathbf{5}$ | $79(\mathrm{a})$ | $81(\mathrm{l})$ |
| $\mathbf{6}$ | $74(\mathrm{a})$ | $76(\mathrm{a})$ |
| $\mathbf{7}$ | $72(\mathrm{a})$ | $83(\mathrm{k})$ |
| Average angle | $73^{\circ}$ | $86^{\circ}$ |

11. As long as the same letters or sets of letters are compared, the angles of slants between the two signatures should be very close. Average angle slants for the lowercase "a" could range between 70 and 90 degrees, depending upon the number of letters averaged for the calculation.
12-16.
Answers will vary, depending upon the letters chosen for measuring. The following letter heights are examples. Ratios should be similar for the sample and questioned signatures.

Table 2

| Sample letters for <br> measurement | Letter height (pixels) |  |  |
| :---: | :---: | :---: | :---: |
|  | Sample <br> signatures | Questioned <br> signature |  |
| Uppercase | Letter 1 | 20 | 19 |
|  | Letter 2 | 17 | 18 |
|  | Letter 3 | 18 |  |
|  | Letter 4 | 23 |  |
|  | Average | 19.5 | 18.5 |
| Lowercase | Letter 1 | 13 | 13 |
|  | Letter 2 | 13 | 12 |
|  | Letter 3 | 12 | 12 |
|  | Letter 4 | 11 | 13 |
|  | Average | 12.3 | 12.5 |
| Ratio (Upper>Lower) |  |  |  |

17. The ratios are close, though not exact, for the questioned and sample signatures.
18. Answers will vary. However, students will be most likely to choose either Dan, Jane, Mark, or Beth since these fingerprints have the same patterns.
19. John and Susan can be eliminated at this point since John's fingerprint pattern is a whorl and Susan's is an arch. The crime scene print was left by someone with a loop pattern.
20. Beth was probably at the crime scene. A comparison of her fingerprint ridge count and marked bifurcations to that of the print obtained from the crime scene print shows a very close match.
21. Carla's hair is similar in color and texture to that of the crime scene hair. Both her medulla and the crime scene medulla are fragmentary. Furthermore, the ratio of the diameter of the hair to the diameter of the medulla is similar for Carla and the crime scene hair. Carla's is 14.2 while the crime scene hair is 13.8. (Ratios will vary according to where measurements are taken along the length of the hair.)
22. Mary's tire tread (Tread 2) matches that of the crime scene tread. Therefore, her car was most likely to have been at the scene of the crime.

## Digital Detective Investigating lookup tables



Lesson

Forensic Science-the science of interpreting or establishing the facts in civil or criminal law cases.

## Process > Enhance Contrast tip

For best results, experiment with different percentages of Saturated Pixels.


A faint receipt from an ATM (Automatic Teller Machine) was found at the home of a suspected burglar. Unable to read the account number or the amount of the deposit, you scanned the receipt to convert it into a digital image. Use the techniques described earlier to reveal this information.

1. What is the account number? How much was deposited?
2. Describe the steps you used to reveal the information.

## ㅁ⿹ㄹ Close 01 ATM Receipt.tif.

## Open 02 Surveillance Camera.tif.

You've asked the bank for photos of anyone using the account number above. They were able to provide one picture, but the quality is poor.
$\square$ Enhance the image to bring out the details.
3. Describe the suspect and how you were able to see him or her.

Close 02 Surveillance Camera.tif.
Q Open 03 License Plate.tif.
The license plate of this car was caught on camera as the suspect was fleeing from a burglary. You believe your burglar has struck again.

E Enhance this image to reveal the license plate number of the car.
4. What is the license plate number?
5. Describe the method you used to reveal the plate number.

Close 03 License Plate.tif.

## Part 2: Examining documents

Documents are another major focus of forensic science. Examiners determine the authenticity of written materials and look for signs of alteration or forgery. They determine the age of documents, examine the printing or writing, and even analyze the paper and ink itself. They work with typewritten, photocopied, and handwritten materials ranging from personal correspondence to legal documents. Validating signatures is an important part of document examination.

## Open 04 Dated Will.tif.

At first, this will appears to be genuine and unaltered. However, you believe it is actually an older will, and that someone changed the date to make it seem to be the most recent version.
6. Can you show that the date has been changed? Describe your technique.

## Close 04 Dated Will.tif.

A woman named Susan Jones has just been arrested for suspected fraud. She is accused of forging checks using several aliases, one of which is Sarah Black. As the document examiner in charge of the case, you have asked Susan to submit a handwriting sample to you. Even though no one signs his or her name exactly the same way each time, there will be many similarities between signatures from the same person.

Open 05 Sample Signatures.tif and 06 Questioned
Signature.tif.

Arrange the images so you can see both of them at the same time．
The questioned signature has faded from exposure to the sun．
包 Activate each image，and enhance it to make the writing as dark and clear as possible．If you enhance the image contrast using Image／Adjust／Brightness／Contrast，be sure to click the Apply button in the $\mathbf{B \& C}$ window before continuing．

Now compare the questioned signature to the sample signature using the following overlay technique．Watch for similarities in the strokes of letters．

## Making transparent overlays

Activate the 06 Questioned Signature．tif image，click $\square$ ，and select a rectangle around the signature．
ㅁopy the selection，activate $\mathbf{0 5}$ Sample Signatures．tif，then paste the selection on top to create a＂floating＂copy of the questioned signature．
Q Open the Paste Control，and try out different transfer modes to find one that makes the pasted signature transparent while allowing you to see the sample signatures beneath it．

7．Which transfer modes make the pasted signature transparent？
Drag the questioned signature over the sample signatures，and look for similarities between letters．

8．What letter or letters share a similar shape and style between the two signatures？
圆 Choose Edit＞Undo to remove the pasted signature．
You will be presenting your evidence to a jury，so you need to gather additional data for comparing the signatures．Document examiners often characterize signatures by measuring the angle of slant and calculating the ratio of uppercase to lowercase letter sizes．For the best results，average the data from at least four of the sample signatures．

## Calculating the average slant angle

Before you can measure the slant angle of the letters in the signature，you need to be sure the baseline－the imaginary line that touches the bottoms of the letters－is horizontal．The questioned signature is tilted slightly．

回 Activate the $\mathbf{0 6}$ Questioned Signature．tif image．
品 Click - then drag a straight line from left to right that touches the bottoms of most of the letters．As you move the line，its angle is displayed in the status bar of the ImageJ window．You can adjust the line after you select it by dragging the small handles at the ends or in the middle．

9．What is the angle of the baseline you selected？
员 Choose Edit＞Selection＞Select All to select the entire image．
믕 Choose Image＞Rotate＞Arbitrarily，and enter the angle of the baseline you recorded．Check the Interpolate option，and click OK．The signature should now be horizontal．（If not，press the R key to revert to the original image and repeat the measurement and rotation process．）

L Image＞Adjust＞ Brightness／Contrast

```
4 Edit > Copy
L Edit > Paste
```

L Edit＞Paste Control．．．

## Paste Control tip

Do not click on the image outside the area you pasted or it will become a permanent part of the image，and you will not be able to reposition it．If this happens， immediately choose Edit＞Undo and repeat the select，copy，paste process．


Use again to measure the slant of several letters in both the sample signatures and the questioned signature. To measure consistently, drag each line selection from the baseline up.
10. Record your angle measurements on your Data Sheet. When you have finished the angle measurements for each signature, calculate and record the average angle for each signature in the table.
11. How does the average slant of the letters in the questioned signature compare to the average slant in the sample signatures?

## Calculating the uppercase-to-lowercase

## letter height ratio

To calculate a ratio of uppercase to lowercase letter sizes, you will measure the heights of the first two letters of the first and last names. Divide the average uppercase height by the average lowercase height to obtain the ratio. Use this technique for both the sample signatures and the questioned signature. (Note: With the questioned signature you will only have two uppercase and two lowercase measurements.)

든 Click $\mathbb{Q}$, then click on the images to magnify both to $200 \%$ or larger to make the letters easier to measure.
믕 Click and hold down the Shift key to make a straight vertical selection from the top to the bottom of each letter. The length of the line is displayed in the status bar of the ImageJ window as you drag the line selection. Record the height of each letter on your Data Sheet.
12. Record the heights of the uppercase letters of the sample signatures and the questioned signature on your Data Sheet.
13. Calculate the average uppercase heights for the sample signatures and the questioned signature, and record them on your Data Sheet.
圆 Repeat the procedure to measure and record the heights of the lowercase letters on your Data Sheet.
14. Record the heights of the lowercase letters of the sample signatures and the questioned signature on your Data Sheet.
15. Calculate the average lowercase heights, and record them on your Data Sheet.
16. Divide the average uppercase height by the average lowercase height for the sample signatures and the questioned signature, and record them on your Data Sheet. This is the letter height ratio.
17. How does the ratio calculated for the questioned signature compare to that for the sample signatures?

Close all images when finished.

## You solve it!

Open any of the cases in the $\mathbf{0 7}$ More Signatures folder, and use the identification techniques of this lesson to solve the crime.


## Part 3: Latent prints (fingerprints)

Latent fingerprints are one of the most common forms of forensic evidence collected at crime scenes. Since every fingerprint is unique, a suspect's fingerprint found at the scene of a crime is an undeniable way to prove the person was there at some time.

## Open 08 Fingerprints.tif.

The set of fingerprints you are looking at was photographed, cut out, pasted onto a card, and then scanned into the computer. The crime scene fingerprint comes from a glass display case at a jewelry store that was burglarized. You also have fingerprints of several suspects.
18. Whose fingerprint do you think the crime scene print most closely resembles?

It is difficult to see details in these fingerprints, so you will use larger versions that have been scanned at higher resolution.

Close 08 Fingerprints.tif.

- Open all the images in the 09 Large Fingerprints folder. (Click Window > Tile to spread them out.)
$\square$ Enhance each fingerprint to obtain the sharpest, most detailed image possible.
One way to begin eliminating suspects is to classify each fingerprint by type. You may want to move the windows around to place fingerprints side by side for comparison.


## Open 10 Fingerprint Types.tif.

Fingerprints are classified into three main types: arches, loops, and whorls.
员 Classify each suspect fingerprint by type. Click A. to label each suspect image with its fingerprint type. Remember to click on the image using the text tool, type the name, then press Ctrl-D (Opt-D on Mac) to print the name on the image. (Double-click to open the Color Picker and change the color of the text.)
19. Which suspects can you eliminate based on the fingerprint type?

Close the fingerprint images of the suspects you have eliminated.
At this point, you may think you know who committed the burglary. To help you make a positive identification, you need to gather more data from the fingerprints. You will mark and label key features and calculate ridge counts.

Open 11 Fingerprint Info.tif. Use the < and > keys or the slider control to move through the slices of the stack as instructed.

When evidence is presented to a jury, labeled diagrams of fingerprints are used. The key features of a fingerprint that used to characterize it include the core, the delta, the ridge count, and several points of comparison.

Move to the second slice of the stack.
The core is the central point in the fingerprint pattern, and the lines are known as ridges. The delta is the point closest to the core where the ridges diverge or pull apart. Cores and deltas are unique features of loop- and whorl-type fingerprints, but arches lack them.

## Latent?

The word "latent" means present but not evident. A latent fingerprint is a fingerprint that is not apparent to the eye, but can be made visible by dusting or some other method to make it identifiable.

## Ten types of fingerprints?

The 10 in the file name means it is the tenth image you open, not that there are ten types of fingerprint patterns! There are only three main types of fingerprint patterns.


Move to the third slice of the stack.
The number of ridges crossed by a line drawn between the core and the delta is called the ridge count.

Move to the fourth slice of the stack.
In a fingerprint, points where ridge lines split are known as bifurcations. Any of these can be marked, as they are unique to the fingerprint. Typically, 5-10 points of comparison are selected for each fingerprint. The legal number of points required to confirm a match vary from state to state.

Label each fingerprint in the manner shown on the fifth slice of the stack. To make the dots, click to use the Point Selection tool, and then click on the image. Click $\square$ to draw the lines and A to insert text labels. Remember to press Ctrl-D (Opt-D for Macs) to "lock" the point or lines on the image!
20. Based on your analysis, which suspect was definitely at the scene of the crime?Close all the fingerprint images.

## Part 4: Trace evidence

Trace evidence includes many kinds of material left at crime scenes, including footprints, tire tracks, hair, blood, bullet markings, clothing, and bodily fluids.

## Hair and fibers

Open all the images in the $\mathbf{1 2}$ Hair folder. (Choose Window > Tile to spread the images out.)
These hair samples were gathered from friends and associates of a murder victim. Each sample is magnified about 400 times actual size.
$\square$ Choose Window > Hair Structure.tif to activate the hair structure image and familiarize yourself with the terminology and criteria for categorizing and comparing hair specimens.

The central core of a hair, the medulla, is made of large cells that are often separated by air spaces. When viewed under a microscope, the medulla is often clearly visible. You will use the features of the medulla to help you describe each hair and try to find a match to the crime scene hair.

Enhance each image to show the most detail.
Label each hair with its medulla classification: none, fragmentary, discontinuous, or continuous.
ㄷompare the suspects' hair to the hair from the crime scene. Close the hair images from the suspects you eliminate.
莡 When you have narrowed down the list of suspects to a few, measure the diameters of the remaining hairs and the diameters of their medullas.
Calculate ratios by dividing the diameter of each hair by the diameter of its medulla.
21. Who was at the murder scene, and what evidence do you have to support your claim?

## Hair structure

Each strand of hair contains three distinct layers. From the outermost layer inward:

- The cuticle is a single layer of scales that interlock with the cells of the hair's inner root sheath to firmly anchor the hair in the follicle.
- The cortex is composed of keratinized cells that are tightly bound around each other. These bands provide the hair with strength.
- The medulla consists of large, loosely connected cells with air spaces between them. By reflecting light, these air spaces help to determine the sheen and color tones of the hair.


Cuticle of a normal hair shaft.Close all the hair images when you are finished.

## Tire tracks

뭉 Open all the images in the $\mathbf{1 3}$ Tire Tracks folder.
Automobile tires also have characteristic patterns that help to determine whether or not a particular vehicle was at the crime scene. The three Tire Tread images show the tire tread patterns from the cars of three suspects, and the Crime Scene Track image is a photograph taken of a track left on a driveway at the crime scene. The lighting was poor when the picture was taken, so the track pattern is nearly impossible to see.

Process the Crime Scene Track image to obtain the clearest possible view of the tread pattern, and compare it to the tire patterns from the three suspects' vehicles.
22. Based on your examination of the evidence, which suspect's vehicle was at the scene of the crime?

등 Close all the tire track images when you are finished.

## On your own

Create your own "forensic evidence" for an imaginary crime. Write a short story describing the crime, the suspects, and any available clues. Challenge others to use image processing techniques to help solve the crime.
$\qquad$
$\qquad$

## Digital Detective



Data
Sheet

1. What is the account number? How much was deposited?
2. Describe the steps you used to reveal the information.
3. Describe the suspect and how you were able to see him or her.
4. What is the license plate number?
5. Describe the method you used to reveal the plate number.
6. Can you show that the date has been changed? Describe your technique.
7. Which transfer modes make the pasted signature transparent?
8. What letter or letters share a similar shape and style between the two signatures?
9. What is the angle of the baseline you selected?
10. Record your angle measurements in the table below. When you have finished the angle measurements for each signature, calculate and record the average angle for each signature in Table 1.

Table 1

| Letter <br> Number | Letter Angle |  |
| :---: | :---: | :---: |
|  | Sample <br> Signatures | Questioned <br> Signature |
| 1 |  |  |
| 2 |  |  |
| 3 |  |  |
| 4 |  |  |
| 5 |  |  |
| 6 |  |  |
| 7 |  |  |
| Average <br> angle |  |  |

11. How does the average slant of the letters in the questioned signature compare to the average slant in the sample signatures?
12. Record the heights of the uppercase letters of the sample signatures and the questioned signature in Table 2.

Table 2

| Sample letters for <br> measurement | Letter height (pixels) |  |  |
| :---: | :---: | :---: | :---: |
|  | Sample <br> signatures | Questioned <br> signature |  |
|  | Letter 1 |  |  |
|  | Letter 2 |  |  |
|  | Letter 3 |  |  |
|  | Letter 4 |  |  |
|  | Average |  |  |
| Lowercase | Letter 1 |  |  |
|  | Letter 2 |  |  |
|  | Letter 3 |  |  |
|  | Letter 4 |  |  |
|  | Average |  |  |
| Ratio (Upper/Lower) |  |  |  |

13. Calculate the average uppercase heights for the sample signatures and the questioned signature, and record them in Table 2. (Divide average uppercase height by average lowercase height.)
14. Record the heights of the lowercase letters of the sample signatures and the questioned signature in Table 2.
15. Calculate the average lowercase heights, and record them in Table 2.
16. Divide the average uppercase height by the average lowercase height for the sample signatures and the questioned signature, and record them in Table 2. This is the letter height ratio.
17. How does the ratio calculated for the questioned signature compare to that for the sample signatures?
18. Whose fingerprint do you think the crime scene print most closely resembles?
19. Which suspects can you eliminate based on the fingerprint type?
20. Based on your analysis, which suspect was definitely at the scene of the crime?
21. Who was at the murder scene, and what evidence do you have to support your claim?
22. Based on your examination of the evidence, which suspect's vehicle was at the scene of the crime?

# More Than Numbers Graphing digital images 

## Overview

Interpreting digital images based on only their appearance can be misleading. Two- and three-dimensional graphs of pixel values can aid in their interpretation and analysis. In this lesson, students create histograms, plot profiles, and surface plots to help them understand the data making up digital images. Histograms show the number of pixels of each value in an image. Density plot profiles graph the pixel values along a given line, and surface plots display pixel values as a three-dimensional projection.

In Part 1, students investigate a Digital Elevation Model (DEM) of a section of Colorado. First, they discover the most frequently occurring elevation value using the histogram function. Next, they create density plot profiles and surface plots to help them visualize the shape of the land. They find that the view from which a surface plot is created affects the manner in which the data are displayed. After gaining a basic understanding of histograms, plot profiles, and surface plots, students use these graphs in Part 2 to help interpret surface sea temperatures and tree ring growth. A comparison of a profile plot for a photographic image of a crater to one for an elevation image of a crater helps to solidify the idea that the information a graph provides is directly dependent upon the data present in the image. In Part 3 students explore synthetic images as a means of visualizing mathematical relationships. In the On Your Own section, students learn how to density calibrate images.

## Objectives

Students will:

- interpret a Digital Elevation Model (DEM) using a histogram, density profile plots, and surface plots
- use the appropriate two- or three-dimensional graph to interpret digital data
- explain the numerical patterns in image data via analysis of histograms, density profile plots, and surface plots
- compare and contrast histograms, density profile plots, and surface plots to evaluate their usefulness for graphing image data


## Related Activities

- See "Pixelated Pictures" to gain an understanding of digital images.
- Refer to the lesson "Photo Effects" to discover how histograms can be used to help with the restoration of old photographs.


Teaching Notes

## Goal

Students create histograms, plot profiles, and surface plots to help them interpret the data contained in digital images.

## IP Skills

- Interpreting histograms
- Interpreting density profile plots
- Interpreting surface plots


## ImageJ Skill Sheets

- Selecting
- Lookup Tables
- Calibrating Images
- Managing Windows


## Vocabulary

- histogram
- calibration


## Career Links

Digital Elevation Models (DEMs) are used in a wide variety of fields such as civil engineering, natural resource management, landscape architecture, space science, and geology. Density plot profiles and surface plots make it possible to see quickly the data of DEMs. Photography is another field in which digital graphs can play a role. Histograms provide useful information about the distribution of light and dark pixels in an image. These data help photographers digitally enhance images.

## Notes

All of the images used in this lesson are calibrated. See the Calibrating Images ImageJ Skill Sheet for more information about image calibration.

Students can make images from any rectangular array of measurements or numbers. The values do not have to be in the range of 0-255. Importing the data as text causes the values to be read in and automatically scaled to the range of 0-255. Measurement results or mathematical functions can be entered into spreadsheets, exported as text, and imported as images.

