ChromaCL Software Manual



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Warnings

Before you attempt to install or operate this equipment for the first time, please make sure that you are aware of the precautions that you must take to ensure your own safety.

Safety

The following safety precautions must be observed during the operation, service and repair of this instrument. It is your responsibility to ensure your own safety, and the safety of the people working around you. If cryogens are used please refer to separate booklet *Safety Matters* that we have supplied with the system. If the system uses any Beryllium components please refer to the attached Be safety sheet

Protective ground

The instrument must always be connected to an electrical ground when it is being used, to reduce the risk of electric shocks. The ground wire (green/yellow), in the instrument power cable, must be connected to the laboratory electrical ground. Only use extension cables if they have an earth conductor. Do not disconnect the protective ground inside or outside the instrument and do not have external circuits connected to the instrument when its protective ground is disconnected.

Danger

The instrument will not stop working if the earth wire is not connected, and there is no indication that you might be in danger. Make sure that it is checked regularly (and at least annually).

Working environment

Warning Do not use electrical equipment in:

- · rain or excessive moisture environments
- the presence of flammable or explosive gases

Unless specifically stated, our equipment is not designed to be water or splash proof, or to be used in areas where there are flammable or explosive gases or fumes.

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Some internal adjustments can be made to electrical equipment supplied by *Gatan UK*. Although we do not encourage you to make these adjustments we try to supply you with enough information to allow skilled/qualified personnel to do it safely.

Danger

Lethal voltages are accessible inside the instrument. Disconnect the AC power supply before you remove the covers or fuses. It is not sufficient to switch off the main power switch. Only do this type of work if you are suitably qualified and sufficiently skilled to understand all the risks you are taking.

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All the cartons comprising the shipment should be inspected for any signs of damage before unpacking. If any severe damage is visible, the following procedures should be carried out **immediately**:

Photograph the extent of the damage. Digital photographs sent by email are often the best method.

Describe the extent of the damage to Gatan UK and / or their appointed agent

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Introduction

The standard *Chroma*CLTM product consists of a detector interfaced to the electron microscope, DigiScan II Digital Beam control system, and computer and DigitalMicrographTM software pre-installed. With ChromaCL, DigitalMicrograph software contains plug-ins for DigiScanTM and ChromaCL. ChromaCL software is written to work in a beta format for Gatan Microscopy Suite (GMS) version 1.4 and in normal configuration for later releases. GMS1.4 is intended to run on a Window PC running XP (service pack 1) or Windows 2000. Customers with earlier versions of DigitalMicrograph or using DigiScan I will need to upgrade to the latest version of hardware and software.

The manual describes software functionality pertinent to the DigiScan and ChromaCL components of DigitalMicrograph only. For details of other functionality, please refer to the DigitalMicrograph manual, tutorials or software help system. No ChromaCL help files are incorporated in GMS1.4 documentation other than this manual.

If ChromaCL is not pre-installed on the computer, special assistance should be requested from GatanUK as there may be special instructions pertinent to the release version. Note installation software is licence controlled and cannot be copied from one storage medium or computer to another.

The computer communicates with the DigiScan using a Firewire card. GMS installation wizard installs GatanFireWire adapter drivers. If this operation is successful the device manager should show the GFA as a functioning device when the DigiScan is turned on and connected to the computer. This is a prerequisite for DigiScan to function.

ChromaCL and DigiScan software for DigitalMicrograph using GMS1.4 does not provide communication to the electron microscope hardware. Hence DigitalMicrograph is only aware of the column settings, e.g. magnification, scale marker bar, kV etc if the user enters this information into dialog boxes.

1.1. Start Up

When DigitalMicrograph software is started the DigiScan should be powered on. If the DigiScan is off, then DigitalMicrograph will start but DigiScan software will not be loaded and ChromaCL will not function. It is not possible to turn the DigiScan on after DigitalMicrograph has started and expect the software to establish communication. If the DigiScan is turned on and DigitalMicrograph starts but the DigiScan communication window is not found in the windows/floating windows drop down menu, then check computer to DigiScan connections and check that the Gatan Firewire Adapter (GFA) is found in the 1394 section of device manager.

