OneView Camera

Model 1095 User Manual

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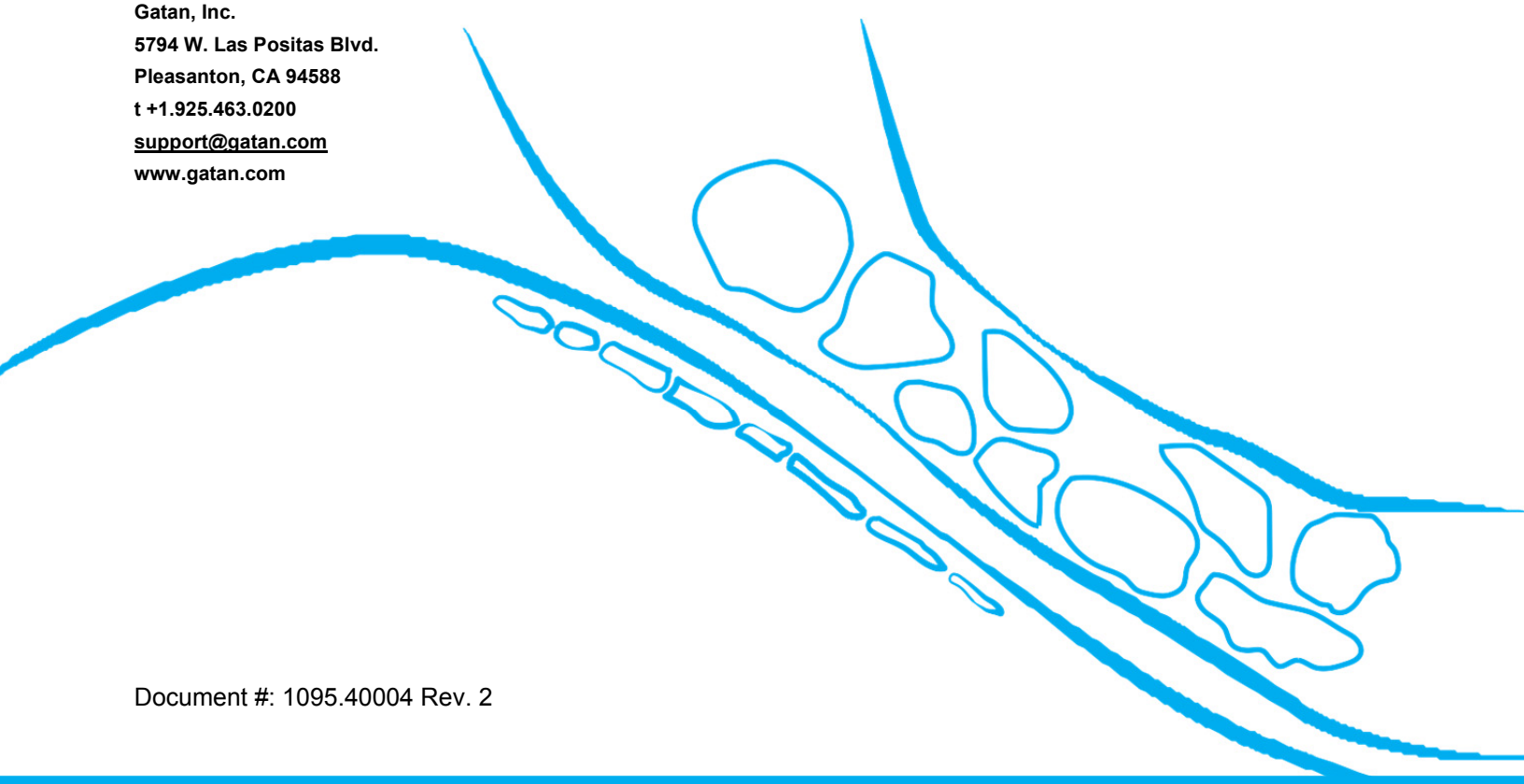

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1 Introduction

1.1 OneView camera system

With the OneView™ you can use only one camera to capture high quality 16 megapixel still images and video in all of your transmission electron microscope (TEM) applications.

Advantages

- Matches the benchmark image quality of the UltraScan® US4000 camera
- Always have a “live” experience with 25 fps at full 4k x 4k resolution, no compromise to resolution between viewing and recording images
- Guarantee optimal image quality with real-time drift correction and outlier removal using in-line data processing
- Detect single electrons with highest signal-to-noise ratio using the most sensitive scintillator and fiber optics available
- Extend the dynamic range beyond 16-bits, no beam stop required

- Increase productivity, even for novice users, with intuitive, built-in workflows to support and optimize recording modes

In-situ option

- Flexibly trade-off resolution against frame rate—from 4096 x 4096 pixels at 25 fps to 512 x 512 pixels at over 300 fps, always at 100% duty cycle
- Video buffer allows you to capture only the video you want; with post-event triggering and LookBack feature
- Never miss the start of an *in-situ* reaction again
- Tailor videos to your unique applications with powerful post-processing tools

1.2 Gatan Microscopy Suite interface

Launching Gatan Microscopy Suite® (GMS) 3.0 brings up the following screen, shown in Figure 1.

