pro PSratiodual2

; 6-26-06 Written by Dan for "classic" excitation-based polarized TIRF.

; This program:

; (a) imports full images of sequentially one p-excited 514p, one s-excited 514s, one different color 442 (cerulean). This is one movie where the first image is cerulean, the second S-pol and the third p-pol

-offcell background subtraction or ROI background subtraction as an option; Subtract the off-cell background mean in each frame from the original floating point stacks.

; (b) takes a ratio of the two images, pixel by pixel (R\_image=P\_image/S\_image) where S\_image is zero or negative or P\_image is negative, then R\_image is set to zero) Ri

R\_Image=Max(R\_imagefloat)

; (c) displays the ratio image and records a 16-bit tif image file of the ratio.

Here is how he further process the p/s ratio we can talk about this over the phone.

if multratioNON lt 10000. then multratioNON=10000.

print,'The recorded 16-bit tif RATIO files have pixel values multiplied by', multratioNON

print,'Now writing RATIO TIF file #...'

for iframe=ifirst,nframes-1 do begin

print,iframe

singlemeanNON=mean(RfloatNON[\*,\*,iframe])

if iframe ge 0 and iframe le 9 then zeroes='000'

if iframe ge 10 and iframe le 99 then zeroes='00'

if iframe ge 100 and iframe le 999 then zeroes='0'

if iframe ge 1000 and iframe le 9999 then zeroes=''

quotsingleNON=REFORM(RfloatNON[\*,\*,iframe]);/(singlemean/stackmean); which can be uncommented for samples with high photobleaching

tifnamestackNON=parentname+'-PSratioNON'+zeroes+STRTRIM(iframe,2)+'.tif'

quotsingleNON=REVERSE(quotsingleNON,2); needed to allow Image J default orientation to agree with IDL

if multwrite eq 0 then WRITE\_TIFF,tifnamestack,FIX(multratioNON\*quotsingleNON),/short,orientation=1

if multwrite eq 1 then begin

tifnamestack=parentname+'-PSratioNON.tif'

if iframe eq ifirst then WRITE\_TIFF,tifnamestack,FIX(multratioNON\*quotsingleNON),/short,orientation=1

if iframe gt ifirst then WRITE\_TIFF,tifnamestack,FIX(multratioNON\*quotsingleNON),/short,orientation=1,/append

endif

endfor

print,'Now writing Ceru file #...'

for iframe=ifirst,nframes-1 do begin

print,iframe

singlemean=mean(Cerufl[\*,\*,iframe])

if iframe ge 0 and iframe le 9 then zeroes='000'

if iframe ge 10 and iframe le 99 then zeroes='00'

if iframe ge 100 and iframe le 999 then zeroes='0'

if iframe ge 1000 and iframe le 9999 then zeroes=''

Cerusingle=REFORM(Cerufl[\*,\*,iframe]);/(singlemean/stackmean); which can be uncommented for samples with high photobleaching

tifnamestack=parentname+'-Cerul'+zeroes+STRTRIM(iframe,2)+'.tif'

Cerusingle=REVERSE(Cerusingle,2); needed to allow Image J default orientation to agree with IDL

if multwrite eq 0 then WRITE\_TIFF,tifnamestack,FIX(Cerusingle),/short,orientation=1

if multwrite eq 1 then begin

tifnamestack=parentname+'-Cerul.tif'

if iframe eq ifirst then WRITE\_TIFF,tifnamestack,FIX(Cerusingle),/short,orientation=1

if iframe gt ifirst then WRITE\_TIFF,tifnamestack,FIX(Cerusingle),/short,orientation=1,/append

endif

endfor

read,'START AGAIN on new images with the SAME NORMALIZATION (1=YES, 0=NO) ?',again

if again eq 1 then goto,startover

print, 'ALL DONE ! Program now ended'

end

; (d) Option to normalize PS ratio to the PS ratio of Rhodamine 6G recorded on the same day.

Normalization modified: if rhod images are differently sized, use average over image. Otherwise, do pixel-by-pixel norm. [1-) Pure diI image dimensions MUST equal data image dimensions for pixel-by-pixel correction. 2-)'This is not the case here, so the correction is a single scalar factor over the whole image.

(e) make movies of P+2S pixel by pixel

Part II

Note that the input PSratNON.tif input file has probably had its values pre-multiplied by 10,000 before

; recording by PSratiodual2.pro. So restore the correct values here

PSratNON=PSratNON/10000.