

Leica DFC Cameras

Image Acquisition



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Symbols



Warning

This symbol indicates especially important information that, if not observed, can lead to malfunction of, or damage to, the equipment.



Useful Information

This symbol indicates additional information or explanation.

Action

This symbol within the text indicates that certain operations actions must be carried out.

Introduction

Leica DFC Cameras are controlled by DFC Twain camera software. DFC Twain supports image acquisition under Leica IM and Leica QWin and image editors such as Imaging ®, Photoshop ® and Paint Shop Pro ®.

The information provided in this Leica DFC Camera Acquisition Manual is subject to change without notification and does not present Leica Microsystems Imaging Solutions Ltd with any binding commitments. The software described here is a licensed product, and may be used only in accordance with the conditions as defined in the license agreement.

Information about DC180 – 480 and DC500 cameras can be found in the Leica DC Manual.

DFC Twain Camera Controls

The Leica DFC Twain user interface will look similar to the screenshot below although the size may be smaller.



The camera control panel is on the left, and the camera live image is on the right.

The name of the camera currently active will appear in the DFC Twain title bar at the top.

Control Structure The graphic control elements laid out in the upper part of the control panel are used to control the standard functions of your image source. These include controls for exposure, color, contrast and image acquisition, and a live image histogram, showing the light intensity across the live image.

In the lower section of the control panel, detailed parameters of the live and captured images including size, color, cropping, shading, sharpening and focusing can be controlled here.

Multiple Cameras If you have more than one camera connected and no live image appears in DFC Twain (or you want to change the active camera), you will need to activate it by right-clicking on the required camera,



or right-click "Active Camera>>Configuration" and highlight the camera you want and click "OK".



Control Panel

The individual functions of DFC Twain are accessible as branch entries in a tree structure. The main branches (Captured Image, Live Image, Extra) are opened/closed by double-clicking or clicking "+" or "-".



Tree structure with main branches (collapsed)



Tree structure with main branches and functions (partially expanded)

Popup menus Many functions or status messages contain further subfunctions or settings. These sub-functions are ordinarily hidden and are accessible by right-clicking on the required function.



Context menu with important functions

Controls within the live image window

The live image window also contains control elements. These controls may be selected from a context menu, activated after drawing a rectangle within the live image area with the mouse (left-click and drag).



Context menu shown after drawing rectangle

Each function has its own rectangle color and is active until switched off again in the control panel. This means there may be more than one rectangle within the live image window.

Rectangle control The position and size of these rectangles can be changed at any time:

- Left-click on the colored rectangle to activate it.
 When active, the rectangle assumes a doted line.
- Move the rectangle to any position by left-click and dragging the dotted line.
- Resize the rectangle by left-clicking on one of the black square dots around it and dragging.



Control structure

Histogram The histogram shows the light distribution in the live image, from the darkest components on the left-hand side to the brightest components on the right-hand side, (indicated in a grayscale bar). The higher a peak in the histogram, represents more pixels of that tonal density. A well-illuminated image should contain all components from dark to light.



Live image histogram

Automatic exposure

The controls described below help to create the best conditions for image acquisition.

DFC Twain uses intelligent automated systems to analyse the current light conditions and control exposure time, image amplification, contrast and gamma values to optimise the resultant image.

There are two controls above the live image histogram: a slider for brightness and an automatic exposure icon.

 To activate automatic exposure, click on the icon above the histogram.

When automatic exposure is switched on, the image brightness is displayed as a percentage. This is a target value that the automatic adjustment of exposure attempts to achieve.



Activation of automatic exposure

Manual exposure

Automatic exposure offers the best overall image quality for most acquisition purposes. However, in situations with difficult light conditions, you may wish to adjust the individual control parameters. If automatic exposure is switched off, the slider determines the absolute exposure time in the range of microseconds and seconds.



Automatic exposure switched off

Manual contrast andIn addition to manual exposure you can switch off thegamma valueautomatic contrast and gamma functions.

 Right-click in the histogram and choose "auto contrast" or "auto gamma" to toggle the functions on or off individually.



Activate contrast and gamma control

A small check-mark indicates the control is active.