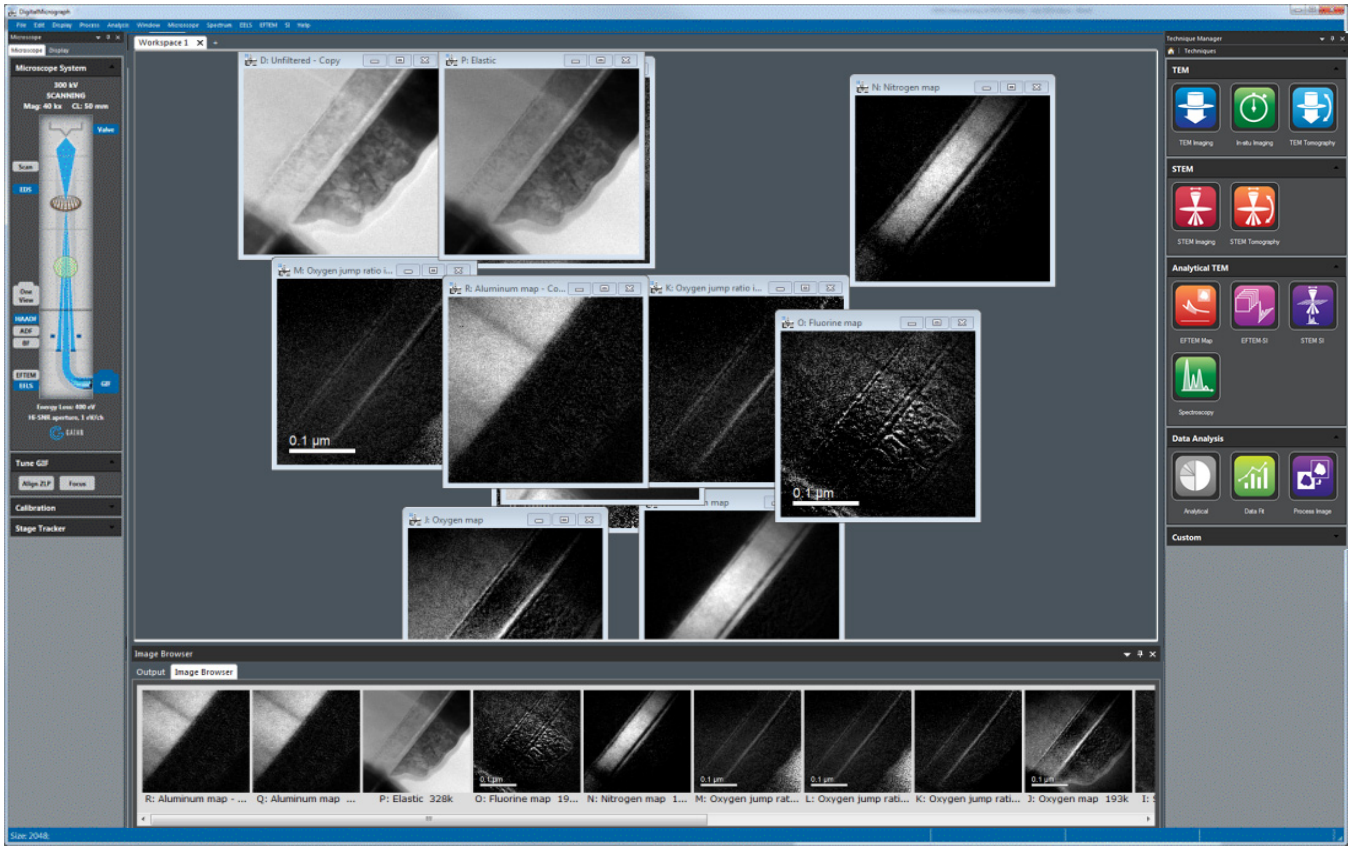

Figure 1: ***The GMS 3 interface, with the Microscope System panel on the left, the Workspace area in center, the Technique Manager panel on the right and the Image Browser and Output areas on the bottom.***

On the left: Microscope System panel, a schematic diagram of the microscope that graphically indicates:

- Whether the camera is inserted or retracted into the beam: Inserted shown
- The current choice of either imaging or diffraction mode: Imaging mode shown
- The presence of a sample: Grid shown
- The status of the viewing screen: Up is shown

In the center is the Workspace where images are shown.

At the bottom of the screen are two tabbed panels, one for text output and another showing recently acquired image icons.

On the right is the Technique Manager, with as many as five groups of techniques (Dependent on licenses):

1 TEM: TEM Imaging; In-situ Imaging; and TEM Tomography

2 Scanning TEM (STEM): STEM Imaging and STEM Tomography

3 Analytical TEM: Energy-filtered TEM (EFTEM) Map; EFTEM spectrum imaging (SI); STEM SI; and Spectroscopy

4 Data Analysis: Analytical; Data Fit; and Process Image

5 Custom

2 Operation of the OneView camera

IMPORTANT
Before installing and operating this produce, and to avoid the risk of injury and potential hazards, read and review the Regulatory Pamphlet and follow all safety instructions.

2.1 OneView imaging

Specimen viewing and single-frame acquisition are organized exactly as for traditional charge coupled device (CCD) cameras. The two modes View and Capture, can each be set up with separate preferences. Subareas can be selected as well.

The TEM imaging application setup has two operating modes which are preset for easy use; View mode allows the user to observe live (continuous) images on the monitor and Capture mode is for standard acquisition of images.

The TEM *in-situ* mode acquires a series of images.

2.2 OneView TEM imaging modes

After starting GMS software, discussed below, the steps to acquire single images is to first prepare the Dark and Gain References, discussed below, and then:

1 Start the TEM Imaging application

2 Select Image Format and Binning

3 Select the Exposure Time

4 Choose View or Capture

Choosing the technique TEM Imaging shown in the upper right of Figure 1 will result with the Camera palette appearing on the upper right panel as detailed in Figure 2. When selecting View, a live image window appears in the View panel. When selecting Capture, an icon representing the image is placed into the Image Browser (bottom panel), and a new Workspace is created with the image in the Workspace.

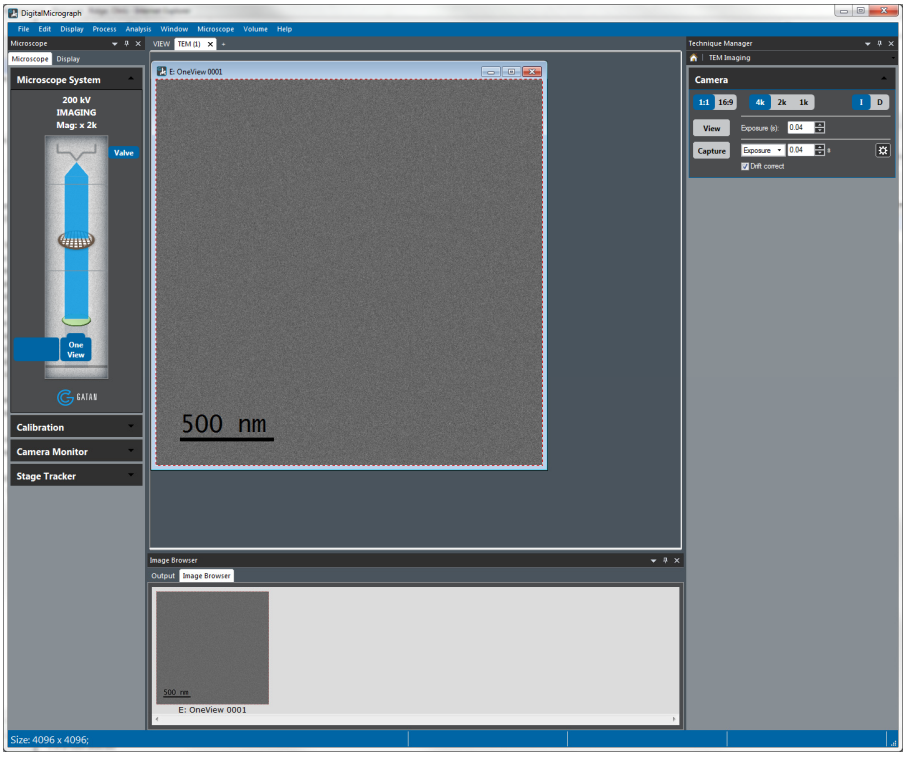

Figure 2: ***GMS screen shot showing captured image in the workspace TEM.***

Once the references are acquired, images can be acquired in two simple ways, View and Capture. In the Camera palette, the top row shows:

- The choice of 1:1 or 16:9 image format
- A binning choice of 4k (4096 x 4096), 2k (2048 x 2048), or 1k (1024 x 1024) image size
- The TEM mode, I (imaging) or D (diffraction)

Note: When diffraction is chosen the schematic display of the TEM on the left side of the GMS window will change the blue depiction of the e-beam to indicate a diffraction setup on the TEM.

