Flicker Vignettes - Short Examples

Vignettes are short examples that pose and answer questions of the form "how do I do xxxx?". We list the vignettes below. Each vignette describes the problem using demonstration data, the method suggested to solve the problem, any additional setups required, and links of related vignettes that may be referenced.

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36. Vignette: How do I calibrate grayscale using a spot list of known calibrations?

Flicker Vignette - How do I on-line help?

This vignette shows how do you can get on-line help from the Flicker Web site.

Method

The reference manual and vignette examples etc. are available on the http://open2dprot.sourceforge.net/Flicker Flicker Web site.

1. You can look at the reference manual on the Web site.
2. If you are running Flicker, you can access the reference manual and other parts of the Web site from the Help menu.
3. Look at the list of Vignettes in the reference manual.

Related vignettes

Flicker Vignette - How do I select an image from the left or right images?

This vignette shows how do you select either the left or right images. In order to do many operations, you must first select an image to operate on.

Method

1. Select the image you want to position by clicking on it. The title of the window will change from black to blue indicating it is selected.
2. To deselect an image so there are no images selected, then click on the Flicker window outside of either the right or left image. Both titles will now appear in black indicating neither is selected.

Setup for the demo

You need to have two images loaded in Flicker. The default when you start Flicker is to load the two human plasma gel images.

Related vignettes

Flicker Vignette - How do I position gel images in the scrollable windows?

This vignette shows how do you position gel images in the left and right scrollable windows so the spot appears in the center of the cross-hairs.

Method

Positioning window and trial object by Control-key/Mouse-Press

1. Select the image you want to position by clicking on it. The title of the window will change from black to blue indicating it is selected.
2. Press the **CONTROL**-key and then click on the spot you want to position in the cross-hairs. This will scroll the image window to the position.
3. This ***DOES*** position the *trial ojbect* used when you flicker.

Positioning image window by image window scrollers

1. Select the image you want to position by clicking on it. The title of the window will change from black to blue indicating it is selected.
2. Move the image window horizontal and/or vertical position scrollers to change the position you want to view.
3. This ***DOES NOT*** position the *trial object* used when you flicker.

Positioning the trial-object by draging the mouse

1. Select the image you want to position by clicking on it. The title of the window will change from black to blue indicating it is selected.
2. Press or drag the mouse to the spot you want to position in the flicker window. It will not change the position in the selected window.y
3. This ***DOES*** position the *trial object* used when you flicker.

Setup for the demo

You need to have two images loaded in Flicker. The default when you start Flicker is to load the two human plasma gel images.

Related vignettes

Flicker Vignette - How do I compare two images?

This vignette shows how do you can compare two images.

Method

1. Load the two images you want to compare (see vignettes to load gel images if you want different images).
2. Position the left or right image to the spot you are interested in the scrollable image window (see vignette for positioning gel images).
3. Then position the other image so it is positioned at the spot you think corresponds or the

region roughly corresponds
4. Enable flickering by enabling the **Flicker (C-F)**), typing (C-F)), or selecting (**View | Flicker images (C-F)**)
5. Fine-tune the position of the left and right images as required.
6. When the two images are aligned, the spots that are optimally aligned may appear to pulse.
7. When you are done flickering, you can disable flickering by the same controls in step [4].

Setup for the demo

You need to have two images loaded in Flicker. The default when you start Flicker is to load the two human plasma gel images.

Related vignettes

- Vignette for loading gel images
- Vignette for positioning gel images in the left and right scrollable windows

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Contact us | Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker | Revised: 01/19/2004

Flicker Vignette - How do I query a spot's putative identity?

This vignette shows how do you can query a spot's putative identity.

Method

1. Load the two images you want to compare (see vignettes to load gel images if you want different images). One of the gels must be an active clickable map linked to a Web server. You could use, for example, (**File | Open demo images | Human Plasma | (Swiss-2DPAGE vs. Merril) gels clickable**).
2. Alternative, you can explicitly load one of the over 30 active map gel images in the left or right image. Select the left or right image. Then select the active map image using the (**File | Open active map image | ...**) command.
3. Flicker align the gels in the region you are interested in.
4. Enable the **Click to access DB** checkbox. IF flickering was enabled, it will disable

flickering.
5. Then select the image with the red "+"'s on spots that are in the active Web database.
6. Click on a red "+" for the spot you are interested in. This will popup a Web browser that links to the spot you have selected in the associated Web database. Each time you click on a spot, it will post the new data in the popup browser window.

