

Using Falcon II camera on *Tecnai F30*

Chen Xu

cryoem@brandeis.edu

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12:08:37 xuchen Exp\$**

Falcon II/Orius camera system is now moved from *Tecnai F20* to *Tecnai F30*. As you probably knew this already, Falcon II has back-thinned chip sensor, comparing to Falcon I. The Tecnai software is also upgraded to version 4.5.1 now. This version of Tecnai software supports intermediate frame (movie) mode. **SerialEM** was also implemented with new features to support the movie mode. With current FEI hardware and software setup, the movie mode supporting is still limited, but it is better than nothing.

You can also get pdf version of this document here (article.pdf).

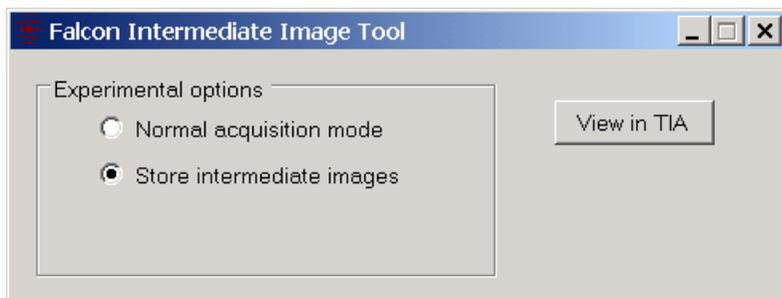
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1 Switching between Normal Image Mode and Intermediate Frame Mode

This is something new.

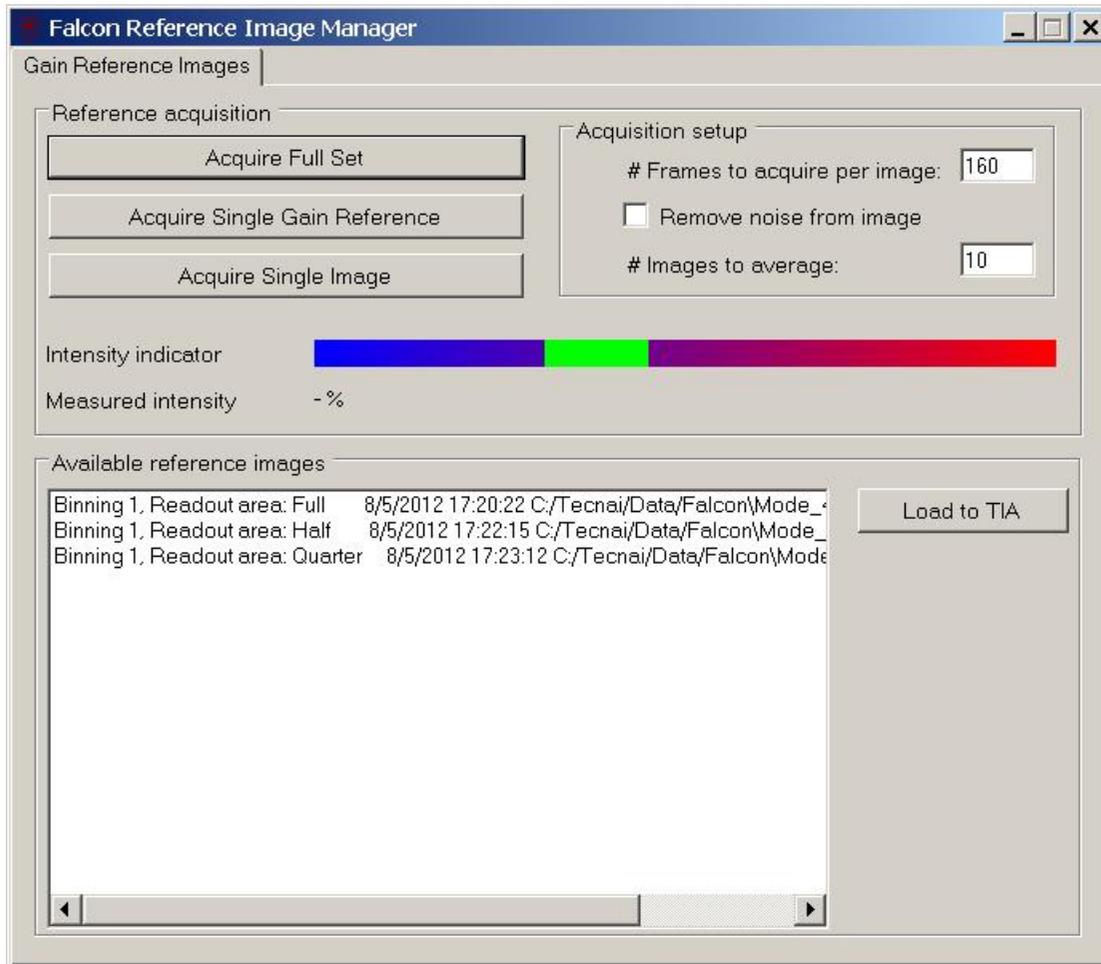
There is now an executable to run so we can switch between the two modes. Here is the window.



