

DeltaVision Ultra

32-Bit Deconvolutions

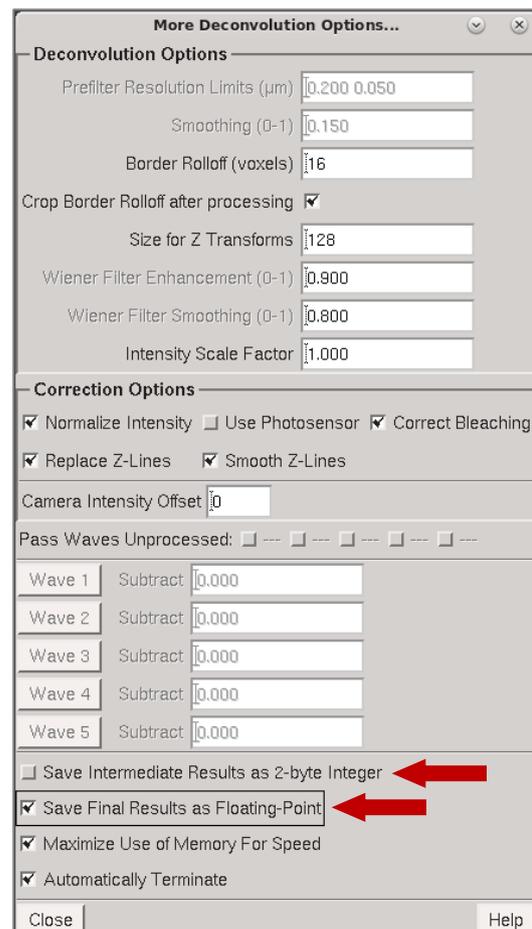
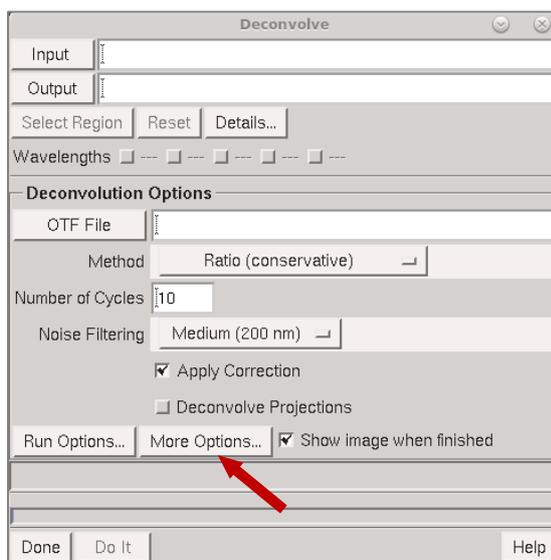


By default, softWoRx converts deconvoluted images to 16 bit. This feature is problematic with the 16-bit images from the sCMOS camera, because deconvolution increases a signal, which can, hence, exceed the 16-bit range. Some images within a series of samples (with some high signal) may be rescaled to fit 16 bits, while others (with lower signal) may be left unchanged, making it impossible to compare them side-by-side or to stitch them. The following instructions describe workflows for deconvolutions in a 32-bit range, circumventing this problem.

1. Manual deconvolutions

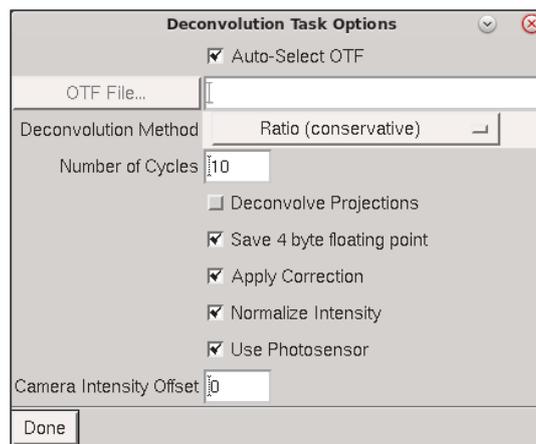
In softWoRx, setup a deconvolution as usual. In the "Deconvolve" dialog, click "More Options...". In the dialog popping up

- uncheck "Save intermediate Results as 2-byte Integer"
- check "Save Final Results as Floating-Point"



