ImageJ Brightfield Cell Segmentation Workflow
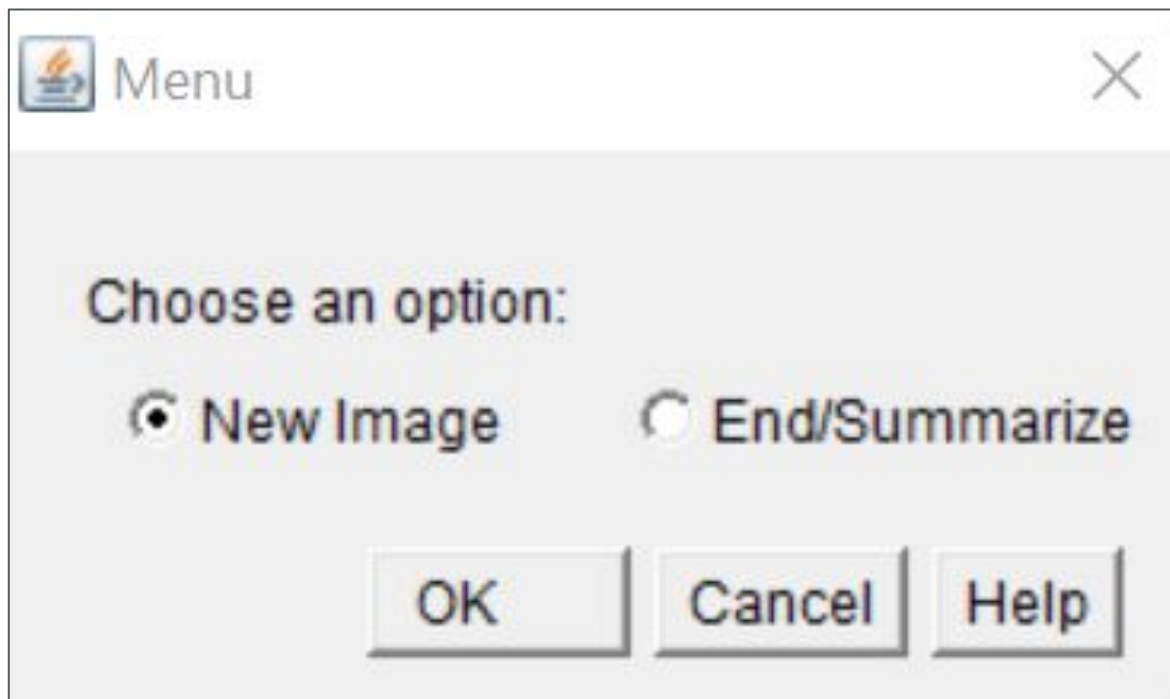
Benjamin Ngu (bngu@usc.edu)
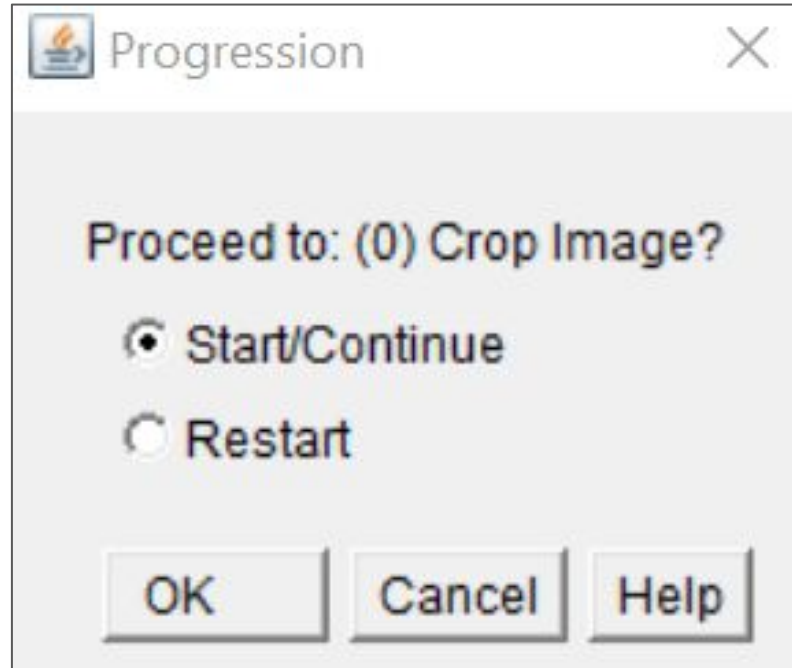
Start-up
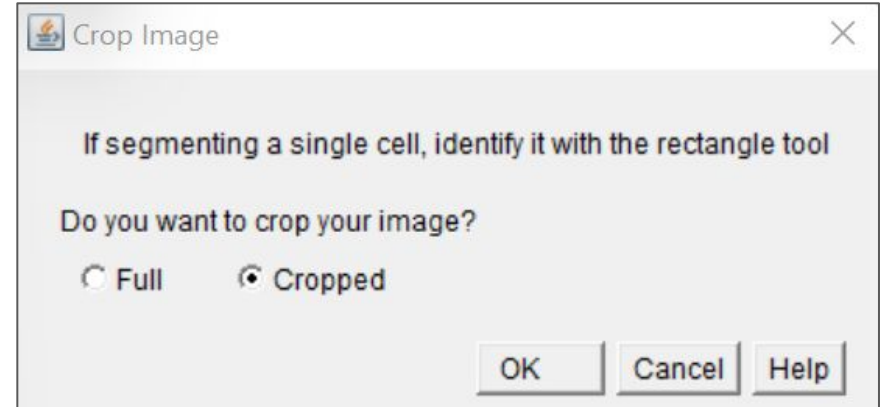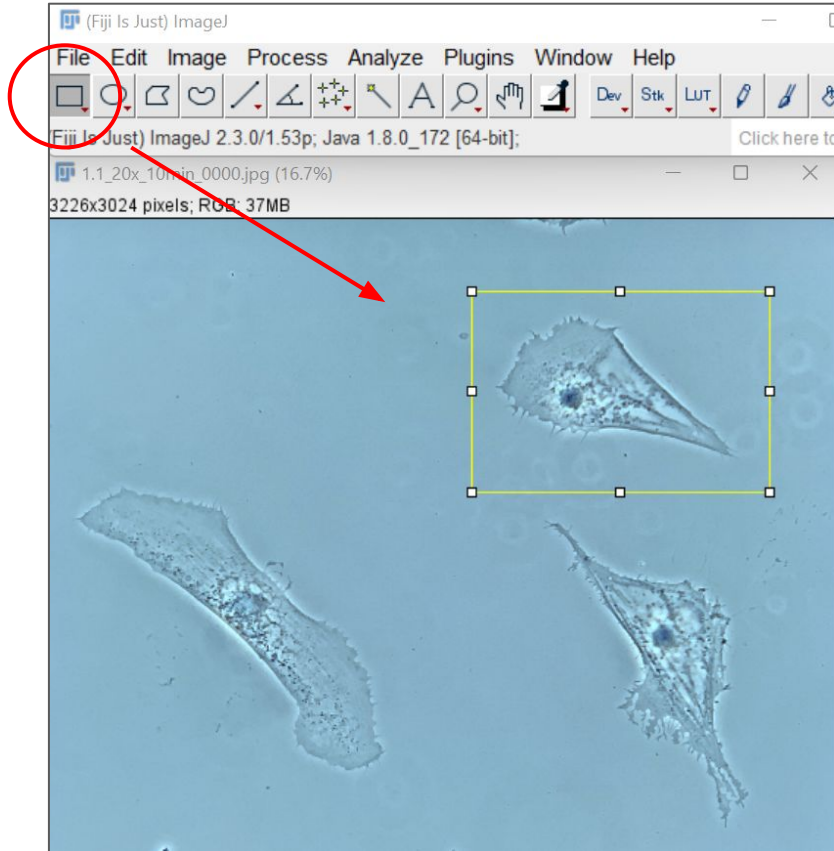