## Acknowledgments

Lesson developed by Carla McAuliffe, Center for Image Processing in Education, Tucson, Arizona. Updated for use with ImageJ by Larry Kendall.

Parts of this lesson are based on "The Plot Thickens," developed by LuAnn Dahlman, "Picture This...," developed by LuAnn Dahlman and Larry Kendall, and "The Snake \& the Rat," developed by Denice Warren, all of the Image Processing for Teaching project, University of Arizona.

Images courtesy of the University of Arizona, NOAA, USGS, and the authors.

## Answers

Note: Several of these questions ask students to speculate, predict, or describe their observations. While there are no "right" answers to these questions, students should demonstrate reasonable effort in their responses.

1. Answers will vary. Students may describe shades of gray and shapes.
2. Answers will vary, depending upon the students' focus. Students will often guess it is some kind of X-ray.
3. Answers will vary. (Pixel values represent elevation.)
4. Answers will vary. The highest elevation is 14,433 feet (raw value $=$ 244 ), and the lowest elevation is 3,350 feet (raw value $=1$ ).
5. 1,588 pixels have a value of 8,002 feet.
6. The highest elevation is 14,433 feet, and the lowest elevation is 3,350 feet.
7. The modal elevation in the DEM is 3,532 feet (frequency $=4,041$ ).
8. The mean elevation is 6,979 feet.
9. The areas of highest elevation are located at the bottom center of the image.
10. Answers will vary, depending upon where the transect was drawn on the image. Elevations can be as high as 14,433 feet or as low as 3,350 feet.
11. Answers will vary. Plots with fixed Y-axis scale can be compared with each other.
12. Answers will vary. Students may describe dips, peaks, and flat spots along the transect, indicating valleys, mountains, and plains or plateaus.
13. Predictions will depend on students' ability to visualize 3-D structures.
14. The land is high and mountainous on the left side of the image and then slopes gently toward the right. Two drainage basins are visible in the foothills area. The Rocky Mountains cover the western two-thirds of the image.
15. The face of the mountain front and the two valleys in the foothills area can be seen more readily in this view.
16. More detail is revealed with a surface plot from small rectangular selections.
17. The parts of the image with a calibrated value of $-3^{\circ} \mathrm{C}$. are land and ice. This is a pre-set value and not the actual surface temperature.
18. The sea surface temperature that occurs most often is $27^{\circ} \mathrm{C}$.
19. Answers will vary. Lines drawn from pole to pole across the equator will show temperatures to be warmest at the equator and coolest at the poles.
20. The peaks represent the light-colored wood, and the dips represent the darker wood. Together, each consecutive set of light and dark wood represents one annual growth ring.
21. Wider sets of annual growth rings represent relatively good growing conditions ("wet" years), and the narrower rings represent years of less growth (possibly drought years).
22. Predictions will vary depending upon whether students recognize the difference between brightness and elevation values.
23. Student should compare prediction and actual plot.
24. You cannot readily see the crater because the graph shows the brightness of the surface, not the elevation.
25. The long, flat section of the profile is the bottom of the lake.
26. Answers will vary. The stepped ramp image has vertical bars of distinct shades of gray, while the smooth ramp has continuously changing shades of gray.
27. Histograms show the number of pixels of each value in an image. Each line in the stepped ramp histogram corresponds with a vertical bar of color. With the smooth ramp the histogram shows that there are equal numbers of pixels of many shades in the image.
28. Answers will vary. The smooth ramp image produces a smooth rampshaped structure, while the stepped ramp gives a plot that looks like a staircase.
29. A surface plot shows the combined result of multiple density plot profiles. A surface plot enables one to view the entire pattern of numbers, while a single profile plot can be very useful for revealing discrete, smaller scale trends.
30. 


31. The snake's body temperature appears to change the most due to the heat lamp.
32. Use the area selection tools to sample and measure the surface temperature of the rat and the snake before and after they were exposed to the heat lamp. Use this information to complete Table 1.

Table 1

| Animal | Body Surface Temperature $\left({ }^{\circ} \mathrm{C}\right)$ |  |
| :--- | :---: | :---: |
|  | Before Heat Lamp | After Heat Lamp |
| Rat | 36.9 | 38.0 |
| Snake | 22.8 | 25.7 |

33. Answers will vary. Mammals maintain a body temperature within a fairly narrow range regardless of the air temperature, while reptiles' body temperature varies with air temperature.

# More Than Numbers Graphing digital images 



Lesson

## Part 1: "See" the numbers

As you look at an image, your eyes give your brain clues about the shape, environment, and meaning of objects in the image. However, some of the data contained within an image may be difficult for your eyes to detect or interpret effectively. Graphing image data can help you obtain useful information you might not otherwise be able to see. In this lesson, you will create and interpret graphs of the numbers that make up digital images.

## Open 1 Colorado DEM.tif.

Many digital images are like photographs; the pixel values in the image are directly related to the brightness in the original scene. In some images, however, the pixel values represent measurements other than brightness.

回 Choose Analyze > Set Measurements..., and set the number of decimal places to zero. This will simplify things by displaying all measurements as whole numbers.

1. Look carefully at this image. Then, describe what you see.
2. What do you think this image shows?

Move your cursor across the image, and read the pixel values in the status bar of the ImageJ window.
3. What do you think the pixel values in the image represent?

This image, called a Digital Elevation Model, or DEM, displays a regular grid of elevation measurements for a section of Earth's surface. It is a type of digital map that is similar to a topographic map since it allows you to read elevation data. The data from a DEM can be used to create maps that show the locations of animal communities, soils, and vegetation types. This particular DEM image shows the state of Colorado.

Move your cursor around the image again as you observe the values in the status bar.

This DEM is an 8-bit image with possible pixel values ranging from 0 to 255 . These values are converted to elevation using an equation that was saved with the image. In the status bar, the calibrated pixel value is followed by the raw, uncalibrated pixel value in parentheses.
4. What are the highest and lowest elevations you can find? What raw pixel values correspond to each elevation?
Rather than simply examining this image with your eyes, you can create a graph to help you "see" the numbers-in this case, the elevation data of the image. A histogram is a graph that shows the frequency distribution of values in the image-in other words, it shows the number of pixels there are for each different value in the image.

[^2]\& File > Open

## The status bar

The status bar is located below the tool bar in the ImageJ window.


## Calibrated and uncalibrated images

In uncalibrated (raw) images, the pixel values are proportional to some measurement but are not measurement values themselves. They have meaning relative to the other pixel values in that image but don't tell you what the actual measurement was at each point in the image.

In calibrated images, the pixel values are converted to approximate measurements using an equation. For this image, the calibration equation is:
elevation $=45.6 \times$ raw value $+3,304.4$
(To see the calibration equation, choose Image > Show Info...)
$\underset{L}{d}$ Analyze > Histogram
$\square$ Move your cursor across the histogram plot, and read the Value (calibrated pixel value, or elevation) and Count (number of pixels) in the bottom part of the histogram window.
5. How many pixels in the image represent an elevation of 8,002 feet?
6. According to the histogram, what are the highest and lowest elevations in the image? (Reported as Max and Min in the Histogram window.)
7. What elevation occurs most often in this DEM? (Reported as the Mode in the histogram window.)
8. What is the average (Mean) elevation in the DEM?

Sometimes, it is useful to produce a detailed list of the information in the histogram.
$\square$ Click the List button in the Histogram window. Scroll down the histogram list, and examine the numbers. Double-check your answer to Question 5.

The histogram list shows the raw pixel values (level), the calibrated pixel value (value), and the frequency (count). This list can be saved or copied and pasted into a spreadsheet for further analysis.

Close the Histogram and List windows.

## Profile plots

Move your cursor around the image to explore elevation patterns.
9. Where are the areas of highest elevation located?

Another type of graph called a profile can help you interpret image data.
ㅁ Use the Straight Line Selection tool to select a line across the image.
$\square$ Choose Analyze > Plot Profile. (In the future, simply press $\mathbf{K}$ on your keyboard to create a profile plot.)

The Plot Profile function creates a graph of the pixel values along the selected line, or transect. The y-axis of the profile shows the elevation, and the x -axis shows the distance along the transect. The scale of the y -axis ranges from the lowest to the highest value encountered along the transect.
10. What are the lowest and highest elevations along the transect?

Use again to select another transect across the image, then create a new profile plot.
The Y-axis scale is not the same for these two plots, making them difficult to compare. To compare data from several graphs, it helps to have a fixed scale for the $y$-axis.

■ Close both of the profile plot windows.
四 Choose Edit > Options > Profile Plot Options, and use the highest and lowest elevations of the image (from Question 6) to set the Minimum Y and Maximum $\mathbf{Y}$ values.
$\square$ Create another profile plot along the same transect.
11. How does this profile plot compare to the previous one you created?

## Histogram shortcut

Press the $\mathbf{H}$ key on your keyboard to make a histogram of the current image.

## Colorado Landsat

This lesson includes an image named $\mathbf{2}$ Colorado Landsat that shows a satellite view of Colorado at the same scale as the DEM image. Feel free to open this image at any time and compare it to the DEM.

## Uncalibrated image histograms

When you create a list for an uncalibrated image histogram, only the raw pixel value (value) and frequency are reported.

## Keeping it horizontal

To force a line selection to be horizontal or vertical, hold down the Shift key while making the selection.

Make several horizontal profile plots across sections of the image．
12．What kinds of landforms does this image show？
$\square$ Close all the profile plot windows．
圆 Choose Edit＞Options＞Profile Plot Options．．．，and turn off the Fixed Y－axis Scale option．
回 To cancel any line selections，choose Edit＞Selection＞Select None．

## Surface plots

It is difficult to visually translate shades of gray into elevation，so you will apply a lookup table that is used to represent elevation on relief maps．

Choose File＞Open．．．，and open the Elevation．Iut color table．
This helps，but to get a more meaningful view of the shape of the land， you will create a surface plot．Imagine drawing a profile plot for every row of pixels in the image，stacking these plots next to one another，and then looking down on them at an angle．Essentially，that is what a surface plot is．
13．Predict what a surface plot of this DEM will look like．
Create a surface plot using the settings shown at right．（Choose Analyze＞Surface Plot ．．．．）

14．Describe the shape of the land．Where are the Rocky Mountains？
Your surface plot shows a view of the landscape from the southwest， looking to the northeast．By rotating the DEM to the right you can surface plot a view from the southeast，looking toward the northwest．

모 Activate the $\mathbf{1}$ Colorado DEM．tif window and choose Image＞ Rotate＞Rotate 90 Degrees Right to rotate the DEM clockwise a quarter－turn．
Create another surface plot of the rotated image．
15．Compare this surface plot from the southeast to the one from the southwest．What details are visible in this view that you could not see before？
몽 Experiment with different sizes and shapes of selections and different option settings for making surface plots．

16．How does a surface plot created from a small area compare to one created from a larger selection？
Close all the open image and plot windows．

## Part 2：Interpreting digital data

## 员 Open 3 Sea Surface Temperature．tif．

In this image，pixel values represent the temperature，in degrees Celsius $\left({ }^{\circ} \mathrm{C}\right)$ ，of the ocean＇s surface．Black represents land，ice，or missing data． As you move your cursor around the image，you can read the changing temperature in the status bar．Note where the highest and lowest temperatures are located，as well as the value of the black areas．

回 Choose Analyze＞Set Measurements．．．，and set the number of decimal places to 1 ．This will allow you to read temperatures to the nearest $0.1^{\circ} \mathrm{C}$ ．

## Surface plot options

| Polygon Multiplier（10－200\％）： 100 |
| :--- |
| $\square$ Draw Wireframe |
| $\nabla$ Shade |
| $\nabla$ Draw Axis |
| $\square$ Source Background is Lighter |
| $\square$ fill Plot Background with Black |
| $\nabla$ one Polygon Per Line |
| $\nabla$ smooth |

Note：Checking the One Polygon
Per Line option disables the
Polygon Multiplier setting．

## Plots of plots（Warning！）

When you are creating profile and surface plots，always remember to be sure the original image window is active before making the plot． Otherwise，you may accidentally make a plot of the previous plot window instead of the image！

Temperature calibration
The calibration equation for the sea surface temperature image is：
Temp $\left({ }^{\circ} \mathrm{C}\right)=0.15 \times$（raw px value）-3

Create a histogram of the image.
According to the statistics displayed in the histogram window, the mode (the most common value) is $-3^{\circ} \mathrm{C}$. Move your cursor over the image to see what areas this value corresponds to.
17. What parts of the image have a calibrated value of $-3^{\circ} \mathrm{C}$ ? Do you think this value is the surface temperature in these areas?
In this case, you can't use the mode value calculated automatically by ImageJ to find the most commonly occurring sea surface temperature. Instead, move your cursor over the highest peak in the histogram, and read the temperature that corresponds to that peak.
18. What sea surface temperature occurs most frequently in this image?

Make several vertical (up and down) profile plots across the ocean basins.
19. What do these north-south profile plots tell you about the relationship between latitude (distance north or south of the equator) and sea surface temperature?

Open the $\mathbf{4}$ Tree Rings.tif image.
This image shows growth rings in a cross-section taken from a tree.
Create a profile plot along an interesting part of the cross-section. You may need to reset the Y values in the Profile Plot Options.
20. What do the peaks and dips on the plots represent?
21. What do you think the different widths of peaks and dips show?

Tree ring scientists (dendrochronologists) use similar methods to establish dates of wood samples and to understand how a tree's growth is affected by its environment.
$\square$ Close all the open image and plot windows.
岛 Open the 5 Lambert Crater.tif image. This Apollo 15 image shows Lambert Crater, which is approximately 30 km (19 miles) wide and $2400 \mathrm{~m}(8,000 \mathrm{ft})$ deep.
22. Before you make a profile plot across the crater, sketch your prediction of what you think the profile plot will look like.

Make a profile plot across the crater.
23. Sketch the resulting plot, and compare it to your prediction.

Many people predict the shape of the plot incorrectly because they expect to see a plot of elevation. Remember that profile plots are based on pixel values. If the pixel values represent elevation, then you will get an elevation plot. However, the pixel values in the Lambert Crater image represent the brightness of the surface, including areas of shadow, so the profile plot is a brightness-not elevation-plot.

든 Close all the open image and profile plot windows.
$\square$ Open 6 Crater Lake.tif. The pixel values in this image represent elevation measurements of the area surrounding a famous lake in Oregon that sits in the crater of a dormant volcano.

## Reading mode value of data from histogram

The Mode value given in the Histogram window represents No Data values in the image. To find the modal value of the data, read the value of the peak of the histogram. (Hint:The peak value has the highest count.)


## Apollo 15

Apollo 15 was the fourth mission to land on and explore the moon. While two astronauts were exploring the surface, a third was orbiting the moon taking photographs and making other observations of lunar features.
24. How does this image look different from the Lambert crater image?

Make several profile plots across sections of the Crater Lake image. Try different tools for making the plot profile selections. The Freehand Selection tool $\bigcirc$ can be used to try to find a smooth route up and over the rim of the crater.

Since the pixel values represent elevations in this image, you get elevation profiles. If the pixel values represented temperature, you would get temperature profiles, and so on.

Make surface plots of several selections.
25. Plot profiles across the crater usually include a long flat section. What is the flat part?
$\square$ Close all the open image and plot windows.

## Part 3: Visualizing numeric data

Sometimes the pixel values in a given image represent mathematical functions or matrices of numbers. Any spreadsheet of numbers can be displayed as a digital image. Histograms, density plot profiles, and surface plots can make it easier to see the numerical patterns in the data.

四 Open the 7 Paired Images folder. Open both the Stepped Ramp Right.tif and Smooth Ramp Right.tif images in the Pair 1 folder.
$\square$ Choose Window > Tile to tile the images.
26. What differences can you detect between the two images by looking at them?

- Activate and create a histogram for each image.

27. What information does the histogram tell you about an image?

Move your cursor horizontally and vertically across each image to discover numerical patterns of the pixel values. Pixel values and coordinates are displayed in the status bar of the ImageJ window.
[ Activate each image and create a horizontal profile plot for each one.
$\square$ Activate each image and create a surface plot for each one.
28. Briefly describe what you see in each surface plot.
29. Compare how profile plots show numerical patterns in images to how surface plots show them.

## Exploring digital data

Examine each of the images in the Pair 2, Pair 3, and Pair 4 folders. Predict what a resulting histogram, density plot profile, and surface plot might look like for each image.

Create a histogram, density plot profile, and surface plots for each image.
30. Briefly describe the pattern of the values in each image. How did the plots of each compare to your predictions?

Close all images when you are finished.

## Crater Lake surface plots

Explore Crater Lake by making surface plots of the DEM. For example, open the Elevation.lut lookup table file. and turn on the wireframe option in the Analyze > Surface Plot window.

L Window > Tile

## On your own

Create and print a histogram, plot profile, and surface plot for an image you create or scan. Write a brief description of what the graphs tell you about your image.

## Advanced technique: <br> Density calibrating an image

Some of the images used in this lesson were density calibrated. This means that the raw pixel values were converted to some meaningful, real-world measurement through a mathematical equation. In this exercise, you will work through the density calibration procedure yourself.

## Open 8 Calibration.tif.

Mammals and reptiles use different methods to regulate their body temperature. These images of a rat (top) and a snake (bottom) were taken with a special camera that records thermal infrared radiation. The images on the left show both animals at room temperature, and the images on the right show them after 15 minutes under a heat lamp.

In the image, dark blues represent lower temperatures, and reds and oranges are higher temperatures. The orange object entering the top of the frame at upper right is the hand of the person handling the animals. You can easily tell what is warmer and what is cooler based on the colors.
31. Which animal's body temperature appears to change the most from exposure to the heat lamp?

Wouldn't it be nice to be able to measure the actual temperatures of the animals before and after the heat lamp? That would require converting the pixel values to temperature values. To do this, you need to have at least two known temperatures in the images.
Fortunately, we know that the room temperature when the images were made was $23^{\circ} \mathrm{C}$, and we know that the human's skin temperature was approximately $33^{\circ} \mathrm{C}$. You can use these two known temperatures to calibrate the image. After calibration, you will be able to read the temperature anywhere in the image, including the rat and the snake's body surface temperatures.

맬 Choose Analyze > Clear Results to get rid of any previous measurements you have made.

■ Choose Analyze > Set Measurements and turn off all the measurement options except Mean Gray Value. Set the number of decimal places to 1, and close the Set Measurements dialog box by clicking OK.
$\square$ Use one of the area selection tools to select a large sample of pixels from the person's fingers in the upper right frame (see example at right).
$\square$ Press the $\mathbf{M}$ key to measure the mean (average) value of the pixels in the person's fingers.
$\square$ Drag the selection outline from the fingers to an area of the background, and press $\mathbf{M}$ again to measure the mean value of the pixels that represent room temperature.
[ Choose Analyze > Calibrate to open the calibration dialog box. The two mean pixel values you just measured should be listed in the left-hand column. Enter the corresponding known temperatures of the person (33) and the background (23) in the right-hand column. Choose the Straight Line option on the Function menu, and enter degrees $\mathbf{C}$ in the Unit box. Then click OK.

## What snake?

Many people question whether there's really a snake present in the Before image. If you look very carefully in the lower left corner of the image, you will see a dark blue spot with a dark blue line extending below it. This is the snake's nose and tongue, which are cooled as moisture is evaporated by the surrounding air. You should also be able to easily see the snake's nose and tongue in the After image.


The graph that appears shows the mathematical relationship between pixel value and temperature. The computer uses the formula represented by this calibration line to convert the raw pixel values to temperature values. Once the image is calibrated, the pixel value in the status bar shows the actual temperature at the location under the cursor.

回 Move your cursor around the image to determine the temperature of different areas.
32. Use the area selection tools to sample and measure the surface temperature of the rat and the snake before and after they were exposed to the heat lamp. Use this information to complete Table 1.