Setup for the demo

Related vignettes

- Vignette for loading gel images
- Vignette for loading active map gel images
- Vignette for positioning gel images in the left and right scrollable windows
- Vignette for flicker aligning two gels

Contact us

Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker

Revised: 01/19/2004

Flicker Vignette - How do I assign a spot's putative identity?

This vignette shows how do you can assign a spot's putative identity from a known gel.

Method

1. Load the two images you want to compare (see vignettes to load gel images if you want different images). One of the gels must be an active clickable map linked to a Web server. You could use, for example, (**File | Open demo images | Human Plasma | (Swiss-2DPAGE vs. Merril) gels clickable**).
2. Alternatively, you can explicitly load one of the over 30 active map gel images in the left or right image. Select the left or right image. Then select the active map image using the (**File | Open active map image | ...**) command.
3. Flicker align the gels in the region you are interested in.
4. Click on the spot in the active image and use ctl-M for spot measurement.
5. Then click on the corresponding spot in the user's gel and use ctl-M for spot measurement. This will also assign the same number to the spot.
6. One can select multiple spots in both images. Selecting one at a time for the corresponding

spots between the right and left images.
7. Then select the active image by clicking on the on a red "+" for the spot you are interested in.
8. Make sure to set (**View | Set view measurement options | Use 'spot identifer' for spot annotations)** to view the spot annotations.
9. Also, enable the **Click to access DB** checkbox. IF flickering was enabled, it will disable flickering.
10. Click on the menu (**Quantify | Measure by Circle| Lookup Protein IDs and Names from acitve map server (select image))**. This will set and display the annotation of the spots from the remote database int the active image.
11. Now you must click on each measured spot in the user gel and click ctl-I or (**Quantify | Measure by Circle | Edit selected spot(s) 'id' fields from spot list(s)**). A popup 'Edit spot annotation' window asks for the annotation 'id' value. A default is provided which can be edited.

Setup for the demo

Related vignettes

- Vignette for loading gel images
- Vignette for loading active map gel images
- Vignette for positioning gel images in the left and right scrollable windows
- Vignette for flicker aligning two gels

Flicker Vignette - How do I load an active map image from the Web?

This vignette shows how do you can load an active map image from the Web.

Method

1. Select the image you want to replace with an active map image
2. Select the active map image with (**File | Open active map image | ...**) command. This will replace the selected image with the Web image. It also enables your access (in step [3]) to the

Click to access DB checkbox.

3. Enable the **Click to access DB** checkbox when you want to access the active Web site.

Setup for the demo

Related vignettes

- Vignette for querying a spot's putative ID

Contact us

Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker

Revised: 01/19/2004

Flicker Vignette - How do I load gel images from my disk or from the Web?

This vignette shows how do you can load a gel image into the selected left or right image window. Individual gel images may reside on your local disk, on the the weg. You can also load pre-defined pairs of gel images into the left and right image windows from the demo gels provided with Flicker or for the user's own gels. The user's gel images are in directories that they should copy to the installation **Image/** directory.

Method

Loading an image into the selected image window

1. Select the left or right image window by clicking on it.
2. Load the image from your file system by the (**File | Open image file**), or
3. Load the image from the Internet by the (**File | Open image URL**), or
4. Load the image from the Internet by the (**File | Open active map image**)

Loading a pair of images into both left and right image windows

1. Load the image from your file system by the (**File | Open demo images | ...**) submenu to select the image pair you want to use, or
2. Load the image from your file system by the (**File | Open user images | ...**) submenu to select the image pair you want to use

Related vignettes

Contact us

Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker

Revised: 01/19/2004

Flicker Vignette - How do I compare my own images?

This vignette shows how you can compare your own gel images using Flicker.

Method

1. Flicker is able to handle GIF, JPEG or B&W TIFF formated images. If your images are in another format, then you must convert them one of these formats.
2. First you must copy the directories of your images to the **Images/** directory where you installed Flicker. For a MS Windows system, it is generally located at
   `C:\Program Files\Flicker\Images\`
3. Then restart Flicker. This will read your image file directories to discover what you are making available.
4. Go to the (**File | Open user images | ...**) to access pairs of your images. Note that the images appear as pairs of images as described in the Reference Manual Section 4.5. Select the pair of images you wish to analyze.
5. After Flicker has loaded the images, proceed to compare them using the method described in the Vignette for comparing gel images.