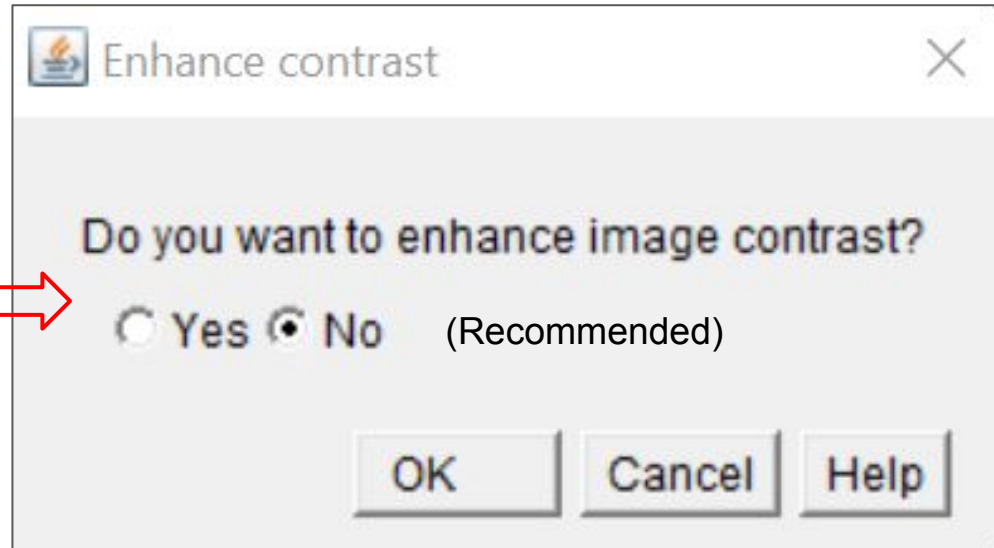# Make sure an image is open before proceeding
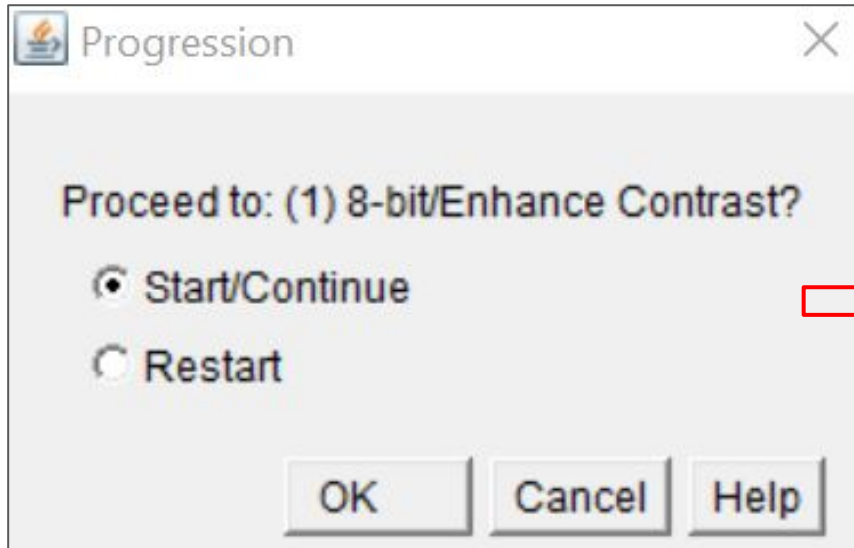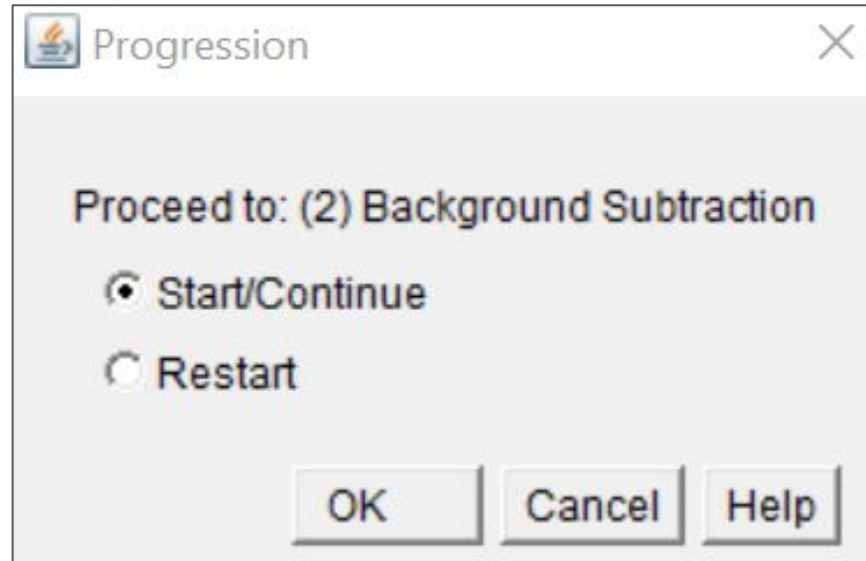
Checkpoint #0: Crop Image