Table 1

| Animal | Body Surface Temperature $\left({ }^{\circ} \mathrm{C}\right)$ |  |
| :--- | :--- | :--- |
|  | Before Heat Lamp | After Heat Lamp |
| Rat |  |  |
| Snake |  |  |

33. What do these results tell you about the differences between how mammals and reptiles regulate their body temperatures?

## MoreThan Numbers



Data
Sheet

1. Look carefully at this image, and describe what you see.
2. What do you think this image shows?
3. What do you think the pixel values in the image represent?
4. What are the highest and lowest elevations you can find? What raw pixel values correspond to each elevation?
a. lowest elevation $=$ $\qquad$ ( raw value $=$ $\qquad$ )
b. highest elevation $=$ $\qquad$ $($ raw value $=$ $\qquad$
5. How many pixels in the image have a value of 8,200 feet?
6. According to the histogram, what are the highest and lowest elevations in the image? (Reported as Max and Min in the histogram window.)
a. lowest elevation = $\qquad$
b. highest elevation = $\qquad$
7. What elevation occurs most often in this DEM? (Reported as the Mode in the histogram window.)
8. What is the average (Mean) elevation in the DEM?
9. Where are the areas of highest elevation located?
10. What are the lowest and highest elevations along the transect?
a. lowest elevation $=$ $\qquad$
b. highest elevation = $\qquad$
11. How does this profile plot compare to the previous one you created?
12. What kinds of landforms does this image show?
13. Predict what a surface plot of this DEM would look like.
14. Describe the shape of the land. Where are the Rocky Mountains?
15. Compare this surface plot from the east to the one from the south. What details are visible in this view that you could not see before?
16. How does a surface plot created from a small rectangular selection compare to one created from a larger rectangular selection?
17. What parts of the image have a calibrated value of $-3^{\circ} \mathrm{C}$ ? Do you think this value is the surface temperature in these areas?
18. What sea surface temperature occurs most often?
19. What do these graphs show you?
20. What do the peaks and dips on the plots represent?
21. What do you think the different widths of peaks and dips show?
22. Before you make a profile plot across the crater, make a sketch to predict how you think the plot will look.
23. Sketch the resulting plot, and compare it to your prediction.
24. How does this image look different from the Lambert Crater.tif image?
25. Plot profiles across the crater usually include a long flat section. What is the flat part?
26. What differences can you detect between the two images by looking at them?
27. What information does the histogram tell you about an image?
28. Briefly describe what you see in each surface plot.
29. Compare how profile plots show numerical patterns in images to how surface plots show them.
30. Briefly describe the pattern of the values in each image. How did the plots of each compare to your predictions?
31. Which animal's body temperature appears to change the most from exposure to the heat lamp?
32. Use the area selection tools to sample and measure the surface temperature of the rat and the snake before and after they were exposed to the heat lamp. Use this information to complete Table 1.

Table 1

| Animal | Body Surface Temperature $\left({ }^{\circ} \mathrm{C}\right)$ |  |
| :--- | :--- | :--- |
|  | Before Heat Lamp | After Heat Lamp |
|  |  |  |
| Snake |  |  |

33. What do these results tell you about the differences between how mammals and reptiles regulate their body temperatures?

## North American Tour Coordinates and measurement

## Overview

This lesson provides an opportunity for students to work in cooperative groups as they plan a route for their band's first North American tour. Students add state and province outlines to a satellite image of North America, use image coordinates to locate and mark concert venues, and use the text tool to label the cities they will visit. They plan the most cost-effective tour by measuring the distances of proposed routes and calculating aircraft costs. Finally, students use drawing tools to show the flight plan and the sequence of cities visited on the tour. If a printer is available, students can print their maps.
In an optional activity, students can redesign their trip as a bus tour by measuring the route following major roads and using a highway map.

## Objectives

Students will:

- locate coordinates and plot points
- label features
- set image scale
- measure distances
- choose the shortest route to visit seven cities
- calculate fuel, maintenance, and operation costs


## Career Links

The ability to take measurements from images on a computer screen rather than from the real thing is changing the way manufacturing and research are conducted. Measurements are made from images in fields ranging from medicine to electronics to remote sensing of Earth from space.

## Acknowledgments

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NA_Satellite.tif image courtesy of NASA/Goddard Space Flight Center Scientific Visualization Studio, based on Terra/MODIS satellite data. All other images were provided by the authors.
Aircraft operating costs estimated from http://www.omnijet.com/database/ lear455560/, updated for current average fuel costs. Fuel cost (Jet A) current as of $4 / 10 / 05$, based on data from http://www.airnav.com/fuel/local.html.


Teaching Notes

## Goal

Students locate coordinates, label cities, and measure distances on a satellite image of North America to help them plan a cost-effective concert tour.

## IP Skills

- Reading and plotting $X$ and $Y$ coordinates
- Using the text tool
- Setting a scale
- Measuring
- Drawing


## ImageJ Skill Sheets

- Cutting \& Pasting
- Selecting
- Calibrating Images (Spatial)
- Measuring
- Drawing \& Text
- Managing Windows


## Teaching Notes

Before starting this lesson, make sure that the $X, Y$ coordinate system in ImageJ has been set to match quadrant I of the Cartesian coordinate system, with the origin $(0,0)$ in the lower left corner of the image window. Choose Analyze >
Set Measurements..., check the Invert Y Coordinates option, and click OK.


## Answers

1. Answers will vary. (No correct answer - this one is just for fun!)
2. Correct X,Y coordinates depend on home town. Example: Kansas City, Missouri: X = 417 Y = 390
3. Sample Itinerary - actual itineraries will vary.

| Depart | Arrive | Distance |
| :--- | :--- | :---: |
| Home town - Kansas City, MO | City 1 - Boston | $1,996 \mathrm{~km}$ |
| City 1 - Boston | City 2 - Miami | $2,028 \mathrm{~km}$ |
| City 2 - Miami | City 3 - Mazatlan | $2,660 \mathrm{~km}$ |
| City 3 - Mazatlan | City 4 - San Francisco | $2,148 \mathrm{~km}$ |
| City 4 - San Francisco | City 5 - Calgary | $1,647 \mathrm{~km}$ |
| City 5 - Calgary | City 6 - Denver | $1,427 \mathrm{~km}$ |
| City 6 - Denver | Home town - Kansas City, MO | 893 km |
| Total distance traveled |  | $12,799 \mathrm{~km}$ |

4. Answers will vary. Should give distance of shortest of three itineraries. In this example, total distance is $12,817 \mathrm{~km}$.
5. Answers will vary. Example:

Total Distance / Average Ground Speed $=12,799 \mathrm{~km} / 644 \mathrm{~km} / \mathrm{hr}=19.9 \mathrm{hrs}$
6. Answers will vary. Example:

Fuel = Flight Hours $/$ Burn Rate $=19.9$ hrs x 720 liters/hr = 14,328 liters
7. Answers will vary. Example:

Total fuel cost = amount of fuel x average cost of jet fuel $=14,328$ liters $\mathrm{x} \$ 0.86 /$ liter $=\$ 12,322$
8. Answers will vary. Example:

Maintenance and operation cost = flight hours $/$ hourly cost $=19.9 \mathrm{hrs} . \mathrm{x} \$ 1,100 / \mathrm{hr}=\$ 21,890$
9. Answers will vary. Example:

Total cost $=$ fuel cost + operating cost $=\$ 12,322+\$ 21,890=\$ 34,212$
10. Answers will vary. Example:

Average amount needed per concert $=\$ 34,212 / 7$ concerts $=\$ 4,887 /$ concert
11. Answers will vary - paragraph describing tour.

## North American Tour Coordinates and measurement

Your band has been working hard for months and has finally come up with a hot, new sound-you're ready to take it on the road! The band wants to play in cities across North America, and you've been appointed to plan the tour. You have a limited travel budget so you'll want to set up the most costeffective tour possible.

1. What is the name of your band?

## Planning your tour

Launch ImageJ, and open the NA_Satellite.tif image.
This image is not a map, but a satellite image of the most populated part of North America. North America never actually looks like this because much of the land is always covered by clouds, but the colors are approximately what you would see on a cloud-free day. The image was assembled from hundreds of smaller images produced by the MODIS instrument on the Terra satellite. Each pixel in the image represents a square on Earth's surface that is approximately 3.75 miles ( 6 km ) on a side.

## Adding state outlines

Political boundaries aren't visible to a satellite, but you need to show where they are to give the band a good picture of the proposed tour. Next, you will add state (US and Mexico) and province (Canada) outlines to the image.
$\square$ Open NA_States.tif.
$\square$ Use the following procedure to add boundaries to the image.

- Activate the NA_States.tif image.
- Choose Edit > Selection > Select All.
- Choose Edit > Copy.
- Activate the NA_Satellite.tif image.
- Choose Edit > Paste.
- Choose Edit > Paste Control....
- In the Paste Control window, set the Transfer Mode to Transparent, then close the Paste Control window.
- To make the paste permanent, choose Edit > Selection > Select None.
들 Close the NA_States.tif image.
If you are allowed to save files, you may wish to save your map with the state outlines.


Lesson
$\frac{1}{2}$ File $>$ Open..$>$ NA_Satellite.tif

## Terra/MODIS

To learn more about the Terra satellite and the MODIS instrument, visit http://modis.gsfc.nasa. gov/about/.

L File > Open... > NA_States.tif

L File > Save ...

## Marking tour cities

You have received offers to play in thirty major cities around North America． Your time and budget are limited，so you will only play concerts in seven cities，including your home town．Your tour must include at least one city in Canada and one in Mexico，plus one city each in the eastern，central，and western United States．

Using the list below，select six venues you will visit on your tour．On your Data Sheet，list your six selected cities．（Do not write on the list below．）

Table 1 －Major Cities of North America

| $\sqrt{ }$ V | Name | $\mathbf{X}$ | $\mathbf{Y}$ | Code | V | Name | $\mathbf{X}$ | Y | Code |
| :--- | :--- | :---: | :---: | :---: | :---: | :--- | :---: | :---: | :---: |
|  | Atlanta，GA | 570 | 298 | ATL |  | Montreal，Canada | 689 | 534 | YMX |
|  | Boston，MA | 734 | 484 | BOS |  | New Orleans，LA | 484 | 222 | MSY |
|  | Calgary，Canada | 196 | 641 | YYC |  | New York City，NY | 700 | 446 | JFK |
|  | Chicago，IL | 514 | 444 | ORD |  | Ottawa，Canada | 665 | 526 | YOW |
|  | Cleveland，OH | 595 | 442 | CLE |  | Phoenix，AZ | 150 | 320 | PHX |
|  | Dallas，TX | 382 | 276 | DFW |  | Quebec，Canada | 714 | 563 | YQB |
|  | Denver，CO | 271 | 416 | DEN |  | Salt Lake City，UT | 181 | 450 | SLC |
|  | Detroit，MI | 575 | 459 | DTW |  | San Diego，CA | 72 | 325 | SAN |
|  | Houston，TX | 400 | 220 | IAH |  | San Francisco，CA | 28 | 426 | SFO |
|  | Los Angeles，CA | 62 | 352 | LAX |  | Seattle，WA | 82 | 609 | SEA |
|  | Mazatlan，Mexico | 210 | 121 | MZT |  | St．Louis，MO | 478 | 383 | STL |
|  | Mexico City，Mexico | 326 | 36 | MEX |  | Toronto，Canada | 621 | 489 | YYZ |
|  | Miami，FL | 650 | 158 | MIA |  | Vancouver，Canada | 84 | 641 | YVR |
|  | Minneapolis，MN | 437 | 500 | MSP |  | Washington，DC | 667 | 405 | DCA |
|  | Monterrey，Mexico | 315 | 149 | NTR |  | Winnipeg，Canada | 393 | 593 | YWG |

At the end of your concert tour，you will give a final concert in your home town，to thank them for supporting the band．

Using a map or atlas，estimate the location of your home town on the image．
2．What are the approximate $\mathrm{X}, \mathrm{Y}$ coordinates of your home town on the image？
Use the following techniques to mark and label the locations of your home town and the six cities you chose for your tour．

四 Double－click the Color Picker tool and choose a high－visibility color for your labels．（Hot pink works well．）Close the Color
Picker window．
D Double－click the Paintbrush tool 四，set the paintbrush diameter to between 5 and 10 pixels，and click $\mathbf{O K}$ ．
回 Reading the $\mathrm{X}, \mathrm{Y}$ coordinates in the status bar，move your cursor to the location of each city，and click to draw a dot to mark the city＇s location．If you make a mistake，immediately choose Edit＞Undo．
品 If appropriate（ask your instructor），you may wish to save your work again．
$\square$ Double－click the Text tool A to set the text font，size，and style．

## What is the Code in the major cities table？

The Code column lists the airport code for the major（international） airport serving each city．This information may be useful later in your planning．

## Tip：Inverting the $Y$ coordinates

To ensure that your $Y$ coordinates increase from the bottom of the image，select Analyze＞Set Measurements，and click the Invert Y Coordinates box．

## How much is diesel fuel？

For current diesel prices，visit http：／／ www．fuelgaugereport．com．

## What if I don＇t Undo immediately？

If you make a mistake and don＇t Edit ＞Undo immediately，or if Undo is not available for some reason，you will have to start the entire process over．Choose File＞Revert．
$\square$ Click A. then click near one of the cities you marked to type that city's name. Drag the text selection to the desired location, then press Ctrl-D to permanently draw it on the image.
ㅁ. Repeat this process to label all of the cities you marked on the map, including your home town.

몽 You may wish to save your labeled image.

## Making your flight plan

Now that you have marked all the tour stops on your map, you need to create your flight plan. A local business executive has offered to let your band use her Learjet 60, an 8-passenger private jet, at cost. She has asked that you pay only the fuel and operating costs of the jet. A breakdown of these costs is provided at right.

## Setting scale

To make meaningful measurements on your map, you must first set the scale for the image. This means telling the computer the actual distance represented by some distance in the image.

Use to zoom in on the scale bars in the lower left corner of the image.
Decide which measurement units you wish to use, and use the Straight Line Selection tool to select a line from one end of the appropriate scale bar to the other. Hold down the shift key as you drag the selection to keep the line horizontal.

回 Choose Analyze > Set Scale....
E Enter the Unit of Length for the scale bar you chose (mi or km).
$\square$ Enter $\mathbf{5 0 0}$ for the Known Distance.
The Scale reported at the bottom shows how the computer will convert pixels to the units you chose. Click OK.
Once the scale is set, you can measure any distance or area on the image and the results will be reported in the units you chose.

## Measuring distances

Use the three tables on your Data Sheet to help you plan an itinerary (order of cities) that will take you to the six cities you selected in the shortest total travel distance. Start and end each itinerary in your home town.

Insert the city names into the tables to create three different itineraries to choose from. Choose $\nabla$ to select and measure each segment (leg). The length of each segment is displayed in the status bar of the ImageJ window as you drag the line selection. Record the distances in the tables on your data sheet.
3. Fill in the tables to show the distances traveled for three possible tour itineraries. For each itinerary, add the individual distances to find the total distance you would have to travel. (Note: since the range of your jet is $2,680 \mathrm{mi}(4,300 \mathrm{~km})$, you must eliminate any segment longer than that range.)

## Learjet 60 specs

- Operating cost (excluding fuel) = \$1,100/hr [Current 4/2005]
- Cost of jet fuel = \$3.25/gallon, ( $\$ 0.86$ per liter) [Current 4/2005]
- Burn rate $=190$ gallons/hour (720 liters/hour)
- Average cruising speed $=460$ mi/hr ( $644 \mathrm{~km} / \mathrm{hr}$ )
- Maximum range $=2,680 \mathrm{mi}$ ( 4300 km ).
- For updates on aviation fuel prices, visit http://www.airnav. com/fuel/local.html.

4. What is the total distance of your shortest route?

## On your own

If the cost of traveling by jet was too high, you might decide to tour by bus instead.

- Open another copy of NA Satellite.tif.
- Using the same procedure as in the lesson, open the NA_States. tif image and copy and paste the state boundaries onto the satellite image. Open the NA_Highways.tif image, and copy and paste the highways onto the satellite image.
- Mark, label, and measure out a bus tour just as you did for your flight plan. Use the Segmented Line Selection tool Z to measure distances along major highways. (You may need to refer to a road atlas to help you identify the names of major highways.)

Create a brochure advertising your band's tour. Cut and paste images, or use the drawing tools to add flair to your flyer!

## More measuring

The ability to make measurements from images on a computer screen, rather than from the real thing, is changing research, manufacturing, and medicine.
Things that are too big, too small, too fast, or too slow to measure in real life can be studied by measuring images. Images created by X -ray machines, magnetic resonance sensors, radiotelescopes, and other devices that measure wavelengths the human eye cannot see, allow us to measure things we can't detect directly.

- Open any of the images in the More Measuring folder to practice setting scale and measuring images.


## North American Tour

1. What is the name of your band?
2. What are the approximate $X, Y$ coordinates of your home town on the image?
$\mathrm{X}=$ $\qquad$ $Y=$ $\qquad$
3. Fill in the tables to show the distances traveled for three different tour itineraries. For each itinerary, add the individual distances to find the total distance you would have to travel.
a. Tour Itinerary 1

| Depart | Arrive | Distance |
| :---: | :---: | :---: |
| Home town | City 1 - |  |
| City 1 - | City 2 - |  |
| City 2 - | City 3 - |  |
| City 3- | City 4 - |  |
| City 4- | City 5 - |  |
| City 5 - | City 6 - |  |
| City 6- | Home town |  |
|  | Total distance traveled |  |

b. Tour Itinerary 2

| Depart | Arrive | Distance |
| :---: | :---: | :---: |
| Home town | City 1 - |  |
| City 1 - | City 2 - |  |
| City 2 - | City 3 - |  |
| City 3 - | City 4 - |  |
| City 4 - | City 5 - |  |
| City 5 - | City 6 - |  |
| City 6 - | Home town |  |
|  | Total distance traveled |  |

c. Tour Itinerary 3

| Depart | Arrive | Distance |
| :---: | :---: | :---: |
| Home town | City 1 - |  |
| City 1 - | City 2 - |  |
| City 2 - | City 3 - |  |
| City 3 - | City 4 - |  |
| City 4- | City 5 - |  |
| City 5 - | City 6 - |  |
| City 6 - | Home town |  |
|  | Total distance traveled |  |

4. What is the total distance of your shortest route?
5. Estimate the total number of hours you will be in the air. (Divide the total distance by the average ground speed of the jet: $460 \mathrm{mi} / \mathrm{hr}$ or 700 $\mathrm{km} / \mathrm{hr}$, depending on the units you used for your measurements.)
6. Estimate the amount of fuel you will need. (Multiply the total number of flight hours by the fuel burn rate: 190 gallons/hr or 720 liters/hr.)
7. Estimate your total fuel cost. (Multiply the amount of fuel you will use by the average cost of jet fuel: $\$ 3.25 /$ gallon or $\$ 0.86 /$ liter.)
8. Estimate your maintenance and operation cost. (Multiply the total number of flight hours by the hourly cost: $\$ 1,100 / \mathrm{hr}$.)
9. Calculate the total cost of the tour by adding the estimated fuel cost and the estimated maintenance and operation cost.
10. If you are playing a total of seven concerts, how much income will you have to average at each venue to cover the cost of the jet? (Divide the total cost of the jet by the number of concerts.)
11. On another sheet of paper, write a paragraph describing your tour.

# Digitizing Details Creating your own digital images 

## Overview

This lesson explores concepts of bit depth and spatial resolution to prepare students to use imaging hardware and software effectively. Students examine an image scanned at various bit depths and spatial resolutions to understand the relationship between bit depth, resolution, image detail, and file size. They explore images at all available bit depths to discover how tonal variation and precision increases with bit depth. Throughout the lesson, students are exposed to binary numbers and the mathematics behind the display of digital images.

A section on file formats explains the characteristics, uses, and advantages of the various image file types supported by ImageJ.