Setup for the demo

Flicker is currently able to handle GIF, JPEG or B&W TIFF formated images. If your images are in another format, then you could convert them using Photoshop, ThumbsPlus (www.cerious.com) or a similar tool to one of these formats.

Related vignettes

- Vignette for comparing gel images

Contact us Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker Revised: 01/19/2004

Flicker Vignette - How do I reset the state for current images?

This vignette shows how do you can reset the state for the currently loaded gel images.

Method

1. Select the (**File | Reset images**) command.
2. This will remove the transformed and brightness/contrast images and any landmarks you may have defined for both the left and right image. It will not change your original preferences.

Setup for the demo

Related vignettes

- Vignette for reseting the view and color preferences.

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Flicker Vignette - How do I change the colors of various overlays?

This vignette shows how do you can change the colors of the various overlays including target, trial object, landmarks and measured spots.

Method

1. You can change the colors for the target, trial object, landmarks and measured spots using the (**Edit | Set colors | *object-type* | *specific colors***)
2. You can reset the colors and view overlay options using the (**Edit | Reset default view**)

Setup for the demo

Related vignettes

- Vignette for reseting the current gel images
- Vignette for saving the Flicker state in a .flk file

Contact us

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Revised: 01/19/2004

Flicker Vignette - How do I set the image window canvas size?

This vignette shows how do you can set the image window canvas size. When you change the canvas size, it changes all three left, right and flicker windows to the same size. It will also resize the main window.

Method

1. You can reset the colors, view overlay options, and canvas size using the (**Edit | Reset default view**)
2. You can change the canvas size for the left, right and flicker windows and measured spots using the (**Edit | Canvas size | Increase size (C-Numpad '+')**) and (**Edit | Canvas size | Decrease size (C-Numpad '-')**) commands.

Setup for the demo

Related vignettes

- Vignette for reseting the current gel images
- Vignette for saving the Flicker state in a .flk file

Flicker Vignette - How do I change parameters?

This vignette shows how do you can change parameters. You can reset and change various parameters.

Method

1. You can reset the colors, view overlay options, and canvas size using the (**Edit | Reset default view**)
2. You can change the colors for the target, trial object, landmarks and measured spots using the (**Edit | Set colors | *object-type* | *specific colors***)
3. You can change the canvas size for the left, right and flicker window and measured spots using the (**Edit | Canvas size | Increase size (C-Numpad '+')**)and (**Edit | Canvas size | Decrease size (C-Numpad '-')**) commands.
4. You can change the transform parameters including the zoom using the slider bars in the upper right part of the main window.
5. You can change the flicker delay rate using the Delay slider bars below the left and right images.

Setup for the demo

Related vignettes

- Vignette for reseting the current gel images
- Vignette for changing the overlay colors
- Vignette for changing the overlay views
- Vignette for changing the canvas size
- Vignette for changing the magnification of the left or right image
- Vignette for saving the Flicker state in a .flk file

Flicker Vignette - How do I change the image overlays preferences?

This vignette shows how do you can change the image overlays preferences.

Method

1. You can reset the colors, view overlay options, and canvas size using the (**Edit | Reset default view**)
2. You can change the overlay for the left, right and flicker windows and measured spots using the (**View | Set view measuremens options | ...**) options.

Setup for the demo

Related vignettes

- Vignette for reseting the current gel images
- Vignette for saving the Flicker state in a .flk file

Flicker Vignette - How do I change the image magnfication (i.e. zoom)?

This vignette shows how do you can change the magnfication (i.e. zoom) of a gel image. Using the zoom mag slider you can magnify the selected image from 1X to 10X or demagnify it from 1X to 1/10X..

Method

1. Select the image you want to zoom.
2. To magnify the image, move the zoom scroller to the right.
3. To demagnify the image, move the zoom scroller to the left.
4. To magnify the other image, select it and then do steps [2-3].

Setup for the demo

Related vignettes

Contact us

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Revised: 01/19/2004

Flicker Vignette - How do I use image processing transforms?