Please note that the triangle position indicators below the histogram are grayed out when automatic contrast and gamma are switched on. When the automatic functions are switched off, the triangles are shown red and will respond to mouse commands.



Automatic functions on: Indicators are positioned automatically



Automatic functions off: Indicators can be moved with mouse

Contrast

To obtain the best image, the contrast indicators (left and right) should be aligned with the limits of the brightness distribution as shown in the example below. (In automatic mode, this is performed constantly on the live image.)



Gamma

The gamma value helps to brighten or darken specific light intensities within an image. Depending on the acquisition situation, dark areas in images often appear too dark for the human eye. The gamma value is displayed as a gray indicator between the two contrast indicators. Moving the gamma value allows selective brightening of these areas.

The gamma value depends on the position of the contrast indicators. A gamma value of 1.0 is set when the gamma indicator is exactly in the middle of the two contrast indicators. Moving it to the left reduces the gamma value, moving it to the right increases the gamma value.

The automatic gamma value function calculates an optimum gamma value from the composition of the current brightness distribution.

- **Display values** The current values for the contrast indicators (black dot, white dot) and the gamma value can be displayed or hidden in the histogram. The values for the contrast indicators vary between 0 and 100, and those for the gamma value vary between 0.1 and 10.
 - Select the context menu by right-clicking in the histogram diagram and select the "Show values" command. A check-mark will indicate it is active.



Managing Configurations

Once you have defined the best source settings for your environment, you can save them and retrieve them at any time. This feature is not only useful for different working and lighting conditions, but can also be used to create reproducible user- specific working conditions.

- To save a configuration, right-click "Configuration" and select the "Save / Delete...".
- Give a name to the current settings and save.



configuration menu

Once a configuration has been saved, it is entered into a list of settings that can be retrieved at any time, as above.

You can thus activate or deactivate setting combinations for contrast, gamma and exposure.

Image acquisition

 To acquire an image with the selected settings for your archive or the current application, click "Acquire".



Depending on the camera type and the settings you have selected, this process may take anything from a few seconds to several minutes. Sometimes the image will be exposed several times, each color will be scanned separately, and a high resolution will be achieved using chip –offset techniques. These processes can take a long time to execute, even on fast computers, and if so, will display a progress bar.

If "Close after Acquire" is checked, DFCTwain will close after acquisition, otherwise the system is ready for the next image acquisition.





If "Close after Acquire" is checked, DFCTwain and the camera will take the same time to reinitialise, as if from the start.

Control Function: Captured image

The properties relating to the captured image can be found in this command section. The live image and captured images are functionally separate and so the live image is not always identical to the captured image.



Properties of the captured image

Resolutions

Image modes include the size and whether the image is high quality or binned or partial image.

🖻 Captured I	mage	▲
1392×1	■ 1392 × 1040.	Eull Frame HO
White Ba	1392 × 1040,	Full Frame
Color de	696 x 520,	Binning 2x2 HQ
Image T	696 x 520,	Binning 2x2
Scaling	348 x 260,	Binning 4x4 HQ
Sharpen	348 x 260,	Binning 4x4
👘 📕 Crop to Å	ROI	

White Balance

The White Balance function performs a color correction in the image source in order to display all neutralcolored components of the image (all gray tones, from black to white) without any color tone. Thus, the term white balance is not strictly correct, neutral balance would be more accurate. However, as the term White Balance is used worldwide, we will continue to use it here.



The White Balance procedure is one of the most important settings for acquisition of a well-balanced color image. It can either be applied to specific detail or region, or to the whole image.

Please note – for brightfield images, colour balance is achieved when a DLF (day light filter) is in the point of illumination.

In order to define the White Balance over a region of interest (ROI), left-click and drag the mouse to define the area in the live image. Release the mouse and select "White Balance".



Live image with ROI for white balance

The live image displays the corrected color.

White Balance may also be applied to the whole image. In this case, you do not need to draw an ROI, but only to click on the White Balance icon at the top of the control panel (see below).



White balance Once you have executed a white balance operation, you can save the settings in order to reactivate them at any time.