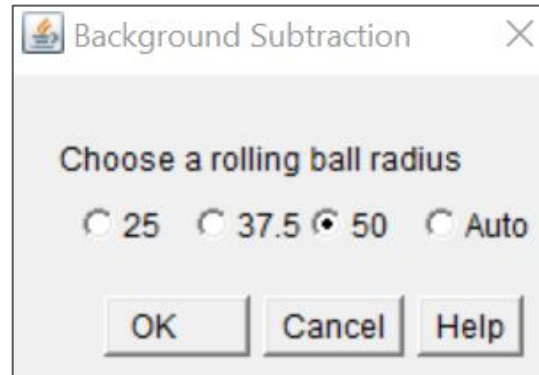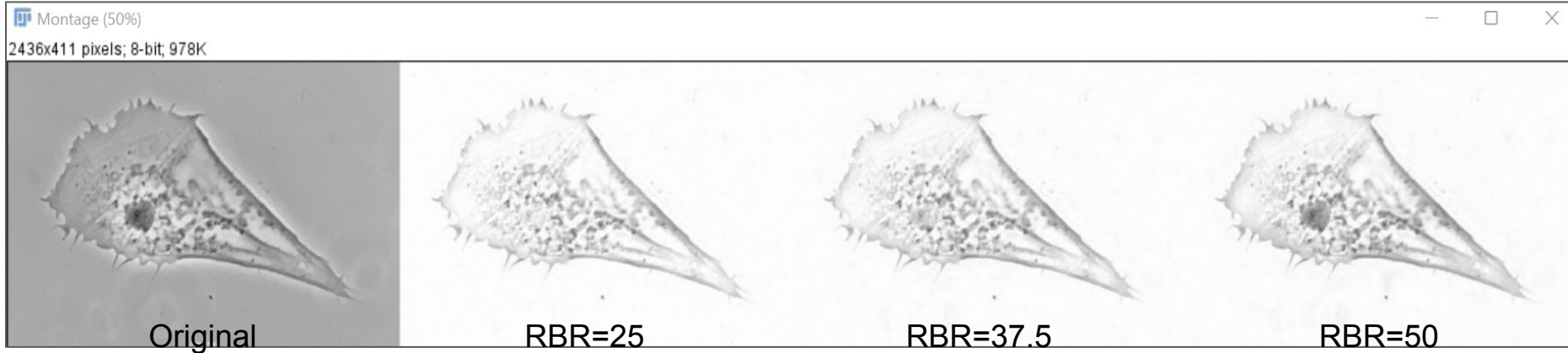# Checkpoint #0: Crop Image