Shown in the lower portion are the View and Capture buttons.

For View, the exposure time shown in the control to the right of the View button shows the minimum exposure time available (0.04 s); longer times will result in two or more images being summed together.

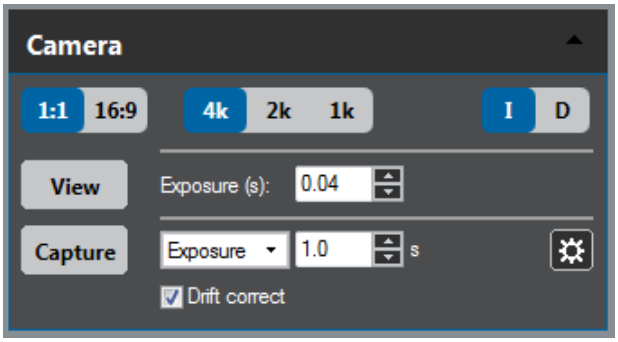

Figure 3: ***The Camera palette.***

For the Capture image acquisition, a drop down menu gives three choices for exposure time, as shown in Figure 4; Automatic; user selected Exposure time; and a user selected Dose calibrated in primary beam electrons (assuming calibration is done).

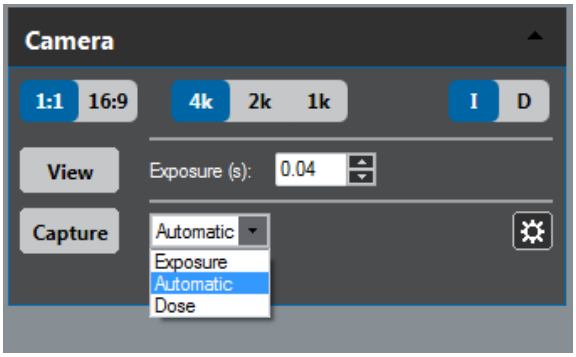

Figure 4: ***The Capture exposure control drop down list.***

The Capture control also has a tool button (the small gear to the right of the Capture button), and when clicked calls up the dialog shown in Figure 5. Options include Remove outliers (statistical outliers in pixel intensity value), Drift Correction, and a sub-option for drift correction, Clip to common area. The latter means only the regions covering the same part of the sample image are retained in the alignment process, and the outer margin that is not used in the aligned, summed images is cropped.

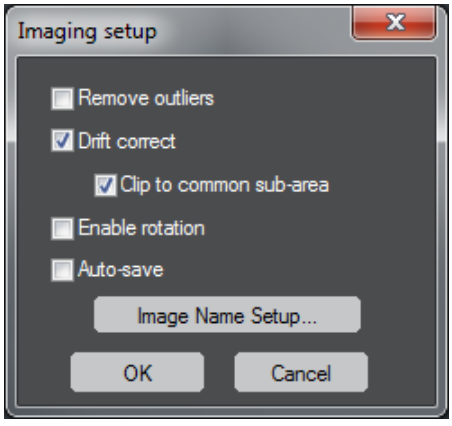

Figure 5: ***Clicking the Capture tool icon brings out these options in imaging setup.***

An example of how effective drift correction can be is shown in Figure 6.

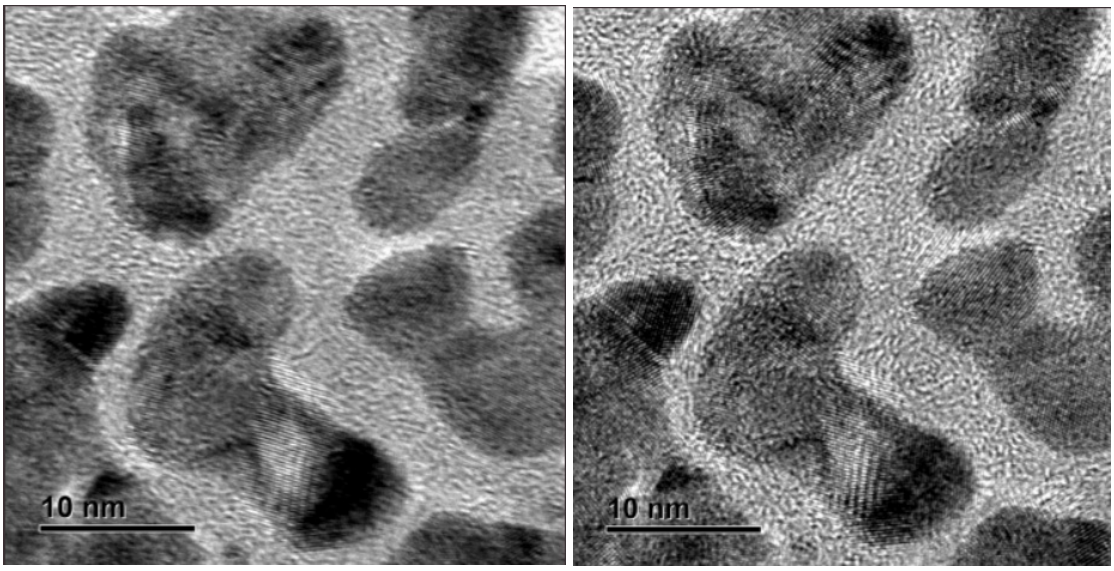

Figure 6: ***Comparison of high resolution of Au nanoparticle images acquired with drift correction on (right) and off (left).***

2.3 OneView camera in TEM *in-situ* mode

The TEM In-situ Imaging technique allows the capture of a series of images, and when the TEM In-situ Imaging technique icon shown in the upper right corner of Figure 1 is clicked, an In-Situ Acquisition and In-Situ Player palettes are created, and the In-Situ Acquisition palette is shown in the left side of Figure 7. The palette, In-Situ Player, will be described below.

The In-Situ Acquisition palette also has the same image formats as the Camera palette (1:1, 16:9, 4k, 2k, 1k, I and D). Other items in this palette are the Record button, along with Lookback Buffer and Disk Space progress bar indicators.

Clicking on the gear icon to the right of the View button brings up the In Situ setup dialog shown on the right side of Figure 7.

