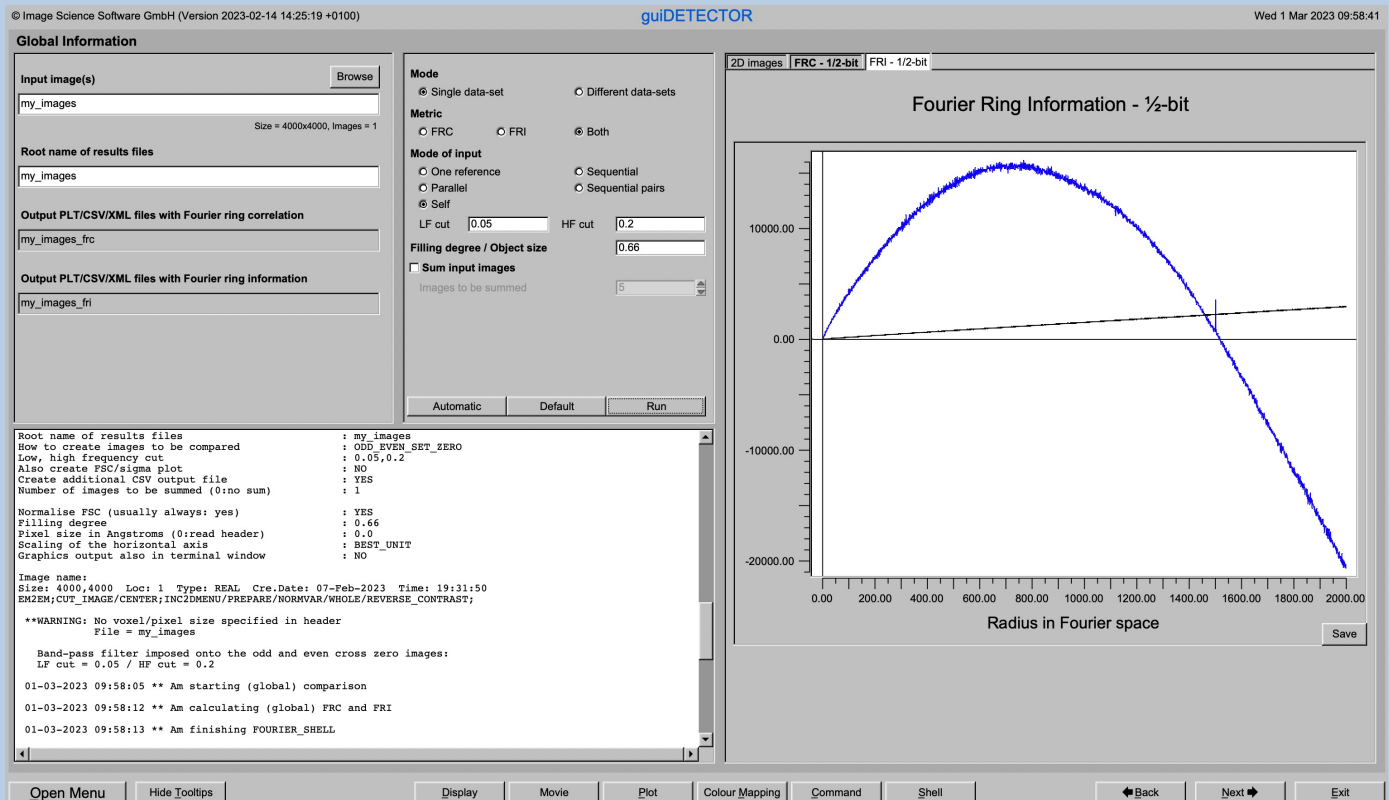


A Brief Introduction

Version 10-Oct-2023
www.ImageScience.de
© Michael Schatz (Image Science)

The IMAGIC guiDETECTOR program



The **guiDETECTOR** program offer a number of options to correct data-sets taken by a detector and/or to estimate the information content of the data and/or the quality of the detector.

This is a brief hands-on on how to use IMAGIC GUI oriented programs and how to work with **guiDETECTOR** :

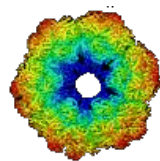
CONTENT:

- IMAGIC GUI programs How to use IMAGIC GUI programs
- **guiDETECTOR** How to use the various options of **guiDETECTOR**
- Error hints How to send us feedback

FOURIER RING/SHELL INFORMATION, FOURIER RING/SHELL CORRELATION:

Refer to our internet pages <https://www.ImageScience.de/metrics> and <https://www.ImageScience.de/fsc>.





IMAGIC

GUI Programs

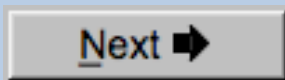


Workflow

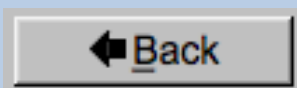
The idea of **guiDETECTOR** is to guide you through a typical camera/detector correction measurement or camera .

The workflow consists of several pages. Each page will perform a specific image processing step.

If the calculations are finished the results are shown and you can press the “Next” button to continue with the next page.



Of course, there is also a “Back” button. But be careful: when leaving a page the results shown on the page may get lost and when coming back you might have to do the calculations once more to get the results printed. The output files do not get lost, of course.



The Working Directory

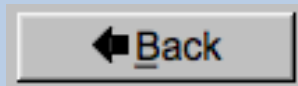
If **guiDETECTOR** is called from the programs list, by using an icon or in a command line the working directory will be your default system directory.

If **guiDETECTOR** is called by an IMAGIC command in a terminal / command window

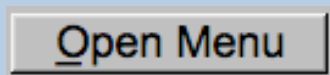
```
IMAGIC-COMMAND : guiDETECTOR
```

the working directory will be the directory used in this window.

If you want to change this directory use the “Back” button(s)



or the “Open Menu” button



to navigate to the “Start” page where you can specify the working directory of **guiDETECTOR**.

All output files will be stored in the working directory which you have specified on the start page.

Input files can be chosen from other directories.



Help

Move the cursor on (nearly) any item (questions, radio buttons, display windows...) shown on the pages and you will get context sensitive help.

Output file:

whgb_micrograph

Name of the output IMAGIC file containing the imported micrographs.

Note that the name of this output file will be created automatically.

Select format ▼

In case of type conflicts

Select the input file format.

Note: Currently only TIFF and MRC files can be imported.

MRC:
This is one of the oldest image formats in use in electron microscopy. One of the philosophies behind this data format is that it is compatible to the CCP4 format in use in X-ray crystallography.

TIFF (Tagged Image Format):
This has become one of the standard formats in desk-top publishing oriented image processing.



Input Files

Usually the input files on each page are output file(s) from the previous page(s) and are suggested automatically.

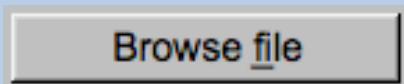
You can, of course, always use other input files names and even use other input directories.

Input file

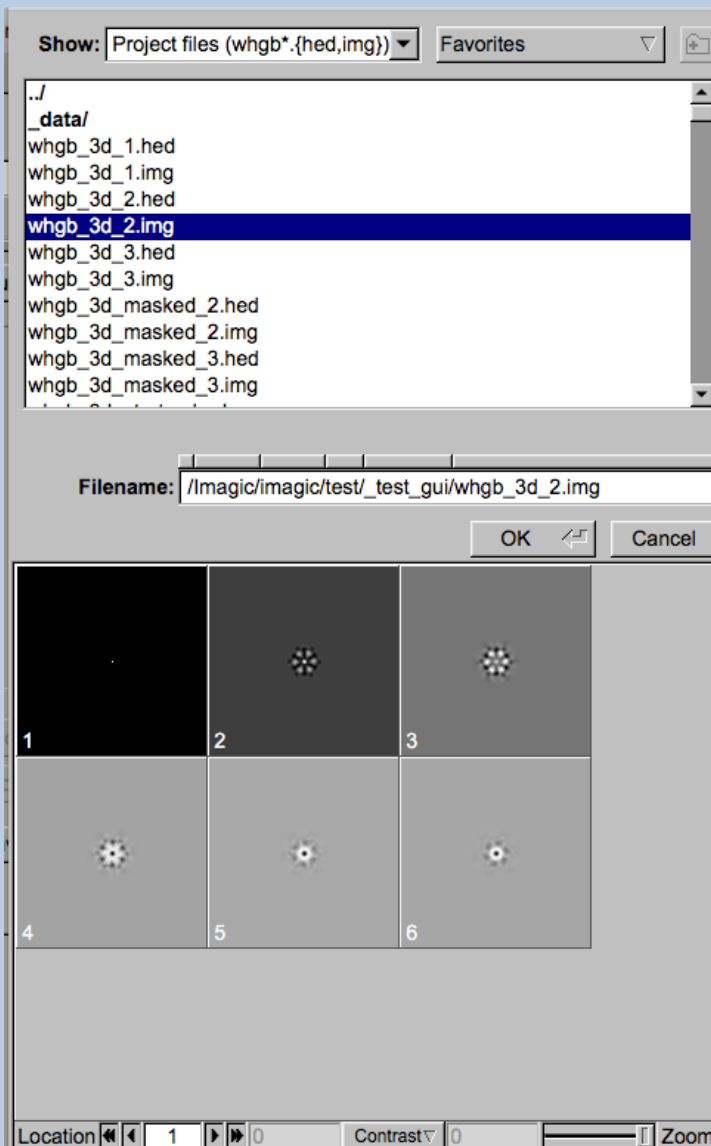


Input File Chooser

In most of the pages you are asked for input file(s) and you will find a “Browse file” button:

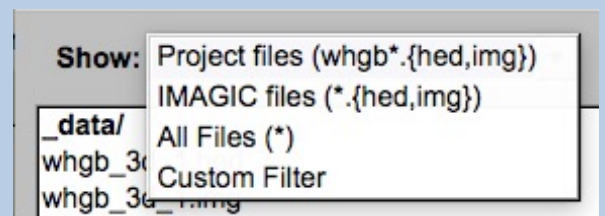


Pressing this button will open the IMAGIC file chooser:

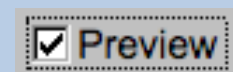


Choose the wanted file by clicking its name

You can use a pre-selection of the files shown:



If the images are in IMAGIC format you can get a pre- view of the images.



Note that you can store your directory in “Favorites”.



Output Files

Usually the names of the output files are suggested but it is your choice, of course. On each page you can specify these output file names on the left hand side.

| Output file | Export |
|-------------|--------|
| my_images | |

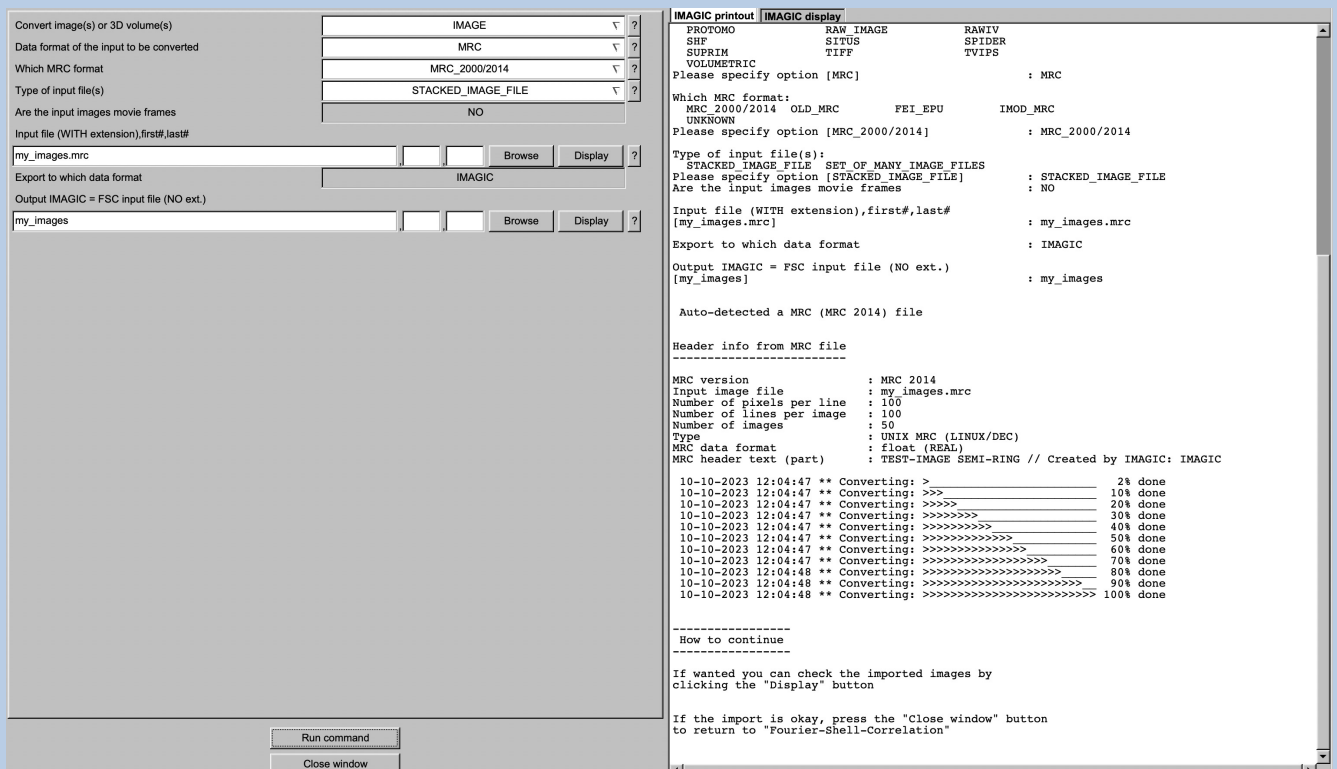


Import Buttons

You do not want to use the “Import page” you can sometimes use an “Import” button to import the input images/3D volumes from any 3DEM format. The “Import” button which is located above the text field specifying the name of the related file.