The ChromaCL detector is not "intelligent" in that there is no handshaking, firewire, serial etc. communication between the ChromaCL detector and the computer or DigiScan. Hence the ChromaCL detector can be turned on and off and this will not affect the software. The only effect on turning the detector off is that the ChromaCL image detection will disappear. If an acquisition is requested with the ChromaCL detector including the HT switch on (green light on), the dark noise from the PMT should be evident. When the HT switch is turned off, there should be no dark noise or any pulse signal.

When DigitalMicrograph software is exited the software remembers which control windows were open and their position by saving information in preference files. This information is recalled when the software is started. ChromaCL includes 3 dialogue windows which are described below. These are found under the windows/floating windows drop down menu. All windows can be moved, and maximized or minimized using the arrow button at the top of each window.

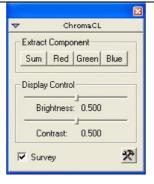
Note, to view and enable the live color mixed ChromaCL image from the DigiScan tools window, the enable tick box in the ChromaCL signal dialogue window as described below needs to be activated. These settings are remembered as saved in the preference files.

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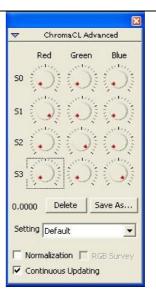
2. Key Commands



This is the standard DigiScan window and is not specific to ChromaCL systems.



This is the standard ChromaCL window.



Press Search, Preview or Record to obtain images as specified using the tools menu.

Restart will restart an acquisition according to the image window which is open.

Stop will stop the acquisition in it's tracks. To stop an acquisition at the end of a scan press the space bar.

The pixel dwell time reports that being used as specified in the tools menu. This value can be altered live and applies to the live image within the limits of the line sync protocol.

If control beam is ticked, the beam is positioned in "cross hairs mode" in a position identified in a live image with a small red cross.

Waveform monitor shows a live representation of the (analogue) signal level. This is specified by the height of the line between the bottom and half way up the screen.

The configure button opens a new dialogue menu covering service options. These values cannot be adjusted without authorization.

The tools button opens the

The ChromaCL window primarily covers post processing options.

When activated on a ChromaCL image, the extract component buttons show a black and white representation of the requested image.

The sum is simply the sum of the raw signals and is equivalent to a panchromatic image. The Red Green and Blue buttons calculate the intensity according to the 12 matrix look up table, i.e. they process the raw inputs S0-3 according the settings in the ChromaCL advanced dialogue button.

The brightness and contrast buttons apply to whichever ChromaCL image is selected, whether for live adjustment or post processing.

The tools button is used to open the ChromaCL signal dialog window as described below. The ChromaCL signals need to be specified the first time the software is used or if the raw signals which make up S0-3 are redefined for other applications.

The survey tick box applies an intelligent "auto-survey" operation either on live or recorded or saved and recalled images. This matches the

The ChromaCL advanced window may not be required for normal operation after installation.

The 12 dial positions influence the weighting given to the input channels S0-S3 to produce the core component RGB images which are mixed to form the color image. The value of each dial position when highlighted is shown.

A factory default setting is provided. These settings are also noted in the manual in case of accidental deletion. Users can save or delete alternative settings. These can be applied to live images or altered as a post processing step to recorded or saved images.

The Normalization and RGB Survey check boxes provide an alternative false colour representation of the colour image. When used the 12 dial matrix buttons act as on – off switches rather than dials with gradations.

The survey tick box on the standard ChromaCL window toggles whether the RGB survey option can be used.

The Normalization and RGB survey options differ only slightly

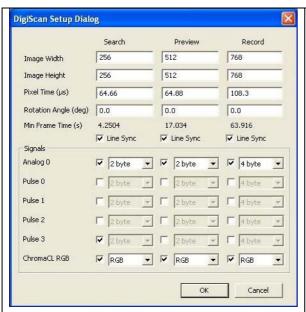
DigiScan dialogue window as described below and covers the settings for Search, Preview and Record and which images are displayed.

