

bAlign Batch 6
Oct 24, 2014

Purpose:

Convert a hard-drive folder of .tif files and perform the following:

- Split 2 channel files into _ch1.tif and _ch2.tif channel separated files.
- Perform slice-by-slice stack alignment on one channel and apply transformations to the other.
This requires 'MultiStackReg' to be installed (<http://bradbusse.net/downloads.html>).
- [optional] For scanimage4 .tif files, remove linear calibration
- [optional] For scanimage4 .tif files, crop all files to a rectangle specified, in pixels, as (left, top, width, height)
- Save 8-bit versions

Installation

- Drag and drop bAlign_Batch_v6.py into your Fiji 'plugins' folder
- Make sure you have 'MultiStackReg' plugin installed, you can download it from:
<http://bradbusse.net/downloads.html>
http://robertcudmore.org/software/download/MultiStackReg1.45_.jar

Running

- Select 'bAlign Batch v6' in Fiji plugin menu
- Select the hard-drive folder with .tif files to convert
- fill in options
- press 'Ok'

Options

Channels

- Get number of channels from scanimage 3.x or 4.x header.
- Otherwise, assume all stacks have this number of channels.

ScanImage4

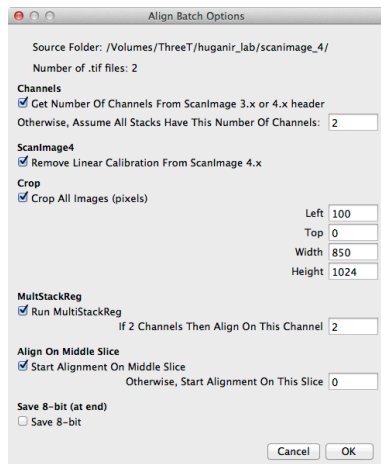
- remove linear calibration
 - crop all images
- Note:** Each image is cropped using the specified rectangle (in pixels).
If your stacks have a mixture of 1024x1024 and 512x512 images you want to put them into separate folders and run bAlignBatch6 on each folder, specifying the proper rectangle (in pixels).

MultistackReg

- Run multistack reg (requires additional plugin)
 - if 2 channels then align on this channel
 - Start alignment on middle slice
 - Otherwise, start alignment on this slice
- Note:** if you specify a slice below the bottom of a stack (for any of the stack you are converting), the FIRST slice of the stack will be selected for alignment.

Save 8-bit

- save another folder with 8-bit copies of your channel separated .tif stack
- Note:** These 8-bit versions are saved in a different folder. The main output folder still contains your channel separated originals with the original bit-depth (usually 11-bit for ScanImage).



The image shows a screenshot of the 'Align Batch Options' dialog box. It has a title bar with standard window controls. The content is organized into sections with checkboxes and input fields. The 'Source Folder' is set to '/Volumes/ThreeT/huganir_lab/scanimage_4/'. The 'Number of .tif files' is 2. Under 'Channels', the checkbox 'Get Number Of Channels From ScanImage 3.x or 4.x header' is checked, and the 'Otherwise, Assume All Stacks Have This Number Of Channels' field is set to 2. Under 'ScanImage4', the checkbox 'Remove Linear Calibration From ScanImage 4.x' is checked. Under 'Crop', the checkbox 'Crop All Images (pixels)' is checked, and the 'Left', 'Top', 'Width', and 'Height' fields are set to 100, 0, 850, and 1024 respectively. Under 'MultiStackReg', the checkbox 'Run MultiStackReg' is checked, and the 'If 2 Channels Then Align On This Channel' field is set to 2. Under 'Align On Middle Slice', the checkbox 'Start Alignment On Middle Slice' is checked, and the 'Otherwise, Start Alignment On This Slice' field is set to 0. At the bottom, there is a 'Save 8-bit (at end)' section with an unchecked checkbox 'Save 8-bit'. 'Cancel' and 'OK' buttons are at the bottom right.

Align Batch Options

Source Folder: /Volumes/ThreeT/huganir_lab/scanimage_4/
Number of .tif files: 2

Channels
☒ Get Number Of Channels From ScanImage 3.x or 4.x header
Otherwise, Assume All Stacks Have This Number Of Channels: 2

ScanImage4
☒ Remove Linear Calibration From ScanImage 4.x

Crop
☒ Crop All Images (pixels)
Left: 100
Top: 0
Width: 850
Height: 1024

MultiStackReg
☒ Run MultiStackReg
If 2 Channels Then Align On This Channel: 2

Align On Middle Slice
☒ Start Alignment On Middle Slice
Otherwise, Start Alignment On This Slice: 0

Save 8-bit (at end)
☐ Save 8-bit

Cancel OK