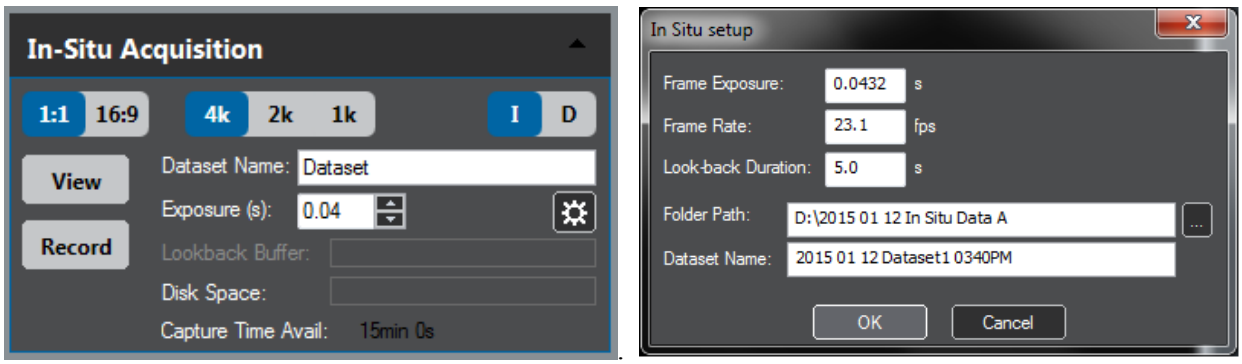

Figure 7: ***The In-Situ Acquisition control palette, on left, and In Situ setup dialog, on right.***

In the In Situ setup dialog, the Frame Exposure time is selected and resulting frame rate is shown. Since images are actually always being acquired, the *in-situ* system allows the Look-back Duration to adjust the amount of time over which images have already been acquired and are stored ahead of time when Record is clicked.

1 In Folder Path, enter desired header name for collected files and the folder where they will be stored

2 In Dataset Name, enter the filename of the image series

3 Set the Look-back Duration (maximum time shown on the graphical user interface)

4 Click OK to use these settings

5 Click on Record button to begin capturing data to the capture PC

6 Wait until finished capturing, then press Stop button

7 To take the next dataset, with the same file name, click on Record again, and with a new file name, change the Folder Path and click Record again.

The In-Situ Player palette is also brought up in the In-situ Imaging technique, and is shown in Figure 8.

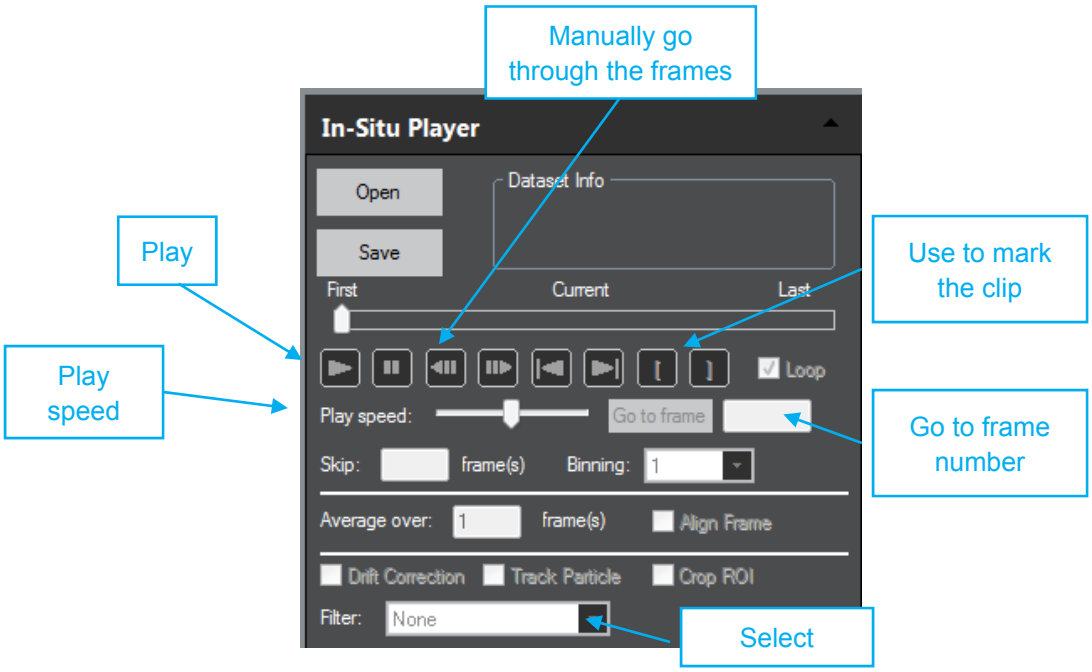

Figure 8: ***The In-Situ Player palette options for playback and post process of data.***

Once the clip is open, you have multiple options for previewing and post processing of the data:

1 Automatically playback the data set by selecting the Play speed and clicking on Play.

Note: Play speed setting here is only for the playback display and does not affect the saved dataset frame rate.

2 Manually move through the frames:

- Use the slider and move it to left and right to view a specific frame

- Set the step size (#frames) in Next/Prev step section and then click on the arrows
  - Setting to 1 means each frame in the dataset will be played
  - Setting it to 3 means playback frames will correspond to every 3rd frame
  - Specify the Frame/Time of interest and click on Go To

Note: The same step size selected here will be used when data is saved.

3 Crop:

To crop in time, use the left and right brackets to specify the section of the frames you are interested in.

To crop in space, use the ROI tool and select the region on the displayed clip. Then check Crop ROI. Once the data is Saved, DigitalMicrograph® (DM) automatically crops each frame to this specified region of interest (ROI).

4 Binning: In order to improve the signal-to-noise ratio (SNR) or to save disc space you can bin these frames by 2, 4 or 8.

5 Average over frames: determines how many frames from your dataset will be averaged to produce one frame of playback.

Setting to 1 means no averaging while setting to 3 means that the n^{th} frame of playback is the average of the $(n-1)^{th}$, n^{th}, and $(n+1)^{th}$ frames from the original dataset.

Note: If you check Average Frame Alignment, with averaging over 3 frames, each 3 frames are aligned before they are averaged to replace the n^{th} frame. This is very useful in cases where the sample drifts in between frames.

Examples

1 If you set Averaging to 3 and the step size is 1, the first frame will be 1/2/3, then the 2nd frame will be frames 2/3/4, then the third frame will be 3/4/5, etc.

2 If you set Averaging to 3 and step size of 3, the first frame will be 1/2/3, then the 2nd frame will be 4/5/6, then the third frame will be 7/8/9.

Note: the same Average settings selected here will be used when data is saved.

3 **Drift Correction:** Compensate for continuous drifting effects in the sample, allowing a drifting sample to appear stationary in processed images. For example, if you set Average Over 3 frames, each 3 frames will be averaged and then averaged frames will be drift corrected. This is especially helpful when the dose in each frame is too low to do drift correction correctly.

4 **Track Particle:** The user draws an ROI on a feature that is moving in the original dataset, DM will drift correct for the selected feature and crop to this specified ROI to keep this feature in the center of the frame in processed images.