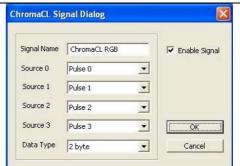
dynamic range of the input to sensible display values but also applies the colour biasing as specified using the advanced ChromaCL 12 matrix look up table. ChromaCL survey overrules Normalization or RGB survey options in the advanced ChromaCL window.

in terms of the stage in the algorithm where equal balancing to the raw components is applied.

The continuous update tick box toggles whether the software is required to update the colour image whilst the dial is turned, or not.

2.1. Tools Buttons





The DigiScan tools button opens the DigiScan Setup Dialog window as shown above. This window allows the user to define the Search, Preview and Record settings for the DigiScan acquisition.

The image width is limited between 16 and 8192 pixels. The image height is limited between 2 and 8192 pixels.

The pixel dwell time is limited between 0.4us and 400ms depending on which Signals and byte allocations are chosen.

Note although ChromaCL images can be obtained at a fast frame time, for previewing a suitable area, long pixel dwell times are recommended to achieve the best quality CL images.

The rotation angle can be specified with a minimum rotation defined by the pixel density.

The minimum frame time is automatically updated.

The line sync option tick box toggles the line sync function. When applied the frame time is automatically updated, and the start of each line frame is synchronized with a periodicity defined by the AC

The ChromaCL tools button opens the ChromaCL Signal Dialog window as shown above.

The ChromaCL RGB tick box in the DigiScan Setup Dialog Window will not appear unless the Enable Signal box is ticked in this window because this Window defines how the ChromaCL signal is comprised.

The default ChromaCL settings are as shown above whereby Source 0-3 is allocated to Pulse 0-3 with the Data Type set to 2 byte.

However the architecture of the ChromaCL RGB software does not limit the live colour mixing functionality to ChromaCL pulse inputs. The user can optionally choose to allocated other signal inputs, either analog or pulse to a particular Source. Examples of other scan synchronous inputs include SE and BS images, whilst pulse inputs may be from X ray detection hardware. (This assumes that such pulses match the TTL pulse input specifications of the DigiScan electronics).

main frequency as specified in the DigiScan configuration settings. When the line sync option is ticked the minimum frame time is limited automatically. The present version of ChromaCL software limits the user to 2 bytes for an analogue and the pulse inputs. The tick boxes allow the user to choose which of the images are displayed live. The default condition for simplicity is to show the analogue signal (e.g. SE electron), together with the ChromaCL signal.

2.2. What the software does not control or register.

Item	Notes				
The retraction or mirror status of the detector.	The retraction position is visible externally because of the position of the tower with respect to the column. A chamber scope is recommended and when turned on, does not unduly influence the ChromaCL image.				
The power to the detector or HT to the PMT.	If a ChromaCL image is requested with the detector or HT turned off, then a black result will be shown.				
The ChromaCL detector dispersion or PMT channel settings.	These are not normally adjusted for standard operation.				
The HT (and therefore gain) applied to the PMT.	This value is fixed and is not intended to be adjusted.				
Relinquish column scan control status.	For FEI microscopes and the current GMS software, the user is required to relinquish the microscope scan control in the FEI software in order to operate DigiScan. Also when the DigiScan operation is terminated, the beam will reside in the centre of the specimen in spot mode unless the FEI acquisition is regained or the column value is closed. With other columns scan control will revert back the microscope by default without user intervention.				
The SEM stage.	Montaging is not currently available.				
The SEM column settings including kV and magnification.	A correct scale marker bar requires the user to input the magnification as requested by the global microscope input dialogue. This dialogue is common the DigitalMicrograph software and is user configured.				
Overload status of the pulse counting electronics.	This is not reported formally but is evident in images because of the common presence of colour inversion as one channel saturates prior to another channel at the periphery of the bright emitting region. If too much light is present on a particular channel, the pulse counting terminates due to pulse pile up. Instead of showing saturation with high brightness, the pulse electronics registers zero signal. This causes contrast reversal and because it is channel specific is often seen as fringes of colour inversion.				

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Recommended Operating Procedure.