This vignette shows how do you can improve the visualization using image processing transforms.

Method

Using the Transform menu commands (C-T)

1. Select the image you want to position by clicking on it. The title of the window will change from black to blue indicating it is selected.
2. Make sure that you have **Allow transforms** checkbox enabled.
3. Make sure that you have **Sequential transforms** checkbox disabled.
4. Perform the transform you want to try in the (**Transform | ...**) submenu. You might try "SharpenGradient", "Average" or some of the other transforms.
5. Now, enable sequential transforms by setting the **Sequential transforms** checkbox to enabled.
6. Then perform the set of sequential transforms you want to try as in step [4].
7. Now turn off the **Sequential transforms** checkbox and you can no longer compute the sequential transform.

Using the repeated Transform menu command

1. Select the image you want to position by clicking on it. The title of the window will change from black to blue indicating it is selected.
2. Make sure that you have **Allow transforms** checkbox enabled.
3. Make sure that you have **Sequential transforms** checkbox ensabled.
4. Perform particular transform you want to repeat in the (**Transform | ...**) submenu. You might

try "SharpenGradient", "Average" or some of the other transforms.

5. Then use the (**Transform | Repeat last transform (C-T)**) command or type **C-T** repeatedly to perform that operation repeatedly as the sequential transform.

Setup for the demo

Related vignettes

- Vignette: to change parameters used in the transforms

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Flicker Vignette - How do I warp a gel image's geometry?

This vignette shows how do you can warp one image to the geometry of the other to make them easier to flicker compare. There are two warping transforms - affine that requires 3 landmarks and 6 for the polywarp. (The polywarp is not available yet- [FUTURE]). You first define the required number of N pairs of corresponding landmarks between the triangular region you are interested in warping. Then select the gel you wish to warp. You then apply the warp transform from the Transform menu. See the example in the Reference Manual.

Method

Defining the N landmarks

1. Select the left or right gel image window by clicking on it. Then select one of the N landmarks you want to use by clicking on it.
2. Select the other gel image window by clicking on it. Then select the coresponding landmark. You may have to Flicker the gels to ensure they are in fact the correct spots.
3. Add the landmark to the landmark database by using the (**Landmark | Add landmark (C-A)**) or type **C-A**.
4. If you do not like the landmark, you can delete the landmark from the landmark database by using the (**Landmark | Delete landmark (C-D)**) or type **C-D**. You only need to do this step if you are not happy with the landmark you just created.
5. Repeats steps [1] through [4] until you have the N landmarks.

Warping the image

1. Select the left or right gel image window you want to warp.
2. Make sure that you have ☐ **Allow transforms** checkbox enabled.
3. Make sure that you have ☐ **Sequential transforms** checkbox disabled.
4. Perform the transform you want to try in the (**Transform | Affine warp**) or (**Transform | Poly warp**) command.
5. Reposition the transformed image and the other image to the spot of interest inside of the landmark region.
6. Enable flickering. It should be now easier to see the corresponding spots.

Setup for the demo

Related vignettes

- Vignette for positioning gel images in the left and right scrollable windows
  See the examplein the Reference Manual.

Flicker Vignette - How do I define landmarks used in image warping?

This vignette shows how do you can define landmarks used in image warping. See the warping vignette for more information on warping.

Method

1. Select the left or right gel image window by clicking on it. Then select one of the landmarks you want to use by clicking on it.
2. Select the other gel image window by clicking on it. Then select the coresponding landmark. You may have to Flicker the gels to ensure they are in fact the correct spots.
3. Add the landmark to the landmark database by using the (**Landmark | Add landmark (C-A)**) or type **C-A**.
4. If you do not like the last landmark you just added, you can delete the landmark from the

landmark database by using the (**Landmark | Delete landmark (C-D)**) or type **C-D**. You only need to do this step if you are not happy with the landmark you just created.

5. Repeats steps [1] through [4] until you have the number of landmarks you want.
6. You can get an idea of the distortion between the two gels by having Flicker compute the least square error between the landmark sets. Use the (**Landmark | Show landmarks similarity**) command.

Setup for the demo

Related vignettes

- Vignette for warping gel images
- Vignette for positioning gel images in the left and right scrollable windows

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Flicker Vignette - How do I display a spot's intensity?