Box cell before hitting "OK"
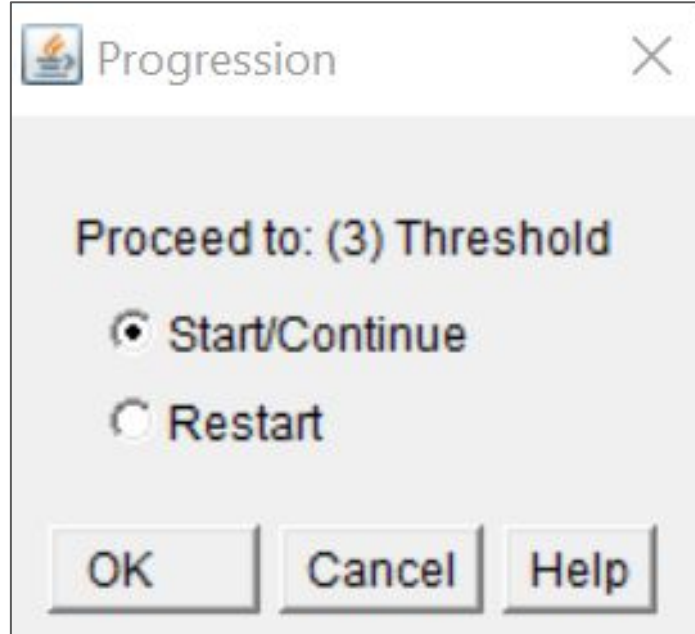
Checkpoint #1: Convert to 8-bit Image/Enhance Contrast