5 **Filter:** Most of the processes explained above are done using cross correlation between consecutive frames. In some cases application of imaging filter may improve the results.

2.4 Power on sequence for OneView

1 Close DM if it is open (or power on the PC if it is off).

2 Turn on the OneView controller by touching the blue Gatan logo shown on the left of the controller in Figure 9; a detailed view of the indicating LEDs is in Figure 10.

Figure 9: ***OneView controller.***

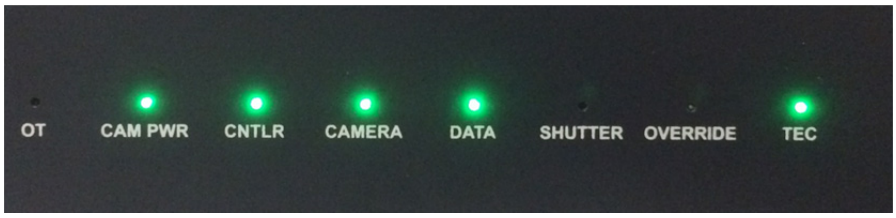

Figure 10: ***OneView controller's indicating LEDs.***

3 Wait at least 3 min after powering on the OneView controller.

4 Launch DM software.

2.5 Power off sequence for OneView

Under normal circumstances, you can leave the camera running. If necessary, you can power down the camera completely.

1 Close DM

2 Turn off controller by touching and holding the blue Gatan logo on the left of the controller for >3 s

3 Turn off the PC if desired

2.6 Setting temperature

For normal use the camera's image sensor must be cooled, and this is done by a menu choice shown below in Figure 11 (Must be in Power User mode).

1 In the Camera menu, select Temperature and enter the set point value. The typical operating temperature is -5 ˚C.

2 If the camera chamber needs to be vented, set this to +20 ˚C

3 Wait for warm-up to complete before venting

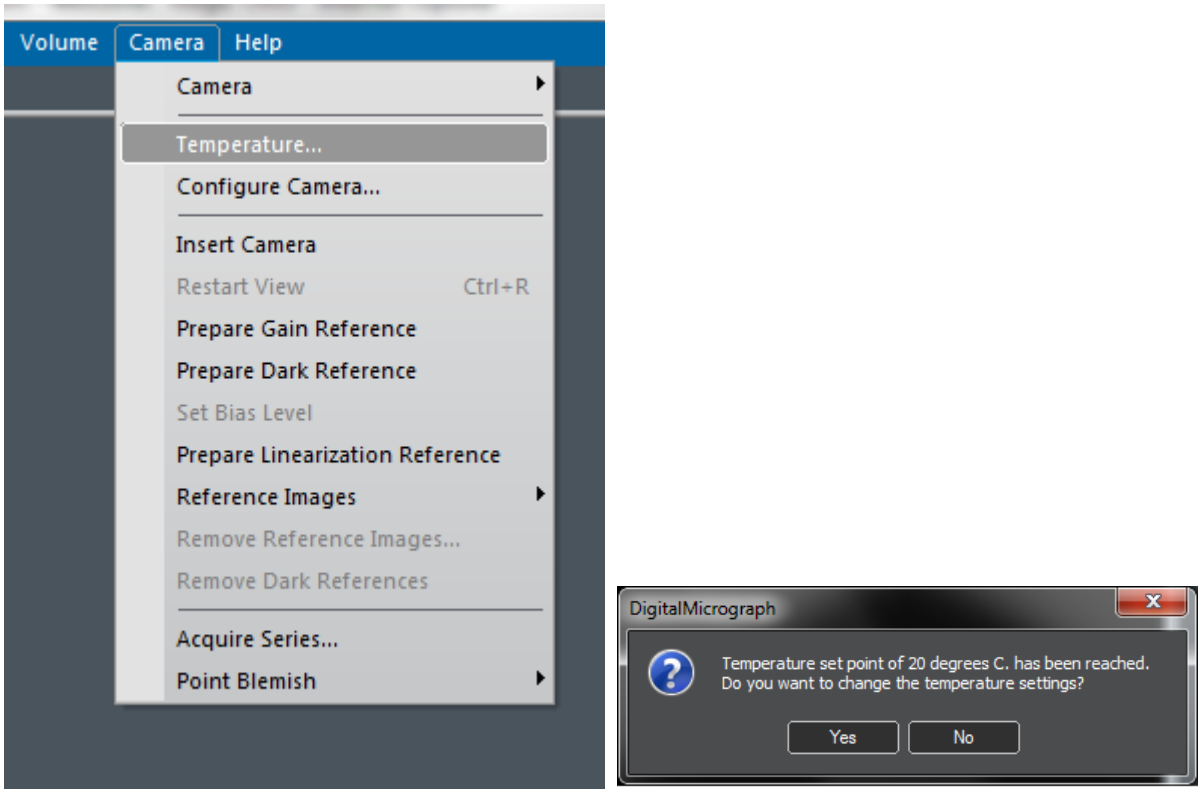

Figure 11: ***Camera menu with the Temperature choice highlighted, and the dialog.***

2.7 Obtaining reference images

Before images can be acquired, both the Dark Reference and the Gain Reference must be prepared. These are accomplished by menu choices shown in Figure 12, where Prepare Gain Reference is shown selected.

For the first one to be done, Dark Reference, the resulting message dialogs will be presented in Figure 13.

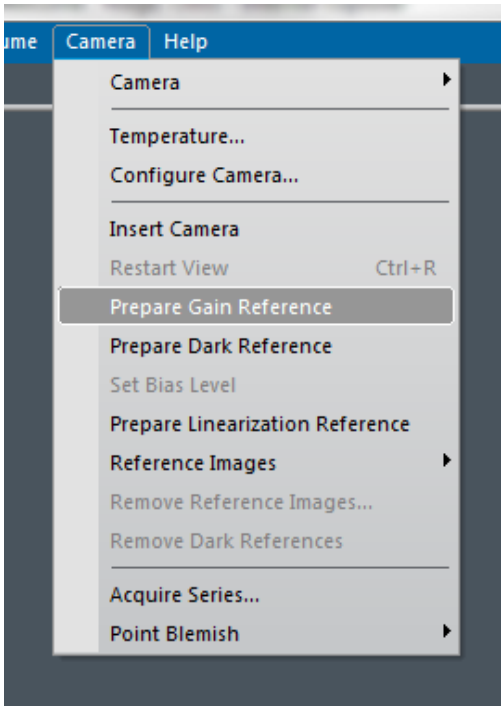

Figure 12: ***Preparing the Dark Reference and, shown selected, the Gain Reference.***