2. Deconvolutions as background task

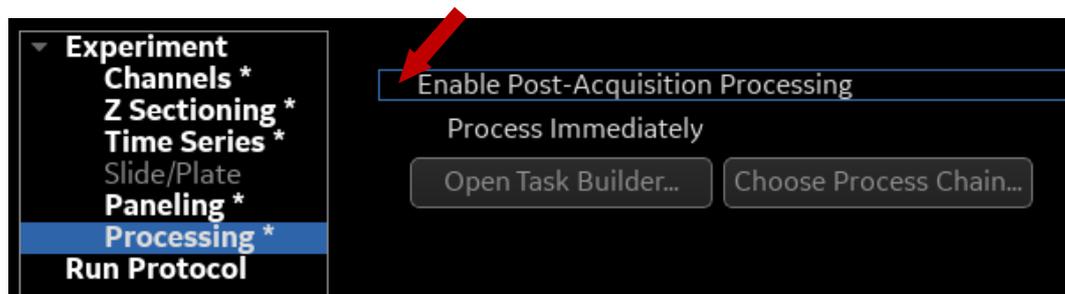
Acquire Ultra allows to perform deconvolutions as background task while acquiring images. The standard Task Builder chain can be used, but the Task Builder GUI exposes only a reduced set of optional parameters. To perform 32-bit deconvolution, select "Save 4 byte floating point" in the "Deconvolution Task Options" dialog:



If you want to have full control on all parameters, use the following procedure instead of the Task Builder:

a) Acquire Ultra

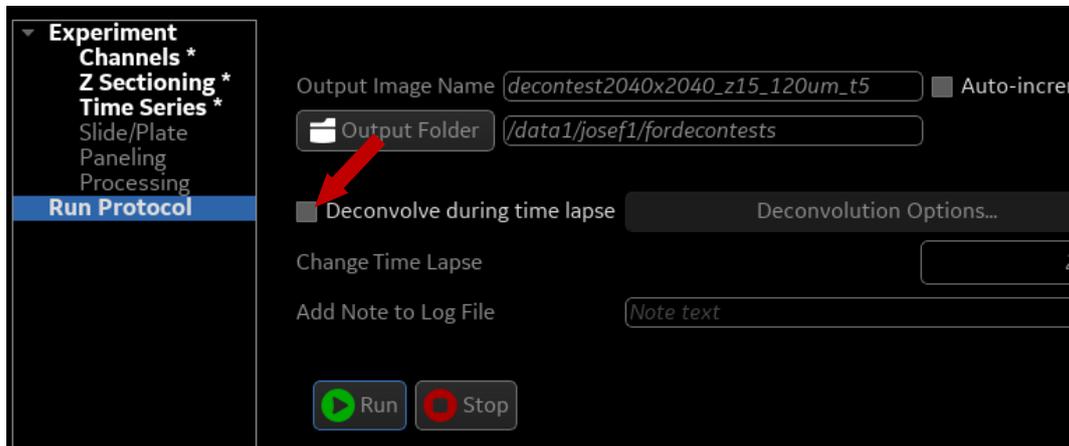
- In the "Processing" tab, *uncheck* "Enable Post-Acquisition Processing"



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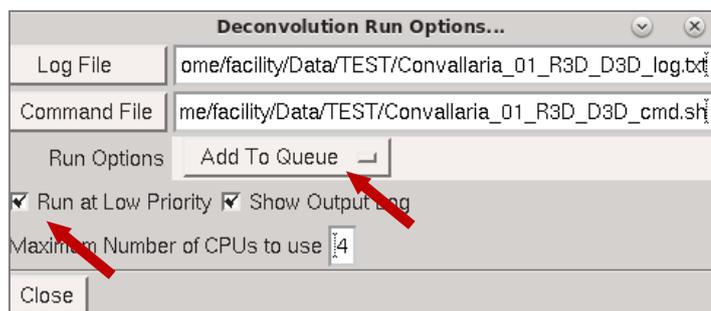
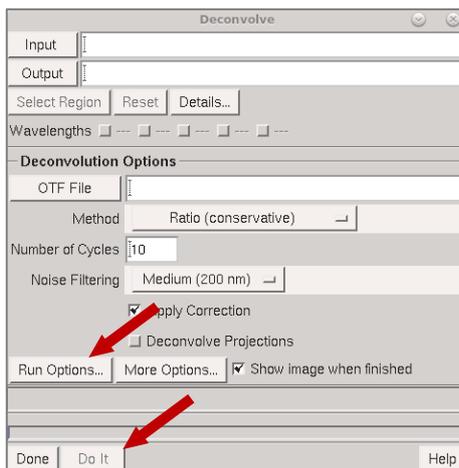
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- In the "Run Protocol" tab, *uncheck* "Deconvolve during time lapse"
- Acquire the first image