Checkpoint #2: Background Subtraction

Progression ✕

Proceed to: (2) Background Subtraction

⊙ Start/Continue

○ Restart

OK   Cancel   Help

Checkpoint #2: Background Subtraction

Checkpoint #3: Threshold
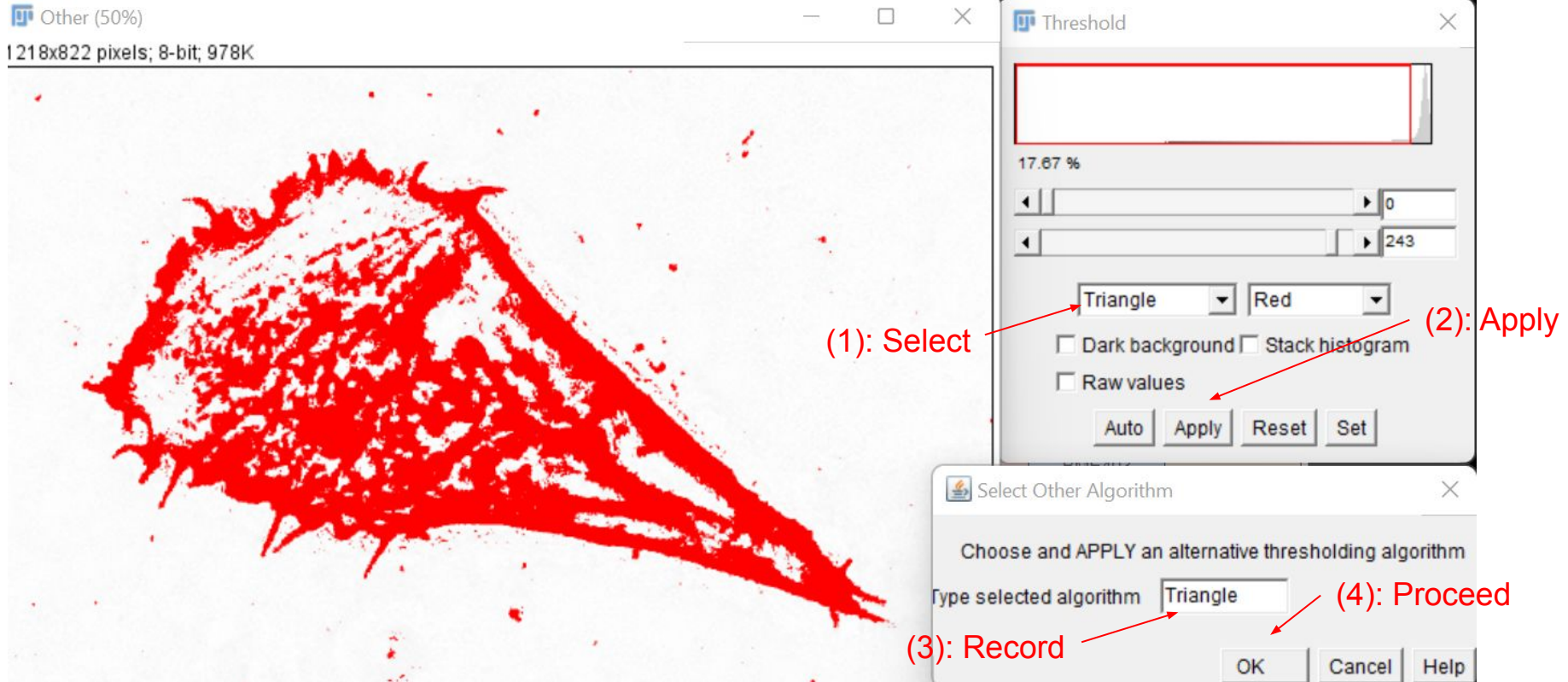
Checkpoint #3: Threshold

Checkpoint #3: Threshold ("Other")
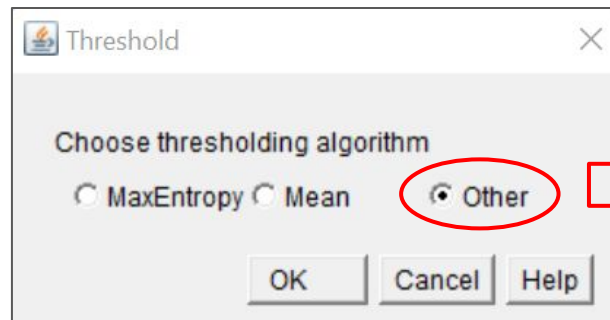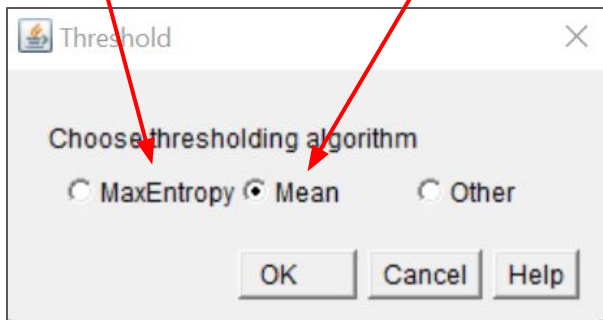
Checkpoint #4: Binary Processing