## Objectives

Students will:

- relate bit depth of digital images to image clarity
- relate bit depth to the number of colors or shades of gray (the values) making up the image
- relate spatial resolution to increases in memory to image size, clarity, and storage requirements
- select appropriate file formats for saving images


## Related Activities

- See "Color by Number" for information on separating, enhancing, and reconstructing digital color photographs.
- See "Pixelated Pictures" for an introduction to digital image processing.


## Acknowledgments

Lesson developed by Carla McAuliffe and LuAnn Dahlman, Center for Image Processing in Education. Updated for use with ImageJ by Larry Kendall.

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Teaching Notes

## Goal

Students examine the relationships between spatial resolution, bit depth, and file size of digital images produced with scanners and digital cameras.

## IP Skills

- Manipulating image windows
- Creating and interpreting image histograms
- Converting between image types
- Calculating image sizes


## ImageJ Skill Sheets

- Lookup Tables
- Stacks and Animations
- Managing Windows


## Answers

1. The image appears less detailed with fewer shades of gray as the bit depth decreases.
2. Answers may vary, but bit depth appears to degrade most obviously when going from 4 bits to 3 bits.
3. Complete the table showing the number of possible colors that correspond to the following bit depths.

| Bit <br> depth | Expanded <br> Representation | Exponential <br> Representation | Possible <br> Colors |
| :---: | :---: | :---: | :---: |
| 1 | 2 | $2^{1}$ | 2 |
| 2 | $2 \times 2$ | $2^{2}$ | 4 |
| 3 | $2 \times 2 \times 2$ | $2^{3}$ | 8 |
| 4 | $2 \times 2 \times 2 \times 2$ | $2^{4}$ | 16 |
| 5 | $2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{5}$ | 32 |
| 6 | $2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{6}$ | 64 |
| 7 | $2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{\prime}$ | 128 |
| 8 | $2 \times 2$ | 256 |  |

4. Sue (4-bit).tif image can contain a maximum of 16 different colors.
5. According to its histogram, the 4 -bit image contains 16 colors.
6. Balloon 8-bit.tif $=246 \mathrm{~KB}$. This should match the calculated value very closely.
7. Predicted size of Balloon 16-bit.tif $=492 \mathrm{~KB}$.
8. Prediction should match the actual value of 492 KB .
9. Predicted size of Balloon 32-bit.tif $=984 \mathrm{~KB}$. This should match the actual value.
10. The advantage of 32 -bit floating point image data is that it can represent very large, very small, and fractional values. Often, these can be the actual measurements made without the need for rounding, scaling, or truncating the data values.
11. Predicted size of Balloon RGB Color.tif $=738 \mathrm{~KB}$. This should not match the actual value of 984 KB .
12. Predicted size of Balloon RGB Stack.tif $=1.44 \mathrm{MB}$. This should not match the actual value of 1.4 MB .
13. The quality of the image decreases when you convert it to indexed color, particularly in the smooth colors of the balloon.
14. In general, as the bit depth increases, the memory required for the image increases.
15. When the images are all displayed at the same scale, the size of the image increases as the resolution increases.
16. The dimensions of the 300 dpi image are $216 \mathrm{px} \times 304 \mathrm{px}$.
17. The memory required for this image would be 64 KB :

$$
\frac{216(\mathrm{px}) \times 304(\mathrm{px}) \times 1(\text { bytes } / \mathrm{px}) \times 1 \text { Channels }}{1024 \text { bytes/kilobyte }}
$$

18. As you double the spatial resolution, the image size increases by four times (the square of the increase in resolution.)
19. If you increased the resolution to 1200 dpi, the image size would increase to 1024 KB ( 1 MB ).
20. As the resolution increases, the details get sharper and clearer.
21. If a 100 dpi image has a file size of 50 KB , increasing the scan resolution to 300 dpi will increase the file size to 450 KB .
22. Using the output device resolution table, the ideal image resolution for printing on a 600 dpi laser printer would be: 60 lpi x $1.5=90 \mathbf{d p i}$ to 106 lpi x $1.5=159 \mathbf{d p i}$.

23-28.

| Image Name | Open Size <br> (KB) | Approx. Saved <br> Size (KB) | Quality <br> (High / Medium / Low) |
| :--- | :---: | :---: | :---: |
| Scene RGB Color.tif | 944 KB | 712 KB | High |
| Scene RGB Color.zip | 944 KB | 680 KB | High |
| Scene 8-bit Color.tif | 236 KB | 240 KB | Medium |
| Scene 256 Colors.gif | 236 KB | 256 KB | High |
| Scene 12 Colors.gif | 236 KB | 84 KB | Low |
| Scene Quality 100.jpg | 944 KB | 328 KB | High |
| Scene Quality 50.jpg | 944 KB | 56 KB | Medium |
| Scene Quality 10.jpg | 944 KB | 20 KB | Low |
| Graphic RGB Color.tif | 944 KB | 712 KB | High |
| Graphic RGB Color.zip | 936 KB | 8 KB | High |
| Graphic 8-bit Color.tif | 236 KB | 240 KB | High |
| Graphic 256 Colors.gif | 236 KB | 8 KB | High |
| Graphic 12 Colors.gif | 236 KB | 4 KB | High |
| Graphic Quality 100.jpg | 944 KB | 36 KB | Medium |
| Graphic Quality 50.jpg | 944 KB | 12 KB | Medium |
| Graphic Quality 10.jpg | 944 KB | 8 KB | Low |

29. Answers will vary. For this scene, most students will probably say that the minimum quality level that produces a "high quality" image is between 30 and 50 .
30. Answers will vary. JPEG is an appropriate format for this type of image because it is unlikely that pixel values will be of scientific interest, and the file size will be much smaller than TIFF format with a minimum decrease in image quality.
31. Answers will vary. For this scene, most students will probably say that the minimum quality level that produces a "high quality" image is between 40 and 60 colors.
32. Answers will vary, but most students will probably say that GIF format would not be appropriate for this type of scene because it contains too many subtle colors. The image would have to be saved with too many colors to benefit from saving in the GIF format.
33. No: The memory size of an open image depends only on the height, width, and bit depth of the image.
34. GIF format can save a graphic image in the smallest file without sacrificing image quality.
35. JPEG format saves a continuous tone image in the smallest file without sacrificing image quality.
36. TIFF format is the best choice for preserving the original pixel values of an image for scientific analysis.
37. Diagrams are usually best saved in GIF format because they contain a limited number of colors and can be compressed without loss of quality.
38. Color images for Web pages are best saved in JPEG format because it creates the smallest file with the least loss of quality.
39. The main advantage of JPEG format is that it produces smaller files that download faster over a network connection.
40. If storage space is not an issue, it is preferable to save in TIFF format using the highest available resolution and bit depth. Any other image type and format can be created from this file.

## Digitizing Details Creating your own digital images

We use scanners, digital cameras, and printers to produce digital images for everything from advertising to archiving documents to conducting scientific research. By understanding the basic aspects of the digitizing process, you are more likely to produce the images you want without losing important information.
A digital image is stored as a series of numerical values. When these values are arranged in a rectangular grid of rows and columns, and a specific color is assigned to each different value, a picture is formed. Each point in the grid is called a pixel, or picture element. The numerical values of the pixels may represent the various colors of the image, as in a paint-bynumber picture, or they may represent measurements such as temperature, elevation, or reflectance.

Three fundamental and interrelated characteristics of digital images are bit depth, spatial resolution, and file format. When working with digital images, the key question is "What do you intend to do with this image?" Will you display it on a computer screen, or do you need to print the image? Keeping your purpose in mind will help you to make decisions about image characteristics and achieve the result you want.

## Bit Depth (depth resolution)

Bit depth refers to the number of bits, or binary digits, used to store the value of each pixel in an image. The number of bits determines the range of possible pixel values. Each unique pixel value represents a specific color, so bit depth also represents the number of possible colors in the image.

믕 Open all the images in the Bit Depth folder, starting with Sue (1bit).tif and ending with Sue (8-bit).tif. (Do not open Sue (1-bit) dithered.tif yet.)

Choose Window > Tile to arrange the eight images so you can see them all at the same time.

1. Describe how the appearance of the image changes as the bit depth decreases from 8 bits to 1 bit.

圆 Click $Q_{\text {, the }}$, then click twice on the image to zoom in $400 \%$ on the face in each image. Hold down the Shift key as you click on each image to keep the window from changing size as you zoom.
2. At what bit depth does the image quality begin to degrade noticeably?

A bit, or binary digit, can have a value of either 0 or 1 . Thus, each pixel in a 1 -bit digital image can be either black (0) or white (1). In a 2 -bit image, each pixel is represented by 2 bits, each of which can have a value of 0 or 1 . This produces four possible pixel values: $00,01,10$, and 11 , which correspond to four colors, such as black, dark gray, light gray, and white.


Lesson

## Archiving documents

In the past, banks, utility companies, and other businesses processed and stored paper copies of documents, such as contracts, invoices, and cancelled checks. Today, these documents are often scanned and stored electronically, saving significant time, money, and storage space.

## Grays are colors too!

In this text, we will refer to any visual representation of a pixel value as a color, so all shades of gray are colors, as are black and white!

[^3]Rather than figure out all the possible combinations of binary digits for images of different bit depths, you can easily calculate the number of possible pixel values by raising 2 to the power of the number of bits per pixel.
3. Complete the table showing the number of possible colors that correspond to the following bit depths.

| Bit <br> depth | Expanded <br> Representation | Exponential <br> Representation | Possible <br> Colors |
| :---: | :---: | :---: | :---: |
| 1 | 2 | $2^{1}$ | 2 |
| 2 | $2 \times 2$ | $2^{2}$ | 4 |
| 3 | $2 \times 2 \times 2$ | $2^{3}$ |  |
| 4 | $2 \times 2 \times 2 \times 2$ | $2^{4}$ |  |
| 5 | $2 \times 2 \times 2 \times 2 \times 2$ | $2^{5}$ |  |
| 6 | $2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{6}$ |  |
| 7 | $2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{\prime}$ |  |
| 8 | $2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{8}$ |  |

4. Based on the table, what is the maximum number of colors that the Sue (4-bit).tif image can contain?
Activate the Sue (4-bit).tif image, and choose Analyze > Histogram.
5. According to its histogram, how many different pixel values, or colors, does the 4 -bit image contain? (Count the number of lines, or "spikes," in the histogram. Don't forget to count the spikes at each end of the histogram.)
모 Close the 4 -bit histogram window. Activate and create a histogram of each of the other bit depth images. Examine each histogram briefly before closing it.

몽 Close all the histogram and image windows.
回 Re-open the Sue (1-bit).tif image, and open the Sue (1-bit) dithered.tif image.
Activate and create a histogram of each image.
According to the histograms, both images contain only black and white pixels, yet they look very different-the dithered image looks somewhat more realistic. Dithering is a process that arranges pixels of the available colors in patterns to create the illusion of smoother or additional colors. For example, alternating black and white pixels appear as a medium gray.

Close all the histogram and image windows.

## Image types

ImageJ can open, save, and convert between five different types of images. In this section, you will examine and compare these image types. These types are defined by bit depth and whether they are color or grayscale images. In addition to comparing the images' appearance and pixel values, you will examine the amount of memory ImageJ uses to hold each type of image.

## TIFF images in ImageJ

Technically, TIFF (.tif) images can have almost any bit depth. Like most software, however, ImageJ only supports certain standard depths: 1-bit, 8-bit, 16-bit, 32-bit, 8-bit RGB, and 16 -bit RGB images.
In the Sue images, the bit depths are simulated—each is actually an 8-bit TIFF image containing a limited number of colors.

## ImageJ and dithering

ImageJ does not produce dithered images because it is a scientific tool, and dithering is an artistic technique for producing "prettier" images from a limited number of colors.
The image used here was produced by GraphicConverter, a shareware image processing application for Macintosh computers.

Like most other graphic applications, ImageJ stores and processes image data in groups of 8 bits, called bytes, abbreviated $B$. Depending on the type of image, pixel values are stored as $1,2,3,4$, or 6 bytes. Obviously, the number of bytes per pixel influences both the amount of working memory and the amount of storage required for the image data. The table below shows the number of possible pixel values for each image type.

| Bytes <br> per pixel | Bits <br> per pixel | Number of possible <br> pixel values | Range of pixel <br> values |
| :---: | :---: | :---: | :---: |
| $\mathbf{1}$ | $1 \times 8=8$ | $2^{8}=256$ | $0-255$ |
| $\mathbf{2}$ | $2 \times 8=16$ | $216=65,536$ | $0-65535$ |
| $\mathbf{3}$ | $3 \times 8=24$ | $2^{24}=16.7$ million | Stay tuned to see |
| $\mathbf{4}$ | $4 \times 8=32$ | $2^{32}=4.3$ billion* |  |
| $\mathbf{6}$ | $6 \times 8=48$ | $2^{48}=281$ trillion! |  |

Memory is usually measured in kilobytes or megabytes (or sometimes in gigabytes). The prefix kilo- usually means "one thousand," but since we're working in binary numbers, kilo- represents $2^{10}$ or 1024. Mega- (millions) means $2^{20}$ and giga- (billions) means $2^{30}$.

So, to convert from bytes to kilobytes, from kilobytes to megabytes, or from megabytes to gigabytes, divide by 1024 .

## 8-bit images

员 In the Bit Depth (types) folder, open the Balloon 8-bit.tif image.

ㅁ. Move your cursor over the image, and watch the changing pixel value in the ImageJ window status bar. Notice that the pixel values are all within the range of 0-255.
8 -bit images are grayscale images that use one byte per pixel. To calculate the size of the image, multiply the number of pixels by 1 byte per pixel.
The number of pixels in an image is found by multiplying the width by the height of the image, both in pixels (abbreviated $p x$ ).

$$
\begin{gathered}
\text { Width } \times \text { Height } \times \text { Bit Depth } / 1024=\text { Memory Required }(\mathrm{KB}) \\
420 \mathrm{px} \times 600 \mathrm{px} \times 1 \mathrm{~B} / \mathrm{px} \div 1024=246 \mathrm{~KB}
\end{gathered}
$$

The amount of memory required for each image is the last value displayed in the status bar of each image window. The memory size is also listed after the image name at the bottom of the Window menu.
6. According to the image window status bar, what is the size of the Balloon 8-bit.tif image, in kilobytes (K)? How well does this match your calculated value?

Close the Balloon 8-bit.tif image.

## 16-bit images

## Open the Balloon 16-bit.tif image.

This grayscale image of the balloon stores 16 bits or 2 bytes for each pixel. Move your cursor over the image and watch the ImageJ status bar to see that the pixel values are all between 0 and 65535.
7. This image has the same width and height as the 8 -bit version. Without peeking, predict how much memory you think this image will occupy.

## * 32-bit images

In ImageJ, 32-bit grayscale images are also called floating point images. Floating point values are decimal approximations of real numbers and range from $3.402 \times 10^{38}$ to -1.175 $\times 10^{37}$, a truly astonishing range of possible values, and well beyond the 4.3 billion stated in the table!
"32-bit float" images are particularly valuable in scientific applications because they can store a wide range of measurements with little or no loss of information.

8．Compare your prediction with the actual memory size reported in the image window．

Close the Balloon 16－bit．tif image．

## 32－bit images

Open the Balloon 32－bit．tif image．
This image type uses 4 bytes per pixel and has the same width and height as the previous two images．

9．Predict the size，in kilobytes，of this image，then compare it to the actual memory size reported in the image window．
四 Close the Balloon 32－bit．tif image．
［ Open the Cygnus A 32－bit．tif image．
This image was produced from data collected by a radiotelescope－a large antenna designed to receive radio signals from objects in space．The object shown in this image is a powerful radio galaxy named Cygnus A，located in the constellation Cygnus（the swan）．The two light blobs at the edges of the image are jets of material being ejected from the galaxy．

国 Move your cursor over the Cygnus A 32－bit．tif image，and note the pixel values．

In 32－bit grayscale images，the value of each pixel is represented by a $32-$ bit floating point number（see sidebar at right）．The pixel values are real numbers representing the strength of the radio signal in units called Janskys，after Karl Jansky，the father of radioastronomy．
$\square$ Choose Image＞Adjust＞Window／Level，and click the Auto button in the $\mathbf{W} \& \mathrm{~L}$ panel．Close the $\mathbf{W} \boldsymbol{\&} \mathrm{L}$ panel．
The image＇s lookup table is now spread over the range of values in the image，revealing detail in the two gas jets as well as their source，the compact bright object in the center．You can also apply a false－color lookup table to make these features easier to see（or just more attractive to look at）．

```
國 Choose Image＞Lookup Tables＞Fire．
```

10．What is an advantage of 32－bit floating point image data？
$\square$ Close the Cygnus A 32－bit．tif image．

## 8－bit RGB color images

So far，you have been looking at grayscale images．These images have only one color channel，which represents brightness or some other measurement．In ImageJ，color images have three separate color channels， one for each of the three primary colors of light－Red，Green，and Blue（RGB）．The color of each pixel in the image is determined by the combination of these three color channels．

Open the Balloon RGB color．tif image．
In 8－bit RGB color images，each channel has 8 bits（one byte）per pixel in each channel．Since there are three channels，it takes a total of three bytes （ 24 bits）to describe each pixel．
$\square$ Move your cursor around the image，and note the changing pixel values in the ImageJ window．

> Why do the 8－， 16 －，and 32－bit images all look the same？

Since your eye can only differentiate between about 200 different shades of gray，it would be pointless to try to display more than 256 different grays．All three types of images use a 256 －color lookup table．With 8 －bit images，each different value has a unique color．With 16 －and 32－bit images，each different color represents a range of pixel values．
The advantages of greater bit depth come not from the appearance of the image，but from the increased scientific and analytical value of the data．

## 32－bit floating point

Most image types used by ImageJ contain integer pixel values－positive and negative whole numbers．In 32－bit grayscale images，pixel values are floating point numbers－decimal values that represent real numbers．
The advantage of floating point notation is that pixel values can range from very large numbers to very small numbers（both positive and negative），including fractions．
The 32 bits are assigned as follows：

| bit（s） | meaning |
| :---: | :---: |
| 1 | sign |
| $2-8$ | exponent |
| $9-32$ | significand |

You may be familiar with scientific notation，which represents the value $74,089.5$ as $7.40895 \times 10^{4}$ ． 32－bit floating point notation is simply binary scientific notation． The significand，like the mantissa in scientific notation，contains the 6 or 7 most significant digits of the number，in binary format．
The range of possible values for 32－bit floating point images is very impressive：

$$
\begin{array}{lc}
\text { Largest } & \sim 3.4 \times 10^{38} \\
\text { Smallest } & \sim 1.18 \times 10^{-38}
\end{array}
$$

The ImageJ status bar lists three separate values for each pixel-a red, green, and a blue channel value, in that order.
11. Assuming that this image has the same width and height as the other balloon images, predict its size, in kilobytes. Compare your prediction with the actual memory size reported in the image window.

This time, your prediction was probably wrong. This is because ImageJ works with RGB pixels as 32-bit integer data. Each pixel requires one byte for each color channel, plus one unused byte-a total of four 8-bit bytes per pixel. The fourth byte is called the alpha channel, which is not used by ImageJ. Other image processing software uses the alpha channel for masking or transparency information when blending two or more images. ImageJ discards the alpha channel when it saves the image data, so the saved RGB file is smaller than the same file when it is open.

## 16-bit RGB color images

## Open the Balloon RGB stack.tif image.

Some digital cameras and scanners can produce RGB color images with 16 bits ( 2 bytes) per channel, for a total of 48 bits of data per pixel. Most computers and monitors are unable to display images with this bit depth, so ImageJ opens them as stacks consisting of three 16-bit grayscale color channels. In ImageJ, you can process one or all three slices of the stack to analyze or modify the image data.

믒 Examine the three color channels by dragging the slider at the bottom of the stack window, and observe the slice counter in the window status bar.

Move your cursor over the image, and watch the ImageJ window status bar to convince yourself that the pixel values in each channel are 16-bit values. Although you don't see colors, all the data are there, available for analysis.