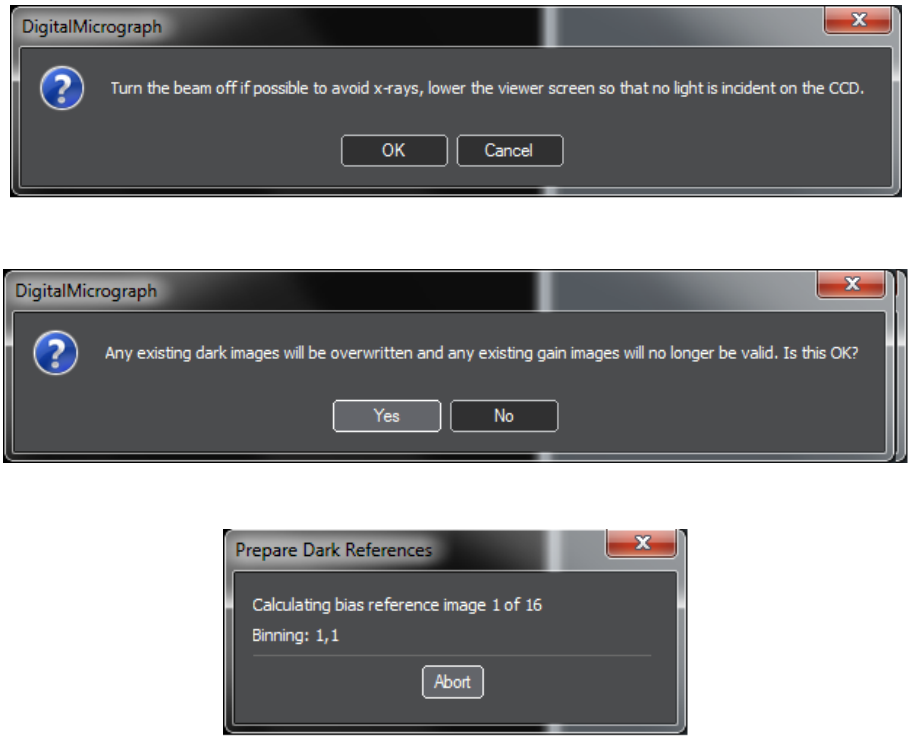

Figure 13: ***Dialogs from the Dark Reference step, asking that the beam be off and viewing chamber be covered, followed by a warning about replacing the old reference. Below that is a progress window showing the acquisition of the various images that are needed.***

For the Gain Reference, a dialog will be presented where the various parameters of the process are determined, shown in Figure 14.

Uniform illumination is required for the gain reference, so some TEM/Sample position adjustments are needed (e.g., finding a hole in the sample).

After the parameters are chosen, an image View will be shown as in Figure 15 with a text annotation in a large font indicating the beam intensity, allowing convenient adjustment on the TEM to get the desired intensity. A dialog will be also shown allowing completion of the process.

After Done is clicked, dialog will be then presented for confirming the choices and allowing confirmation of the choices, or canceling, shown in Figure 16.

When the parameters are confirmed by clicking OK, the gain reference is acquired, and a progress window is shown in Figure 17.

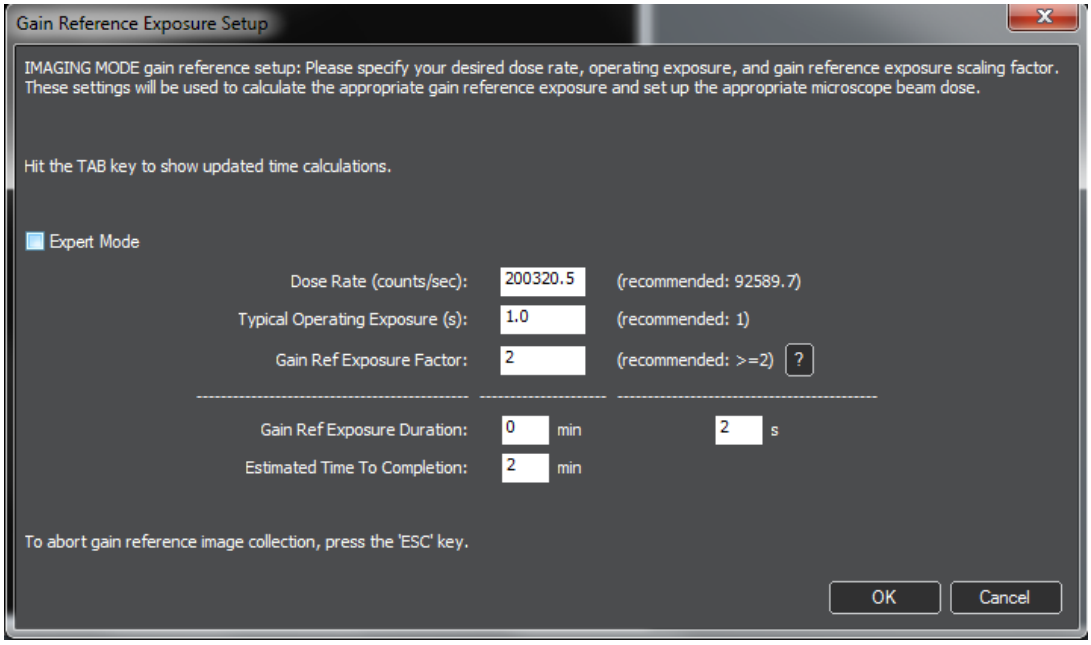

Figure 14: ***Gain Reference Exposure Setup dialog.***

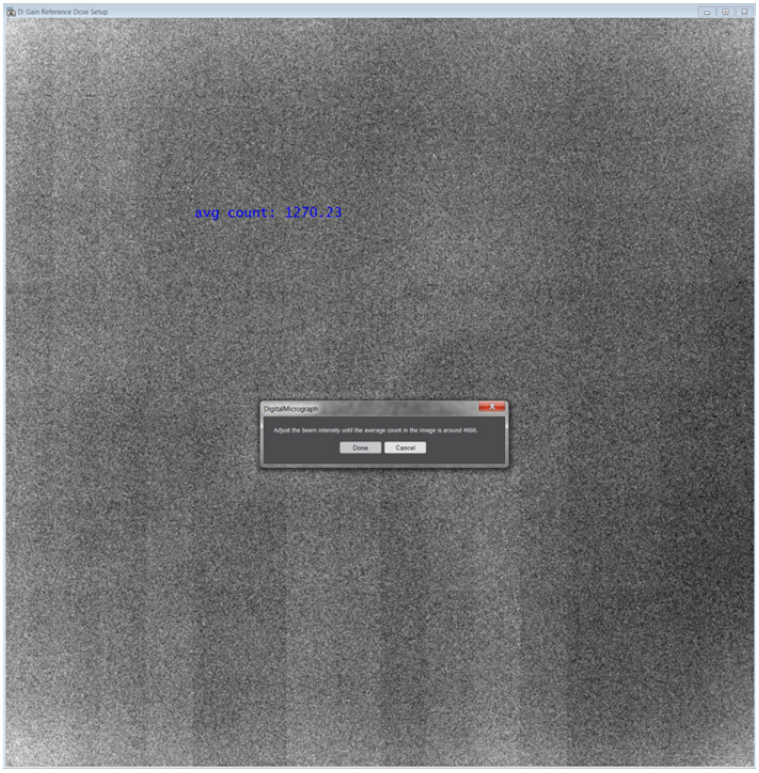

Figure 15: ***During the acquisition of a Gain Reference, an image view is shown, with the resulting intensity written as an annotation, and with a dialog for completion of the adjustment.***