- Right-click "Captured Image>>White Balance" for choices.
- Use saved White Balance entries by clicking on the appropriate name.
- Save / Delete Use this to save the current white balance settings under a name for retrieval or deletion at any time.



Saving a white balance

Reset This resets the image source to its default settings.



The reset command may cause the image to suffer a severe color tone shift. If this happens, you should execute another white balance operation.



White balance is not possible if there are too many under-exposed or over-exposed pixels in the measuring area. A message will inform you if this is the case and the white balance values will be reset. Change the brightness of the picture or select a different measuring area and repeat the white balance.

	White balance failed!
lmage o overexp	contains too much under- or losed pixels!
Change again.	brightness or exposure time and try

Shading Correction

The brightness distribution over the whole image area is seldom homogeneous (because of the lighting, optics, camera adapter, etc.) and Shading Correction corrects this unevenness. To create a perfectly corrected, homogeneous image, a white image, ie an image of the exposure light only and no subject, is taken as reference and calculated into the live and captured image.



Without shading correction



With shading correction

Some cameras require a black image to be acquired followed by a white image before shading correction can be activated.



Select black reference

Shading correction Black Reference

- Select "Get black reference" and ensure that no light falls on the camera. For example, cover the camera lens with its cap.
- Press "Start".

Get black	Reference
Ensure that no light I	falls onto the camera
sensor while acquirir	ng the black reference
sensor while acquirir	ng the black reference

"Get black reference" in progress

If successful, click "Close", otherwise an error message will be displayed if any light has entered the camera, and you should repeat the process.

Get bla	ack Reference
Black reference h acquired.	as been successfully
	<u>C</u> lose

Shading correction Get the white reference in a similar way. White Reference

Captured Image	
1392×1040	Full Frame HQ
White Balance	(Temporary)
Shadin - Color (Get sl	hading Reference
Image • (None Scaling	e)

Follow the onscreen instructions to set the white reference. Avoid any under or over-exposed areas in the image. Change exposure or illumination brightness if necessary.

The window will freeze until shading references for all image resolutions are complete – this may take a few minutes.

If the white shading reference was successful, chose a name for the shading reference, and save it to finish the white shading reference procedure.

Save / Del Shading re	lete ference
<u>N</u> ame: Shading 1	-
Proceed	
Delete	

You can now choose and activate the shading correction you have just created from the Shading menu or record further shading references:

White Bala	nce «Unknown»
Shadipo	Shading 1
Color	Get <u>b</u> lack Reference
Image	Get <u>w</u> hite Reference
Image Sharp	Shading 1
Crop t	« None »
Live Image Extra	

Activate shading correction



Note that image acquisition using shading correction takes a little longer than normal image acquisition, as the black and white references must be calculated into the original image.



Shading corrections become invalid if you change illumination, objective, zoom position or focus! Once shading is set, you should not change any of these parameters.



Image needs shading correction



Improved by applying shading correction



You can apply the shading correction to the live or captured image.

Check "Apply Shading Correction"



or select "Proof" mode in the live image section.



If the Proof Mode is activated all image correction and parameters are applied to the live image. This mode is very slow but allows you to check crop, shading, sharpening etc, as applied to the captured image.

Color Depth

Depending on the camera type, an image can be acquired and saved at various color or grayscale resolutions. Most image sources only offer a color depth of 8 bits for grayscale images and 24 bits for color images. However, when using professional cameras, it is possible to acquire images with higher dynamic ranges, e.g. with 12, 14 or 16 bit resolution.

Selecting the color depth sets the image source to the corresponding image acquisition mode. The live image does not reflect these settings.



Color depth selection



Only a few image file formats such as TIFF, permit storage of dynamic ranges higher than 8 bits (B/W) or 24 bits (color). Other formats, e.g. JPEG, (compressed), or BMP, do not support these dynamic ranges. If an image is acquired in a higher dynamic range and is saved in one of these formats, the following dialog box is displayed.

nvalid image fo	rmat 🔀
The image you we the chosen file for	ould like to save, can not be saved in rmat.
_Image in the me	emory
Color	48 bits / pixel
File format: Win	dows bitmap
– File format: Win	dows hitman
Color	
C Grayscale	³² <u>▼</u> bits / pixel
	3
	Yes No 1

You must decide whether the color depth should be reduced to 8 or 24 bits, or whether to discard the image. Choose a compatible image data format or reduce the color depth in the image settings.