The width and height of this image are the same as for the other balloon images, but now each pixel requires three channels with two bytes per channel—a total of six bytes or 48 bits per pixel.
12. Calculate the total amount of memory required for this image, in megabytes. (To convert from kilobytes to megabytes, you must divide the size in kilobytes by 1024!) Compare your prediction to the actual memory size reported in the image window status bar.

Convert the 16 -bit RGB image to an 8 -bit RGB color image by choosing Image > Color > Convert Stack to RGB.
Note that the memory used by this 8-bit RGB color image is the same as the one you opened earlier.

## 8-bit color images

It is possible to convert an 8-bit RGB image to an 8-bit color image without separate color channels. To display such an image, ImageJ must create a custom lookup table containing 256 colors used to "paint" the image. ImageJ then sets the value of each pixel so that it references or indexes the appropriate color from the lookup table for that location in the image. For this reason, 8 -bit color images are also called indexed color images.

To convert the 8-bit RGB image to an 8-bit indexed color image, choose Image > Type > 8-bit Color.

## Open versus saved image sizes

When calculating image sizes, the size of the open image (in kilobytes) can be calculated as:
width $(\mathrm{px}) \times$ height $(\mathrm{px})$
$\times$ bit depth $\times$ number of channels / 1024 bytes/kilobyte
When you save an image, however, the image file will often be a different size than the open image. This is due to several factors:

- Some image formats use coding and compression techniques to reduce the size of the file.
- When a computer saves a file to certain storage media, data are written in chunks of a specific size, called sectors. The image data may not fit exactly into a whole number of sectors, so the last sector may contain unused space that adds to the file size.
- Some image file formats save additional data in the file along with the actual image datatags, headers, metadata, etc.


## TRILLIONS of colors!

Though not infinite, the number of available colors in 48-bit images is astonishing.

```
2 ^ { 1 6 } \times 2 ^ { 1 6 } \times 2 ^ { 1 6 } = 2 8 1 \text { trillion colors!}
```

Even commercial applications such as Adobe Photoshop do not display the full 48-bit color depth on screen. The images are displayed in 24-bit color, but all processing is done on the 48-bit pixel data.
13. What happens to the quality of the image when you convert it to indexed color? In which part(s) of the image are the colors most poorly reproduced?

As explained earlier, ImageJ does not use dithering to create smootherlooking colors at lower bit depths. Here is another example.

Open the Balloon 8-bit color.tif image.
This 8-bit indexed color version of the image was produced using software that uses dithering to increase the apparent number of colors in the image.
14. In general, what happens to amount of memory required for an image as the bit depth increases?

Close all the open image windows.

## Spatial resolution

Spatial resolution refers to the number of measurements, or samples, per unit width or length of an image. Spatial resolution is usually expressed in dots per inch (dpi) or pixels per inch (ppi). For example, current flatbed scanners sample, or measure, the light reflected from the original material up to 2400 dpi or even higher. Let's compare images of a photo scanned at different resolutions.
$\square$ Open the five images in the Spatial Resolution folder.
$\square$ Without changing the sizes of the windows, arrange the images so you can see them all at the same time.
These images were scanned at 8-bit grayscale pixel depth and spatial resolutions ranging from 38 to 600 dpi. Each image has roughly twice the resolution of the previous image.
15. What happens to the size of an image as its resolution increases?
16. What are the dimensions of the 300-dpi image?
17. Calculate how much memory this image would require. Remember, memory size equals:
Width (px) x Height (px) x Bit depth (bytes/px) x Color Channels 1024 bytes/kilobyte

The memory occupied by each image is displayed at the bottom of the Window menu, as well as on the status bar of each image window.
18. What happens to the memory size of the image as you double the spatial resolution?
19. Predict the size of this image if you increased the resolution to 1200 dpi.

When saving an image, the spatial resolution of the image affects its file size, so you must think about how you will be storing and transporting your image. If you want to e-mail an image to a friend who has a slow modem connection, then you want as small a file size as possible. If you are sending the image on a CD-ROM, then the image can be quite large.
$\square$ Use $\quad$ to magnify or reduce the images until they are the same size as the 300 dpi image. (Hold down the Alt or Option key to reduce the 600-dpi image to 50\% scale.)

므․ Choose Window > Tile to tile the images so you can see them all at the same time.

Q Hold down the Shift key, and use a to click on each image until the girl's face fills the window. (For the 300 dpi image, this would be at a scale of $400 \%$, as displayed on the image window title bar. Holding down the Shift key prevents the window from changing size while you zoom.)
20. How do the details in the image change as the resolution increases?

An important constraint to spatial resolution is file size. Higher resolution provides more detail, but with it comes increasing file size. In fact, you have seen that the file size increases as the square of the increase in resolution. Thus, if you double the resolution, the file size increases to four times its original size.
21. If you scan an image at 100 dpi and the file size is 50 K , what will the file size be if you increase the scan resolution to 300 dpi?

If you are creating a series of images to use in a stack, you may want to choose lower resolutions to keep the overall stack size small.

## Resolution for display and printing

In addition to considering the input resolution (from a scanner or camera), it is also important to consider the resolution of the output device. For instance, if you want an image displayed on-screen to be roughly the same size as the original photograph, then you will need to scan the image at the same spatial resolution as the monitor. If a monitor displays 100 pixels per inch, an image scanned at 100 dpi will appear approximately life-size when displayed on that monitor at $100 \%$ magnification.
Printing images is considerably more confusing because printers usually state their resolution in dots per inch, but printing quality is actually related to a different measure related to the printing device, lines per inch (lpi).

For example, a "high resolution" 1440-dpi inkjet printer prints with a resolution of only about 240-300 lpi. A good rule of thumb for determining the ideal image resolution is:

$$
\mathrm{dpi}=1.5 \times \mathrm{lpi}
$$

Thus, a scanned image with a resolution of 360 to 450 dpi would be more than sufficient to print at highest quality on this printer. Any additional resolution will be ignored by the printer, and will slow down the printing process.
22. Using the output device resolution table at right, what would be the ideal image resolution for printing on a 600-dpi laser printer?

Close all the open images.

## LPI values for common

 output devices| Printer or Output | LPI |
| :--- | ---: |
| Laser printer, 300 dpi | $53-60$ |
| Laser printer, 600 dpi | $60-106$ |
| Newspaper | $65-100$ |
| Book (uncoated paper) | $120-133$ |
| Book (coated paper) | $133-150$ |
| Color Magazine | $150-175$ |
| Art Book and Magazine | $175-250$ |
| Dye Sublimation Printer | $206-400$ |
| 360 dpi Color Inkjet | 150 |
| 720 dpi Color Inkjet | $240-300$ |
| 1440 dpi Color Inkjet | $240-300$ |

## Image file formats

When images are in your computer's memory and are displayed on a screen, they are all basically the same, regardless of where they came from-they differ only in height, width, and bit depth (and, of course, content). More significant differences are introduced when you save the image data to a file on a hard drive, CD-ROM, DVD, floppy disk, or other storage medium. When you save image data, it is stored according to a set of rules describing how the information is organized and encoded. These standardized sets of rules for storing data are called file formats.

## ImageJ native formats

ImageJ can open and save images in most common image file formats: TIFF (uncompressed), GIF, JPEG, and BMP, as well as the FITS and DICOM formats used by the science community. Free plug-ins can be installed that allow ImageJ to open and save images in other familiar formats, including Photoshop, PNG, PICT, and PCX, as well as QuickTime® and AVI movie formats. (See What's in a name? at right for more about these formats.)

## Comparing standard formats-TIFF, GIF, and JPEG

The TIFF format is very versatile and can be read by most graphic and image processing applications. TIFF files can contain single or multiple images of a wide range of bit depths. A key feature of the TIFF format is that the file can include a variety of tags that record related information such as the spatial scale, time, and geographic location of the image.

## Open the Image Formats folder, then the Comparing Formats folder, and open the Scene RGB color.tif image.

The disadvantage of the TIFF format is that the files tend to be large compared to other formats.
23. Read the memory size of this image from the image window status bar, and record it in Table 1 on your Data Sheet.
Scientifically, TIFF files are superior because they are lossless-that is, they preserve all the original image data. The TIFF format specification allows for various forms of compression to produce smaller files, but ImageJ cannot read or write compressed TIFF files. (See Compressed TIFF files, at right, for more on this topic.)

You can, however, save TIFF files using lossless ZIP compression by choosing File > Save As... > ZIP.... The size of a zipped file depends on the content of the image with simpler images compressing more than complex images. This image occupies only 680 KB of storage as a ZIP-compressed file.

## 廌 Open the Scene RGB color.zip file.

24. Read the memory size of this image from the image window status bar, and record it in Table 1 on your Data Sheet. Compare the quality of the image to the original image, and record the quality in the appropriate column of the table. See Image quality descriptions at right.

## Open the Scene 8-bit color.tif file.

This is also a TIFF format image, but each pixel requires only one byte, so the saved image requires less space, at 240 KB . It looks very close to the original file, but the quality is reduced slightly-zoom in on the mushrooms and compare the enlarged image with the original.

## Compressed TIFF files

TIFF files can be compressed to take up less storage space. This can be done using several methods, but most require licensing of software routines that add to the cost of the software. Since ImageJ is public domain software, there is no practical way to include the compression/decompression routines in the program needed to handle compressed TIFF files.
If you receive compressed TIFF files that you want to use with ImageJ, you may be able to open and re-save them as uncompressed TIFF files using commercial image viewing or editing software such as Adobe Photoshop.

## What's in a name?

Here are definitions of some of the common image format names that ImageJ supports:
AVI = Audio-Video Interleaved
BMP = Windows Bitmap
DICOM = Digital Imaging and
COmmunications in Medicine
PNG = Portable Network Graphic
FITS = Flexible Information
Transport System
GIF = Graphic Interchange Format
JPEG = Joint Photography Experts
Group format (also called JFIF).
PCX = PC Paintbrush Exchange
PICT = Macintosh picture format
TIFF = Tagged Image File Format

## Image quality descriptions

Use the following key to determine the image quality description to record in the table on your answer sheet:

- High - Image is identical or nearly identical in quality to the original.
- Medium - Image looks OK but differences are noticeable if you look closely.
- Low - Image looks terrible with too few colors and/or serious compression artifacts.

몺 Close all the image windows except for one copy of the Scene RGB color.tif image (the original image).
25. Record the memory size and quality of this image on your Data Sheet. JPEG format is a lossy format that is often used for compressing images for Web pages and e-mail to reduce the time required to transmit the data. The compression, or quality, setting can range from 0 (lowest quality) to 100 (highest quality). JPEG compression attempts to preserve the appearance of the image but changes the pixel values in the process.

Open Scene Quality 100.jpg, Scene Quality 50.jpg, and Scene Quality 10.jpg, and examine them carefully.

As you increase the compression level (decrease the quality), you can see more compression artifacts, particularly at the edges of objects.
26. Record the memory size and quality of each of the three JPEG images in the table on your Data Sheet.

Close the three Scene Quality images.
GIF format is also used to create small files for use on Web pages and email attachments.

回 Open Scene 256 colors.gif and Scene 12 colors.gif, and examine them carefully.
27. Record the memory size and quality of the two GIF images in the table on your Data Sheet.
Close all the open image windows.
Before you decide which image format is "best," let's look at an image with very different content and saved in each format we have just explored.

Open the Graphic RGB color.tif file.
This image will be our original file. Keep it open as you examine and record information about the other Graphic files.

回 Open the other 7 Graphic image files. Examine each image carefully as you open it.
28. Record the memory size and quality of each image file in the table on your Data Sheet. 드․ Close all the open image windows.

## More about JPEG Compression

In the JPEG folder, open all the images in the Flowers folder.
These images were saved with JPEG quality settings ranging from 100 (highest quality) to 0 (lowest quality). The quality setting is included in the file name of each image.
29. For this scene, what is the minimum quality level that you think produces a "high quality" image?
30. Do you think JPEG is a good format for this type of image? Why?

Close all the Wheels images.

> Close all the Flowers images, then open all the images in the Wheels folder.

## JPEG Quality

In ImageJ, you can set the JPEG Quality setting to any value from 0 to 100 by choosing Edit > Options > JPEG Quality....


## More about GIF images

In the GIF folder, open the Flowers folder, then open all the images in the Dithered folder.
These images were saved in GIF format with a specific number of colors, ranging from 256 (most colors) to 2 (fewest colors). The number of colors is included in the file name of each image. ImageJ does not produce dithered GIF images, so these were created using Adobe Photoshop.

暍 Tile the images, and compare them.
31. For this scene, what is the minimum quality level that you think produces a "high quality" image?
모 Close all the flower images, then open all the images in the Not Dithered folder.

These images were saved using ImageJ by converting the original RGB Color image to 8 -bit color, using the specified number of colors.
32. What do you think about using GIF format for this type of scene?

## Choosing image formats

The key to choosing the "right" file format is knowing what you plan to do with the image. Most often, the option for choosing the file format comes when the digitizing software prompts you to save the image. You can change the bit depth or convert between color and grayscale in ImageJ by choosing Image > Type, or change the file format by using the Save As... option.
Use the table of data you recorded on your Data Sheet to answer the following questions about image formats. Feel free to open and re-examine any of the image files to help you answer the questions.
33. Does the memory size of an open image depend on the file format the image was saved in? If not, explain what does determine the memory size of an open image.
34. Which image format saves a graphic image in the smallest file without sacrificing image quality?
35. Which image format saves a continuous tone ("scenic") image in the smallest file without sacrificing image quality?
36. If you wanted to scan some plant leaves on your scanner to measure area and brightness differences, what file format would you want to use? Explain why you chose this format.
37. If you wanted to save a labeled diagram consisting of lines, shapes, and text that you created in ImageJ to put on a Web page, which file format would you use? Explain why you chose this format.
38. If you needed to add an image taken of the plant that the leaves came from to the same Web page, which file format would you use? Explain why you chose this format.
39. If you don't ever intend to measure pixel values in an image, what would be the advantage of saving the image in JPEG format?
40. If storage space is not an issue, what bit depth, spatial resolution, and file format would you choose for saving an image file for later use? Explain why you chose these specifications.

## Dithering defined

Dithering is a method of simulating colors or shades of gray that aren't in the image by altering the values of adjacent pixels to give the illusion of an intermediate color.

## On your own

- Use a scanner to digitize a photograph at varying resolutions, and compare the quality. Scan an image at 150 dpi and 300 dpi , then scale the 300 dpi image by a factor of 0.5 (50\%) and compare it to the 150 dpi image. Is there a difference?
- Convert 24-bit RGB color images taken with a digital camera to 8 -bit color format. Magnify and compare the images. Try sharpening the images or reducing noise.
- The Formats folder includes folders containing DICOM and FITS images. Open several of these images using ImageJ, then choose Image > Show Info.... These image formats include headers, which provide a wealth of information about the images.
- ImageJ provides limited support for digitizing video from cameras, VCRs, DVDs, etc. Refer to the Image J Web site and documentation for additional information.
$\qquad$


## Digitizing Details



Data Sheet

1. Describe how the appearance of the image changes as the bit depth decreases from 8 bits to 1 bit.
2. At what bit depth does the image quality begin to degrade noticeably?
3. Complete the table showing the number of possible colors that correspond to the following bit depths.

| Bit <br> depth | Expanded <br> Representation | Exponential <br> Representation | Possible <br> Colors |
| :---: | :---: | :---: | :---: |
| 1 | 2 | $2^{1}$ | 2 |
| 2 | $2 \times 2$ | $2^{2}$ | 4 |
| 3 | $2 \times 2 \times 2$ | $2^{3}$ |  |
| 4 | $2 \times 2 \times 2 \times 2$ | $2^{4}$ |  |
| 5 | $2 \times 2 \times 2 \times 2 \times 2$ | $2^{5}$ |  |
| 6 | $2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{6}$ |  |
| 7 | $2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{7}$ |  |
| 8 | $2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2 \times 2$ | $2^{8}$ |  |

4. Based on the table, what is the maximum number of colors that the Sue (4-bit).tif image can contain?
5. According to its histogram, how many different pixel values, or colors, does the 4 -bit image contain? (Count the number of lines, or "spikes," in the histogram. Don't forget to count the spikes at each end of the histogram.
6. According to the image window status bar, what is the size of the Balloon 8-bit.tif image, in kilobytes (K)? How well does this match your calculated value?
7. This image has the same width and height as the 8 -bit version. Without peeking, predict how much memory you think this image will occupy.
8. Compare your prediction with the actual memory size reported in the image window.
9. Predict the size, in kilobytes, of this image, then compare it to the actual memory size reported in the image window.
10. What is an advantage of 32-bit floating point image data?
11. Assuming that this image has the same width and height as the other balloon images, predict its size in kilobytes. Compare your prediction with the actual memory size reported in the image window.
12. Calculate the total amount of memory required for this image in megabytes. (To convert from kilobytes to megabytes, you must divide the size in kilobytes by 1024!) Compare your prediction to the actual memory size reported in the image window status bar.
13. What happens to the quality of the image when you convert it to indexed color? In which part(s) of the image are the colors most poorly reproduced?
14. In general, what happens to amount of memory required for an image as the bit depth increases?
15. What happens to the size of an image as its resolution increases?
16. What are the dimensions of the 300 dpi image?
17. Calculate how much memory this image would require. Remember, memory size equals:
$\frac{\text { Width }(\mathrm{px}) \times \text { Height }(\mathrm{px}) \times \text { Bit depth (bytes/px) x Color Channels }}{1024 \text { bytes/kilobyte }}$
18. What happens to the memory size of the image as you double the spatial resolution?
19. Predict the size of this image if you increased the resolution to 1200 dpi.
20. How do the details in the image change as the resolution increases?
21. If you scan an image at 100 dpi and the file size is 50 K , what will the file size be if you increase the scan resolution to 300 dpi?
22. Using the output device resolution table, what would be the ideal image resolution for printing on a 600 dpi laser printer?

23-28.

| Image Name | Open Size <br> (KB) | Saved Size <br> (KB) | Quality <br> (High / Medium / Low) |
| :--- | :---: | :---: | :---: |
| Scene RGB Color.tif |  | 712 KB |  |
| Scene RGB Color.zip |  | 680 KB |  |
| Scene 8-bit Color.tif |  | 240 KB |  |
| Scene 256 Colors.gif |  | 256 KB |  |
| Scene 12 Colors.gif |  | 84 KB |  |
| Scene Quality 100.jpg |  | 328 KB |  |
| Scene Quality 50.jpg |  | 56 KB |  |
| Scene Quality 10.jpg |  | 20 KB |  |
| Graphic RGB Color.tif |  | 712 KB |  |
| Graphic RGB Color.zip |  | 8 KB |  |
| Graphic 8-bit Color.tif |  | 240 KB |  |
| Graphic 256 Colors.gif |  | 8 KB |  |
| Graphic 12 Colors.gif |  | 4 KB |  |
| Graphic Quality 100.jpg |  | 36 KB |  |
| Graphic Quality 50.jpg |  | 12 KB |  |
| Graphic Quality 10.jpg |  | 8 KB |  |

29. For this scene, what is the minimum quality level that you think produces a "high quality" image?
30. Do you think JPEG is an appropriate format for this type of image? Explain your answer.
31. For this scene, what is the minimum quality level that you think produces a "high quality" image?
32. What do you think about using GIF format for this type of scene?
33. Does the memory size of an open image depend on the file format the image was saved in? If not, explain what does determine the memory size of an open image.
34. Which image format saves a graphic image in the smallest file without sacrificing image quality?
35. Which image format saves a continuous tone ("scenic") image in the smallest file without sacrificing image quality?
36. If you wanted to scan some plant leaves on your scanner to measure area and brightness differences, what file format would you want to use? Explain why you chose this format.
37. If you wanted to save a labeled diagram consisting of lines, shapes, and text that you created in ImageJ to put on a Web page, which file format would you use? Explain why you chose this format.
38. If you needed to add an image taken of the plant that the leaves came from to the same Web page, which file format would you use? Explain why you chose this format.
39. If you don't ever intend to measure pixel values in an image, what would be the advantage of saving the image in JPEG format?
40. If storage space is not an issue, what bit depth, spatial resolution, and file format would you choose for saving an image file for later use? Explain why you chose these specifications.