An additional “IMAGIC EM2EM” page will open. Specify all parameters needed and click the “Run command” button to import the images / 3D volumes:

A screenshot of the IMAGIC EM2EM software interface. The left pane shows a configuration window with several dropdown menus and text fields. The right pane shows a terminal window with the output of the 'Run command' button. The terminal output includes a table of MRC formats, a list of input files, and a progress bar for the conversion process. The progress bar shows that the conversion is 100% done. The terminal also includes instructions on how to continue and how to check the imported images.

Convert image(s) or 3D volume(s) IMAGE ?
Data format of the input to be converted MRC ?
Which MRC format MRC_2000/2014 ?
Type of input file(s) STACKED_IMAGE_FILE ?
Are the input images movie frames NO
Input file (WITH extension),first#,last#
my_images.mrc Browse Display ?
Export to which data format IMAGIC
Output IMAGIC = FSC input file (NO ext.)
my_images Browse Display ?

Run command
Close window

IMAGIC printout | IMAGIC display
PROTOMO RAW_IMAGE RAWTV
SHP SITUS SPIDER
SUPRIM TIFF TVIPS
VOLUMETRIC
Please specify option [MRC] : MRC
Which MRC format:
MRC_2000/2014 OLD_MRC FEI_EPU IMOD_MRC
UNKNOWN
Please specify option [MRC_2000/2014] : MRC_2000/2014
Type of input file(s):
STACKED_IMAGE_FILE SET OF MANY IMAGE FILES
Please specify option [STACKED_IMAGE_FILE] : STACKED_IMAGE_FILE
Are the input images movie frames : NO
Input file (WITH extension),first#,last#
[my_images.mrc] : my_images.mrc
Export to which data format : IMAGIC
Output IMAGIC = FSC input file (NO ext.)
[my_images] : my_images

Auto-detected a MRC (MRC 2014) file

Header info from MRC file

MRC version : MRC 2014
Input image file : my_images.mrc
Number of pixels per line : 100
Number of lines per image : 100
Number of images : 50
Type : UNIX MRC (LINUX/DEC)
MRC data format : Float (REAL)
MRC header text (part) : TEST-IMAGE SEMI-RING // Created by IMAGIC: IMAGIC

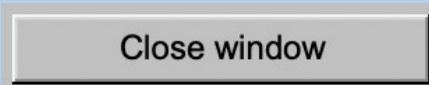
10-10-2023 12:04:47 ** Converting: > 2% done
10-10-2023 12:04:47 ** Converting: >>> 10% done
10-10-2023 12:04:47 ** Converting: >>>> 20% done
10-10-2023 12:04:47 ** Converting: >>>>>> 30% done
10-10-2023 12:04:47 ** Converting: >>>>>>>> 40% done
10-10-2023 12:04:47 ** Converting: >>>>>>>>>> 50% done
10-10-2023 12:04:47 ** Converting: >>>>>>>>>>>> 60% done
10-10-2023 12:04:47 ** Converting: >>>>>>>>>>>>>> 70% done
10-10-2023 12:04:48 ** Converting: >>>>>>>>>>>>>>>> 80% done
10-10-2023 12:04:48 ** Converting: >>>>>>>>>>>>>>>>>> 90% done
10-10-2023 12:04:48 ** Converting: >>>>>>>>>>>>>>>>>>>> 100% done

How to continue

If wanted you can check the imported images by clicking the "Display" button

If the import is okay, press the "Close window" button to return to "Fourier-Shell-Correlation"

Click the “Close window” button to exit this additional window:

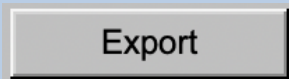


Refer to the **guiEM2EM** manual to get further help.

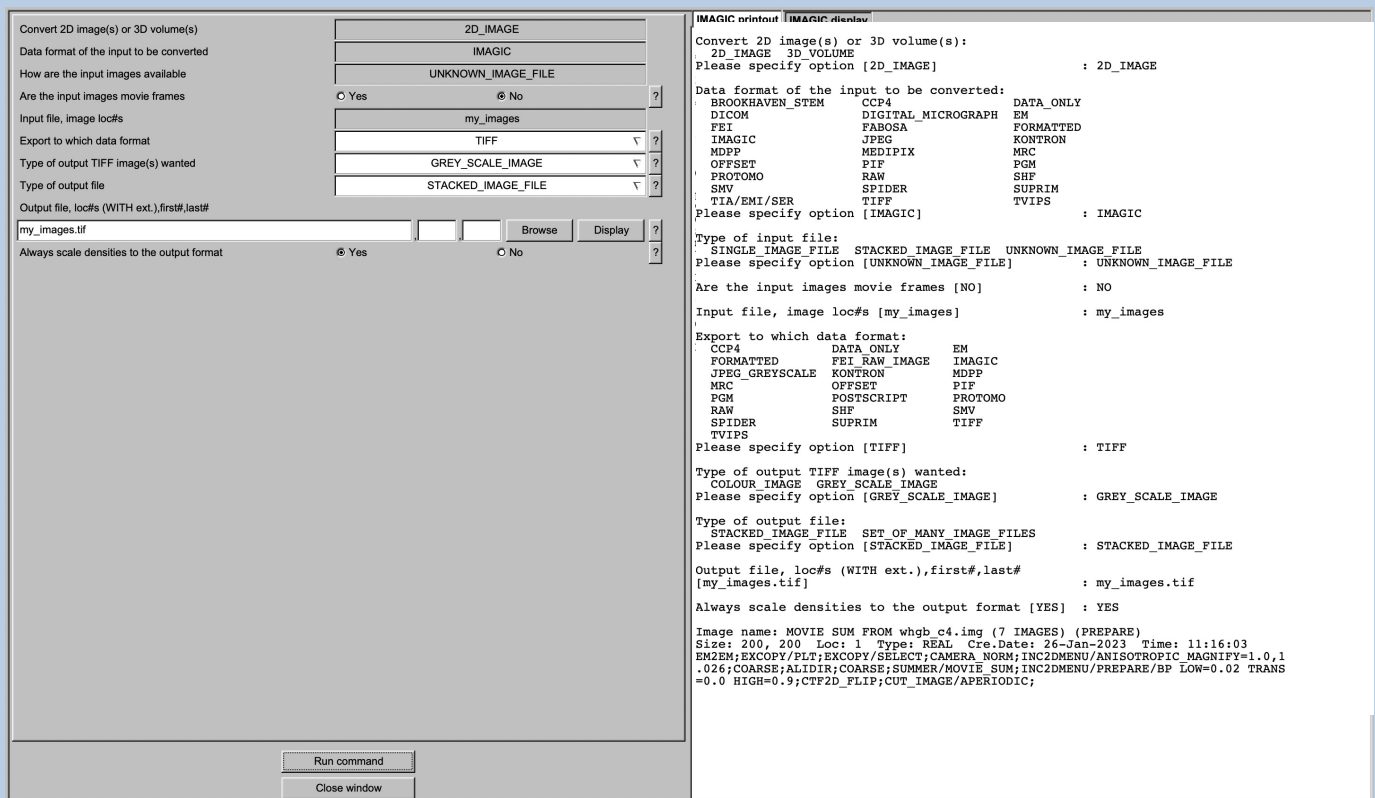


Export Buttons

You can export output images/3D volumes to any 3DEM format. Click the “Export” button which is located above the text field specifying the name of the related file.



An additional “IMAGIC EM2EM” page will open. Specify all parameters needed and click the “Run command” button to export the images / 3D volumes:

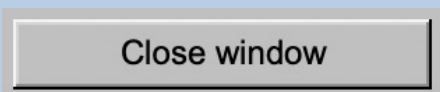
A screenshot of the IMAGIC EM2EM software interface. The window is split into two panes. The left pane is a configuration dialog with various input fields and radio buttons. The right pane shows a terminal window with the command-line output of the software. The configuration dialog includes fields for input file name, output format, and data format. The terminal window shows the execution of the 'EM2EM' command with various options and parameters, resulting in a list of image files and their properties.

Convert 2D image(s) or 3D volume(s) [2D_IMAGE]
Data format of the input to be converted [IMAGIC]
How are the input images available [UNKNOWN_IMAGE_FILE]
Are the input images movie frames [Yes No]
Input file, image loc#s [my_images]
Export to which data format [TIFF]
Type of output TIFF image(s) wanted [GREY_SCALE_IMAGE]
Type of output file [STACKED_IMAGE_FILE]
Output file, loc#s (WITH ext.),first#,last# [my_images.tif]
Always scale densities to the output format [Yes No]

IMAGIC printout | IMAGIC display
Convert 2D image(s) or 3D volume(s):
2D_IMAGE 3D_VOLUME : 2D_IMAGE
Please specify option [2D_IMAGE]
Data format of the input to be converted:
BROOKHAVEN_STEM CCP4 DATA_ONLY
DICOM DIGITAL_MICROGRAPH EM
FEI FABOSA FORMATTED
IMAGIC JPEG KONTRON
MDPP MEDIPIX MRC
OFFSET PIF PGM
PROTOMO RAW SHF
SMV SPIDER SUPRIM
TIA/EMI/SER TIFF TVIPS
Please specify option [IMAGIC]
Type of input file:
SINGLE_IMAGE_FILE STACKED_IMAGE_FILE UNKNOWN_IMAGE_FILE
Please specify option [UNKNOWN_IMAGE_FILE] : UNKNOWN_IMAGE_FILE
Are the input images movie frames [NO]
Input file, image loc#s [my_images] : my_images
Export to which data format:
CCP4 DATA_ONLY EM
FORMATTED FEI_RAW_IMAGE IMAGIC
JPEG_GREYSCALE KONTRON MDPP
MRC OFFSET PIF
PGM POSTSCRIPT PROTOMO
RAW SHF SMV
SPIDER SUPRIM TIFF
TVIPS
Please specify option [TIFF] : TIFF
Type of output TIFF image(s) wanted:
COLOUR_IMAGE GREY_SCALE_IMAGE
Please specify option [GREY_SCALE_IMAGE] : GREY_SCALE_IMAGE
Type of output file:
STACKED_IMAGE_FILE SET_OF_MANY_IMAGE_FILES
Please specify option [STACKED_IMAGE_FILE] : STACKED_IMAGE_FILE
Output file, loc#s (WITH ext.),first#,last# [my_images.tif] : my_images.tif
Always scale densities to the output format [YES] : YES
Image name: MOVIE SUM FROM whgb.c4.img (7 IMAGES) (PREPARE)
Size: 200, 200 Loc: 1 Type: REAL Cre.Date: 26-Jan-2023 Time: 11:16:03
EMEM;EXCOPY/PLT;EXCOPY/SELECT;CAMERA_NORM;INCDMENU/ANISOTROPIC_MAGNIFY=1.0,1
.026;COARSE;ALIDIR;COARSE;SUMMER/MOVIE_SUM;INCDMENU/PREPARE/BP_LOW=0.02 TRANS
=0.0 HIGH=0.9;CTF2D_FLIP;CUT_IMAGE/APERIODIC;

Run command
Close window

Click the “Close window” button to exit this additional window:



Refer to the **guiEM2EM** manual to get further help.



A Typical Page

A typical **IMAGIC GUI program** page has three columns.

The left part contains the file information and a kind of terminal window showing the print-out of the currently running IMAGIC program(s). In additional tabs you can find the control windows to adjust the displays on the left hand side.

The middle part usually contains parameters to be specified and a single or a number of “Run” buttons to start the calculation(s).

The right part displays input and output images. Sometimes it can also contain additional follow-up calculations and the related “Run” buttons.