It is important to switch to "Normal acquisition mode" in the end of your session. In fact, when you exit from **SerialEM**, it will pop up a message window to remind you of this.

2 Prepare Gain Reference Files from Tecnai Interface

To prepare gain reference for Falcon camera, we have to use the "Falcon Reference Image Manager". Its interface is as below.



The procedure to obtain gain reference for Falcon is as:

1. Select "Normal acquisition mode" from Falcon Intermediate Image Tool window.
2. Launch "Falcon Reference Image Manager" from Tecnai → CCD Camera → (flap out) Bias/Gain
3. Adjust magnification so that it satisfies pixelsize at specimen level and adjust Spotsize(C1) and Intensity(C2) to a level that screen current reading is about 1nA. Lift large screen.
4. Click on "Acquire Single Image" from Falcon Reference Image Manager. A black vertical line should end in green region. Otherwise, adjust C1 or/and C2 and do it again.
5. Now, click on "Acquire Full Set" from Falcon Reference Image Manager. Use 160 for "# Frames to acquire per image" and 30 for "# Images to average", for a decent gain reference.
6. With newly file highlighted in "Available reference images" area, click on "Load to TIA" button on right side of Falcon Reference Image Manager window.

7. You should see two files, one is RAW and the other is XML, in the folder called C:\Tecnai\data\Falcon\Mode_4 get updated at this point.
8. Manually copy the above two files into the parallel folder Mode_7. This makes sure that the updated gain reference files are used for intermediate frames, as well.