- If the specimen is electrically insulating, ensure that the specimen has suitable coating as required for the high vacuum and electron injection conditions of the electron microscope. Also make sure the coating is earthed to the stage. This is not necessary if working in VP mode. If required, the specimen coating should be thin enough to allow the electron beam to penetrate at a reasonable kV, and also allow CL to pass through. Note working at low kV or very low accelerating voltages to minimize charging effects as practiced for normal SEM imaging will likely be unsuitable for CL where reasonable to high injection conditions are often required to obtain the best colour contrast information.
- Turn the DigiScan and ChromaCL PC on and start DigitalMicrograph software. Ensure that the DigiScan and ChromaCL GUI are open.
- Use the SEM monitor or DigiScan analogue input to image the SE or BS image to locate the approximate area of interest. The specimen should be planar and not tilted.
- Insert the ChromaCL detector and mirror assembly as detailed in the product manual.
- Ensure the specimen is at the correct working distance with respect to the CL mirror. This should be at approximately 1mm or slightly more beneath the bottom surface of the mirror. This can be checked with a chamber scope. The chamber scope illumination may need to be turned off after use to avoid light contamination.
- Turn the power to the ChromaCL (if not already on) and HT switch on the top of the ChromaCL detector on. The green light should illuminate.
- Press the tools buttons on the DigiScan GUI. Ensure the Search, Preview and Record settings are appropriate. Ensure the Enable signal box is ticked in the ChromaCL signal dialogue window. Ensure the Global Microscope dialogue is set appropriate to the users needs. (e.g. frequency of prompting for entering magnification, kV etc).
- It may make sense at the outset to view the Pulse Inputs, for example in Search mode. Turn off the autosurvey function for pulse inputs (right mouse click on an image), if the z height and hence collection efficiency needs fine tuning. The same argument applies if a ChromaCL RGB acquisition is requested. The user should turn the ChromaCL survey feature on initially, and then off again if the z height needs optimising. This is because it is easier to optimise collection efficiency if the dynamic range of the display is approximately correct, but if the survey features which compensate for intensity changes, are disabled.
- When acquiring ChromaCL images, the user has a choice of whether to keep the Survey button on, or off. If off, the dynamic range setting used will match that previously employed. For better quality images, increase the pixel dwell time and density. If using Preview, press the space bar to terminate the acquisition at the end of a scan. It is not essential to use the Record feature in order to acquire an image worth saving.
- If saturation is occurring, consider reducing the CL intensity by using a smaller spot size, or kV. Saturation should be easy to identify because of an unexpected reversal of contrast. If unsure, reduce the injection conditions and compare the recorded image. It is best to start at lower injection conditions and work upwards (larger spot sizes) than the reverse procedure, because any saturation is easier to identify. If the user does not want to reduce the injection conditions, the collection efficiency can be reduced by increasing the working distance with respect to the mirror, and refocusing. Note, in some insulating specimens, especially those where luminescence is associated with rare earth impurities a slower scan rate will reduce the CL intensity. This effect can be substantial and is not associated with the detector or electronics, but the physics of electron beam stimulation and relaxation of the impurity species.
- The contrast and brightness of ChromaCL images can be adjusted live, or after an acquisition or after a save and recall operation. In addition, the survey, RGB Survey and Normalization functions can also be applied in addition to the contrast and brightness settings.
- Save the ChromaCL images using the file drop down menu, or automated file saving window. This automatic saving is configured using the tools button. Note analogue images and ChromaCL images are not automatically linked in any acquisition and if required will need saving separately.

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- Only export images into an alternative format if happy with the contrast and brightness settings.
- Note, do not save alternative Advanced ChromaCL dial settings using the same name as the factory
 provided name. The user can choose alternative colour biasing and save these settings using the
 advanced ChromaCL dial.

3.1. General Advice.

The cathodoluminescence spectrum and intensity emitted by a specimen prior to it reaching a detector is a complicated function of many factors and variables. For example,

Specimen, including local density, atomic weight, microstructure, topography, relative concentration and distribution of luminescence centres and luminescence killers. Also, surface quality, contamination, coating type thickness and quality.