© Image Science Software GmbH (Version 2022-11-30 18:18:14 +0100) **guiCNORM** Fri 9 Dec 2022 11:35:40

Camera Correction

Input file with (raw) micrographs
my_micrographs
Size = 4096x4096, Images = 70

Input camera statistics average file
my_micrographs_cnorm_average
Size = 4096x4096, Images = 1

Input camera statistics sigma file
my_micrographs_cnorm_sigma
Size = 4096x4096, Images = 1

Output file with camera corrected micrographs
my_micrographs_cnorm

Output good camera corrected micrographs
my_micrographs_cnorm_good

Camera Normalisation

Measure
 Correct
 Measure and Correct

Input Micrographs | Corrected Micrographs | Average | Sigma

Extract micrographs
 Use all
 Use 'good' micrographs only

Ignore micrographs which show
 too extreme sigma of densities
 too extreme min/max difference of densities
Ignore if 1.5 times sigma away from mean value

IMAGIC output | Micrograph | Corrected | Average | Sigma

```
Output file, image loc# : my_micrographs_cnorm
Input average file : my_micrographs_cnorm_average
Input sigma file : my_micrographs_cnorm_sigma
Reverse contrast in camera corrected images : NO
09-12-2022 11:33:58 ** Am correcting/normalising images
09-12-2022 11:33:58 ** Correction: _____ 1% done
09-12-2022 11:34:01 ** Correction: >>>> 10% done
09-12-2022 11:34:03 ** Correction: >>>>> 20% done
09-12-2022 11:34:06 ** Correction: >>>>>> 30% done
09-12-2022 11:34:09 ** Correction: >>>>>>> 40% done
09-12-2022 11:34:12 ** Correction: >>>>>>>> 50% done
09-12-2022 11:34:15 ** Correction: >>>>>>>>> 60% done
09-12-2022 11:34:18 ** Correction: >>>>>>>>>> 70% done
09-12-2022 11:34:21 ** Correction: >>>>>>>>>>> 80% done
09-12-2022 11:34:24 ** Correction: >>>>>>>>>>>> 90% done
09-12-2022 11:34:27 ** Correction: >>>>>>>>>>>>> 100% done
09-12-2022 11:34:27 ** Correction/normalisation done
Image name:
Size: 4096,4096 Loc: 70 Type: REAL Cre.Date: 09-Dec-2022 Time: 11:34:27
EMZEM;HEADERS/ACTIVE;EXCOPY/SELECT/SIGMA/SET_INACTIVE;CAMERA_NORM/REVERSE_CONT
RAST;
```

Open Menu | Hide Tooltips | Display | Movie | Plot | Command | Shell | Back | Next | Exit



A Typical Page - MPI Parallel

If calculations can run in parallel mode the left part of a typical **IMAGIC GUI program** page also shows the buttons to specify the related parameters.

| Run in parallel mode | | Specify path and name of MSA scratch file | |
|--------------------------------------|--------------------------------|---|-------------------------------------|
| <input checked="" type="radio"/> Yes | <input type="radio"/> No | <input type="radio"/> Yes | <input checked="" type="radio"/> No |
| Number of nodes: | <input type="text" value="3"/> | MSA scratch file: | <input type="text"/> |



A Typical Page - Program Parameters

Mode of preparation

Pretreat images

Normalise amplitude spectra (NAS)

Pretreat images

Band-pass Filter

LF cut

Rem. LF

HF cut

Normalisation

Sigma

Mask

Radius

Drop off

Test loc. # to

Run for all particles


Centre particles

Self rotate Self

Total sum Mass center

Test loc. # to

Run for all particles



In the middle part of a typical **IMAGIC GUI program** page you will find the program parameters to be used.

Radio Buttons are showing options. One option only has to be used.

Self rotate Self


Total sum Mass center

Click buttons are showing options which you can use or not.

Band-pass Filter

In text fields you can type in the wanted value. If the needed value is a number you can also move the cursor into this field, press the mouse key and keep it pressed and move the cursor to change the value.

There are also boxes where you can use up and down arrows to change the value.





A Typical Page - Automatic / Default

Resize/Coarsen micrographs
Summing parameter 2

Create patches
Size of patches 4096

Prepare micrograph
Low freq. cut 0.0200
Remaining low frequency 0
High freq. cut 0.9000

Remove outlier pixels
Outlier is 4.50 sigma off the mean value

Invert densities

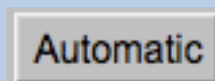
Resize/Coarsen prepared micrographs
Summing parameter 2

Automatic Default

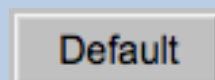
Run

In the middle part of a typical **IMAGIC GUI program** page you will also find “Automatic” and “Default buttons.

Pressing the “Automatic” button will fill in the values suggested by IMAGIC.



Pressing the “Default” button will fill in the values which you have used during the last “Run”.



The values shown when entering a page are the default values (your last values given) if they are available. Else the automatic values are shown.



A Typical Page - Run buttons

Create prepared amplitude images

Filter micrographs

Low freq. cut

Remaining low frequency

High freq. cut

Filter amplitude images

Low freq. cut

Remaining low frequency

High freq. cut

Coarsen filtered amplitude images

Yes No

Summing parameter

MSA options

MSA eigenfilter amplitudes

MSA classify amplitudes

MSA

Inner radius of ring mask

Outer radius of ring mask

Number of eigenimages

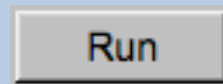
Number of iterations

Classification

Use how many eigenimages

Number of classes

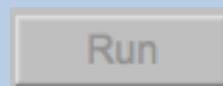
To run the calculations press the “Run” button.



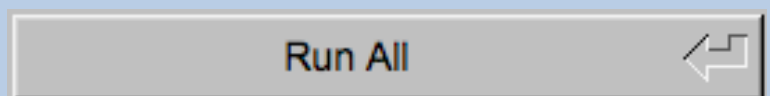
On a number of pages the calculations can be split. In this case you will find more than one single “Run” button.

Not running everything at once can be helpful when testing parameters.

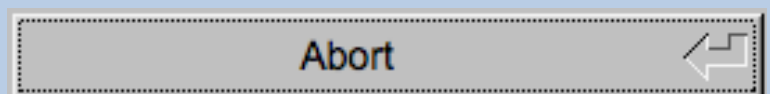
Maybe a certain “Run” button is not yet activated because it needs the results of calculations not yet done.



Pressing the “Run All” button starts all calculations currently activated on the page.

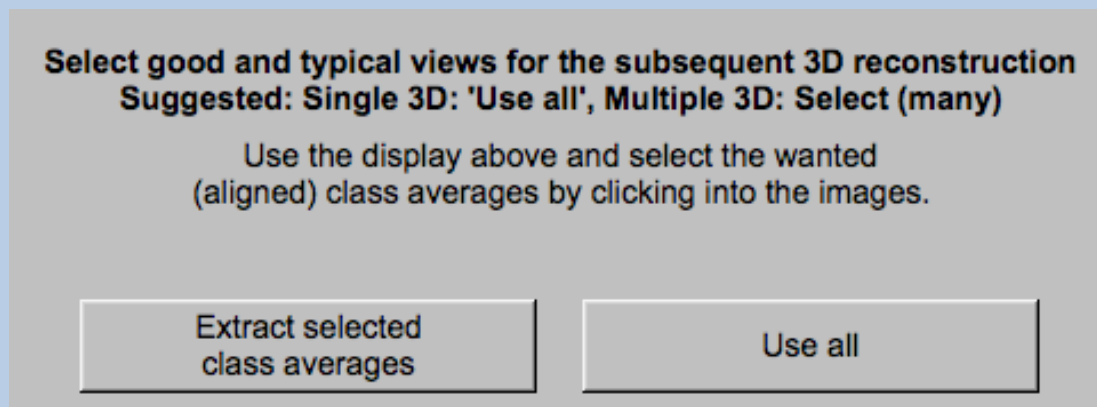


You can abort a running program by pressing the “Abort” button.



A Typical Page - Additional Tasks

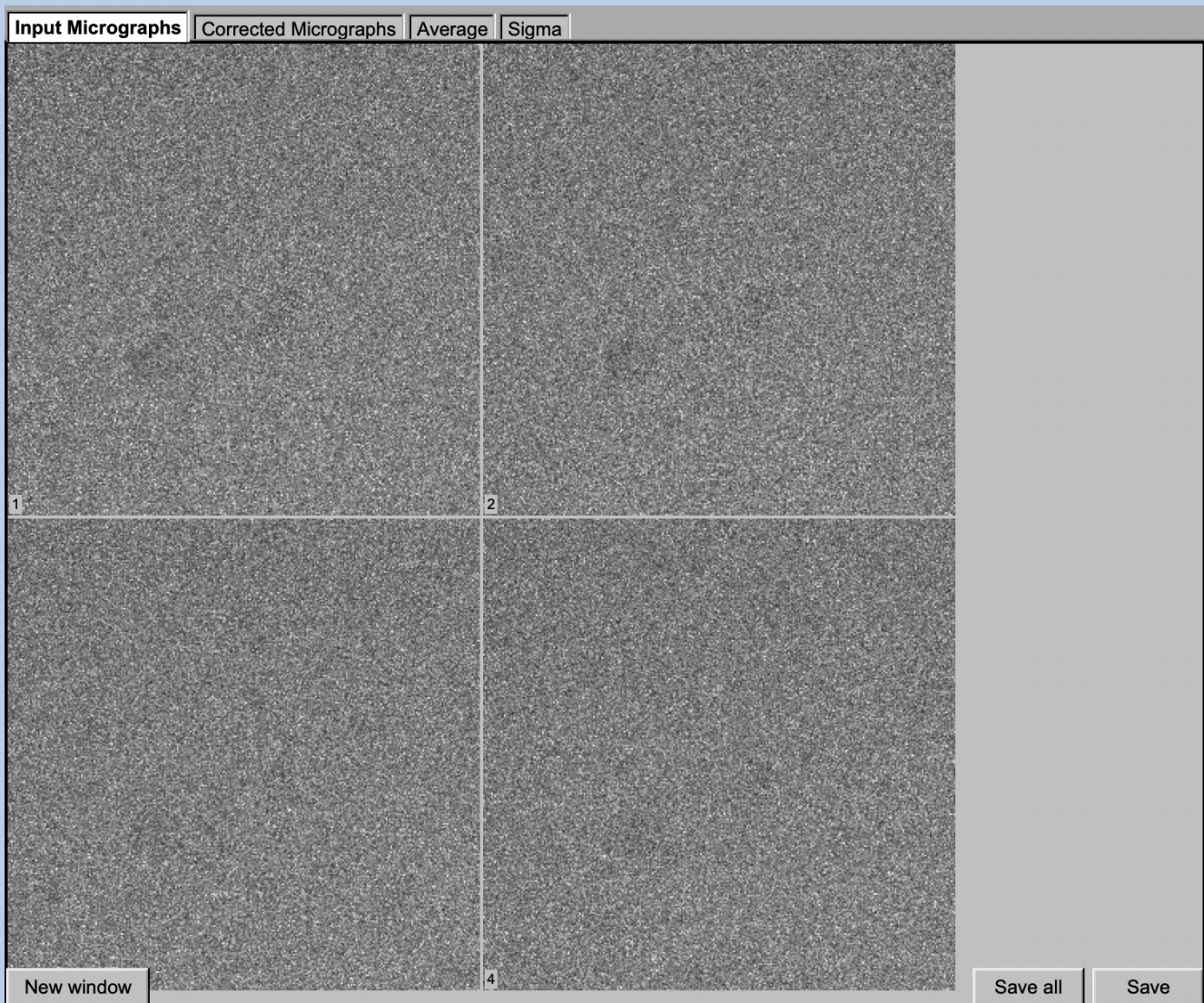
The main calculations on the page are done using the middle part of an typical **IMAGIC GUI program** page. But on a number of pages some additional calculations have to be done. Please follow the instructions given.



Note that the new output images are usually shown in a new display tab.



A Typical Page - Display



In the right part of a typical **IMAGIC GUI program** page you will find displayed images - usually the input and the output images.

You can press the tabs to toggle between the various displays.

Double click into the wanted images or use the "New Window" button to get an enlarged display window. Use "Save" to store the display (JPG).

To adjust the display settings use the related display control tab on the left hand side of the page. Refer to **guiDISPLAY**.



A Typical Page - “Display Control” Tabs

The visualisation settings of the images shown on the right-hand side of each **IMAGIC GUI program** page can be adjusted in its own related “Display control” tab on the bottom left part of each page. Also refer to **guiDISPLAY**.

Grey value scaling: Adjust the contrast

Min/Max: Scale the grey-values to minimum/maximum

Interactive: Set the limits by giving numbers

Sigma: Use an amount of sigma to set the limits

Contrast

How to calculate the grey value scaling

Local: Calculated in each image separately

Global: Calculated using all image densities
(as displayed in the histogram)

Gallery: Calculated in the currently displayed images

Inverse contrast:

Use one of the radio buttons

Zoom

Enlarge the displayed images

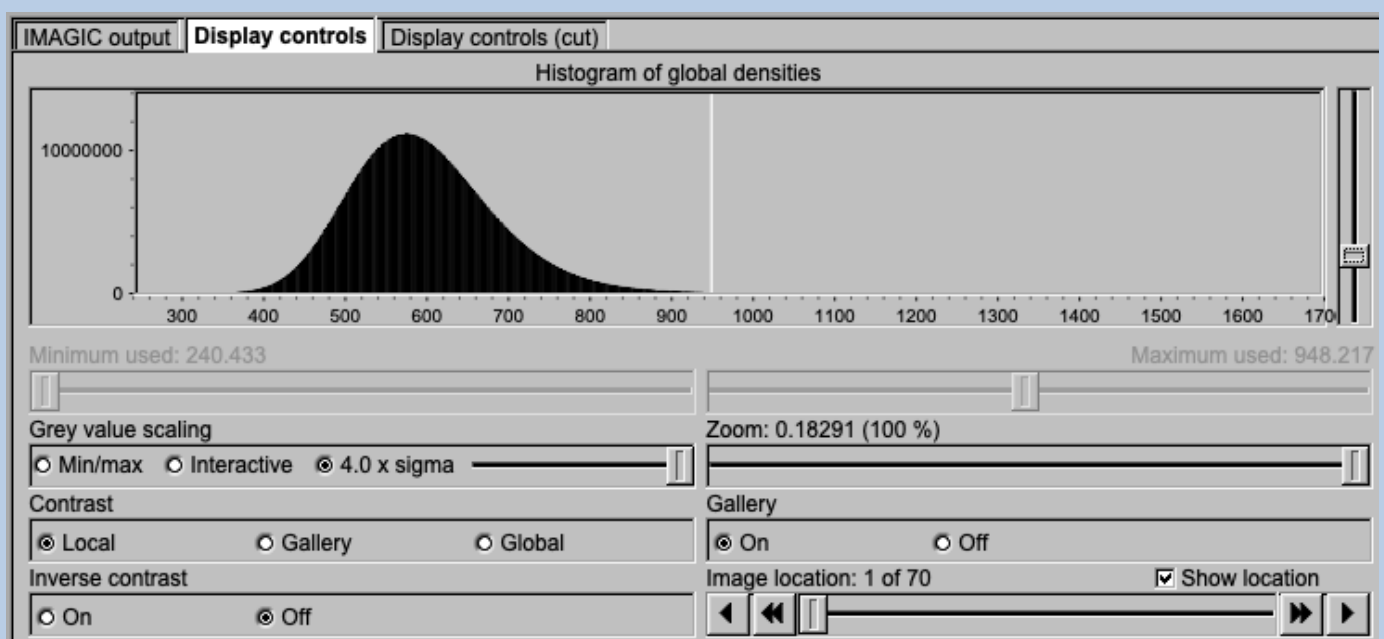
Gallery

On Display the images in a gallery
(may be you need another zoom to see more than one image)

Off Show only one image

Image Locations.