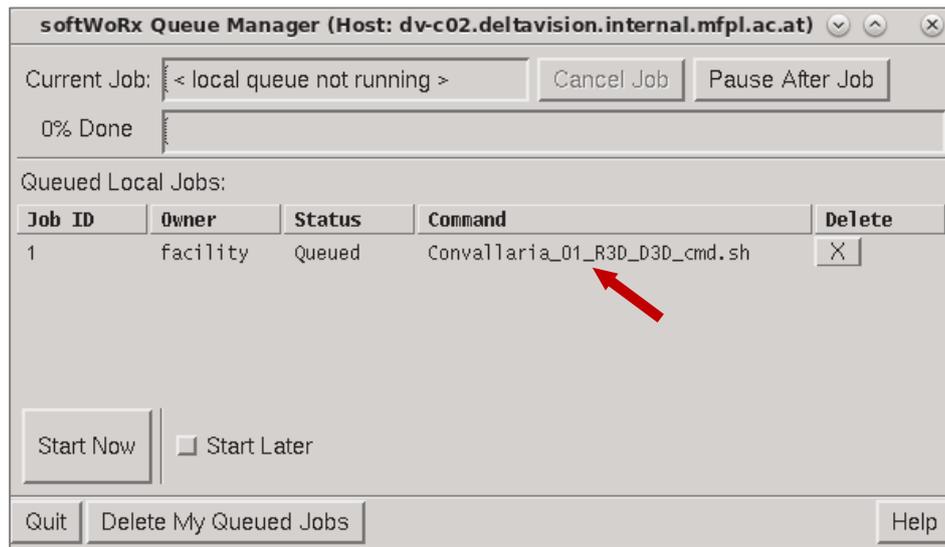


b) softWoRx

- The first deconvolution has to be done manually, including the adjustments for 32-bit deconvolutions as outlined in chapter 1 (Manual deconvolutions)
- Once all parameters are set, click "Run Options". Check "Run at Low Priority". In the "Run Options" combo box, select "Add To Queue", then "Close". In the "Deconvolve" Dialog, click "Do It"

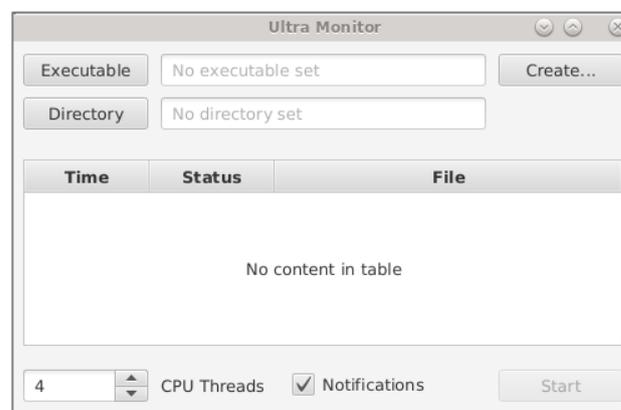
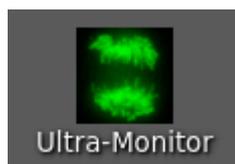


- The "Queue Manager" dialog should pop up and display the job submitted, including the name of the command file (ending with "_R3D_D3D_cmd.sh"), which is required in the next step.



c) Ultra-Monitor helper program

- On the desktop, click on the "Ultra-Monitor" icon
- Click "Create...". First select the command file from the previous step and then choose a file name for the executable to be created. The file name will end with "_R3D_D3D_exe.sh". By clicking "Executable...", you can reuse a previously created executable without creation of a new one (as long as your acquisition protocol remains unchanged)
- Click "Directory" and select the folder in which Acquire Ultra saves your images



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- Click "Start" to monitor the selected directory. If the "Notifications" check box is checked, a notification will appear upon completed deconvolutions (in the lower right corner of the screen)
- The control "CPU Threads" allows to adjust the number of CPU kernels used (a high value interferes with fast acquisitions). The maximum is 8, the default is 4

d) Limitations

The helper program tolerates different slice numbers or time points in time series, as well as changes in exposure time or intensity. All other changes of the acquisition protocol (in particular addition or removal of channels) may require creation of a new executable from a softWoRx deconvolution command file. Deconvolutions of running time series is not possible (they will be processed automatically once a series has ended). Multi-image acquisitions ("Paneling") have not yet been tested.