This vignette shows how do you can display a spot's intensity in the title bar of the left or right window. The default is to just show the (x,y) coordinates (raster coordinate system with (0,0) in the upper left hand corner). The density measurement is taken under the circular mask. So that needs to be set accordingly.

Method

1. Enable displaying the grayscale using the (**View | Display gray values (C-G)**).
2. This will then change the title display for the selected image to show for example,
   plasmaH.gif (105, 45) 469 tot gray-value
   or
   plasmaH.gif (177, 84) 53 mn gray-value
3. To show the total integrated gray-value, (**Quantify | Use sum density else mean density**) is enabled.
4. To show the mean integrated gray-value, (**Quantify | Use sum density else mean density**) is disabled.
5. To show the circular mask, make sure the two options (**View | Set view measurement**

options | View measurement circle). Make sure the (**Quantify | List-of-spots else trial-spot measurement-mode (C-J)**) is set to list-of-spots mode. This area is the area of pixel integration used in computing the integrated density measurement.

6. To change the new circular mask size, use the **measure circle (diameter)** slider. Note: the *NxN* is one of 1x1, 3x3, 5x5, ..., 51x51 options which is the size of the rectangle enclosing the circular mask.
7. As you move the mouse in the selected image, it will display the data in the image at that position.

Setup for the demo

Related vignettes

Flicker Vignette - How do I measure background intensity near a spot?

This vignette shows how do you can measure background intensity near a spot. The background estimate is computed under the circular mask. A separate background estimate is defined for the left and right images. Note the background measurement is used for computing background corrected density for measurements made by circular mask **(C-M)** and by region of interest **(C-R)**. It will be used for multiple measurements until you change it. If you are measuring a series of spots by circular mask, then if you change the background intensity measurement, it will be associated with all spots measured since then and saved in the spot list until (or if) you change it again.

Method

1. To show the circular mask, make sure (**View | Set view measurement options | View measurement circle**) is enabled.
2. Select the new circular mask size in the **measure circle (diameter)** slider. Note: the *NxN* is one of 1x1, 3x3, 5x5, ..., 51x51 options which is the size of the rectangle enclosing the circular mask.
3. Select the spot you want to use to estimate the background by clicking on it in either the left or right image.
4. Capture the backround estimate using the (**Quantify | Measure by circle | Capture**

background (C-B)).

Setup for the demo

Related vignettes

- Vignette for measuring circular mask intensity
- Vignette for measuring ROI intensity

Contact us

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Revised: 01/19/2004

Flicker Vignette - How do I measure spot intensity?

This vignette shows how do you can measure spot intensity under the circular mask. The background corrected measurement estimate is computed under the circular mask. Note the separate background measurement **(C-B)** is used for computing background corrected density for measurements made by circular mask **(C-M)**. The background estimate will be used for multiple measurements until you change it. If you are measuring a series of spots by circular mask, then if you change the background intensity measurement, it will be associated with all spots measured since then and saved in the spot list until (or if) you change it again. You can number the spots if the you have enabled the (**Quantify | List-of-spots else trial-spot measurement-mode (C-J)**) option should be enabled. You may also list the saved spots in the report window. Note that if you save the flicker state, then the spot list will be saved in the .flk file state and an associated .spt spot list file in the **spt/** directory. You can restore the spot list if you start flicker on the saved .flk startup file.

Method

1. To measure the total integrated gray-value, (**Quantify | Use sum density else mean density**) is enabled.
2. To measure the mean integrated gray-value, (**Quantify | Use sum density else mean density**) is disabled.
3. To show the circular mask, set (**View | |Set view measurement options | View measurement circle**) to enabled.
4. Select the new circular mask size in the **measure circle (diameter)** slider. Note: the *NxN* is one of 1x1, 3x3, 5x5, ..., 51x51 options which is the size of the rectangle enclosing the

circular mask.