Use the slider or the arrows to select image locations



“Plot Control” Tabs

The visualisation settings of curves/spectra is shown on the right-hand side of an **IMAGIC GUI program** page can be adjusted in its own related “Plot control” tab on the bottom left part of each page. Also refer to **guiPLOT**.

Style, Colour, Grid: Adjust the curve line style, the colour and add a grid if wanted

Horizontal, vertical scaling: Set minimal and maximal horizontal or vertical limits

Plot title Set the text of the plot title

Text along ... Set the text along the given axis

Use for all plots: Use the setting for all plots in a file independent of what is input in the PLT file

Reset:. Reset to the automatic values

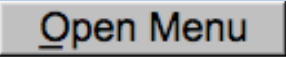
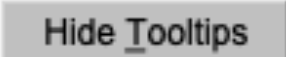
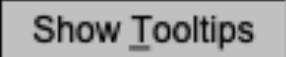
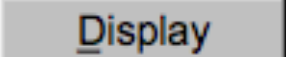
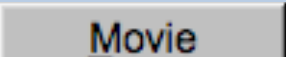
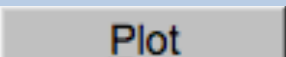

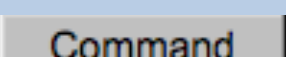
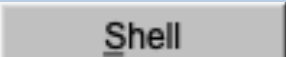
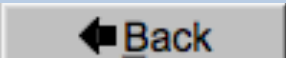
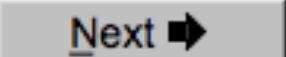
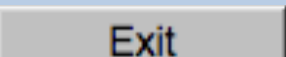
| | | |
|---|---|-----------------------------|
| Style Select curve style ▾ | Colour Select curve colour ▾ | Grid Select curve grid ▾ |
| Horizontal scaling 1.00 | <input type="checkbox"/> Use for all plots 32.00 | Reset |
| Vertical scaling -19.21 | <input type="checkbox"/> Use for all plots 17.00 | Reset |
| Plot title Fourier Ring Information - 1/2-bit | <input type="checkbox"/> Use for all plots | Reset |
| Text along horizontal axis Radius in Fourier space | <input type="checkbox"/> Use for all plots | Reset |
| Text along vertical axis | <input type="checkbox"/> Use for all plots | Reset |



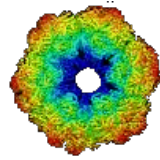
The Toolbar

There is a toolbar at the bottom of each **guiDETECTOR** page.

The toolbar buttons:

| | |
|---|---|
|  | Open the MENU to navigate to each page wanted |
|  | Show or hide the context sensitive tooltips (the help text may sometimes disturb) |
|  | |
|  | Open a DISPLAY page to visualize IMAGIC images. Refer to guiDISPLAY . |
|  | Open a MOVIE page (display in an endless loop). Refer to guiDISPLAY |
|  | Open a PLOT page to show IMAGIC curves. Refer to guiPLOT |
|  | Open a DISPLAY page to visualize IMAGIC images using a colour map stored in another input. |
|  | Open a list to run any IMAGIC command. Refer to guiIMAGIC . |
|  | Run a shell / terminal page. command |
|  | Go to the previous page |
|  | Continue with the next page |
|  | Exit guiDETECTOR |



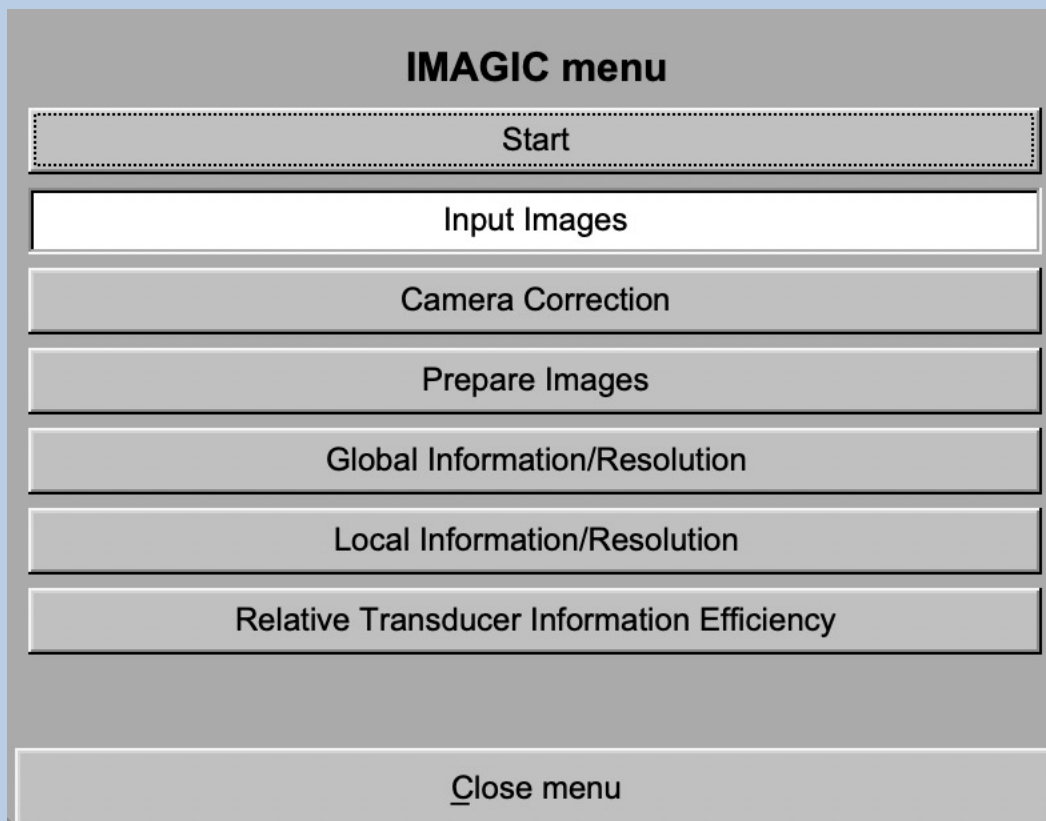


IMAGIC

guiDETECTOR



The guiDETECTOR Menu



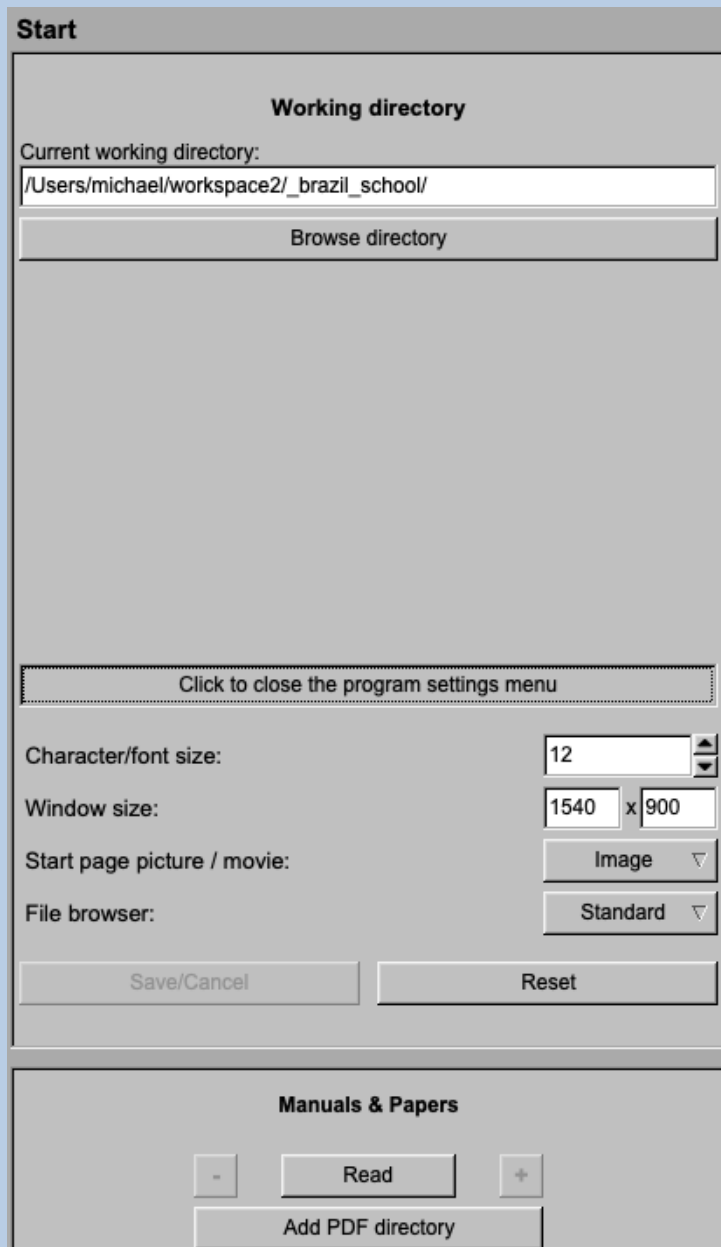
PAGES:

| | |
|-------------------------------|--|
| Start | Page to adjust guiDETECTOR program parameters |
| Input Images | Import or specify the input images. Cut out a part, if wanted. |
| Camera Correction | Correct for camera errors/properties |
| Prepare Images | Pre-treat images: Mask, filter, normalise variance, resize, sum ... images |
| Global information/Resolution | Calculate the global information content / resolution of the input data |
| Local information/Resolution | Calculate the local information / resolution maps of the input data |
| Relative Transducer ... | Compare two transducers, cameras imaging tools... |



The “Start” Page

This page is not part of the **guiDETECTOR** workflow and can only be reached using the “Back” or the “Open Menu” button(s).



On this page you can set some program parameters:

- a) the working directory
- b) the size of the **guiDETECTOR** program windows and/or text
(a re-start is needed)
- c) the type of file browser



Start Working

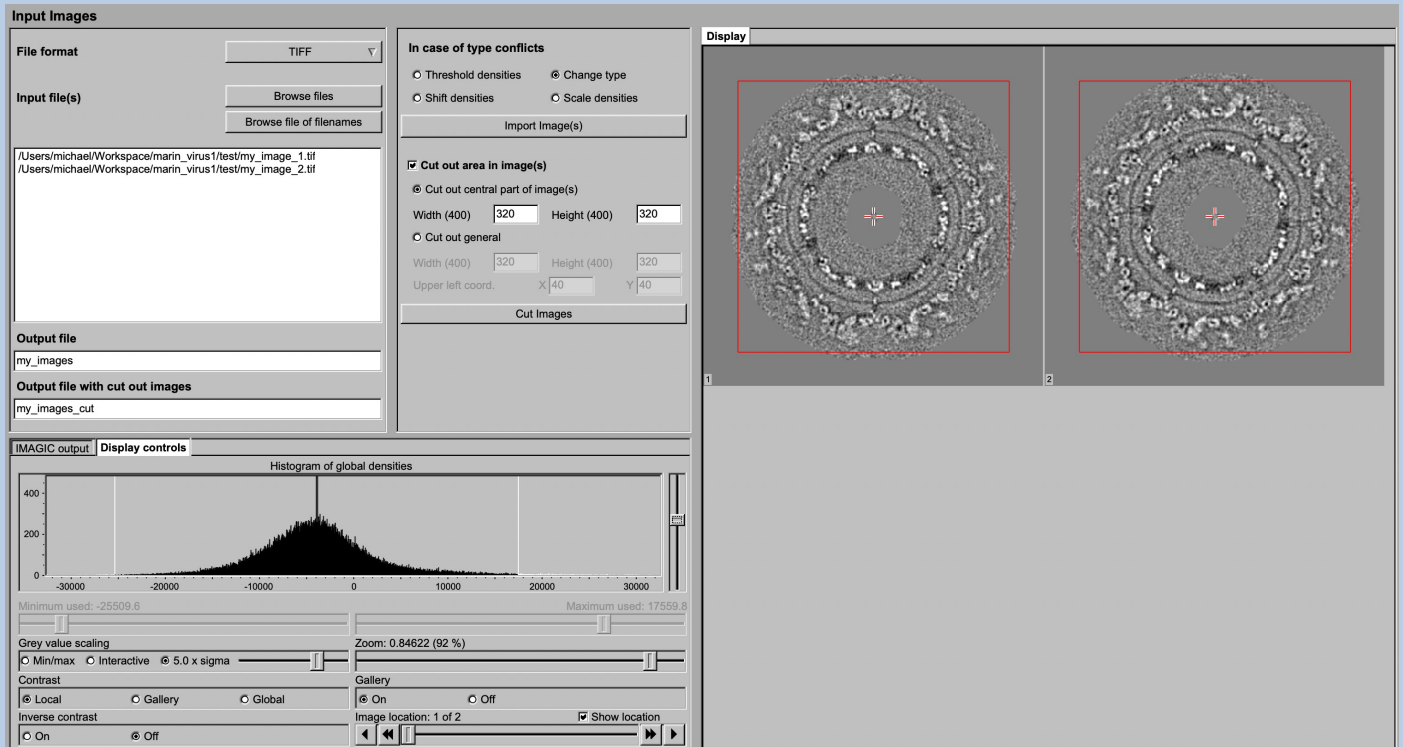
The page **guiDETECTOR** starts with the “Import Images” page.