In addition, there are more complex factors which may affect the probability of a photon of a particular wavelength being emitted.

kV, injection density and spatial resolution

Beam current, injection density affects intensity and spectrum.

Focus, injection density.

Absorption. Probability of photon of certain wavelength reaching surface

Refractive index. Probability of photon of certain wavelength leaving specimen

Temperature. Affects physics of light emission, competitive processes vary with temperature.

Time since illumination started stopped. (Phosphorescence)

Flux, beam induced changes, also for insulators.

Scanning speed. Filling emptying of traps.

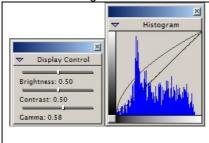
This list indicates that there are many factors, some within the control of the microscopist, and some out of his / her control, which may alter the luminescence detected. If a result is initially not understood, it is worth considering some of the above factors before assuming that the ChromaCL system is not functioning.

The most likely source of confusion is that the colour of the luminescence emitted is likely to be strongly influenced by the injection conditions. These include kV, beam current (spot size), focus, as well as scanning speed. Given the complicated nature of CL, it is wrong to assume that the colour detected should be standard no matter what the injection conditions. There is no reason to assume that the injection conditions applied, and the bias of the spectral response applied in recording CL images on film or otherwise in a flood gun system should be the gold standard for defining the expected colour of a mineral. However, as many microscopists may wish to work to this standard, the ChromaCL software allows alternative biasing using the Advanced panel.

A secondary source of confusion is that the ChromaCL image will show contrast inversion if saturation conditions are met.

4. ChromaCL images.

ChromaCL images differ from standard analogue or pulse map DigiScan images in a number of important ways.



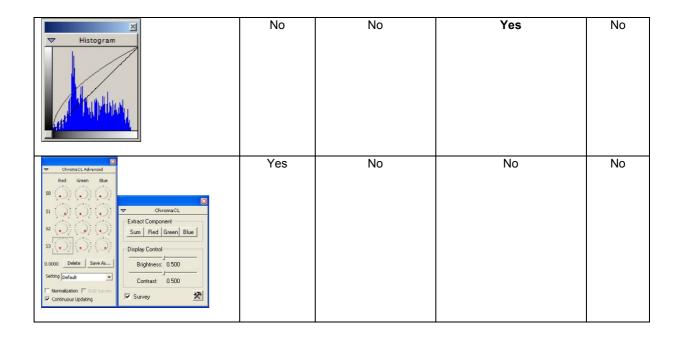
Standard DigiScan images (analogue or pulse images; not including images created using the color mix routine) can have their brightness, contrast and gamma function adjusted using the Display Control window, found under the Floating Windows drop down menu. In addition the histogram tool shows a display of the intensity distribution and this histogram can be used as a tool for adjusting the dynamic range of the image. Drawing on the histogram using the mouse can set the upper and lower limits to the intensity dynamic range, whilst clicking on the lower portion of the histogram applies the Autosurvey function as configured in the Image properties tool.

Standard DigiScan images can be shown in false color representation, (e.g. thermal or rainbow representation) as specified using the Image Display Info dialogue, or by creating using a specific color look up table using the vertical axis of the histrogram tool. Also RGB images can be created in DigitalMicrograph using the color mix function (from Analysis drop down menu), and color images such as jpegs, etc can be imported in DigitalMicrograph. In all cases, these colour representations are different to, and are not related to the ChromaCL functionality. This is because the ChromaCL and Advanced dialogue windows are specific to the large file format of ChromaCL images only.

Furthermore ChromaCL images (and in this case, similar to images created using the color mix routine) do not have their intensity distribution displayed using the histogram tool, and the standard Display Control window for contrast, brightness and gamma does not function. Instead, the contrast and brightness of ChromaCL images can only be adjusted using the Display Control sliders on the ChromaCL window as shown above. The survey functions can appear to adjust the brightness and contrast of images as would be expected from a surveying tool

These distinctions are summarized as below.