5. To sequentially number and capture a series of spots, enable list-of-spots mode by using the (**Quantify | ☑List-of-spots else trial-spot measurement-mode (C-J)**) checkbox. Then it will number a series of **(C-M)** spot measurements as as "+1", "+2", "+3", etc. overlays. Otherwise, the measurement will be numbered as a "+M" overlay and data is not saved in the spot list.
6. If you want to do background correct, capture the backround estimate using the (**Quantify | Measure by circle | Capture background (C-B)**). This is valid for multiple **(C-M)** measurements.
7. Select the spot you want to use to estimate the background by clicking on it in either the left or right image.
8. Capture the background corrected estimate using the (**Quantify | Measure by circle | Capture measurement (C-M)**).
9. To measure a set of spots, just repeat subset of these steps for each new spot.
10. To list the set of saved spots in the popup report window, use the (**Quantify | Measure by circle | List spots in the spot list**). Hint: clear the popup report window first. You can save this list from the popup report window using its **SaveAs** button.
11. To export a spot list to Excel, after clearing the report window, use the (**Quantify | Measure by circle | List spots in the spot list (tab-delimited)**). Then cut and paste the data into Excel.

Setup for the demo

Related vignettes

- Vignette for measuring background intensity
- Vignette for setting the circular mask

Flicker Vignette - How do I generate a list of all measured spots?

This vignette shows how do you can mark spots and generate a list of all measured intensity values for these spots. It is similar to the vignette for measuring spot intensity for a single spot.

Method

1. Enable the multiple spot list measurements using the (**Quantify | List-of-spots else trial-spot measurement-mode (C-J)**) menu checkbox.
2. Set the measurement circle size for the size spots you want to measure using the **meas circle diameter** scroller. This sets the box around the circle as *NxN* to 1x1, 3x3, 5x5, ..., or 51x51.
3. Select the (**Quantify | Measure by circle | Capture background (C-B)**) or type (**C-B**) to capture the background value
4. Select the (**Quantify | Measure by circle | Capture measurement (C-M)**) or type (**C-M**) to capture the spot measurement value. You may use (**ALT-key** click) to both select the spot and add it to the measurement list in one operation.
5. To delete a spot, click on the spot. Then use the (**Quantify | Measure by circle | Delete selected spot from spot list(C-K)**) command. The next spot you measure will get the next spot measurement number - it does not reuse measurement numbers.
6. To edit a spot's annotation 'id' data, click on the spot. Then use the (**Quantify | Measure by circle | Edit selected spot(s) 'id' field from spot list(s) (C-I)**). If spots are selected in both images, then you can edit both spots together.
7. To edit all of a spot's data, click on the spot. Then use the (**Quantify | Measure by circle | Edit selected spot(s) from spot list(s) (C-E)**). If spots are selected in both images, then you can edit both spots together.
8. Repeatedly measure the spots you want using steps [2-7] as required.
9. See the discussion on the various spot overlay options.
10. You can review the list by first clearing the popup report window and then doing a (**Quantify | Measure by circle | List spots in the spot list**). You can also view the list as tab-delimited data that you can then either cut and paste into Excel.
11. See the discussion on the various spot overlay options.
12. You can review the list by first clearing the popup report window and then doing a (**Quantify | Measure by circle | List spots in the spot list**). You can also view the list as tab-delimited data that you can then either cut and paste into Excel.

Setup for the demo

Related vignettes

- Vignette for measuring background intensity
- Vignette for measuring spot intensity under circular mask
- Vignette for setting the circular mask

Flicker Vignette - How do I review a list of all measured spots?

This vignette shows how do you can review a list of previously measured spots. Data for each spot includes position of the spot and background circular masks, density, background density, area, means background corrected density, and circular mask size. It is similar to the vignette for measuring multiple spot intensities.

Method

1. Either load a previously saved Flicker .flk state where you had defined multiple spots, or
2. Define multiple spots using the methods described in measuring multiple spot intensities
3. You can now review the list by first clearing the popup report window and then doing a (**Quantify | Measure by circle | List spots in the spot list**). Note you can cut and paste data from the report window or save it as a text file.

Setup for the demo

Related vignettes

- Vignette: mark and measure intensity for multiple spots
- Vignette: saving text data in the popup report window

Flicker Vignette - How do I set the circular mask size?

This vignette shows how do you can set the circular mask size. The circular mask is used in measuring spot intensity.