The workflow using the “Next” button will guide you through all **guiDETECTOR** pages.

Use the “Back”, “Next” or “Open Menu” buttons to skip a page or to choose the wanted page.



The “Import Images” Page



DESCRIPTION:

Convert import image files using any 3D-EM format (or TIFF) into a single (stacked) IMAGIC image file.

The page can be skipped if your input images are already stored in IMAGIC format.

If wanted you can cut-out parts of the input images. Not suggested for camera correction.

Also refer to program **guiIMPORT**.



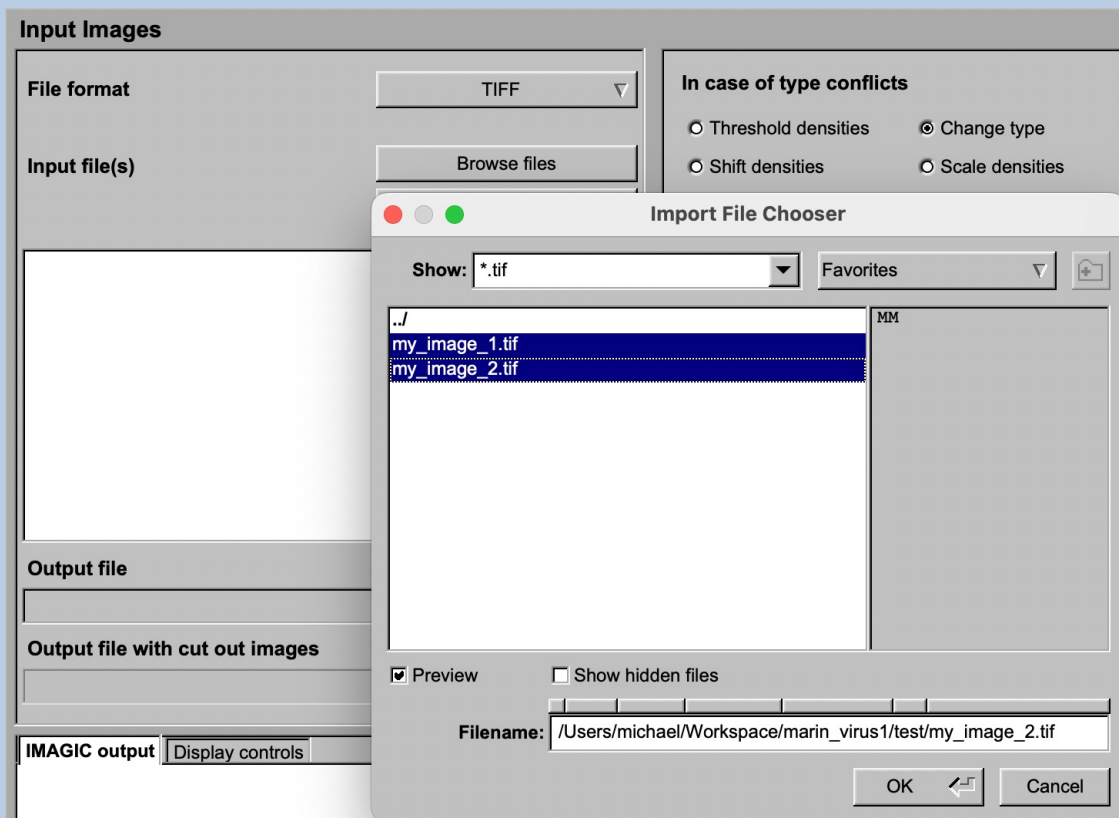
IMPORT IMAGES:

Specify the file format in which your input micrographs/images are stored. Click the “Select format” button



and choose one of the formats in the listing.

Now you can specify the input image files or a “File of filenames” text file(containing the names of the wanted input image files) with the “Browse” button. Refer to chapter “Input Files” and “Input. File Chooser” for help.



If wanted you can edit the list of files. But be careful there is no automatic control of file names in this list.

```
/Users/michael/Workspace/marin_virus1/test/my_image_1.tif  
/Users/michael/Workspace/marin_virus1/test/my_image_2.tif
```

Next, you need to specify the name of the output file which is the IMAGIC image file which will contain the imported image(s).

Depending on the format of the input images you have to specify a number of parameters or options.

Format TIFF, for example:

In case of type conflicts

- Threshold densities
- Change type
- Shift densities
- Scale densities

Having specified every information needed click the “Import Micrograph” button to start the import of the image(s).

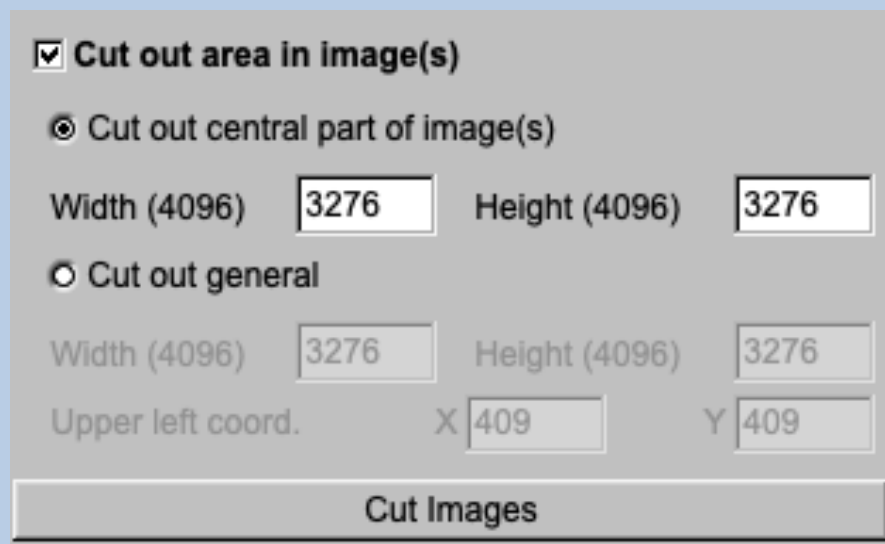
The imported images are shown in the display tab on the right-hand side. See chapter “A Typical Page - Display control tabs”.



CUT MICROGRAPHS / IMAGES:

Having imported the input images, you may want to not use the full size of the images but only a part of them.

Clicking the “Cut out area of image(s)” option you can cut-out parts of the imported images:



Cut out area in image(s)

Cut out central part of image(s)

Width (4096) Height (4096)

Cut out general

Width (4096) Height (4096)

Upper left coord. X Y

Cut Images

The chosen part is shown in the display window. You can cut-out a central part or any part wanted. The cut-out part is the same in all images, of course.

The name of the output file containing the cut-out images is suggested on the left-hand side. As usual you can change this name, of course.

Having specified everything click the “Cut Images” button to run the calculations.



The “Detector Correction” Page

The screenshot displays the 'Camera Correction' page in the guiCNORM software. The interface is organized into several sections:

- Camera Correction:** Contains input fields for raw micrographs, camera statistics average file, camera statistics sigma file, camera corrected micrographs, and good camera corrected micrographs. Each field has a 'Browse file' or 'Export file' button.
- Camera Normalisation:** Includes radio buttons for 'Measure', 'Correct', and 'Measure and Correct', along with a 'Correct' button.
- IMAGIC output:** A text area showing the progress of the correction process, including file paths and a percentage-based progress log.
- Image Display:** A large window showing a noisy micrograph. Above it are tabs for 'Input Micrographs', 'Corrected Micrographs', 'Average', and 'Sigma'. Below the image are options to 'Extract micrographs' and 'Ignore micrographs which show' (with checkboxes for 'too extreme sigma of densities' and 'too extreme min/max difference of densities').
- Bottom Panel:** Contains navigation buttons like 'Open Menu', 'Hide Tooltips', 'Display', 'Movie', 'Plot', 'Command', 'Shell', 'Back', 'Next', and 'Exit'.

DESCRIPTION:

Get the detector statistics and /or detector correct/normalize the input images. Each output image is the input image minus the average image calculated from all images and divided by the standard deviation (again calculated from all images).



Choose one of the options

Detector Normalisation

Measure

Correct

Measure and Correct

Correct

- Measure:** Measure the detector statistics and create the detector statistics images needed to detector correct images taken with this detector.
- Correct:** The detector statistics images are already available. Detector correct the input images using these detector statistics images.
- Measure and Correct:** Do both, measure the detector statistics and correct for it.



MEASURE:

Measure the detector statistics and create the detector statistics images needed to detector correct images taken with this detector.

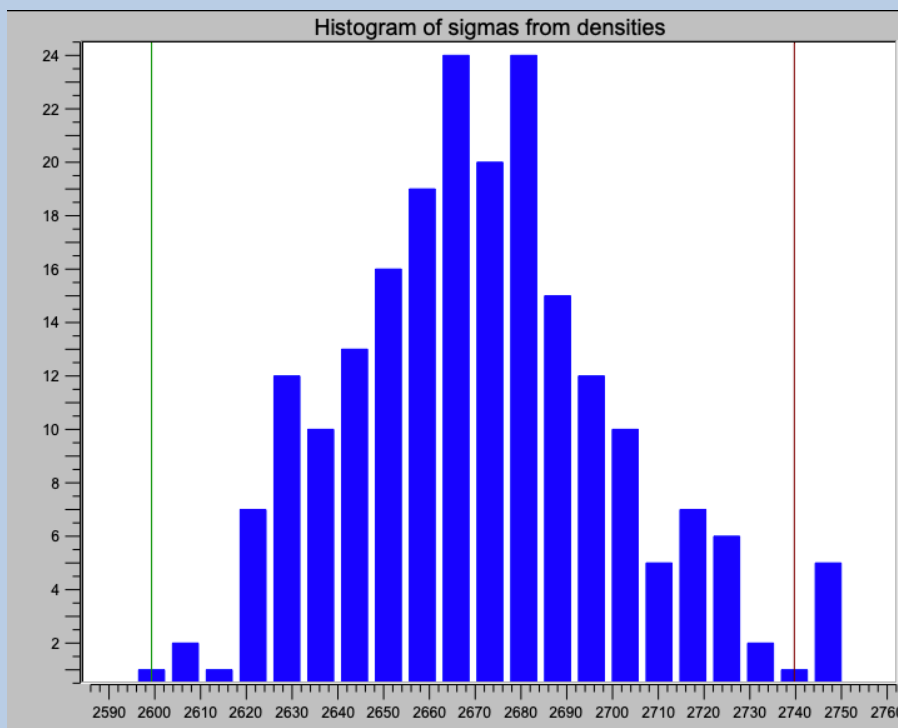
Mode

- Measure
- Correct
- Measure and Correct

Note that you usually need a huge number of input images to get a good statistics and a good subsequent detector correction.

First the statistics of the input image densities is needed. Normally you will use the “Calculate Statistics” button to calculate this statistics.

The histogram of sigma values is shown on the right-hand side. For a “good” dataset this histogram usually has a Gaussian like shape.



If the shape looks correct you can “Use all images”.

Find Detector Statistics

- Use all images
- Ignore outliers

If wanted you can, of course, remove “outliers”.

Find Detector Statistics

- Use all images
- Ignore outliers

Minimum sigma 4279.2400 

Maximum sigma 4334.6800 

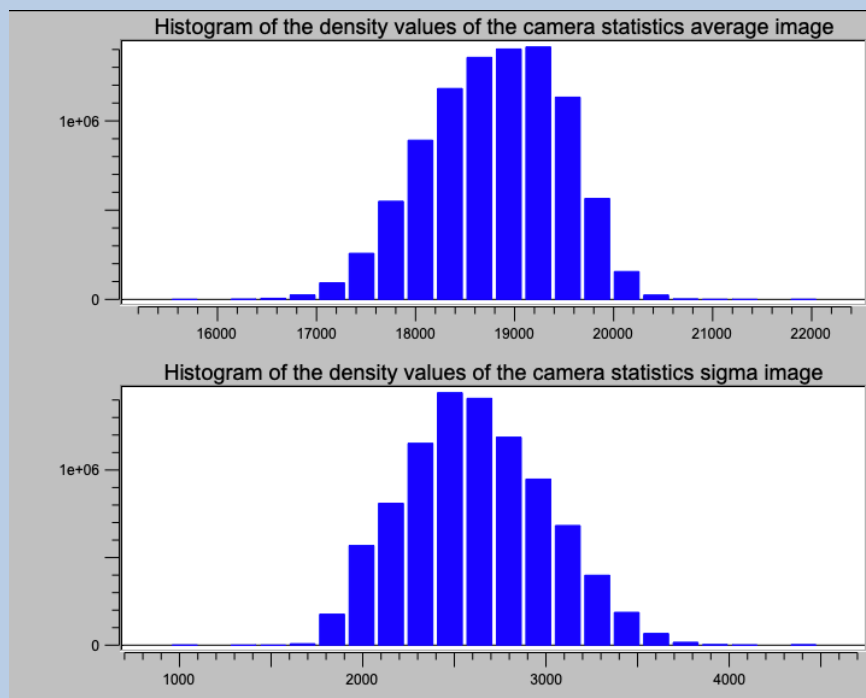
Use the up and down arrows to define a minimum and a maximum value for sigma (do NOT type the values). Only images corresponding to the histogram part between the red lines will be used for the detector correction. The red vertical lines in the histogram will help you to check the chosen values.