Only a few image manipulation software packages, such as Photoshop, are able to handle color depths greater than 8 bit or 24-bit respectively. If an image is acquired with a color depth greater than the image editor allows, an error message may appear, the captured image may be ignored or the software may even crash. DFCTwain does not detect whether or not the image editor can handle higher color depths correctly.

Image Type

Image Type determines whether the image is acquired as a grayscale or color image.



Selection of image type

Image size The image resolutions available depend on the camera model. Special cameras are capable of achieving true image sizes that are multiples of the actual chip size by means of CCD chip movement. Other cameras are able to combine several pixels to form a new pixel, thus increasing sensitivity but at a smaller image size (binning).



Selection of the image resolution

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Higher image resolutions require more memory and recording, reading and writing times are substantially longer.



Captured Image resolution settings do not affect the live image and vice-versa.



Depending on the camera model, binning mode may be available when selecting an image resolution. Binning is a special technique in which several pixels are combined and thus a very fast data download is possible. However some color information is lost and the size of the image is limited.



With short exposure times the live image refresh rate can be almost doubled by activating binning. With long exposure times, however, the faster read out of the sensor data allowed by binning does not affect the live image refresh rate so much.

Scaling

Scaling allows you to scale or enlarge an image by a certain factor. Scaling should not be applied if some additional image manipulation will be applied to the captured image. If however the captured image is going to be printed or used in some other layout software, you may apply scaling and / or sharpening as it is convenient for you. Please note that scaling by a factor 1.5 enlarges the size of an image by factor two. An image of size 4 MB for example will become approx. 9 MB!



Sharpen

Images of certain subjects can be technically correct, but can give a rather dull impression. Sharpened pictures, however, often appear crisper for focusing and for viewing.

Select ROI Activate Sharpen by right-clicking on "Sharpen" and selecting the appropriate level of sharpness.





The effects of sharpening are not visible in the live image, but only in a 1:1 scale reproduction of the image acquired. Each pixel in the image corresponds exactly to each pixel displayed on the monitor.



2:1 representation of an unsharpened image



2:1 representation of the same image, with sharpening

Crop to ROI

Every image source has different aspect ratios for image acquisition. The Crop function allows you to acquire any image size or format.

Select ROI Activate the "Crop to ROI" by the check-box on the control panel.

You will see a rectangle on the live image and this is the area that will be acquired.

The whole image will be acquired if "Crop to ROI" is not checked.



ROI for image acquisition

There are several ways of selecting the size and position of the Crop window:

Manual ROI definition

▶ Select "Set Rectangle...".

Captured Image	
670 x 604	Full Frame HQ
White Balance	(Temporary)
Shading	(None)
Color depth	8 Bit/Channel
Image Type	Color
Scaling	1.00
Sharpen	Off
Crop to Cost	Pectanole
🗄 Live Imag	Coccangion
🛨 Extra	

 Enter the values for the position (left, top) and size (width, height) in the appropriate fields.

<u>L</u> eft:	357	-
<u>T</u> op:	334	*
<u>W</u> idth:	621	*
<u>H</u> eight:	782	*

Manual ROI definition

The ROI rectangle is updated automatically on the live image.

Drawing ROI The Crop area may also be drawn directly on the live **on the live image** image using the mouse.

 Click on the frame of the Crop area, this marks it so that it can be moved.

or

 Draw a new rectangle at the position required using the mouse and select "Region of Interest".



Draw new ROI using mouse on the live image

Control Function: Live image

The live image shows the image as supplied by the camera. Depending on the type of camera, the live image may differ from the subject significantly, for example, if the camera supports various different resolutions. The main task of the live image is a visual check on the image quality, i.e. focusing, framing, etc.

The quality of the live image display is heavily dependent on the camera's specification. Each camera specification is different.