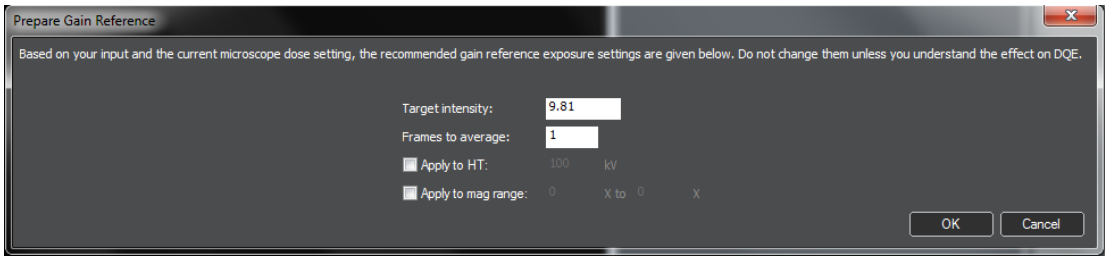

Figure 16: ***After the intensity, adjustment, a confirmation dialog will come up showing the result of the parameter choices, and clicking OK will acquire the gain reference.***

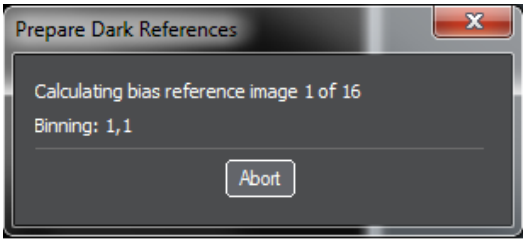

Figure 17: ***The gain reference progress window.***

2.8 Care of detector

2.8.1 Temperature

Typical operating temperature is -5 °C. Monitor the temperature; the Camera Monitor palette indicates the camera temperature.

Check the flow of cooling water periodically. If the flow rate of the cooling water deviates significantly from the value originally set (~15 L/h) make sure the lines are not obstructed and adjust the pressure regulator to bring the flow back to the original level. If the water flow stops while the Peltier cooler is on, damage to the camera may result.

2.8.2 Insertion indication

The blue and white Gatan logo on the side of the camera lights up continuously when powered. When the Camera Inserted box is checked in the Camera View pallet and the camera is inserted, the Sensor Inserted green LED on the bottom of the camera is illuminated, as shown in Figure 18.

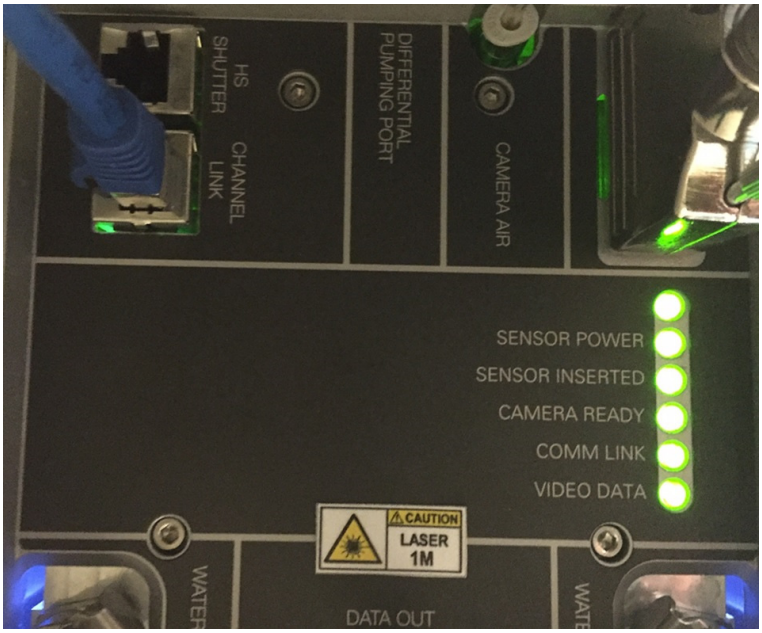

Figure 18: ***The OneView camera Sensor Inserted lights up when the camera is inserted (bottom of camera).***

2.9 Magnification correction/calibration

The displayed nominal magnification on TEM is for photographic film and has an accuracy of 5 – 10%. The OneView camera is located on a different plane (height wise) respect to the film camera. As a consequence, the magnification must be calibrated. The calibration is done using Reference calibration samples.

At low magnifications: Use a cross grating sample or any sample with known spacing.

At high magnifications: Use graphite or any crystalline samples with known lattice spacing and use the FFT method.

It is very important to make sure DM software correctly reads the TEM magnification. If the communication between the computer and the TEM is established, the magnification is read automatically. Otherwise, make sure DM software is set to prompt the user to enter a value for TEM magnification every time an image is to be acquired. This can be set by choosing the Global Microscope Info window under the Microscope menu.

2.9.1 Low magnification

Record an image of a cross grating replica, such as shown in Figure 19.

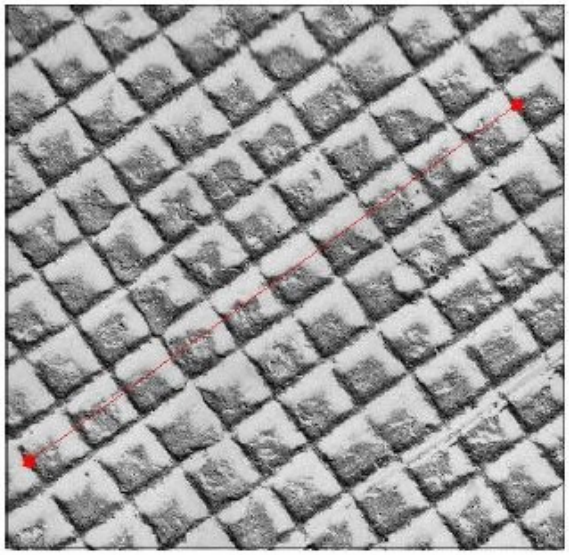

Figure 19: ***Example of marking a known distance during magnification calibration.***