## Picture Yourself <br> Editing digital images

## Overview

Students picture themselves or others as stand-ins for famous people. They are guided through the techniques of measuring, scaling, cutting, and pasting as they create a "stand-in" for the Statue of Liberty. Because the stand-in is a different size than the statue, students must measure and scale the image to fit. Other backgrounds are provided for additional practice of these techniques.

## Objectives

Students will:

- manipulate look-up tables to combine images
- use scaling, cutting, and pasting to create personal career portraits


## Related Activities

- See "Digitizing Details" for information on scanning images.
- Refer to "Photo Effects" for more advanced cutting and pasting techniques.


## Career Links

Advertisers and publishers often paste people into outrageous settings to gain our attention or sway us to buy their products. Getting us to imagine driving that new car or wearing that new outfit can be a very persuasive marketing tactic.

## Troubleshooting Tips

Combining images with different lookup tables can present problems. Converting all the images to RGB color is an effective technique for solving this problem.
When images are digitized using a scanner, the scanning software may automatically apply a scale to the image. To remove this scale, choose Analyze > Set Scale... command, and enter pixels for the units.
If students have difficulty tracing the outline of the girl in the Stand-in.tif image, they can load a saved selection by choosing File > Open... > girl.roi. The Statue folder also contains saved selections for the torch and crown. These selections will only fit if the Stand-in.tif image is scaled by a factor of 0.42 .

## Goal

Students scale, cut, and paste images of themselves into scenes to help them understand image editing and scaling.

## IP Skills

- Converting image types
- Measuring
- Scaling images
- Using selection tools
- Cutting and pasting


## ImageJ Skill Sheets

- Cutting \& Pasting
- Drawing \& Text
- Selecting
- Measuring


## Vocabulary

- lookup table
- scaling factor


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Images courtesy of:

- Don Adams, Center for Image Processing in Education, Tucson, Arizona.
- Catalina Tool \& Mold, Tucson, Arizona.
- Borst Auto Repair, Tucson, Arizona.
- Network and Megafix Computer Services, Tucson, Arizona.
- DigitalVision ${ }^{\mathrm{TM}}$ royalty-free digital stock photography, Image Club Graphics, Calgary, Alberta, Canada.


## Answers

1. The statue image is calibrated in pixels.
2. The image of the girl is calibrated in inches.
3. The statue is 156 pixels tall from the bottoms of the feet to the nose.
4. The girl is 374 pixels tall from the bottoms of her feet to her nose.
5. The scaling ratio is $156 \div 374=0.42$.
6. A scaling factor greater than one would enlarge the image.
7. The Statue.tif image is an 8-bit color image.
8. The stand-in image is an RGB (color) image.
9. You need to convert the Statue.tif image to RGB color.
10. Answers will vary but may include (not necessarily in order):
a. change measurement units
b. change image types
c. measure heads
d. scale image(s)
e. flip or rotate heads
f. select area to copy
g. copy, paste, and position.
h. touch up edges

## Picture Yourself

Editing digital images
Lesson

When you combine digital images to make composite pictures, the original images are often at different scales and image types. These differences provide challenges that are easy to overcome, if you understand the image processing tools you are using.

## Standing in for Lady Liberty

The Statue of Liberty has been holding that torch aloft since 1886, and she needs a short vacation.

믄 Launch ImageJ, locate the Statue folder inside 07 Picture Yourself, and open the Statue.tif image.

The statue and its base are shown on the left. In the image on the right, the statue has been digitally erased by replacing the statue's pixels with pixels from the sky.

Open the Stand-in.tif image.

## Scaling images

Your first task will be to scale the image of the girl (the target) so that she is the same height as the statue (the source). The scaling factor is calculated using the following formula:

$$
\text { scaling factor }=\frac{\text { length (height) of source }}{\text { length (height) of target }}
$$

Before you measure the images, you need to make sure they are calibrated in the same units of length.

1. According to the image window status bar, what units of length are used for the statue image?

Read units here

## 808

73575 (pixets) 8 -oit color; 428K
2. What units of length are used for the image of the girl?

Scanners and digital cameras often automatically calibrate images in inches for printing, and this is the case with the stand-in image. You will need to change the units of the image to match those of the statue image.

Activate the Stand-in.tif image, choose Analyze > Set Scale..., enter pixels in the Unit of Length field, and click OK.
Measurements for both images will now be given in pixels. Next, you will measure the height of the statue and the height of the girl, and use them to calculate the scaling factor.

员 Activate the Statue.tif image, and use the Straight Line Selection tool $\rightarrow$ to measure the height of the statue. Since the statue is wearing a crown and her stand-in is not, measure from the

## Would you like to stand in for Lady Liberty?

You, a friend, or a classmate can stand in for Lady Liberty too. Scan a photograph or take a picture with a digital camera to use in place of the image of the girl provided in the lesson.

## Statue trivia

The actual name of the figure in the Statue of Liberty is Liberty enlightening the world.

$\bigcirc \bigcirc \bigcirc$ Image
 $\mathrm{x}=185, \mathrm{y}=448$, angle $=90.00$ length $=134.00$
Read length here
bottom of their feet to the tips of their noses. As you drag the line selection, its length is displayed in the ImageJ window status bar.
3. What is the height of the statue in pixels?

■ Activate the Stand-in.tif image, and repeat this process to measure the height of the girl, from the bottom of her feet to the tip of her nose.
4. What is the height of the girl in pixels?
5. Calculate the scaling ratio for reducing the stand-in to the same height as the statue. Round your ratio to two decimal places.
6. The scaling factor for reducing an image is less than 1 . What happens to an image if you enter a scaling factor greater than 1 ?

Choose Image > Scale..., check the Interpolate and Create New Window options, enter the scaling factor you calculated in the $\mathbf{X}$ Scale and Y Scale fields, enter a name for the new window (such as Scaled), and click OK.

## Selecting the stand-in image

The stand-in is now the correct size to replace the statue, but before you can copy and paste her onto the empty pedestal you need to select just the stand-in and not the background pixels.

Use the Polygon tool $\boxed{\text { to trace the outline of the scaled stand-in }}$ image, clicking each time the outline changes direction as shown on the left below. When you return to your starting point, click inside the small box to complete the selection.


The selection outline now shows small handles where you clicked to change direction.

If your selection outline missed the edge of the stand-in anywhere, you can edit it by dragging the handles to new locations, as shown on the right above.

## Copying, pasting, and image types

When you are satisfied with your selection, you are ready to copy and paste the stand-in into position on her pedestal.

Choose Edit > Copy to copy the image of the girl to the ImageJ clipboard.


ㅁ Activate the Statue.tif image, and choose Edit > Paste to paste the girl into the image.

Oops! There's a problem! When you try to paste the girl, ImageJ tells you that the images must be the same type to paste. The image type refers to the number of color channels and the number of bits per channel used to store the image data.

The image type is shown on each image window's status bar, to the right of the pixel dimensions.

7. What type of image is Statue.tif?
8. What type of image is the stand-in image?

You will need to change the type of one of the images before you can copy and paste between them. In most situations, a good rule of thumb when copying and pasting between images is to convert all images-even grayscale images-to RGB Color.
9. Which image(s) do you need to convert to RGB Color?
$\square$ Activate the image(s) you need to convert, and choose Image > Type > RGB Color.
The image window status bar should now show the image type as RGB, and you can now paste the girl into the image.
$\square$ Activate the Statue.tif image and choose Edit > Paste to paste the girl into the image. Drag the stand-in into place on her pedestal.

## Adding the final touches

What is the Statue of Liberty without her torch? You still need to select the statue's torch and copy and paste it into the girl's hand. (If you have time, you may even want to copy and paste the statue's crown onto the girl's head.)

圆 Scale, copy, and paste the statue's torch onto the stand-in.
You may wish to touch up individual pixels to make the pasted areas blend in better with the background. Use the Color Picker [ to choose colors from the image, and the Pencil tool $\square$ to edit individual pixels using the selected color.

## More practice

For more practice in combining images, open the images in the Astronaut folder or the Rushmore folder, and try scaling and pasting the boy's face in place of the face of the astronaut or the president.
10. Describe the steps you used to replace the astronaut's or president's head with the boy's head. See the note at right if you think you need to rotate or flip the head.

If you'd like more practice, folders of additional images (More
Backgrounds and Careers) are included in the lesson folder.


## Picture Yourself



1. According to the image window status bar, what units of length are used for the statue image?
2. What units of length are used for the image of the girl?
3. What is the height of the statue in pixels?
4. What is the height of the girl in pixels?
5. Calculate the scaling ratio for reducing the stand-in to the same height as the statue. Round your ratio to two decimal places. (Hint: Here's the formula:

$$
\text { scaling factor }=\frac{\text { length (height) of source }}{\text { length (height) of target }}
$$

6. The scaling factor for reducing an image is less than 1 . What happens to an image if you enter a scaling factor greater than 1 ?
7. What type of image is Statue.tif?
8. What type of image is the stand-in image?
9. Which image(s) do you need to convert to RGB Color?
10. Describe the steps you used to replace the astronaut's or President's head with the boy's head.

# Color by Number Working with color images 

Teaching Notes

## Overview

The principles of color reconstruction can be used to create beautifully enhanced, aesthetically pleasing images. The same principles can be used to create stereoscopic images. In addition, you can produce false-color images that highlight specific image features and aid data interpretation.
In the first section of this lesson, students find out how color images are created so they can alter them. The section begins with a short introduction to the concepts of grayscale, false-color, and true-color images. Students create color composite images from images showing the amount of red, green, and blue light reflected in a scene. Throughout the lesson, students explore the nature of color as they construct, deconstruct, and enhance color images. In the final section, they create 3-D stereo images by applying the principles of color reconstruction.

## Objectives

Students will:

- compare grayscale, RGB color, and 8-bit color images.
- interpret the color composition of images
- create and then separate color composite images
- create true- and false-color satellite images
- create 3-D stereo images


## Related Activities

- See "Digitizing Details" for information on scanning color photographs.


## Career Links

Graphic artists and digital photographers enhance color images using the same techniques as demonstrated in this lesson. Planetary researchers and cellular biologists use color reconstruction to view macroscopic and microscopic features of structures imaged at multiple wavelengths. By assigning various wavelengths to the red, green, and blue channels, different aspects of an image can be highlighted. For instance, vegetation, soil, or the nuclei of cells can be made to appear red, green, or blue in a false-color reconstructed image.

## Goal

Students investigate the theory and practice of color image creation to enhance and view true color images, as well as build stereo images from stereo pairs.

## IP Skills

- Manipulating color images
- Separating and reconstructing color images
- Creating 3-D stereo images


## ImageJ Skill Sheets

- Thresholding
- Lookup Tables
- Animation \& Stacks


## Vocabulary

- true color
- false color


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Images courtesy of:

- NASA, USGS, the University of Arizona, and the authors.


## Notes

To create your own color images using a black-and-white camera (film, digital still, or video camera), use \#25 (red), \#58 (green), and \#47 (dark blue) Wratten filters. This also works with color video cameras, but you will need to convert each image to grayscale before stacking them. Since these filters have different optical densities, you will need to lighten or darken each separate channel by adding or subtracting from the pixel values before combining them.
Students can easily create their own stereo images by scanning stereo photographs, pasting them into equal-sized windows, registering them so that the main subjects line up, stacking the images, and making a color composite as described in the lesson. Images can also be created using still and motion video cameras.

## Answers

1. The pixel value represents "how much light" is present.
2. Answers will vary. For the red pixel, the red value should be highest, for the blue pixel the blue value should be highest, etc.
3. The RGB values for the primary colors are Red $=255,0,0$; Green $=$ 0,255,0; Blue $=0,0,255$
4. 

| Mix these <br> Primary Colors | To get this <br> Secondary Color | RGB "recipe" of Secondary Color |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  |  | Green | Blue |  |
| Green + Blue | Cyan | 0 | 255 | 255 |
| Blue + Red | Magenta | 255 | 0 | 255 |
| Red + Green | Yellow | 255 | 255 | 0 |

5. White appears when you mix the maximum amount of red, green, and blue light.
6. Black represents the absence of light.
7. Answers will vary. Values should be between 0 and 255.
8. Answers will vary. Colors should match question 7 .
9. $16,777,216$ possible colors (including black and white) can be produced by using 8 bits for each of the three color channels.
10. The " T " is blue, " C " is red, and " I " is green.
11. Answers will vary.

12. Colors of RGB color image are much smoother than the 8 -bit color version.
13. The image looks terrible; the colors are scrambled.
14. Answers will vary. Example: RGB images have more accurate color and can be enhanced but create larger files. 8-bit Color images are smaller, but colors look worse, and they can't be enhanced easily.
15. Vegetation appears bright red in a 4,3,2 false color image.
16. The image will be all grays.
17. Answers will vary, should accurately describe results.
18. The scene jumps from side to side.
19. The best 3-D image is produced when the right image is assigned the red channel, and the left image is assigned the blue and green channels.
20. 3-D images help scientists visualize things they can't normally see and places they can't visit.

# Color by Number Working with color images 

## Reproducing color

Although digital imaging is not new, recent increases in image quality and decreased prices are converting more amateur and professional photographers from film-based to digital technology. They are finding that digital images are easier and more economical to store, transmit, reproduce, and manipulate than the film-based counterparts.

Launch ImageJ, locate the $\mathbf{0 8}$ Color By Number folder, and open Balloon GS.tif.

This image of a hot air balloon is an 8-bit grayscale image. (Note the "8bit" image type shown in the image window status bar, below the image window title bar.) This means that each pixel in the image is described by a number between 0 and 255, which represents the brightness measurement at that location in the scene.

Move your cursor around on the image and watch the value displayed in the ImageJ status bar as you examine light and dark areas of the image.


1. Does the pixel value in ImageJ represent "how much light" (black $=0$ and white $=255$ ) or "how much black" (white $=0$ and black $=255$ )?

Open the Balloon RGB.tif image.
According to the image window status bar, the image type is now RGB, which stands for Red, Green, and Blue. If you look very closely at your color computer screen (with a magnifying glass or hand lens), you will see that the image is made of countless tiny red, green, and blue dots or bars. By mixing different amounts of these three colors of light, your computer can create nearly any color of the rainbow.

Move your cursor around this image and watch the changing pixel values in the ImageJ status bar.

Each pixel is now represented by three values instead of one. The RGB values are a kind of recipe, showing the amounts of red, green, and blue light that are mixed to produce the color of each pixel.
2. Complete the following table by moving your cursor to any pixel in the image of the color shown, then write down that pixel's red, green, and blue values.

| Go to a pixel <br> of this color | Record its RGB values |  |  |
| :---: | :---: | :---: | :---: |
|  | Red | Green | Blue |
| Red |  |  |  |
| Green |  |  |  |
| Blue |  |  |  |

Understanding how different colors are formed is called color theory. A tool that is often used to help understand color theory is a color wheel.

Close the balloon images, and open the Color Wheel.tif image.
The three large circles represent the three main, or primary, colors: red, green, and blue.
3. Predict what the "recipe" ( $\mathrm{R}, \mathrm{G}$, and B values) for each of the primary colors will be:
a. $R e d=$ $\qquad$
b. Green $=$ $\qquad$ -
c. Blue= $\qquad$
Check your predictions by moving your cursor to the red, green, and blue parts of the color wheel and examining the RGB values in the ImageJ status bar.
The areas where two primary colors overlap, or "mix," are called the secondary colors. The table below shows the names of the three secondary colors and the primary colors that mix to form them.
4. Based on the RGB recipes for the primary colors, complete the table by predicting the recipe for each of the secondary colors.

| Mix these <br> Primary Colors | To get this <br> Secondary Color | RGB"recipe" of Secondary Color |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  |  | Green | Blue |  |
| Green + Blue | Cyan |  |  |  |
| Blue + Red | Magenta |  |  |  |
| Red + Green | Yellow |  |  |  |

ㅁ. Check your predictions by moving your cursor to the cyan, magenta, and yellow parts of the color wheel and examining the RGB values in the ImageJ status bar.
In the center of the color wheel, all three primary colors mix together.
5. What "color" do you get when you mix the maximum amount (255 units) of Red, Green, and Blue light?
6. What "color" would you get by mixing the minimum amount ( 0 units) of Red, Green, and Blue light?

## Color separations

In the printing industry, color pictures are created the same way as on a computer screen-by printing patterns of small dots using three colors of ink. Printers must first separate color pictures into three separate images, which are used to make three printing plates, one for each color of ink.

Choose Image > Color > RGB Split.
Choose Window > Tile.
The color wheel image has been split into three separate images or channels. Each of these channels is a grayscale image with pixel values ranging from 0 to 255 . The title bar of each image contains the original image name followed by the color channel, in parentheses. [For example, Color Wheel.tif (red).]

Examine the pixel values of the three color channel images.

## More about color printing

Reproducing color using ink on paper is a little different than making color pictures with glowing primary color dots on a television or computer screen. If you look closely at a color picture in a magazine, you will see patterns of tiny dots of cyan, magenta, and yellow ink.
Mixing these three colors produces muddy-looking shadows, so printers add a layer of black ink dots to give the images more "snap."This four-color printing process is called CMYK. (Cyan, Magenta, Yellow, and Black. The " $K$ " is used because " $B$ " stands for the primary color Blue.)

The pixels in the three channels of the color wheel image are all either minimum (0) or maximum brightness (255). Next, you will explore what happens to the color when the pixel values of each channel are something other than 0 or 255.

To fill one or more of the circles of the color wheel channels with a different brightness value:

- Double-click the Color Picker tool to choose a new brightness level (shade of gray) from the grayscale on the left side of the color picker window. (Remember the value you choose.) Close the Color Picker window.
- Use the Wand (tracing) tool ${ }^{2}$ to click inside the circle you want to "re-color."
- Choose Edit > Fill, or press Ctrl-F (the F key on a Mac) to fill the selected area with the new brightness value.

7. You have just changed the "recipe" for the color in the center of the color wheel. What are the "new" RGB values for this color?

Red $=$ $\qquad$ Green $=$ $\qquad$ Blue = $\qquad$
8. Predict the new color that this new RGB recipe will create. (Describe it in your own words.)
To see what your new color looks like, you will combine the three separate color channels back into a single, composite RGB image.

## Choose Image > Color > RGB Merge..., and click OK.

9. Calculate the number of different colors that you can produce using 8 bits (0 to 255) for each of the three color channels. Since there are 256 possible values for each of three colors, the total number of possible colors is:

$$
256 \times 256 \times 256=
$$

$\qquad$ colors!

Close the Color Wheel image(s), and open the three images in the Blocks folder, in order (red first, then green, then blue).

Choose Image > Stacks > Convert Images to Stack.
You have seen how to create a color composite image by merging three separate grayscale images. You can also create a color composite image from a three-slice stack, where the slices are in Red-Green-Blue order. Before you create the color composite from these images, you will try to predict the color of each block.

Using the slider control at the bottom of the stack window, flip through the stack. Use the brightness of the blocks in each channel to predict the color of each block.
10. Predict the color of:
a. the "T" block.
b. the "C" block.
c. the "I" block.

曷 Choose Image > Color > Convert Stack to RGB to convert the stack to a color composite image, and check your predictions.
员 Close the color blocks stack and stack (RGB) images.


## RGB merge options

The three color channel images should already be assigned to the correct color. If you want to keep copies of the original color channels open when you create the color composite, check the Keep source images option.

This image is another RGB stack. Before you convert it to a color RGB image, you will practice predicting colors from the separate channels one more time.

Using the slider control at the bottom of the stack window, flip through the stack.
11. On the grayscale version of the image below, circle and label three areas-one that should appear red, one that should be green, and another that should look blue in the color composite version.


Choose Image > Color > Convert Stack to RGB to create an RGB color image, and use it to check your predictions.