Do not forget to specify the names of the output files on the left hand side of the page,

Press the “Measure” button to start the calculations.

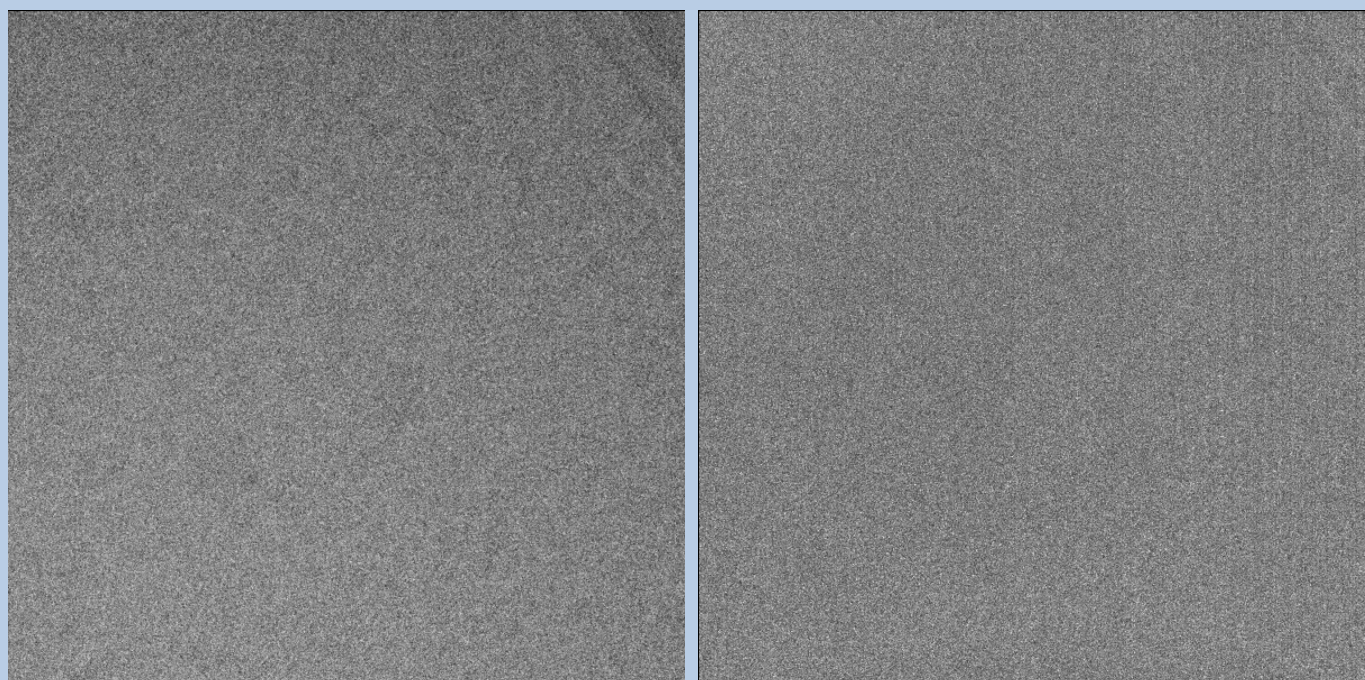
The measured detector statistics is shown in two histograms using the sigma of the densities in the detector statistics average and in the detector statistics sigma image, respectively.





The histograms usually have a Gaussian like shape. In case of detector errors (blind or dark pixels, for example) these can easily be seen as vertical lines.

The detector statistics average and sigma images are also displayed in tabs on the right hand side of the page. Always use a zoom factor of 1 (refer to chapter "A Typical Page - Display control" tabs) to make sure that you can check single pixels.



CORRECT:

Once having the detector statistics average and sigma images available you can correct all images taken with this detector (also the ones not used for getting the statistics).

Detector Normalisation

Measure

Correct

Measure and Correct

Correct

Of course, you need to specify the input detector statistics average and sigma image file needed for the detector correction

Input detector statistics average file Browse file

my_images_cnorm_average

Size = 400x400, Images = 1

Input detector statistics sigma file Browse file

my_images_cnorm_sigma

Size = 400x400, Images = 1

and, as usual, the output file name

Output file with detector corrected images Export file

my_images_cnorm

Click the "Correct" button to start the detector correction.

The detector corrected images are displayed on the right hand side of the page.

Note that the detector corrected images can be converted to any 3DEM format by clicking the "Export file" button which opens a separate "EM2EM" page.



The “Prepare Images” Page

Prepare Images

Input file
my_images
Size = 400x400, Images = 2

Output file with resized images
my_images_c2

Output file with prepared images
my_images_c2_pret

Output file with image sum(s)

Run in parallel mode
 Yes No Number of nodes: 3

Histogram of global densities
Minimum used: -25509.6 Maximum used: 17559.8

Zoom: 1 (100 %)

Grey value scaling
 Min/max Interactive 5.0 x sigma

Contrast
 Local Gallery Global

Inverse contrast
 On Off

Image location: 1 of 2 Show location

Resize images (by binning)
Summing parameter: 2

Prepare images
 Pretreat images (filter, masks...)
 Normalise amplitude spectra (NAS)

Pretreat images
 Band-pass Filter
LF cut: 0.100
Rem. LF: 0.000
HF cut: 0.800

Normalisation
Sigma: 10.000

Mask
 circular rectangular Gaussian
 center off-center
Radius: 0.9 Drop off: 0.050

Reverse contrast

Sum Images
 Odd-even sum Half-half sum
 Sequence sum 2 Images
 Total sum

Input Images | **Prepared images**

Automatic Default Run

DESCRIPTION:

It can be helpful to pre-treat the input images by imposing a band-pass filter, normalise the variance, impose a mask and...

NOTE:

Of course, you can skip this page if no such treatment is wanted/needed.



You can resize the images by binning

Resize images (by binning)

Summing parameter

You can pre-treat the images. Options are

Prepare images

Pretreat images (filter, masks...)

Normalise amplitude spectra (NAS)

Pretreat images

Band-pass Filter

LF cut

Rem. LF

HF cut

Normalisation

Sigma

Mask

circular rectangular Gaussian

center off-center

Radius Drop off

Reverse contrast

Options are band-pass filtering

Band-pass Filter

LF cut

Rem. LF

HF cut



normalise the variance in each image

Normalisation

Sigma

imposing a mask

Mask

circular rectangular

center off-center

Radius Drop off

if wanted you can also reverse the contrast.

Reverse contrast

Instead of using these pre-treatment options you can apply a NAS filter which means that the amplitude spectra of the images are normalised:

Prepare images

Pretreat images (filter, masks...)

Normalise amplitude spectra (NAS)

Mask

circular rectangular

center off-center

Radius Drop off

Reverse contrast

NAS Filter

LF cut

Rem. LF

HF cut

Imposing a mask and reversing the contrast are also options here.



As usual, specify the names of the input and the output files:

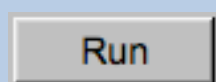
Input file

Size = 400x400, Images = 2

Output file with resized images

Output file with prepared images

Start the calculations by clicking the “Run” button:



You can also create various image sums which you may need for the subsequent calculations of the global and local information content in your images.

Sum images

Odd-even sum Half-half sum

Sequence sum images

Total sum

Output file with image sum(s)



The “Global Information/Resolution” Page

Global Information/Resolution

Input image(s)
my_image_1
Size = 400x400

Input image(s) to be compared
my_image_2
Size = 400x400

Root name of results files
my_image_1_2

Output PLT/CSV/XML files with Fourier shell correlation
my_image_1_2_frc

Output PLT/CSV/XML files with Fourier shell information
my_image_1_2_fri

Mode
 Single data-set Different data-sets

Metric
 FRC FRI Both

Mode of input
 One reference Parallel

Fourier shell correlation/information
 Also create 'anisotropic' curves
Missing cone angle
Point-group symmetry
2D images
Filling degree / Object size

How to continue

- Check the FRC and the 1/2-bit curve (first plot) as well as the FRI and the 1/2-bit threshold (third plot). Does the FRC curve (red) and the threshold curve (blue) intersect at a reasonable value? Does the FCI curve (red) and the threshold line (blue) intersect at a reasonable value? In both, FRC and FRI, is the resolution cross point 1/3rd away from the Nyquist frequency? (You should never claim any resolution level beyond 2/3rd of the Nyquist frequency = undersampling)
- Do both, FRC and FRI, fluctuate around zero close to the Nyquist frequency (if not: overfitting or sharp masks used?)
- Also check the FRC (red) and 3-sigma (blue) curves (second plot). If the FRC and 3-sigma curves do not intersect you did not collect a sufficient amount of data to allow a direct structural interpretation at that resolution level.
- Also have a look at the radially weighted FRI curve (4th plot). It does not give a resolution value but shows the information content.

First image file | Second image file | **FRC - 1/2-bit** | FRI - 1/2-bit

Fourier Ring Information - 1/2-bit

Spatial frequency [1/Å]

Resolution: 11 Å

DESCRIPTION:

Calculate the global information content (using the Fourier ring or shell information metric (FRI/FSI) and/or the global resolution (using the Fourier ring or shell correlation metric (FRC/FSC)).

The comparison(s) can be calculated between different images / 3D volumes or by comparing each image / 3D volume with itself.



Specify if input are two different or a single data-set. Note that these two different option are not relevant for the calculations but to give better help and tool tips.

Mode

Single data-set Different data-sets

In “Single data-set” you have to choose which of the following input modes is to be used

Mode of input

One reference Sequential
 Parallel Sequential pairs
 Self

in “Different data-set” the input mode can be one of the following:

Mode of input

One reference
 Parallel

Modes of input are:

➤ One reference:

The input images / 3D volumes are compared with a single reference

Input image(s) to be compared Import Browse

my_images

Size = 400x400

Reference image Import Browse

my_reference



➤ Parallel:

The images / 3D volumes in the first input file 1 are compared one-by-one with the images / 3D volumes in the second input file. Both input files are expected to contain the same number of images / 3D volumes.

Input 1st half data-set 3D volume(s) Import Browse
my_images_1
Size = 400x400
Input 2nd half data-set 3D volume(s) Import Browse
my_images_2
Size = 400x400

➤ Sequential:

The images / 3D volumes to be compared are stored in a single input file. The comparison is in sequential order which means The image / 3D volume #1 is compared with #2, #2 with #3 etc.

Input images to be compared Import Browse
my_images
Size = 400x400

➤ Sequential pairs

Same as sequential but the comparison is done pair-wise: The image / 3D volume #1 is compared with #2, #3 with #4, #5 with #6 etc.

➤ Self

In case of a “Single dataset” there is an additional mode of input option “Self” which allow to calculate the information content/resolution using single images / 3D volumes which are only compared with itself but NOT with any other images / 3D volumes.

Input image(s) Import Browse
my_image
Size = 400x400



Note that in case of option “Self” you have to specify the parameters of a band-pass filter which is applied to remove high frequency information introduced by this very specific “self compare” technique:

Mode of input

One reference Sequential
 Parallel Sequential pairs
 Self

LF cut HF cut

Next you are expected to choose the wanted metric:

Metric

FRC FRI Both

Options are:

- | | | |
|------------|------|--|
| Images | FRC | Global resolution using the Fourier Ring Correlation |
| | FRI | Global information using the Fourier Ring Information |
| | Both | Calculate both, FRC and FRI |
| 3D volumes | FSC | Global resolution using the Fourier Shell Correlation |
| | FSI | Global information using the Fourier Shell Information |
| | Both | Calculate both, FSC and FSI |

In case of input images you have to specify (or check) the filling degree (move the cursor to the input field to get help) and the pixel size:

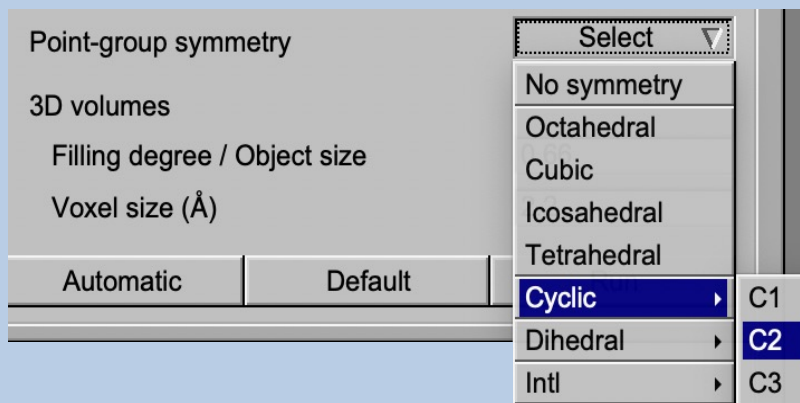
2D images

Filling degree / Object size

Pixel size (Å)

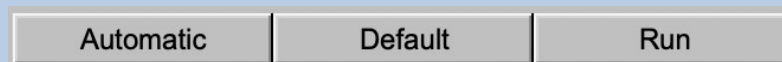


In case of input 3D volumes you also have to specify the symmetry:



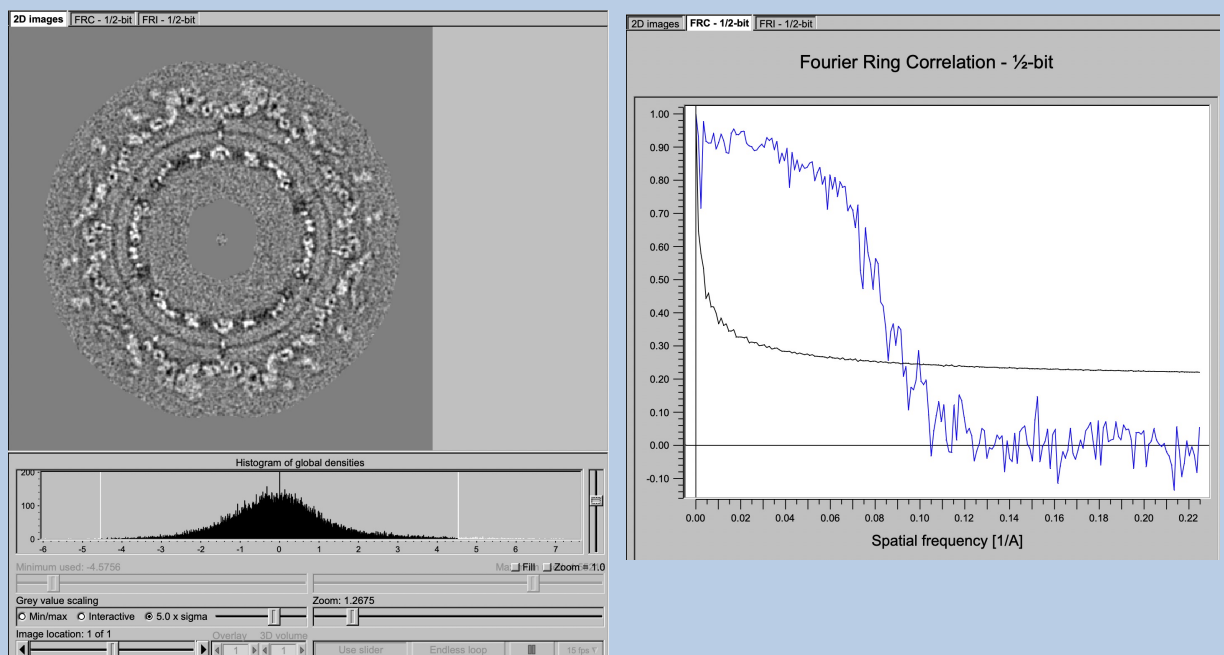
Move the cursor to an input fields to get context sensitive help.