1 Choose Microscope > Calibrate Image

2 Follow the instructions on screen, as shown in Figure 20 and a red line will appear on the image

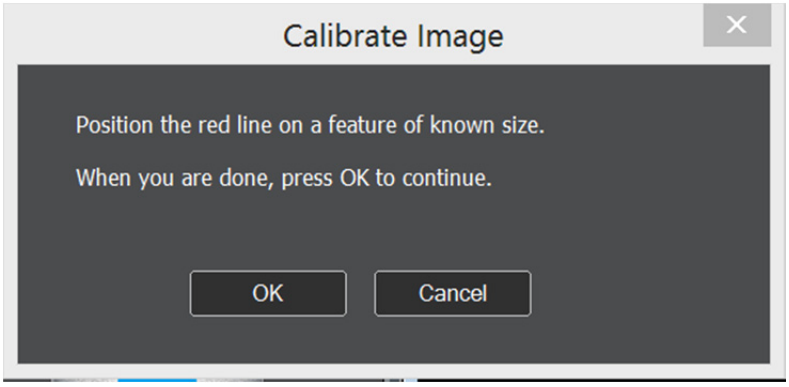

Figure 20: ***Magnification calibration instructions.***

3 Position the red line on a feature of known size

4 Press OK on the Calibrate Image window

5 Enter the correct distance for the selected feature (for example 10 line pairs of cross grating sample where the distance = 10 x 0.463 µm) in the Calibration window and select the units. Select the distance marked in the previous figure to perform the magnification calibration, in the Calibration dialog, as shown in Figure 21.

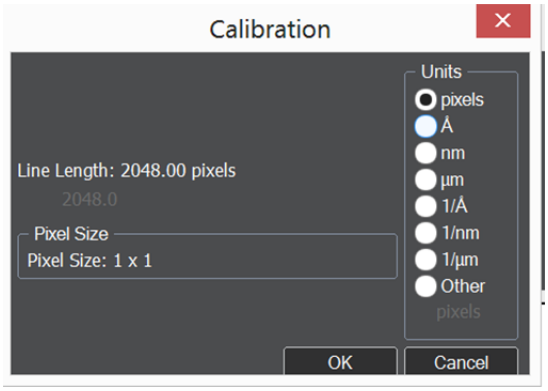

Figure 21: ***Calibration dialog.***

6 Press OK

7 The calibration can be checked on the calibration table containing pairs of value, the nominal microscope magnification and the calibrated value.

8 To view the magnification table, select Microscope > Calibrations

- The microscope calibration dialog shows the table of magnification calibrations stored for the current imaging device

2.9.2 High magnification

Record a lattice image of the crystalline sample.

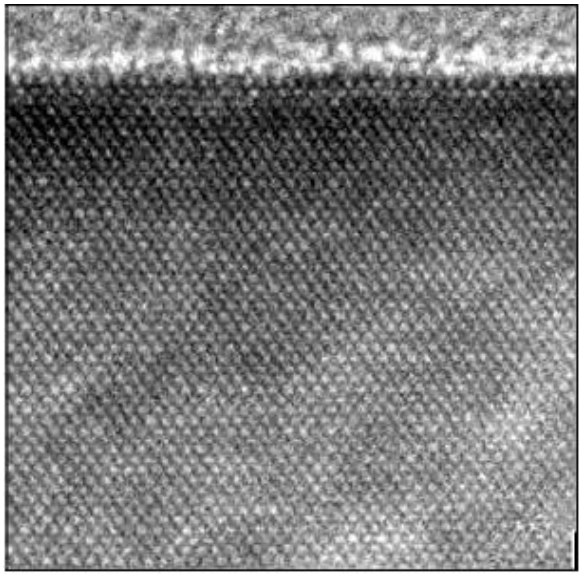

Figure 22: ***High resolution image of sample to be used in the magnification calibration.***

1 Select Microscope > Calibrate image from Diffractogram

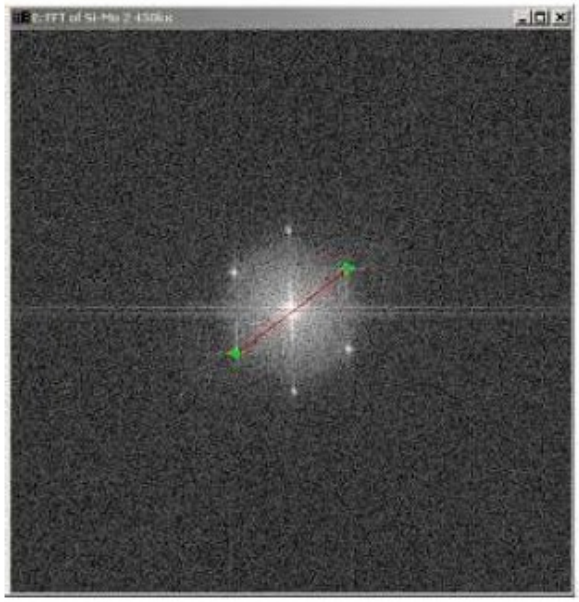

Figure 23: ***Distance between peaks in the calculated diffractogram.***

2 To calculate the diffractogram, follow the on screen instructions

3 A red line appears on the diffractogram indicating the distance between peaks

4 Position the endpoints of the red line on two symmetrical diffraction peaks

5 Press OK to specify the reciprocal unit and the d-spacing (in the corresponding real units) in the next window.

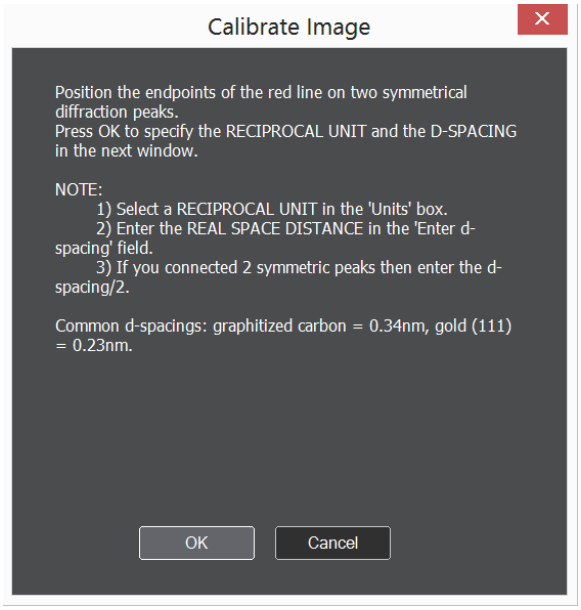

Figure 24: ***Calibration instructions.***

6 Read the calibration instructions and click OK, then enter the known spacing between peaks in the magnification calibration in the Calibration settings window.

2.10 Compatibility with other Gatan cameras and GIFs

All OneView cameras are fully supported within Gatan's industry leading software platforms, DM and GMS. GMS software gives you access to a wide range of additional applications to make electron microscopy easier and more efficient.

The OneView camera is retractable and is mounted below the TEM, in a standard Gatan camera housing, which allows simultaneous fitting of other cameras, Gatan imaging filters (GIFs) or electron energy loss spectrometers, to cover the entire range of imaging requirements.