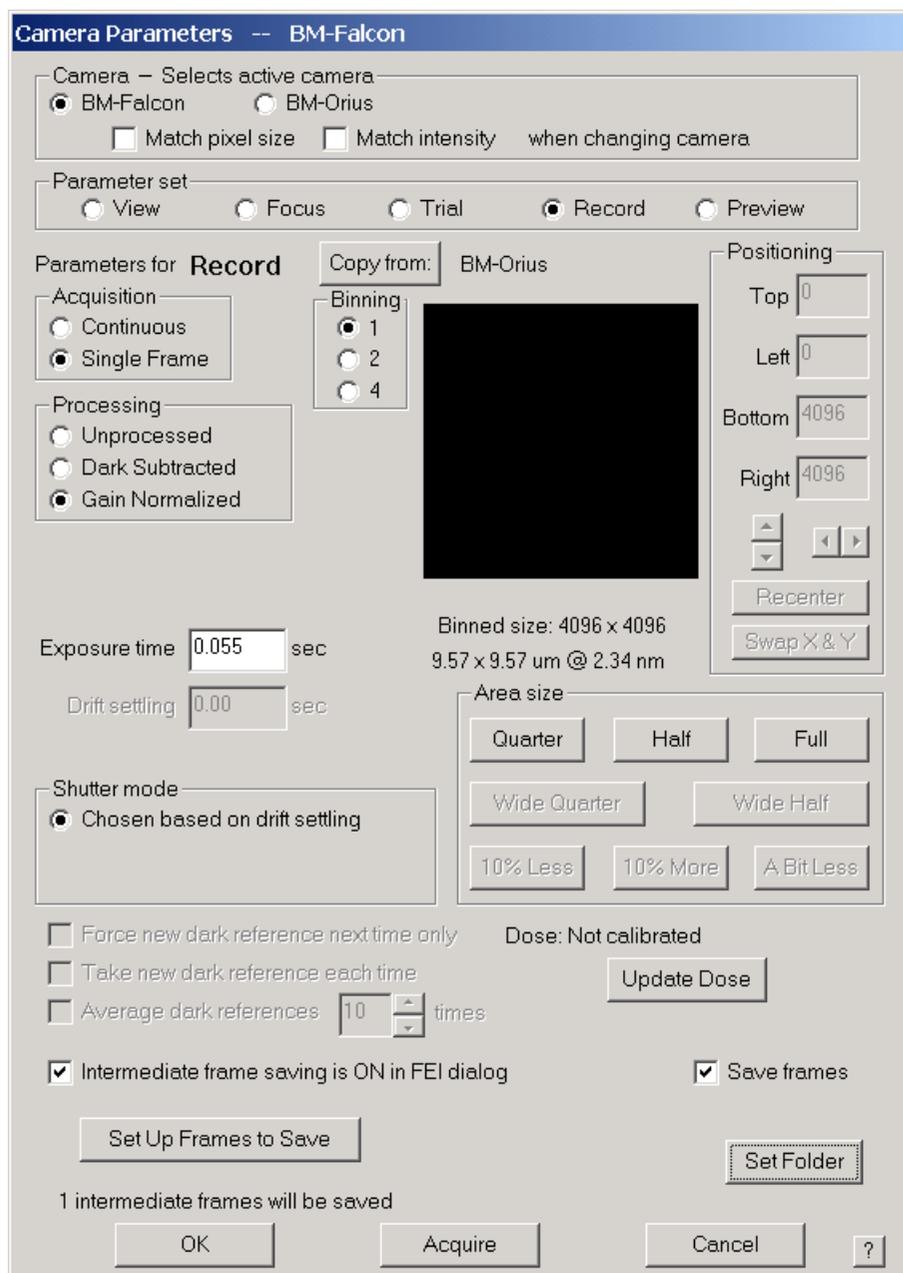
3 Intermediate Frame Mode with SerialEM

Currently, Falcon camera can output frames at rate about 17~18 frames per seconds. If one defines a very long exposure, the camera system can generate many frames. However, possibly due to the limitation of computational power of Tecnai XP computer, the maximum numbers of chunks can be saved to disk drive is 7, for any single exposure. In the world of **SerialEM**, they are called frame and sub-frame, equivalent to FEI's term - chunk and frame. Which frame containing which sub-frames are defined by an XML file. From Tecnai User Interface, one has to edit this file manually, which is annoying.

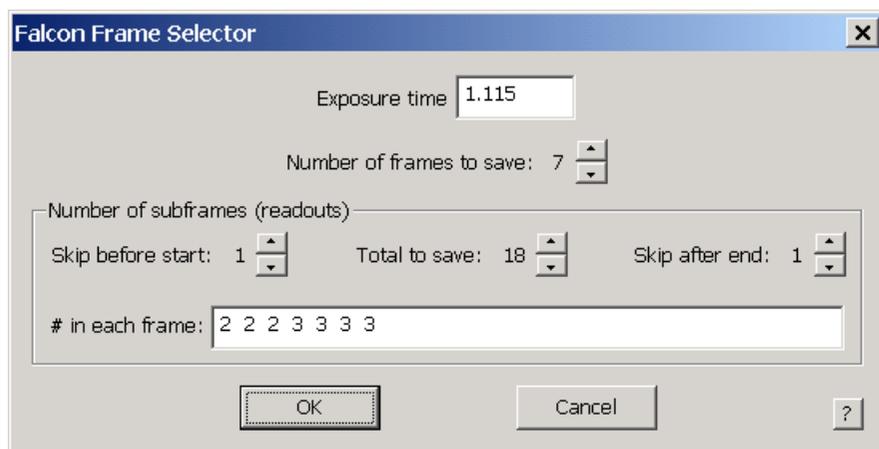
SerialEM tries to work around this. Put following three lines to general section of your property file.

```
MaxFalconFrames          7
FalconFrameConfigFile    C:\tecna\data\Falcon\IntermediateConfig.xml
LocalFalconFramePath     C:\Temp\Falcon-dump
```

SerialEM tries to simplify user input by using a frame selector dialog window which can be launched from Camera Parameter Setup window, as shown below.



When the check box next to "Intermediate Frame Saving is ON in FEI dialog", then the button below it "Setup Frames to Save" becomes clickable. Clicking on this button will bring up a frame selector dialog window, as shown below.



This frame selector dialog window makes our life a lot easier!

The frames/chunks written to harddrive are in format of raw, 4-bytes integer, with 49 bytes header. They can be viewed via TIA interface, by clicking "View in TIA" on Falcon Intermediate Image Tool window and then select the file.

SerialEM can stack the series of raw frames together to a single MRC stack file, with 16-bit as default. Therefore, it is recommended because it takes less disk space. This stacking process writes frames to stack and deletes the frames, one by one. **SerialEM** can even take a few exposure and combine all the frames into a single MRC stack. This has to be done from macro command.

```
DeferStackingNextShot  
R  
DeferStackingNextShot  
R  
R
```

The above macro will make three exposure and output a single MRC stack file.