As usual you can reset all parameters to the last values you have used by clicking the “Default” button, clicking the “Automatic” button will reset all parameters to the values suggested by **guiFSC**.

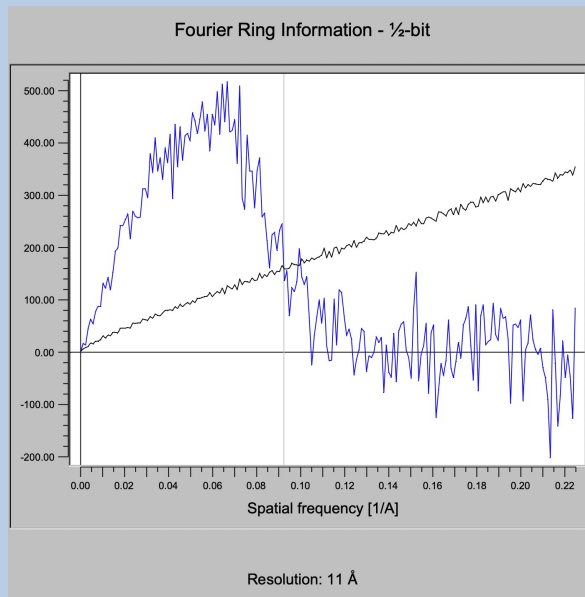


Clicking the “Run” button will start the calculations.

As usual the input images and the resulting information and/or resolution curves are displayed in the display/plot tabs on the right-hand side.

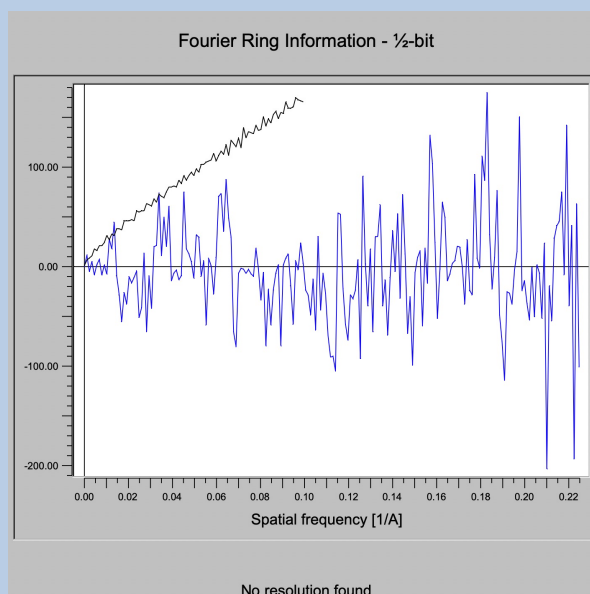


In contrast to the FRC/FSC curve the FRI/FSI plots also show the estimated resolution value in case FRI/FSI curve and the ½-bit threshold curve intersect:



The ½-bit information curve indicates where a sufficient amount of data is collected to allow a direct interpretation at that resolution level. The overall resolution achieved is estimated by the intersection of the FRI/FSI curve (blue) and the 1/2 Bit curve (black).

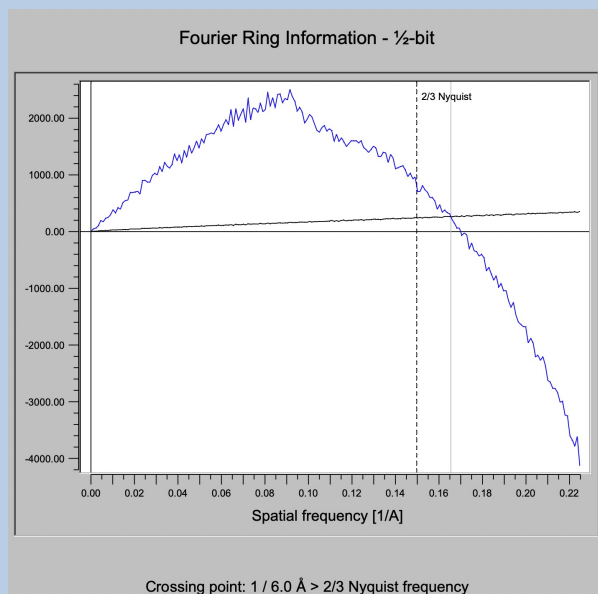
No resolution is printed if the estimated resolution value is too small :



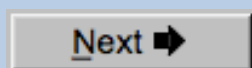
No resolution but a warning is printed if the estimated resolution value is too small or too close to the Nyquist frequency.

PLEASE NOTE:

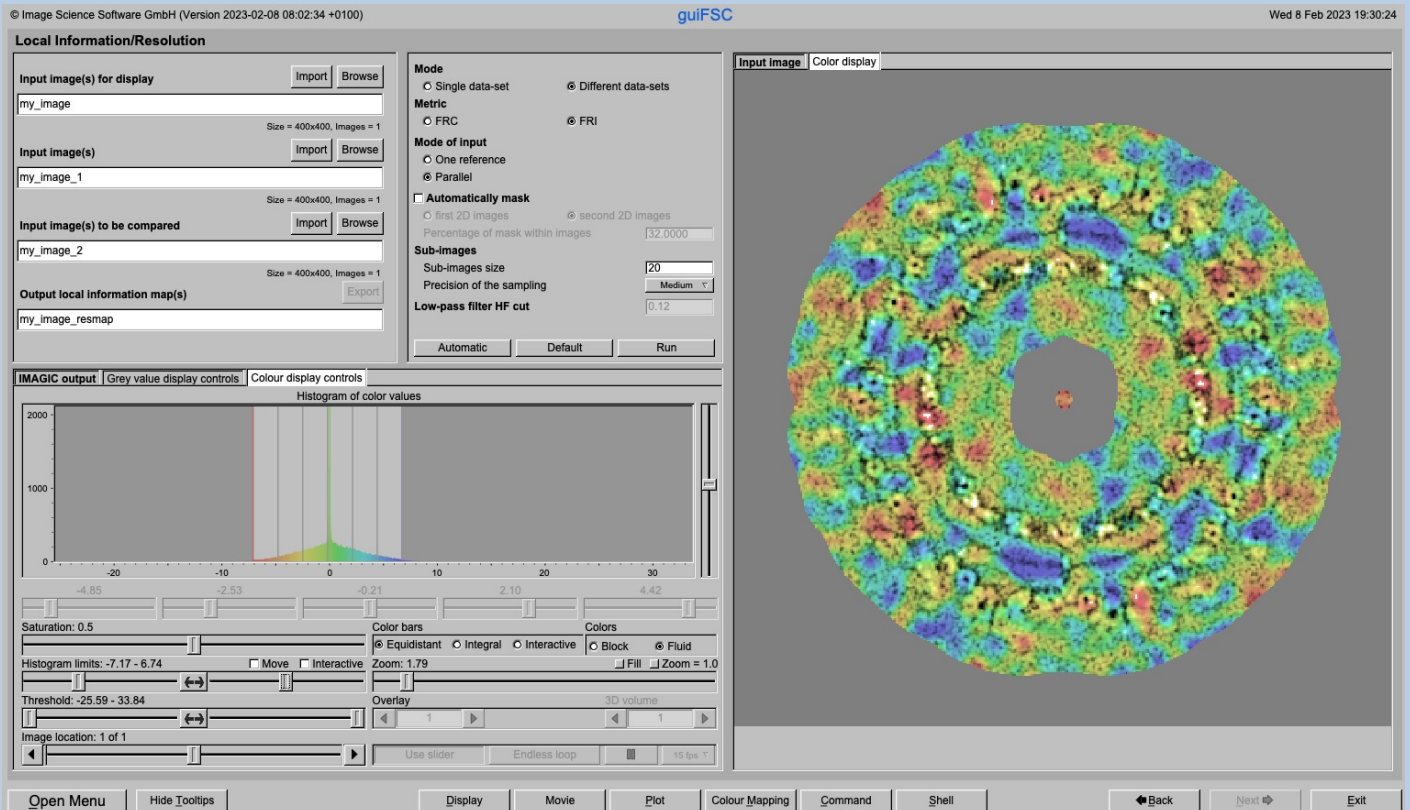
Under-sampling remains one of the worst sins one can commit in estimating the resolution / information content in images / 3D volumes. You should never claim any resolution level beyond 2/3rd of the Nyquist frequency.



As usual you can click the "Next" button to navigate to the next page which is the "Local Information / Resolution" page.



The “Local Information/Resolution” Page



DESCRIPTION:

Calculate the local information content (using the Fourier Ring or Shell Information metric (FRI/FSI) and/or the local resolution (using the Fourier Ring or Shell Correlation metric (FRC/FSC)).

The comparison(s) can be calculated between different images / 3D volumes or by comparing each image / 3D volume with itself.



Specify if input are two different or a single data-set. Note that these two different option are not relevant for the calculations but to give better help and tool tips.

Mode

Single data-set Different data-sets

In “Single data-set” you have to choose which of the following input modes is to be used

Mode of input

One reference Sequential
 Parallel Sequential pairs
 Self

in “Different data-set” the input mode can be one of the following:

Mode of input

One reference
 Parallel

Modes of input are:

➤ One reference:

The input images / 3D volumes are compared with a single reference

Input image(s) to be compared Import Browse

my_images

Size = 400x400

Reference image Import Browse

my_reference



➤ Parallel:

The images / 3D volumes in the first input file 1 are compared one-by-one with the images / 3D volumes in the second input file. Both input files are expected to contain the same number of images / 3D volumes.

Input 1st half data-set 3D volume(s) Import Browse
my_images_1
Size = 400x400
Input 2nd half data-set 3D volume(s) Import Browse
my_images_2
Size = 400x400

➤ Sequential:

The images / 3D volumes to be compared are stored in a single input file. The comparison is in sequential order which means The image / 3D volume #1 is compared with #2, #2 with #3 etc.

Input images to be compared Import Browse
my_images
Size = 400x400

➤ Sequential pairs

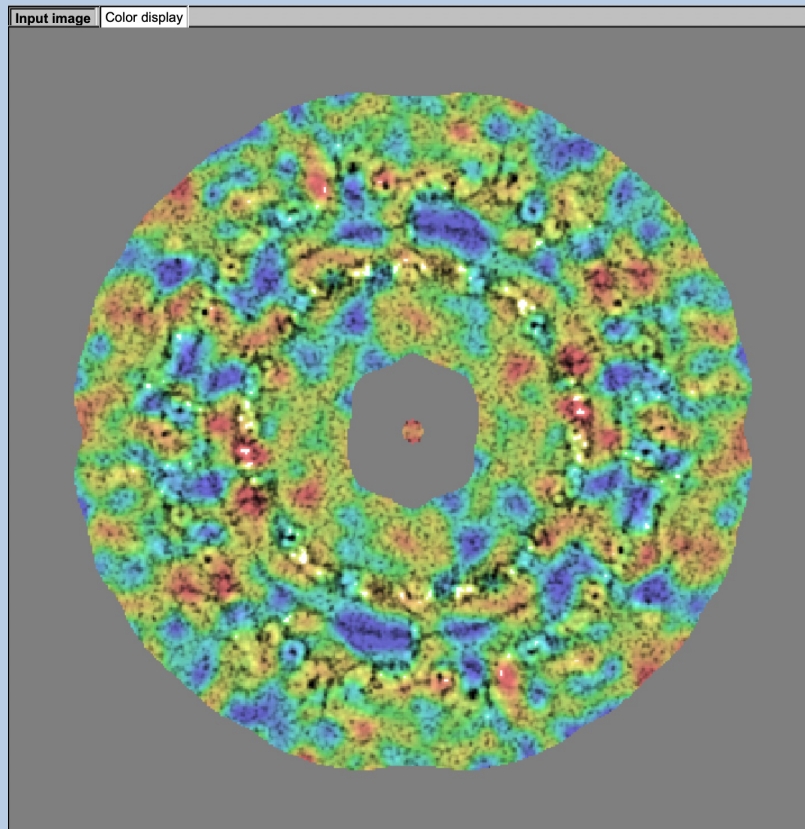
Same as sequential but the comparison is done pair-wise: The image / 3D volume #1 is compared with #2, #3 with #4, #5 with #6 etc.