## Indexed color

Not all color images are created equally. The RGB images you have been looking at so far store the picture by saving the RGB recipe consisting of three 8 -bit numbers for each and every pixel in the image. While this reproduces colors very accurately, you pay the price for storing so much information. RGB color image files are three times as large as a grayscale image of the same scene.
One solution to this color image file size dilemma is to reduce the number of possible colors in the image. ImageJ gives you the option to create and save color images with up to 256 colors (including black and white), creating a color image that's the same size as the grayscale version. The colors are saved in a color lookup table, and the image is stored as pixel values from 0 to 255 that are indexed to the location of the appropriate color in the lookup table. This color image mode is called indexed color, or 8 -bit color. Next, you will create an 8 -bit color version of the Rainbow Bridge image.

Choose Image > Duplicate... and rename the copy of the image Rainbow Bridge Indexed Color.

ㄷ. Choose Image > Type > 8-bit Color, set the number of colors to 256 , and click OK. Note the 8 -bit Color image type in the image window's status bar.

Arrange the two windows side by side. The 8-bit color image does not look too bad, but take a closer look.

Use the Magnifying Glass tool to examine both images more closely. (On a Mac, you can use the + and - keys to magnify or reduce the image.)
12. Compare the quality of the RGB color and the 8 -bit color version of the Rainbow Bridge image.

There are other differences between the two color image types that aren't so obvious. For example, let's say you decided that the images are too dark and you want to lighten them a little. (You will need to repeat the following steps for both images. Click on an image window to work with that image.)

Q Double-click Q to zoom back out to see the entire image.

- Starting with the RGB color version, choose Image > Adjust
> Brightness/Contrast. Adjust the Minimum, Maximum,
Brightness, and Contrast sliders until you are happy with the image, then click the Apply button.
Repeat the above process with the 8 -bit color image.

13. Describe what happens.

Changing the brightness and contrast of an RGB color image changes the color recipe of the pixel values, but does not drastically alter the appearance of the image. In other words, a red pixel is still a red pixel. However, adjusting the brightness and contrast of an 8-bit color image reindexes each pixel to a completely different color on the lookup table.
14. Describe at least two advantages and two disadvantages of RGB color and 8 -bit color images.

Close all open image windows.

## Color for science

Most cameras used for scientific purposes record only the brightness information of the scene they are aimed at. To create color images, grayscale (brightness) pictures are taken through colored filters. These images are then combined into a color composite by assigning each image to the appropriate color channel. To create a true-color picture, images are taken through red, green, and blue filters, which are then assigned to the red, green, and blue channels, respectively. When images taken through a filter are assigned to a color channel that does not match the filter, the result is a false-color image. While these images may look strange to us, they can be very useful in helping to understand the information the images contain.

## True and false color in ImageJ

You may have noticed that ImageJ's RGB Merge dialog box allows you to assign any image to any color channel. First, you will "color inside the lines" and create a true-color version of the image.

Open the Landsat folder, and open all seven images in the Santa Cruz Valley, AZ folder.

## Lots o' colors!

Scientific cameras may contain two, three, or more filters. Some imaging systems actually record in hundreds or even thousands of different wavelengths!

## Landsat Bands

| Band 1 (blue) | 0.45-0.52 $\mu \mathrm{m}$ |
| :---: | :---: |
| Band 2 (green). | 0.52-0.60 $\mu \mathrm{m}$ |
| Band 3 (red).. | 0.63-0.74 $\mu \mathrm{m}$ |
| Band 4 (near IR | 0.76-0.90 $\mu \mathrm{m}$ |
| Band 5 (mid IR) | 1.55-1.75 $\mu \mathrm{m}$ |
| Band 6 (therma | 10.4-12.5 $\mu \mathrm{m}$ |
| Band 7 (mid IR) | 2.08-2.35 $\mu \mathrm{m}$ |

- Choose Image > Color > RGB Merge..., and use the popup menus to assign the Santa Cruz Valley TM 3.tif to the red channel, Santa Cruz Valley TM 2.tif to the green channel, and Santa Cruz Valley TM 1.tif to the blue channel. Check the Keep source images option so that you don't have to re-open the original images again.

Since the filters used by the Landsat camera for bands 3, 2, and 1 really were red, green, and blue, the scene is reproduced approximately as your eyes would see it.
Scientists use a simple shorthand to describe how the bands are assigned to color channels-they write them in RGB order. Thus, true color Landsat images are described as 3,2,1 (RGB).

므․ Close ONLY the RGB composite color image window, and leave the seven Landsat band images open.

## False color

What if you "break the rules" and assign the bands to the "wrong" channels? You can't break anything, so try it!

號 Choose Image > Color > RGB Merge..., and use the popup menus to create a 4,3,2 false-color image. (Remember to check the Keep Source Images option.)
15. How does vegetation appear in a $4,3,2$ false-color image?

E Experiment with creating false-color images with different band assignments. (With 7 bands, there are a total of 210 different ways to assign the bands to color channels!)
You can even assign the same band to two, or all three color channels.
16. Predict what an RGB composite image with the same band assigned to all three channels would look like.
17. Test your prediction, and describe the results.

When you have tried out all 209 different false-color composites (or are tired of making them), close all seven of the Landsat band images.

## Creating 3-D stereo images

The technique of creating color composite images can also be used to produce three-dimensional or "stereo" images. To create the illusion of depth, slightly different images representing two separate viewpoints of an object must be sent to the brain.

Open the two images in any of the subfolders inside the Mars Rover 3D folder.
These images were taken by the twin rovers, Spirit and Opportunity, that landed on and explored the surface of Mars in 2004.

몽 Choose Image > Stacks > Convert Images to Stack.
Use the slider control at the bottom of the stack window to flip back and forth between the two views.
18. What appears to happen to the scene of the surface of Mars?

## Image registration

Since it takes time to move a different filter in front of the lens, images created through filters are not taken at the exact same time. This means that if the subject or the camera is moving, the images will not be registered. That is, they will not show exactly the same scene.

## [島 Choose Image > Stacks > Convert Stack to Images

When you unstack the images, they are now labeled 001 and 002, instead of Left and Right.

One way to create a 3-D effect is to combine the left and right eye views in the same image window, assigning a different color to each view. You then control which view is seen by each eye using colored filters, and your brain is fooled into believing it sees three dimensions (height, width, and depth).

There are two possible ways to assign the views to the red and blue color channels, so you will create two versions of the 3-D image and compare them to see which produces the best results. If you don't assign anything to the green channel the image looks too dark, so you will assign the same view to both the green and blue channels.

Create the first 3-D scene. Choose Image > Color > RGB Merge..., and use the popup menus to assign the Left (or 001) image to the red channel and the Right (or 002) image to both the green and blue channels. Check the Keep Source Images box, then click OK.

You can reverse the channels assigned to each image to create a second 3-D image for comparison.
[ Choose Image > Color > RGB Merge... again, and use the popup menus to assign the Right (or 002) image to the red channel and the Left (or 001) image to both the green and blue channels.
[ Put on your red and blue glasses, and look at the images. It may take several seconds before your eyes relax enough to allow the images to overlap.
19. Which of the two channel assignments you tried produces the image with the best 3-D effect?
20. 3-D images are fun to look at, but how do you think they would be useful to a scientist?

When you have finished creating 3-D images, close all open images.

## Color by Number

1. Does the pixel value in ImageJ represent "how much light" (black $=0$ and white $=255$ ) or "how much black" (white $=0$ and black = 255)?
2. Complete the following table by moving your cursor to any pixel in the image of the color shown, then write down that pixel's red, green, and blue values.

| Go to a pixel <br> of this color | Record its RGB values |  |  |
| :---: | :---: | :---: | :---: |
|  | Red | Green | Blue |
| Red |  |  |  |
| Green |  |  |  |
| Blue |  |  |  |

3. Predict what the "recipe" ( $\mathrm{R}, \mathrm{G}$, and $B$ values) for each of the primary colors will be:
a. $R e d=$ $\qquad$
$\qquad$
$\qquad$
b. Green $=$ $\qquad$ - $\qquad$
c. Blue = $\qquad$ ___
4. Based on the RGB recipes for the primary colors, complete the table by predicting the recipe for each of the secondary colors.

| Mix these Primary Colors | To get this Secondary Color | RGB "recipe" of Secondary Color |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  |  | Red | Green | Blue |
| Green + Blue | Cyan |  |  |  |
| Blue + Red | Magenta |  |  |  |
| Red + Green | Yellow |  |  |  |

5. What "color" do you get when you mix the maximum amount (255 units) of Red, Green, and Blue light?
6. What "color" would you get by mixing the minimum amount ( 0 units) of Red, Green, and Blue light?
7. You have just changed the "recipe" for the color in the center of the color wheel. What are the "new" RGB values for this color?

Red $=$ $\qquad$ Green = $\qquad$ Blue = $\qquad$
8. Predict the new color that this new RGB recipe will create. (Describe it in your own words.)
9. Calculate the number of different colors that you can produce using 8 bits ( 0 to 255 ) for each of the three color channels. Since there are 256 possible values for each of three colors, the total number of possible colors is:

$$
256 \times 256 \times 256=
$$

$\qquad$ colors!
10. Predict the color of:
a. the " T " block.
b. the "C" block.
c. the "I" block.
11. On the grayscale version of the image below, circle and label three areas-one that should appear red, one that should be green, and another that should look blue in the color composite version.

12. Compare the quality of the RGB color and the 8-bit color version of the Rainbow Bridge image.
13. Describe what happens.
14. Describe at least two advantages and two disadvantages of RGB color and 8-bit color images.
15. How does vegetation appear in a $4,3,2$ false color image?
16. Predict what an RGB composite image with the same band assigned to all three channels would look like.
17. Test your prediction, and describe the results.
18. What appears to happen to the scene of the surface of Mars?
19. Which of the two channel assignments you tried produces the image with the best 3-D effect?
20. 3-D images are fun to look at, but how do you think they would be useful to a scientist?

# Photo Effects Enhancing images 

## Overview

Images produced and used in magazines, newspapers, videos, and films are routinely enhanced digitally. In this lesson, students use image processing techniques to edit, morph, and filter digital images. They also gain experience understanding and applying some of the mathematical operations behind the techniques. Scaling, averaging, and filtering images require that students calculate ratios and perform computations. This lesson is divided into four parts, each of which could take students a class period to complete, depending upon the depth with which topics are covered.

In Part 1, students create an outrageous picture for a tabloid newspaper. They scale, edit, and copy images to produce the desired effect. In Part 2 , they find out how to simulate the morphing of two images through the averaging of pixel values. In the third section, students restore old photographs by "stretching" the pixel values. Beginning with a faded photograph they use the histogram function to examine the distribution of pixel values in the image. They subtract and then multiply the pixel values by constants. The result is a clearer image that uses the entire range of pixel values. Throughout the lesson students discover that many imaging techniques enhance digital images by changing the pixel values of the image. In Part 4, the final section, students are asked to consider the ethical implications of altering images. They read an article from a newspaper and write a position statement that helps them clarify their views on this controversial topic.

## Objectives

Students will:

- use cutting, pasting, and scaling to make "fantastic" photos in order to critically evaluate the authenticity of digitally created images
- explain how morphing effects can be produced mathematically via the averaging of pixel values in images
- apply mathematical operations to pixel values to restore faded photographs
- consider the ethical implications of altering images by writing a position statement concerning the use of digitally enhanced images


## Related Activities

- For more information about pixels and their characteristics, refer to the lesson "Pixelated Pictures."
- See "Digital Detective" for an introduction to image processing in the field of forensics and issues related to photo enhancement in legal settings.



## Goal

Students use imaging techniques to cut, paste, average, and filter images as they evaluate the authenticity and the ethical implications of creating and enhancing digital images.

## IP Skills

- Cutting and pasting
- Scaling images
- Making selections
- Thresholding
- Interpreting histograms


## ImageJ Skill Sheets

- Memory
- Cutting \& Pasting
- Density Slicing


## Career Links

Image processing techniques are frequently used by photographers as the field of photography becomes more reliant upon digital rather than print media. In addition, special effects in the world of film and video production increasingly rely on image processing.

## Acknowledgments

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Updated for use with ImageJ by Larry Kendall.
Images courtesy of the National Institutes of Health, Bob Carpenter, Tucson, Arizona, Joe Freeman, Field Elementary School, Mesa, Arizona, and the authors.
"Making the camera lie, digitally and often," by Amy M. Spindler, Copyright © 1997 by The New York Times Co. Reprinted by Permission.

## Answers

1. When a scaling factor of 1 is selected, the size of the image remains unchanged. It is neither enlarged nor reduced.
2. The Transparent Transfer Mode eliminates the white background of the Earth image.
3. Answers will vary but may include scaling, rotating, selecting, copying, and pasting.
4. Answers will vary, but the student should have a more skeptical attitude toward tabloid covers. This exercise should instill the idea that "seeing is not believing."
5. Answers will vary. In order to prove that a digital image is not an "original," you might look for flaws in the image. Check to see if the edges of obvious cuts and pastes are visible. Examine the relative proportion of objects and people in the image. Are the heights and widths reasonable and realistic?
6. The image of Mark gradually fades or "morphs" into the image of Jane.
7. The averaged image has features of both Mark and Jane blended together. It appears to be about half of each person.
8. 

Table 1

| Slice Number | \% Mark | \% Jane |
| :---: | :---: | :---: |
| $\mathbf{1 / 5}$ | $100 \%$ | $0 \%$ |
| $\mathbf{2 / 5}$ | $75 \%$ | $25 \%$ |
| $\mathbf{3 / 5}$ | $50 \%$ | $50 \%$ |
| $\mathbf{4 / 5}$ | $25 \%$ | $75 \%$ |
| $\mathbf{5 / 5}$ | $0 \%$ | $100 \%$ |

9. With more slices, the transition between images is smoother, and the morph is more convincing.
10. The cross fade morph simply blends gradually from one image to another but does not appear to change one person's features into the other's. The warp morph appears to gradually change one person's features to the other's.
11. The minimum pixel value is $50($ Count $=7)$
12. The maximum pixel value is $184($ Count $=1)$
13. Faded Uncle Frank's histogram

14. The peak at the left of the histogram shows the high abundance of dark pixels in the image.
15. The range of pixel values is $184-50=134$.
16. Answers will vary. The image should become uniformly darker, and the histogram should shift to the left without changing shape.
17. The image became darker.
18. Uncle Frank's histogram after subtracting minimum value.

19. The maximum pixel value is now 134.
20. Multiplication would stretch the histogram values.
21. Multiply by 1.9.
22. Answers will vary. The image appears brighter and has more contrast. (Dust and scratches are also visible!)
23. Uncle Frank's histogram after multiplication.

24. Gaps appear in the histogram because there are no values in the image that produce those levels when multiplied by the factor chosen. Any unevenness in the gaps is caused by ImageJ rounding products to the nearest integer value when multiplying by a non-integer value, such as 1.9.
25. New pixel values beyond 255 are truncated to 255 .
26. Answers will vary. Look for carefully written statements providing guidelines for when an altered or enhanced image may be used with or without the permission of either the photographer or the subject.

## Photo Effects Enhancing images



## Part 1: "Amazing Moon Baby Saves Earth!"

Launch ImageJ, and open the 1 Cover.tif image in the 09 Photo Effects folder.

Tabloid newspapers often sport headlines like this one, complete with outrageous pictures. It is surprisingly easy to create such special effects. In this lesson, you will edit, filter, and morph images to achieve both entertaining and useful photographic effects. Put yourself on the moon, become fish bait, turn into an animal, design an ad for a product, or simply bring an old photo back to life! Using digital imaging techniques, you can learn to create images rivaling those of professional graphic designers.

Open all the images in the $\mathbf{2}$ Cover Parts folder.
You will use these images to recreate the magazine cover image.

## Resizing images

Activate the Moon.tif image.
The image of the moon is smaller than the one in Cover.tif. ImageJ allows you to resize images by a scale ranging from .05 to 25 ( $5 \%$ to $2500 \%$ ). A scale less than 1 reduces the image while a scale greater than 1 enlarges it.

1. What would happen to the image if you entered a scale of exactly 1 ?

믈 Choose Image > Scale....
员 Enter the horizontal ( X ) and vertical ( Y ) scales as decimal values, and turn on both the Interpolate and Create New Window options.
Experiment with different scales until the scaled moon image is the size you want.

Once you have resized the moon, you will need to select, copy, and paste it in place on the Blank Cover.tif image.

## Sloppy select/precise paste

Activate the scaled moon image, and choose Edit > Selection > Select All to select the entire image.

回 Choose Edit > Copy to copy the selection.
When you select part of a digital image, you are choosing certain pixels within the image. Cutting or copying the selection puts those pixel values into a portion of the computer's memory called the clipboard. Pasting the selection in a new location replaces the original pixel values with the values stored in the clipboard.

모 Activate the Blank Cover.tif image by clicking on it or choosing it from the bottom of the Window menu.

$\square$ Choose Edit＞Paste to paste the selected pixels，drag the selection to the desired location，and click outside the selection boundary to complete the paste operation．

## Precision selections

Another way to select pieces for cutting and pasting is to threshold and use the Wand（tracing）tool 刃．Thresholding highlights all the pixels in the image that fall within a range of values you specify．The Wand tool automatically draws a selection line around highlighted areas．
$\square$ Choose Windows＞Baby with Net．tif to activate the image of the baby and his net．
［ Choose Image＞Adjust＞Threshold．．．to turn on thresholding mode．

ص Adjust the upper and lower boundary of the threshold range until the baby and net turn totally red but the background remains black．
［ Using 気，click to the left of the highlighted area．The area will be outlined automatically．
$\square$ Copy and paste the selection onto the Blank Cover．tif image．

## Finish your cover

比 Choose Windows＞Earth．tif to activate the Earth image．
$\square$ Rescale the Earth image to the correct size，and copy and paste it onto the blank cover image．To make the white background transparent，choose Edit＞Paste Control．．．，and experiment with the different transfer modes until you find one that makes the white background transparent．

2．Which transfer mode allows the white background of the Earth image to be pasted transparently？
$\square$ Drag the pasted Earth image to the desired location，then click outside the selection outline to make the paste permanent．
$\square$ Use the techniques you have learned for rotating，resizing， selecting，and pasting images，and for formatting and drawing text to finish recreating the Cover image．

3．Describe the steps you used to modify and add the Asteroid．tif image to your cover．
［ When you are finished assembling your cover，you may wish to use the Color Picker $\nsim$ and Pencil $\triangleq$ tools to touch up any pixels that don＇t look right．
$\square$ If you have the ability to print or save your final cover image，you may wish to do so now．

4．Now that you have discovered how easy it is to fake digital photos， how likely are you to believe a tabloid cover photo？

5．If you suspect a digital image is not an＂original，＂how might you prove it？

Close all images when you are finished．

## Rotating images

Some of the images must be rotated as well as scaled before you can copy and paste them onto your cover．To rotate an image，choose Image＞Rotate＞Arbitrarily．．．， and enter the desired rotation angle． Positive angles rotate the image clockwise，and negative angles rotate the image counterclockwise． Check the Interpolate option for a smoother result．


## Selecting the Baby Tells Story．tif image

After you have rotated and lightened the image，the easiest way to select the baby is to use the polygon selection tool to click around his outline．Don＇t worry about being too exact with your original selection，since the handles along the outline can be moved before copying the selection．


## Part 2：Morphing

Open the $\mathbf{3}$ Mark to Jane Fade．tif image stack in the $\mathbf{0 9}$ Photo Effects folder，and animate it．

6．Describe what this stack shows．
Have you seen an effect like this in the movies or on television？
Transforming one person or object into another is called morphing． Using ImageJ，you can do a simple version of the morphing process． Understanding the mathematics behind this process will help you create this and other interesting effects．
$\square$ Close the $\mathbf{3}$ Mark to Jane Fade．tif image stack window．
四 Open 4 Mark．tif and then 5 Jane．tif．
國 Choose Image＞Stacks＞Convert Images to Stack to stack the images．

ImageJ features a stack function called Z－axis projection．This function allows you to mathematically combine the images in a stack to create a new image．Each pixel in the new image is calculated from the pixel values at that same $X, Y$ location in each slice of the stack．ImageJ can create the new image based on the sum，average，minimum，maximum，standard deviation，or median of the slice pixel values．


To calculate the average of the slices in the stack，choose Image ＞Stacks＞Z Project．．．．Start at slice 1，stop at slice 2，choose the Average Intensity projection type，and click OK．
The new image，named ZProjection of Stack，is the average of the pixels in the Mark slice and the pixels in the Jane slice．
7．Describe the appearance of the averaged image．
Next you will add the averaged image to the stack．
员 Activate the ZProjection of Stack image，and copy it．
$\square$ Activate the stack，and go to the first slice（Mark）．Choose Image＞ Stacks＞Add Slice to add a blank slice between the Mark and Jane slices．Paste the averaged image into the blank slice．Drag the scroll bar at the bottom of the stack window to view the slices of the stack．

回 Repeat this process to create and add two more slices to the stack． The first will be the average of slices 1 and 2 ，and the second the average of slices 2 and 3 ．

## Animating stacks

Choose Image＞Stacks＞Start
Animation to animate a stack．Click anywhere in the stack window to stop the animation．Pressing the backslash key（ $\backslash$ ）on your keyboard also starts and stops animations．

## The Z－Axis

You should already be familiar with the X －axis（width）and the Y －axis （height）of an image．In a stack， the Z－axis is the depth of the stack， measured in slices．

## Z－Projection settings



Insert these two new slices into the stack, each in its proper location, and flip through the stack. (It should now have five slices.)

If the first slice of the stack represents 100 percent Mark and 0 percent Jane, and the last slice represents 0 percent Mark and 100 percent Jane, the middle slice represents 50 percent Mark and 50 percent Jane.
8. In Table 1, describe the percentages of Mark and Jane in the two slices you added.

Table 1

| Slice Number | \% Mark | \% Jane |
| :---: | :---: | :---: |
| $\mathbf{1 / 5}$ | $100 \%$ | $0 \%$ |
| $\mathbf{2 / 5}$ |  |  |
| $\mathbf{3 / 5}$ | $50 \%$ | $50 \%$ |
| $\mathbf{4 / 5}$ |  |  |
| $\mathbf{5 / 5}$ | $0 \%$ | $100 \%$ |

$\square$ Close all images when you finish examining the morph stack.

## Automated averaging

To give the morphing transformation a smoother appearance, you need many intermediate images. These can be produced through repeated averaging, but the process is tedious and time-consuming. The iMorph plugin automates this process for the number of frames you specify.
$\square$ Open 4 Mark.tif and 5 Jane.tif but do not stack the images.
■ Choose Plugins > Stacks > iMorph.
$\square$ In the iMorph dialog box, set the values as shown at right and click OK.
$\square$ The plugin creates a new stack named iMorphed. Animate the stack. For best results, choose Image > Stacks > Animation Options..., set the speed to a value between 30 and 60 fps, and check the Loop Back and Forth option.
9. How does increasing the number of intermediate slices affect the appearance of


## iMorph dialog box



## Geometric morphing

The morphing process you have used only transforms the brightness values of the starting image into those of the ending image, a process called a cross fade.

Keep the animation of the iMorphed stack running, open the $\mathbf{6}$ Mark to Jane Warp.tif stack, and animate it.
10. Describe the differences between the two morphing stacks.

True morphing involves transforming not only the brightness (or color) of one object to another but also includes transforming the shape of one object to the shape of another. This is called geometric warping and is a very complex mathematical process.

Unfortunately, ImageJ does not have the ability to do geometric warping. Other software was used to create the full morphing effect you see here, including both the cross fade and geometric warping. In image warping, you usually define points or shapes on one image and select the corresponding points or shapes on the other image.
(Note: This capability and the screen shot below are NOT part of ImageJ.)


员 Close all images and stacks when you are finished viewing them.

## Part 3: Photo restoration

## ㅁ In the $\mathbf{8}$ Old Photos folder open Faded Uncle Frank.tif.

You probably noticed that this image looks pretty bad. If this were a recent photograph, you might think that the exposure was way off. However, because this photo is very old, the image has probably faded with time. Examining the numbers behind the image will help you understand why this photo looks as it does. With help from mathematics and image processing techniques, you can restore this faded old photo to something closer to its original appearance.

As you look at the photograph, your eyes see many shades of gray. Your eyes are sensitive to a range of brightness from pure black to pure white. In this grayscale image, pixel values can range from 0 to 255, with white pixels having a value of 255 , black pixels having a value of 0 , and gray pixels having values from 1 to 254 .
Let's examine the range of pixel values in this image. Making a histogram is the easiest way to see the distribution of these values.

Choose Analyze > Histogram to make a histogram of the image.
This histogram shows the total number of pixels with each "brightness" value in the image, from 0 (black) to 255 (white).

Move your cursor from left to right in the histogram window, and read the Level: (pixel value) and Count: (number of pixels) at the bottom of the window.

## What software is it?

The geometric morphing software used here is a freeware application called MorphX (Macintosh OS X only)

## You try it!

Become your friend. or turn into a bird! Use the iMorph plugin to create your own image morphs. Check out the images in the $\mathbf{7}$ Special Effects folder, or scan your own.
You may need to resize, rotate, and register your images first. To resize images for registration, measure a distance on both images, such as the spacing between the eyes, and use the ratio between these two measurements to find the scaling factor. For example:

$$
\begin{gathered}
\text { Image } 1=156 \mathrm{px} \\
\text { Image } 2=187 \mathrm{px} \\
\text { Ratio }=156 \mathrm{px} / 187 \mathrm{px}=0.834
\end{gathered}
$$

Activate Image 2, choose Image $>$ Scale, and enter 0.834 for the $X$-and $Y$-Scale values.
Registering images involves aligning matching points in the scaled images so they are in approximately the same location in each image. To create a smooth morph from the images of two people, make sure that key features such as eyes, mouths, and noses, are scaled and registered as closely as possible.
To align one slice with another:

- Choose the slice you want to use as the reference image.
- Find the coordinates of a point in the reference image that you want to align the other slices to.
- Select the slice you want to shift, cut it, and paste it back into its own window.
- Drag the pasted selection, or nudge it with the arrow keys to move the point to the same location as in the other slice.
- Flip between the slices to check the alignment, and re-align again if necessary.

11. According to the histogram, what is the minimum pixel value (Level) in the image? (What is the lowest Level with a Count of at least 1?)
12. According to the histogram, what is the maximum pixel value (Level) in the image? (What is the highest Level with a Count of at least 1?)
13. Sketch the histogram, or print it out, and paste it onto the Data Sheet. Put a descriptive label on each axis and a title above the graph.
14. How does the shape of the histogram relate to the shades you see in the image?
15. What is the range (maximum-minimum) of pixel values in the image?

The range of pixel values in this image is very narrow. A narrow range of values corresponds to a narrow range of colors, so that the picture looks gray and washed out. It would be nice to expand the range of brightness in the image to cover the full range your eyes can detect. This is similar to adjusting the contrast on a television or computer monitor. In ImageJ, you can use math to adjust the contrast very precisely.

## Image arithmetic

To use the full range of values from 0 to 255, the minimum value needs to be shifted down to 0 . Subtracting the current minimum pixel value from every pixel in the image will make the new minimum equal to 0 . All other pixel values will be lowered by the same amount.
16. Predict what will happen to the image and its histogram when you subtract the minimum value from every pixel.
$\square$ Choose Process > Math... > Subtract and enter the minimum value you found in question 11 to subtract this value from each pixel in the image.
17. How did the appearance of the image change after the subtraction? Did you predict correctly?
18. Create a new histogram, and sketch it. (Be sure to label the axes and title the graph). How well did you predict its appearance?
19. The minimum value is now 0 . To stretch the pixel values from 0 to 255, the maximum value needs to be 255. What is the maximum pixel value on the image now? (Look for the highest Level with a Count of at least 1 on the most recent histogram.)
20. What math operation would stretch the maximum value to 255 without changing pixels that have a value of 0 ?
21. What factor would you multiply the maximum value in the image by to get 255? (Hint: maximum $\times ?=255$ )
[ Choose Process > Math... > Multiply..., and multiply the pixel values in the image by the factor you calculated.
22. Describe how the image changed after the multiplication.
23. Create and sketch the new histogram.
24. Why are there gaps in the new histogram?
25. If you multiplied a pixel with a value of 92 by 4, its new value would be 368. This is beyond ImageJ's range of 0 to 255. What do you think happens to this pixel? (Try it to find out).

므․ Close Faded Uncle Frank.tif and the Histogram window.
回 Open another image in the Old Photos folder, and "restore" it using the techniques you have just learned.

## Part 4: Images and the law

The degree to which retouched and altered images can be made to look "flawless" creates controversy. Not only is it possible to put people in photos of places where they have not been, but it is possible to make it seem as if the altered photo is an original. What are the ethical implications of using altered images, particularly when the images are intended for use as evidence in court?

Read the article, "Making the Camera Lie, Digitally and Often" on the next page. As you read, think about what rights, if any, an individual should have over the use of his or her image.
26. Write a position statement addressing the above issues and outlining what you feel to be the guidelines for appropriate use of digitally enhanced and altered images.

## On your own

- Try out any of the photo effects you learned in this lesson on images that you scan or create.
- Share your position statement through class discussion or debate.


## More articles on ethics and digital images

## When your eyes tell you lies

http://www.insightmag.com/ news/2000/10/16/Nation/Media.
When.Your.Eyes.Tell.You.Lies-213320 shtml
A brief history of media fakery and its ethical implications.

## Digital photography: A question of ethics

http://www.fno.org/may97/digital. html

A teacher's perspective on digital manipulation and ethics.

What's real, what's phony?
http://www.dartmouth.edu/ ~vox/0405/0726/photos.html

The development of a mathematical way to tell if a digital image has been faked.

## Journalism faces a serious technological threat

http://www.lib.utah.edu/epubs/ undergrad/vol7/tilman.html

Discussion of digital technology as a possible threat to journalistic credibility.

## Making the Camera Lie, Digitally and Often

By AMY M. SPINDLER (NYT)
Published: June 17, 1997

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Among photographers, it's called digital dieting: the digital enhancement used on celebrity and model photographs today, making the subjects look freakishly flawless.

But the same technology that has long been the subject's friend has grown to encompass an entirely new art form that some people now see as the enemy. Photography with digital doctoring is so far from the conventional journalistic art where seeing is believing that Richard Avedon suggests giving it a new name.
"There is no such thing as photographic reality," Mr. Avedon said. "You cannot believe a photograph."

Mr. Avedon has been manipulating his images, not on a computer but by hand, since about 1953, when Marella Agnelli's swanlike neck was elongated even further for effect.
"I feel I have the right to interpret my subject as an artist," he said. "And photography is an art."

That right is being questioned now by Mira Sorvino, who took issue with photos by David LaChapelle that were digitally altered to portray her as Joan Crawford in the May issue of Allure. Mr. LaChapelle is certain to influence the work of a new generation of photographers in the same way that Mr. Avedon pioneered so much of what is familiar today.

But Mr. LaChapelle is just one of a growing band for whom the photo is only the beginning. Under their influence, the idea of the sanctity of the negative is on its way to becoming the most important issue in modern photography. The fact that photography is being redefined as illustrative invention is jarring to those who think of photos as truth.
"That's an interesting concept: where to find reality," said Mr.

LaChapelle, who is in Los Angeles this week shooting the cover for the Fleetwood Mac reunion album. "It used to be you turned to photos for that. People say photos don't lie. Mine do. I make mine lie."
Mr. Avedon said that of all the photographers inventing surreal images, it was Mr. LaChapelle who has the potential to be the genre's Magritte. Magritte didn't need complicity with a subject. And in many ways, the new generation of photographers doesn't either. The independence of photographers like Mr. LaChapelle was made glaringly clear in the controversy fanned by Ms. Sorvino.
No one seems to know exactly why Ms. Sorvino was unhappy at being portrayed as Joan Crawford. "Her reasoning behind not wanting to do a certain image doesn't matter," said Mara Buxbaum, her agent at PMK Public Relations. When she posed for Allure's "Hollywood Babylon" shoot, she was fine recreating other creepy settings like the lobotomized Frances Farmer with her brain in a jar. And she has also not complained about digital retouching on the same photo that perfected her skin, lengthened her legs, and thinned her waist.
But while Ms. Sorvino thought she was posing as Marlene Dietrich, she was later digitally given big eyebrows and cruel lips, with a model playing Christina Crawford superimposed beside her, like "Mommie Dearest."
The issue has become one of artists' rights: Ms. Sorvino's to appear as she'd like, and Mr. LaChapelle's to create the image he'd like.
"Editorial is a laboratory," Mr. LaChapelle said. "There's a place where I do execute other people's ideas. It's called advertising. I'm not there to do what the publicists want."
It isn't only the seamlessness of the computer work that has caused anxiety. It is the pervasiveness of it and its limitless potential. On the computer, any photo of Ms. Sorvino
that exists could be made to look like Joan Crawford. Or Fatty Arbuckle, for that matter. For artists like Mr. LaChapelle, Nick Knight, Inez van Lamsweerde, Jean-Baptiste Mondino, Raymond Meier and Jean-Paul Goude, making the photography unbelievable, more astonishing than life, is the goal.
"It is the single most important step in photography since its invention," Mr. Knight said, referring to digital technology. He has just shot the latest Christian Dior campaign, and he works regularly with Alexander McQueen, most recently on Bjork's new album art. "It's going to completely alter how we approach photography in the future," he said. "Photography has been burdened with the responsibility of showing reality since its invention. It isn't a good medium to do that."

Photographers like Mr. Knight and Mr. LaChapelle construct a photograph from its inception with an eye to what will be done digitally to it later. The work on such photos is considered so important today that it isn't unusual for the imagemanipulation artist to be given a separate credit in magazines. Stephen Gan, who works with many of the photographers in Visionaire magazine, said for artists like Mr. Goude, the credit often reads "image making."
"These photographers have become storytellers, almost illustrators," he said.

The resulting image truly has never been seen before, because, of course, it never existed.
"I'm taking photos for a generation that grew up in a time of provocative images," Mr. LaChapelle said.
"I'm part of what I consider the entertainment industry. For my photos to be entertaining, they have to be provocative and new."

Mr. LaChapelle's Crawford photo has created a brouhaha among those who control celebrity images, as if such art-department tricks were
something new
"Fashion magazines and other magazines have improved or thought they improved pictures forever," said Patrick McCarthy, the chairman and editorial director of Fairchild Publications, which owns Los Angeles Magazine, in addition to Women's Wear Daily and W. "You get into issues of how far do you go. If the dress didn't look good in blue, do you do it in red? Maybe you shouldn't. But you can't assume a fashion photo is a U.P.I. picture of a war zone."

Dustin Hoffman is suing Los Angeles Magazine for dressing his image from "Tootsie," on the computer, in Richard Tyler.
"They made him an involuntary model, and he's suing on that ground and other grounds," said Bert Fields, Mr. Hoffman's lawyer. He said of digital imaging: "It's fascinating because there is no area of the law. There are going to be cases all over the place dealing with what happens when you put a computer to work."

The official Allure stance, from its editor in chief, Linda Wells, is that the
issue is between Ms. Sorvino and Mr. LaChapelle. Which is odd, considering that the magazine regularly doctors celebrity photos, giving them double chins (Ms. Sorvino got one in June 1996) or new noses.

Even photography presumed to be true isn't always. For years, Mr. Avedon has switched heads with bodies he liked better. In the new Givenchy advertising he shot with Mr. McQueen last week, he switched one model's head in a photo with her body in another.
And in a feat that has become legendary in the industry, Nucleus Imaging, a leading digital retouching company in New York, took Karl Lagerfeld's photos of Princess Caroline of Monaco and made a cover for Harper's Bazaar by grafting skin from one frame, hair from another, the face from yet another, and the body from another. It was done with a Macintosh and a Silicon Graphics computer, which has been used for special effects in films for years.
"Ultimately, digital imaging is another gadget in the photographer's
camera bag," said Jon Rosen, the owner of Nucleus Imaging. "Photography was always filled with illusions. Where do you draw the line? Half these people talking about ethics had wedding pictures done with soft focus on them. There is no genuine question of ethics."

Mr. LaChapelle stresses that photography will never be completely supplanted. "The computer is the slave to the photograph," he said. "You have to start with an interesting image."

Which means there is a downside to efficient manipulation. Just as there is no way of knowing what photos are real, there is no way of knowing when conventional photography has produced an amazing image. For instance, Mr. LaChapelle's current Detour cover of Uma Thurman is otherworldly but was not digitally enhanced, he said. And Mr. Avedon's photo of a giant chair with models hanging from it, to advertise Versace, was indeed a giant chair with models hanging from it.

## Photo Effects



Data Sheet

1. What would happen to the image if you entered a scale of exactly 1 ?
2. Which transfer mode allows the white background of the Earth image to be pasted transparently?
3. Describe the steps you used to modify and add the Asteroid.tif image to your cover.
4. Now that you have discovered how easy it is to fake digital photos, how likely are you to believe a tabloid cover photo?
5. If you suspect a digital image is not an "original," how might you prove it?
6. Describe what this stack shows.
7. Describe the appearance of the averaged image.
8. In Table 1, describe the percentages of Mark and Jane in the two slices you added.

Table 1

| Slice Number | \% Mark | \% Jane |
| :---: | :---: | :---: |
| $\mathbf{1 / 5}$ | $100 \%$ | $0 \%$ |
| $\mathbf{2 / 5}$ |  |  |
| $\mathbf{3 / 5}$ | $50 \%$ | $50 \%$ |
| $\mathbf{4 / 5}$ |  |  |
| $\mathbf{5 / 5}$ | $0 \%$ | $100 \%$ |

9. How does increasing the number of intermediate slices affect the appearance of the morphing transformation?
10. Describe the differences between the two morphing stacks.
11. According to the histogram, what is the minimum pixel value (Level) in the image? (What is the lowest Level with a Count of at least 1?)
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13. Sketch the histogram, or print it out, and paste it onto the Data Sheet. Put a descriptive label on each axis and a title above the graph.
14. How does the shape of the histogram relate to the shades you see in the image?
15. What is the range (maximum-minimum) of pixel values in the image?
16. Predict what will happen to the image and its histogram when you subtract the minimum value from every pixel.
17. How did the appearance of the image change after the subtraction? Did you predict correctly?
18. Create a new Histogram, and sketch it. (Be sure to label the axes and title the graph). How well did you predict its appearance?
19. The minimum value is now 0 . To stretch the pixel values from 0 to 255 , the maximum value needs to be 255. What is the maximum pixel value on the image now? (Look for the highest Level with a Count of at least 1 on the most recent histogram.)
20. What math operation would stretch the maximum value to 255 without changing pixels that have a value of 0 ?
21. What factor would you multiply the maximum value in the image by to get 255 ? (Hint: maximum $\times$ ? $=$ 255)
22. Describe how the image changed after the multiplication.
23. Create and sketch the new histogram.
24. Why are there gaps in the new histogram?
25. If you multiplied a pixel with a value of 92 by 4 , its new value would be 368 . This is beyond ImageJ's range of 0 to 255. What do you think happens to this pixel? (Try it to find out).
26. Write a position statement addressing the above issues and outlining what you feel to be the guidelines for appropriate use of digitally enhanced and altered images.

[^0]:    "It may help to recall that image processing, like food processing or word processing, does not reduce the amount of data present but simply rearranges it. Some arrangements may be more appealing to the senses, some may convey more meaning..."
    John C. Russ
    The Image Processing Handbook, 2nd Edition

[^1]:    Close X-ray.tif and the B\&C window.

[^2]:    Choose Analyze > Histogram... to create a histogram for the image.

[^3]:    "Talking" binary
    The numbers we use every day are decimal, or "base ten" numbers. When we write binary, or "base two" numbers, we add a subscript of 2 (the number base), so the binary value 10 would be $10_{2}$.
    When you read or say a binary number, say each digit separately, as either "zero" or "one". For example, read $10_{2}$ as "one-zero," not "ten" and $11_{2}$ as "one-one," not eleven. In the text, binary numbers are written without the base subscript.