Method

1. Select the new circular mask size in the **measure circle (diameter)** slider. Note: the *NxN* is one of 1x1, 3x3, 5x5, ..., 51x51 options which is the size of the rectangle enclosing the circular mask.
2. To show the circular mask in the left and right images, make sure (**View | Set view measurement options |** ☐ **View measurement circle)** option is enabled.
3. You can view the size of the actual circle by turning on the trial-spot mode by disabling the (**Quantify |** ☑ **List-of-spots else trial-spot measurement-mode (C-J)**) checkbox.
4. Then click on a spot and type (**C-M**) to show the circle.
5. Adjust the circle until the spot fits just inside it.

Setup for the demo

Related vignettes

Contact us

Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker

Revised: 01/19/2004

Flicker Vignette - How do I set and measure a Region Of Interest (ROI)?

This vignette shows how do you can set and measure a Region Of Interest (ROI). If you have defined the circular mask background density estimate, using **C-B**, then it will be used to do the background correction for the ROI measurement by subtracting the ROI area*MeanBackgroundDensity.

Method

Setup for the demo

1. Select the (**Quantify | Measure by circle | Capture background (C-B)**) or type **C-B** to capture the background value
2. Select the (**Quantify | Region Of Interest (ROI) | Set ROI ULHC (C-U)**) or type **C-U** to define the upper left hand corner.
3. Select the (**Quantify | Region Of Interest (ROI) | Set ROI LRHC (C-L)**) or type **C-L** to define the lower right hand corner.
4. Select the (**Quantify | Region Of Interest (ROI) | Capture measurement by ROI (C-R)**) or type **C-R** to capture the background corrected density within the ROI.

Related vignettes

- Vignette for measuring background intensity
- Vignette for setting the circular mask

Contact us

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Revised: 01/19/2004

Flicker Vignette - How do I save the state of Flicker?

This vignette shows how do you can save the state of Flicker so you can continue your data-mining session at a later time. The state is saved in a .flk file in the **FlkStartups/** subdirectory in the Flicker installation directory. You can later restart Flicker to return to this state by either clicking on the .flk file which starts Flicker on this data, or using the (**File | Open state file**) command which requests the name of the .flk state file. In addition, every time you exit Flicker, it automatically saves the current user preferences in the **Flicker.properties** file. This file is read each time Flicker is restarted. If you are starting Flicker with a .flk startup file, it will overide these user preferences and use the values in the .flk file.

Method

1. Select the (**File | SaveAs state file**) command
2. Specify a .flk file (e.g., "MyData.flk").
3. When you are done, exit Flicker.
4. You may restore when the state when Flicker is run in the future either by clicking on the "MyData.flk" or by specifying the file in (**File | Open state file**) command.

Setup for the demo

Related vignettes

- Vignette: restarting Flickering using the .flk startup file

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Contact us

Flicker is a contributed program available at open2dprot.sourceforge.net/Flicker

Revised: 01/19/2004

Flicker Vignette - How do I restart Flicker on a saved state?

This vignette shows how do you can restart Flicker on a previously saved session so you can continue your data mining session. The state should have been saved in a .flk file in the **FlkStartups/** subdirectory in the Flicker installation directory using the (**File | Save(As) state file**) commands.

Method

1. Save the state of a Flicker session in a named file using the (**File | Save(As) state file**) command. It will ask you for a file name (e.g., "MyData.flk").
2. You may restore when the state when Flicker is run in the future either by clicking on the "MyData.flk" or by specifying the file in (**File | Open state file**) command.

Setup for the demo

Related vignettes

- Vignette: saving the Flickering state in a .flk startup file

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Contact us

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Revised: 01/19/2004

Flicker Vignette - How do I save transformed or overlay images?

This vignette shows how do you can save transformed or overlay images.

Method

Save the transform image

1. Select the (**File | Save transformed image**) command. This saves the transformed image in **tmp/** as a .gif with the same name of the input image.

Save the overlay image

1. Select the (**File | SaveAs overlay image**) command to save the overlay image as a .gif image. You must specify the .gif image file name.

Setup for the demo

Related vignettes

Flicker Vignette - How do I save text in the popup report window?

This vignette shows how do you can save text in the popup report window.

Method

1. Press the **SaveAs** button. This will request a file for you to save it as. The default file is **tmp/FlickerReport.txt**.

Setup for the demo

1. If the popup report is not visible, select (**View | Popup scrollable report**) to pop up the window.

Related vignettes

Contact us

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Revised: 01/19/2004