🖃 Live Image		
2088×19	Gull George	
Mode	• 2088 × 1550,	Full Frame
🗌 🗌 Under/ov	2088 x 1550,	Full Frame HQ
📃 Find Foci	696 x 516,	Progressive VGA
📃 Check Co	1044 x 772,	Color Binning 2x2
Apply sh-	1044 x 772,	Color Binning 2x2 HQ
🖃 Extra	696 x 514,	Color Binning 3x3
🔲 Always lir	696 x 514,	Color Binning 3x3 HQ
Flip Verti	520 x 384,	Color Binning 4x4
Flip Horiz	520 x 384,	Color Binning 4x4 HQ
Color circ	1044 x 775,	Progressive Red
Close aft	1044 x 775,	Progressive Red HQ
EP About	1044 x 775,	Progressive Green
	1044 x 775,	Progressive Green HQ
	1044 × 775,	Progressive Blue
	1044 x 775,	Progressive Blue HQ

Some resolution modes offer color or monochrome binning, sub-sampling, high/low bit depth and color separation.

Some cameras may offer several live image formats that are not available for capturing and vice-versa.



On some cameras, the brightness of live image and capture image may appear to be different. In such cases make sure to select the same image format in live and in capture image, eg. 2x2 binned for live and for captured image.

Special Image Formats

🖃 Live Image		
640 x 480	4000 - 4004	5.85. S
Mode	1280 x 1024,	Full Frame
Under/ov	1280 x 1024,	Full Frame HQ
📕 Find Focu	640 x 512,	Subsample VGA
📃 📕 Check Co	640 x 512,	Subsample VGA HQ
🗌 Apply sha	● 640 x 480,	Center Scan VGA
🖃 Extra	640 x 480,	Center Scan VGA HQ
📄 Always liv	800 x 600,	Center Scan SVGA
📄 Flip Vertic	800×600 ,	Center Scan SVGA HQ
📃 Flip Horiza	1024×768 ,	Center Scan XGA
Color circl	1024×768 ,	Center Scan XGA HQ
Close afte	r acquire	

Some cameras offer special image formats for live and / or for capture images:

Center Scan Modes: display only the center areas of the CCD for better focusing.

Progressive (Fluoro) Modes: The Progressive modes only display pixels with a certain color filter. The Progressive Red mode, for example, discards all the green and blue pixels of a CCD and combines the remaining red pixels to form a grayscale image.



Mode

This function lets you select different live viewing modes:



Standard: default setting which should be used most times. In Standard mode, the live image fits-to-window when the window is resized.

Proof: In Proof Mode all captured image correction and parameters are applied to the live image. This mode is slow but allows you to check crop, shading, sharpening etc.

Fullscreen: Displays the live image in fullscreen on the monitor. Press ESC to return to Standard mode.

Standard – Pan: The live image displayed remains the resolution selected even when the window is resized. Navigate around the live image using scroll bars, or leftclick and drag within the live image window.

Overexposure/underexposure

This function helps to find areas of the image which are underexposed or overexposed. In conjunction with the live histogram, this function assists in adjusting the lighting conditions to the dynamic range for the camera.



Activation of over- / under-exposure display

Underexposed areas of the live image appear blue and overexposed areas appear red.

Find focus

This function helps you find the best focus at a point in the image. It uses a user-drawn rectangle within the image to find the optimum focus and displays the focus in the form of a yellow vertical bar. The better the focus in the area selected, the higher the mark within the bar. A maximum level indicator helps you return to an optimum focus previously established.



Focus window with activated focus finder

 Define the position and size of the focus-measuring area, by drawing a rectangle and selecting the "Find Focus" from the menu.



Focus window, created with the mouse

Focus measurement and its measuring window can be switched on and off at any time at the "Find Focus" check box in the control panel.

Zoom Focus

Zoom Focus allows the setting of a region of interest for fast focusing. Draw a rectangle and select "Zoom Focus". The live image will be frozen apart from the selected area for fastest focusing. Note that Zoom focus is not available in binning modes.