Tool.	ChromaCL RGB	Color mix RGB	Standard DigitalMicrograph analogue or pulse images.	Imported images formats, e.g. bmp, jpg.
Top of the property of the pro	No	No	Yes	No
Display Control Brightness: 0.50 Contrast: 0.50 Gamma: 0.58	No	No	Yes	No



4.1. The ChromaCL Survey and Normalization Tick Boxes.

When the Survey box option on the ChromaCL dialogue window is ticked, the Survey function is applied afresh to the selected ChromaCL image. This also applies to images which have been saved and recalled. If there is no noticeable difference in an image when the survey box is ticked, this can mean that the survey function has already been applied, or that the dynamic range of the image is sufficiently close to ideal that no noticeable change has been applied.

When the survey button is ticked, an increase in brightness, for example because the beam current, (or spot size) has been increased, will not cause a significant difference in the brightness of the image displayed. However, if the button is not ticked, then an increase in signal during a live acquisition will result in an increase in brightness.

If the Survey box is not ticked, and if a live image is being recorded, or a new live image requested, the survey settings from the last time the survey function was applied will be used. In this way the Survey function can be set to a useful setting, and then turned off if the user desires and is confident that live surveying is not required. In this way all variations in intensity if other variables are altered will be reflected in the display of the image. Furthermore the user can always request the Survey function once an acquisition has been completed.

The RGB Survey function on the advanced panel is servant to the Survey function and is disabled if the Survey function is applied. The normalization function can be applied on it's own or in tandem with the Survey function. As stated above the 12 matrix dials behave as on-off switches with the Normalization function and RGB survey functions. This is because these functions give equal weight to the either the raw components or the RGB contributions.

4.2. Image calibration.

Correct calibration of the scale marker bar requires the DigiScan to be calibrated. This is normally performed at installation but can be performed simply by the user at any time using a simple routine. The Global Microscope Information dialogue gives the user the option to be prompted for the microscope magnification as never, once, or always. There is no option for automatic communication between the scanning electron microscope column (e.g. magnification, kV settings) and DigitalMicrograph.

The calibration routine requests the user to position a line on an image of known dimension and to label the length of this line in a choice of units. The scale marker bar is requested using "edit / data bar / add scale marker

bar". This can be rescaled, reformatted and positioned accordingly.

Digital Micrograph presents the user with the option of "critical dimension" calibration. The user is able to calibrate the SEM image using a calibration standard for each kV of the microscope. For most microscopes and external scan control options, the magnification is assumed to be calibrated accurately and to remain independent of kV. In reality this is an approximation, and if required DigiScan can be calibrated for a range of accelerating voltages. This requires the user to configure and utilize the global microscope information dialogue and indicate both the kV and reported SEM magnification manually.

4.3. Image saving.

ChromaCL RGB images are comparatively large in terms of bytes. The reason for this is because the images comprise all the raw component pulse maps. This is the reason why ChromaCL images can be opened and reprocessed under different matrix or normalization options. DigitalMicrograph results (saved as *.dm3 files) cannot normally be opened using common image processing programs. To achieve this the images need to be exported to a chosen format. This is achieved using the "Save Display As" option in the File drop down menu. Files exported in this manner can be chosen as screen display or original image resolution with a choice of including annotation and specifying grey scale or color.

ChromaCL images saved in *.dm3 format cannot be opened using software other than GMS1.4 installations where ChromaCL has been installed as an on-line or off-line version. Off-line licences for ChromaCL are a cost effective way of working with ChromaCL images in the DigitalMicrograph user environment using a computer other than that attached to the hardware.

Note the present release of GMS1.4 provides a batch covert routine to export *.dm3 into common display formats. In the present release the default batch convert performs this routine in greyscale only and hence is not suitable for ChromaCL images to be converted into a format keeping their colour. This issue will be addressed in future releases.

4.4. Help

DigitalMicrograph provides a range of help files which can be accessed and searched by pressing F1, or using the Help drop down menu. At present this help file system does not include details of the ChromaCL software. Documented help regarding software operation specific to ChromaCL is limited to this manual.

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Appendix 1

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Operators Handbook

Supplied to:

Project number: