Image-Pro Plus Product Note
Sequence Gallery

Introduction

Creating and viewing stacks of images and time-lapse sequences is commonplace in microscopy. Both confocal and widefield microscopy applications utilize through-focus stacks to create image volumes. Time-lapse studies also create stacks by acquiring images at predefined intervals. It is then possible to play these acquired images as a movie to view the motion and other activities of the system being studied.

There are times when viewing all the image planes from a through-focus stack or all time points in a time-lapse sequence is of interest. An example is comparing various images in a stack to judge which is most in-focus, or the time point when a change of interest occurs. A method for comparing all the images in the sequence is very useful for this operation. By viewing all images at once, more qualitative and quantitative judgments can be made concerning specific points in a time-lapse, or individual planes of interest in a stack file.

An additional benefit exists- viewing an image stack as a sequence gallery is useful for examining the effects of image filters and look-up tables on the stack.

Applications and Examples

The **Process|Sequence Gallery…** tool has been included in Image-Pro Plus to highlight and view individual planes in a stack or time-course of images. Sequence Gallery requires an image volume comprised of at least two image planes. The planes may be of any image format supported by Image-Pro Plus, including those generated by many third-party software packages (Table 1).

- Molecular Dynamics (GEL)
- Universal Imaging (STK)
- BioRad (PIC)
- Zeiss (LSM)
- AutoQuant (DEB, AVZ)
- Leica (LEI)
- Gatan Digital Micrograph (DM3)
- Fuji (INF)
- Nikon C1 (ICS)
- NIH (RAW)

Table 1. Third-Party File Formats Supported in Image Pro Plus v. 4.5

In the following example, a through-focus stack of a fetal mouse heart is displayed in a sequence gallery (Fig. 1). The purpose is two-fold: first to determine which image planes contain useable information, and second to isolate and observe structure within the heart.

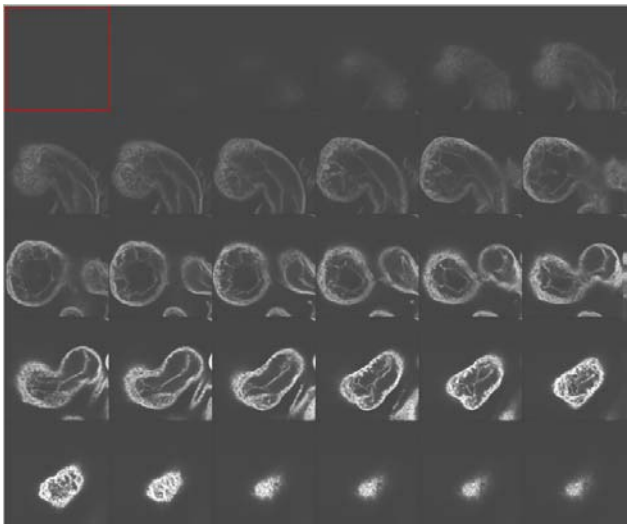


Fig. 1- Sequence Gallery of FITC-Labeled Fetal Mouse Heart. Courtesy of Bob Zucker- USDA, Beltsville, MD

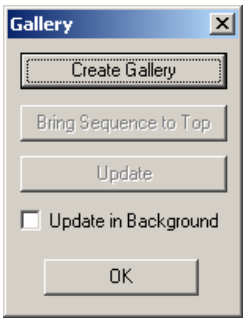
With the image stack displayed as a gallery it is obvious the first four planes do not contain useable information. They may be removed from the stack at this point by using the **Acquire|Sequence Toolbar…** to reduce the overall size of the image and to provide a better image if the stack is going to displayed using the 3D Constructor™ plug-in module. The remaining image planes may now be observed for their structural qualities.

Implementation

Process|Sequence Gallery…

- Using Image Pro, acquire through focus or time lapse series of images. Save as either sequence (.seq) or .tif stack image file. Alternatively, acquire the image through third party hardware (confocal microscope, etc.) and save as a stack file.

- Open the stack file and create an image montage using **Process|Sequence Gallery…** The following menu appears:



- Click 'Create Gallery' to create a sequence gallery. It will look something like this (Fig. 2):

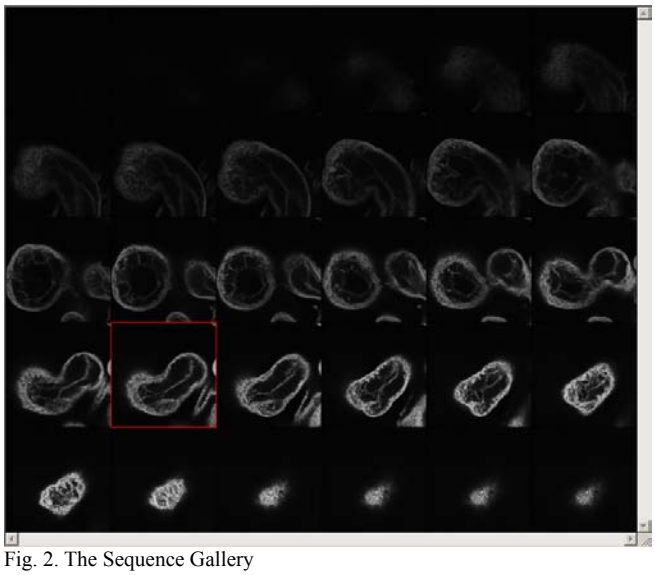


Fig. 2. The Sequence Gallery

- To highlight an image plane, click the plane of interest in the montage. The red box will appear around the plane, and the image plane will appear as the active plane in the stacked image (Fig. 3).

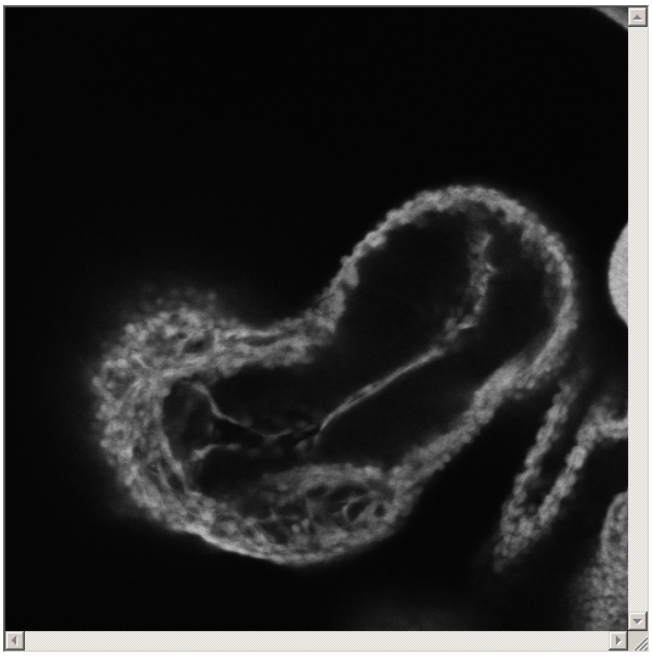


Fig 3. Image Plane Corresponding to Red Outlined Frame in Fig. 2.

- Click 'Bring Sequence to Top' to display the image stack on top of the gallery.

- Click 'Update' to show the frame in the gallery corresponding to the presently active image in the stack. This tool is useful especially after playing through the stack using the Sequence Toolbar.

- Click the 'Update in Background' check box to update the outlined frame in the gallery to the corresponding frame in the image stack, without bringing the image stack to the 'front' of the work space.

See Also

Media Cybernetics Product Note- "Extended Depth of Field"

How To Order

For more information on Image-Pro Plus, and to locate a Media Cybernetics' reseller in your area, visit our website at www.mediacy.com.

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