➤ Self

In case of a “Single dataset” there is an additional mode of input option “Self” which allow to calculate the information content/resolution using single images / 3D volumes which are only compared with itself but NOT with any other images / 3D volumes.

Input image(s) Import Browse
my_image
Size = 400x400





Use the “Colour display controls” to adjust the coloured display on the right-hand side.

IMAGIC output | Grey value display controls | **Colour display controls**

Histogram of color values

2000
1000
0

-20 -10 0 10 20 30

-4.31 -1.82 0.68 3.17 5.67

Saturation: 0.5

Color bars

Color options: Equidistant Integral Interactive Block Fluid

Zoom: 1.79 Fill Zoom = 1.0

Histogram limits: -6.81 - 8.17 Move Interactive

Threshold: -25.59 - 33.84

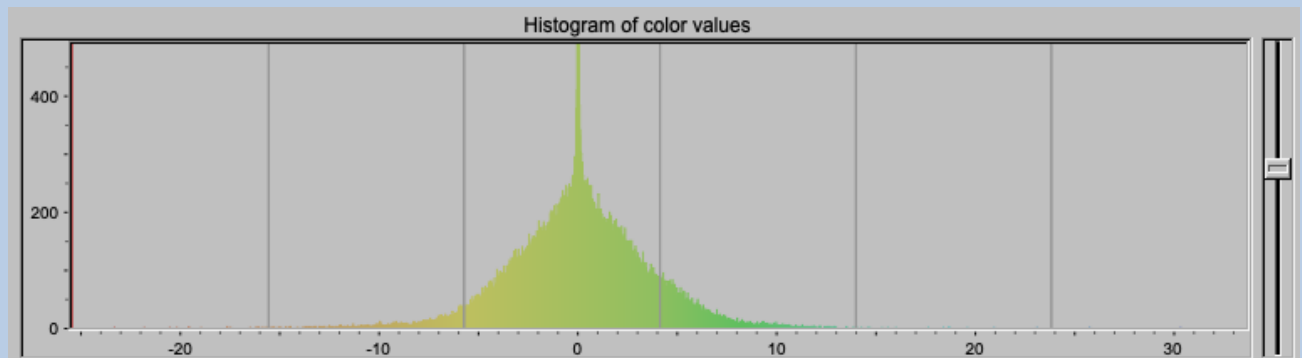
Overlay 3D volume

Image location: 1 of 1

Use slider Endless loop 15 fps



The histogram shown in the “Colour display controls” is the histogram of colour values used.



Use the slider to adjust the vertical scaling of the histogram.

The colour mapping can be adjusted by a number of parameters and options. Refer to the “**guiColourMapping** manual” for details.

- | | |
|-------------------|--|
| Saturation: | Adjust the saturation |
| Histogram limits: | Use the two sliders to adjust between which values the colour palette is used. Interactive: Set the limits by giving numbers Move: Move the chosen limits through the palette |
| Colour bars | How to use the colour palette Equidistant: The colour palette is used linearly Integral: The colour palette is squeezed according to the number of histogram values Interactive: Use the histogram sliders to set the colour bars |
| Colours: | Block: Fixed colour between two colour bars Fluid: The colours are changing continuously |
| Zoom | Enlarge the displayed image using the slider Fill: Fit image size to window size Zoom = 1.0. Display image 1:1 |
| Threshold: | Adjust the threshold limits with the two sliders. Colours below the threshold are displayed in grey, colours above the threshold are displayed white |
| Image Locations. | Use the slider or the arrows to select image locations |



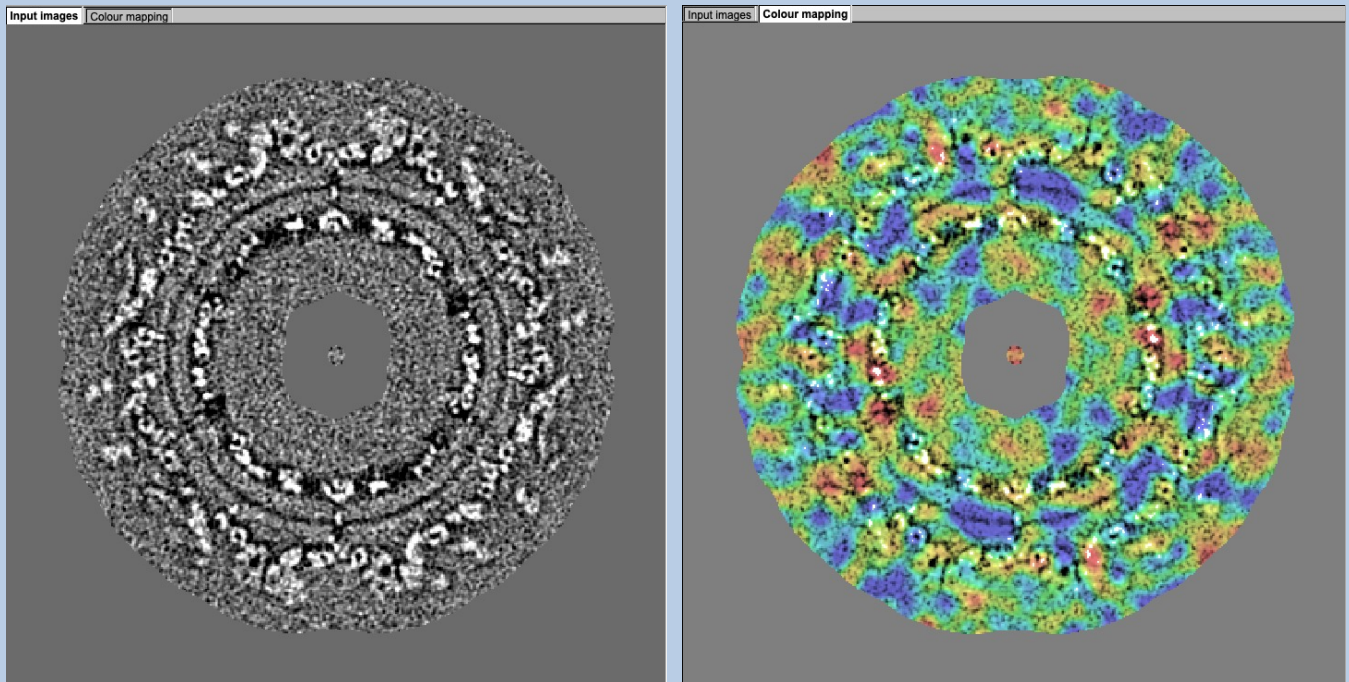
COMPARE TWO DIFFERENT LOCAL INFORMATION / RESOLUTION MAPS:

Note that the value of each colour is an absolute value. If you want to compare different information / resolution maps make sure that the same "Histogram limits" are used. Activate the "Interactive" option and specify the same limits in both colourised displays for a correct comparison.


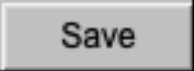
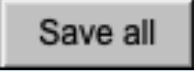


Display Windows

The image(s) are displayed in the display windows at the right hand side. Click the related tab to get the wanted display window.

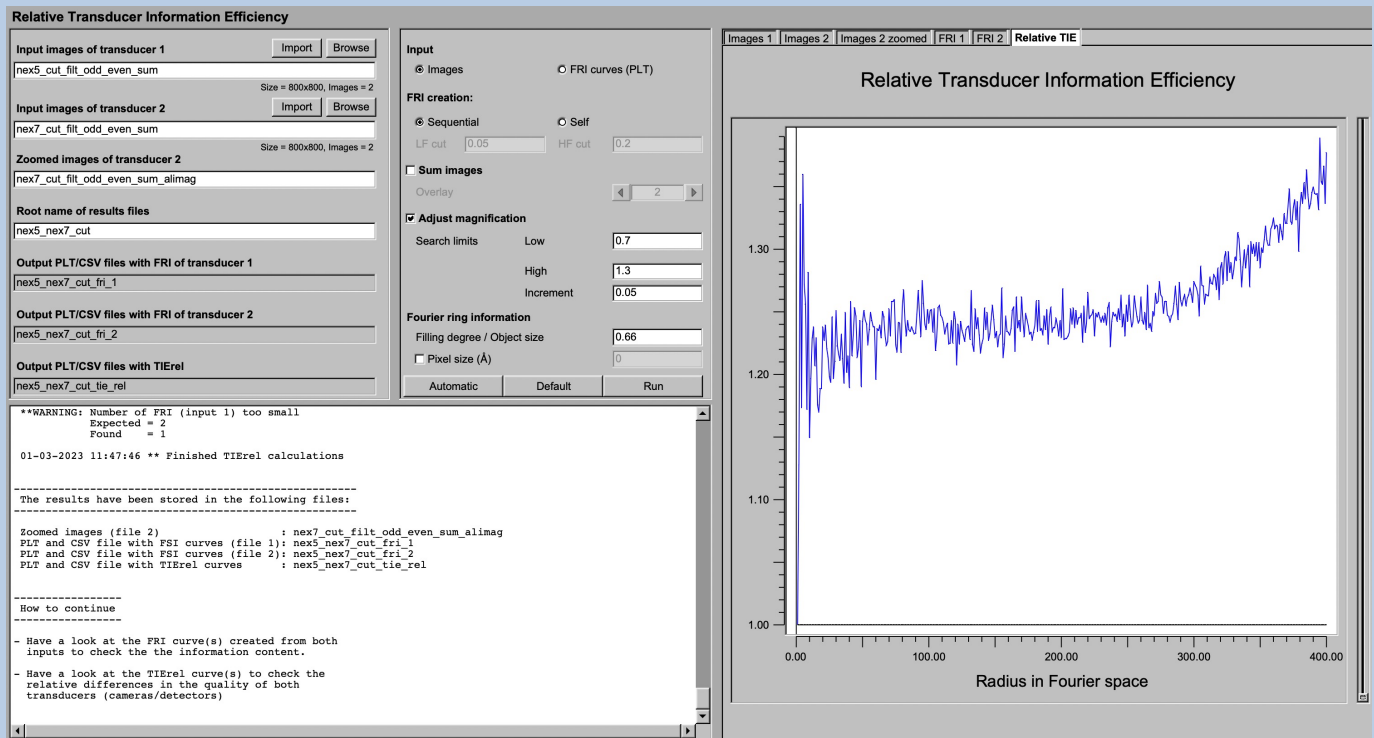


Moving the cursor into the display window there are a few options:

- | | |
|---|--|
|  | Open the display window a larger separate display window |
|  | Save the current displayed image in a JPG image |
|  | Save the whole window in a JPG image |



Relative Transducer Information Efficiency (TIE_{rel})



DESCRIPTION:

Calculate the relative transducer information efficiency to compare (the quality of) two detectors.



Input are two data sets each one taken by another detector. Usually the input are images or images sums which are used to calculate the global information (FRI) in each of the input data-sets

Input

Images

FRI curves (PLT)

As usual, specify the related file names:

| | | |
|---|---------------------------------------|---------------------------------------|
| Input images of transducer 1 | <input type="button" value="Import"/> | <input type="button" value="Browse"/> |
| <input type="text" value="nex5_cut_filt_odd_even_sum"/> | | |
| Size = 800x800, Images = 2 | | |
| Input images of transducer 2 | <input type="button" value="Import"/> | <input type="button" value="Browse"/> |
| <input type="text" value="nex7_cut_filt_odd_even_sum"/> | | |
| Size = 800x800, Images = 2 | | |

Also specify the root name to be used to create the names of the various output files.

| |
|--|
| Root name of results files |
| <input type="text" value="nex5_nex7_cut"/> |
| Output PLT/CSV files with FRI of transducer 1 |
| <input type="text" value="nex5_nex7_cut_fri_1"/> |
| Output PLT/CSV files with FRI of transducer 2 |
| <input type="text" value="nex5_nex7_cut_fri_2"/> |
| Output PLT/CSV files with TIErel |
| <input type="text" value="nex5_nex7_cut_tie_rel"/> |



There are a few parameters and options you can adjust:

Fourier ring information

Filling degree / Object size

Pixel size (Å)

If wanted input images can be summed before the FRI is calculated

Sum images

Overlay

The magnification (the pixel size) in the input files can be different. In this case a magnification alignment is useful:

Adjust magnification

Search limits

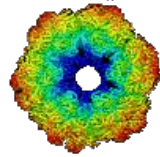
Low

High

Increment

As usual, the resulting FRI and the TIE_{rel} curves are shown on the right hand side.





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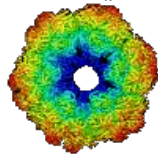
guiDETECTOR

Not (yet) possible

The following options are not (yet) possible:

- Run in batch mode.
- Store output files and results of different pages in different sub-directories of the working directory.





IMAGIC

guiDETECTOR

[Feedback / Error hints](#)

We intensively tested the **guiDETECTOR** program and tried to find all possible errors and inconsistencies. But the current program is very complex and still in progress. So you may still find some problems.

We are happy to get feed-back. Please send your comments, error hints etc. to

imagic@ImageScience.de

THANK YOU VERY MUCH.



Image Science

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