Note that activating the zoom focus will also activate the yellow focus bar and that the live image resolution within the area is displayed in full resolution)! This change of resolution may result in a temporary gray screen until the live image is refreshed, especially if long exposure times are set! Return to normal viewing mode by removing the checkmark in the "Find focus":



Draw a region and select "Zoom Focus"



Set the best focus inside the region

Color balance

This function is used for fine-tuning the white balance. Depending on the lighting and the camera characteristics, fine correction of the image may be necessary after a white balance to compensate for any color shift. Use the mouse to drag the small square in the color wheel displayed towards the color balance you require.

From the control panel:

- Activate the Color Wheel and the measuring rectangle on or off at any time using the "Check Color" checkbox.
- Using the mouse, drag the small square to a color value and intensity and observe the small dot. The dot represents the current color within the selected area of the image.



Color fine-tuning after white balance

On the Live Image:

Adjust the measuring rectangle for the color fine-tuning by drawing a rectangle in the live image and then selecting "Check Color".



Measuring area for color fine-tuning

Control Function: Extra

Functions that support specific camera model properties are listed under in the "Extra" branch.



Always live

This function allows you to display the live image even if DFCTwain is running in the background. This may be useful when other software applications need to be running in the foreground on the computer while the live image is still displayed. In most cases however this function is not necessary and can be switched off. This means that the software running in the foreground is automatically allocated the main resources of the computer.

 Activate or deactivate by the "Always Live" checkbox.

Flip Horizontal / Flip Vertical

Depending on the installation conditions, it may not be possible to connect the camera to a microscope in a way that results in the correct image orientation. With these two functions, any camera position can be corrected by flipping the image horizontally and/or vertically as required. Activate or deactivate the functions by clicking on the appropriate check box.



Color Circle Always Visible

This turns the Color Wheel on or off, depending on how frequently it is required for color correction.

 Activate or deactivate the function by the "Color Circle Always Visible" check box.



Close after acquire

This function defines whether or not DFCTwain closes automatically after an image is acquired. This may be useful, for example, if you would like to add database information for the image immediately after acquisition.

 Activate or deactivate the function by the "Close After Acquire" check box.



About

This displays information about DFCTwain.



Hidden Service Functions

Depending on the camera, seldom-used functions may be available in a hidden menu. They are required for service purposes and contain calibration functions for which special maintenance accessories are needed. They also switch on and off cooling on certain cameras, and allow a shortened list of resolutions for the standard user.

These functions should only be accessible to trained personnel.

 Access to these functions is available by pressing the CTRL+ SHIFT keys and simultaneously rightclicking on "Extra".
 Select from the hidden function menu.

Color calibration

The color calibration function allows perfect color tuning of your camera by using a Macbeth ® ColorChecker as reference.



Color calibration

Proceed as follows:



Macbeth ColorChecker

- Place the Macbeth ColorChecker in front of the lens of your camera so that it fills as much of the image as possible. Align the columns vertically.
- Set the exposure values so that no part of the image is overexposed or underexposed.
 Do NOT perform a white balance manually as automatic color balance incorporates a white balance.
- Left-click and drag to show the calibration template in the live image. Cover the 24 color fields preciselyif not, redraw the template, and then click "Color Calibration".

Once color balance is completed, you can name the color balance setting and save it. This setting can be activated or deactivated in the white balance menu for the image acquisition parameters (see white balance functions).



Cooling

Some cameras have the ability to turn off the cooling device inside the camera. This may become necessary if you are working in a very humid environment where condensation may occur on the CCD.

Activate or deactivate by selecting "Cooling".



If the cooling is turned off, it remains off until it is turned on again.

Do not run the camera for long with the cooling turned off – it may damage the camera.



Brightness Correction -High Sensitivity Binning

When the image in is a binned mode, the intensity of the binned pixels is added together, hence increasing the apparent sensitivity of the camera. The intensity (brightness) of the image is greater than if binning were not used.

In order to keep the resulting image similar to the original as if it were not binned, we can adjust the brightness by checking "High Sensitivity Binning". (This is checked by default.)



Show All Resolutions

The number of image resolution modes varies with each camera. You can select which modes are displayed. For the advanced user, all modes are displayed, while for the standard user the set of modes is restricted depending on the camera.

Check "Show All Resolutions" to display all modes. Unchecking displays a (shorter) standard list of modes.



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