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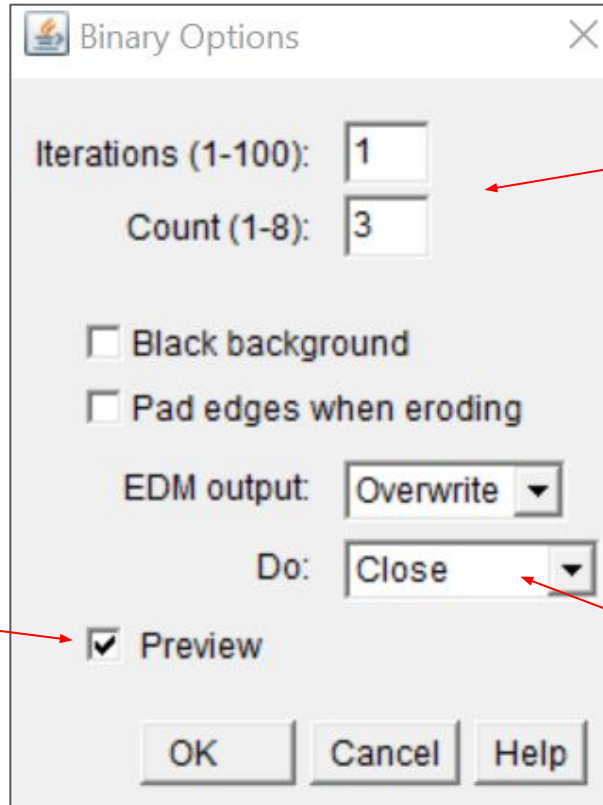

Program will "fill in" the initial binary image produced by the thresholding algorithm. "Check to start" if it is not satisfactory. Else, you will proceed to checkpoint #5

Checkpoint #4: Binary Processing

Binary Options

Iterations (1-100): [1]

Count (1-8): [3]

☐ Black background

☐ Pad edges when eroding

EDM output: [Overwrite ▼]

Do: [Close ▼]

☑ Preview

[OK] [Cancel] [Help]

Iterations   Specifies the number of times erosion, dilation, opening, and closing are performed. Iterations can be aborted by pressing [Esc].

Count   Specifies the number of adjacent background pixels necessary before a pixel is removed from the edge of an object during erosion and the number of adjacent foreground pixels necessary before a pixel is added to the edge of an object during dilation.
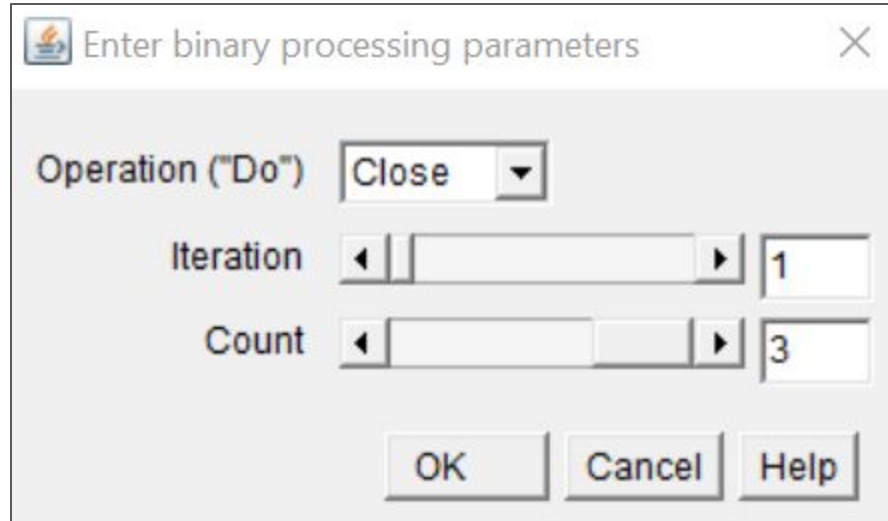
Recommendation:
- Iteration: 1-10
- Count: 3

Select to preview the effects of your binary operation

Select binary processing operation

Recommendation: "Close" and "Open"
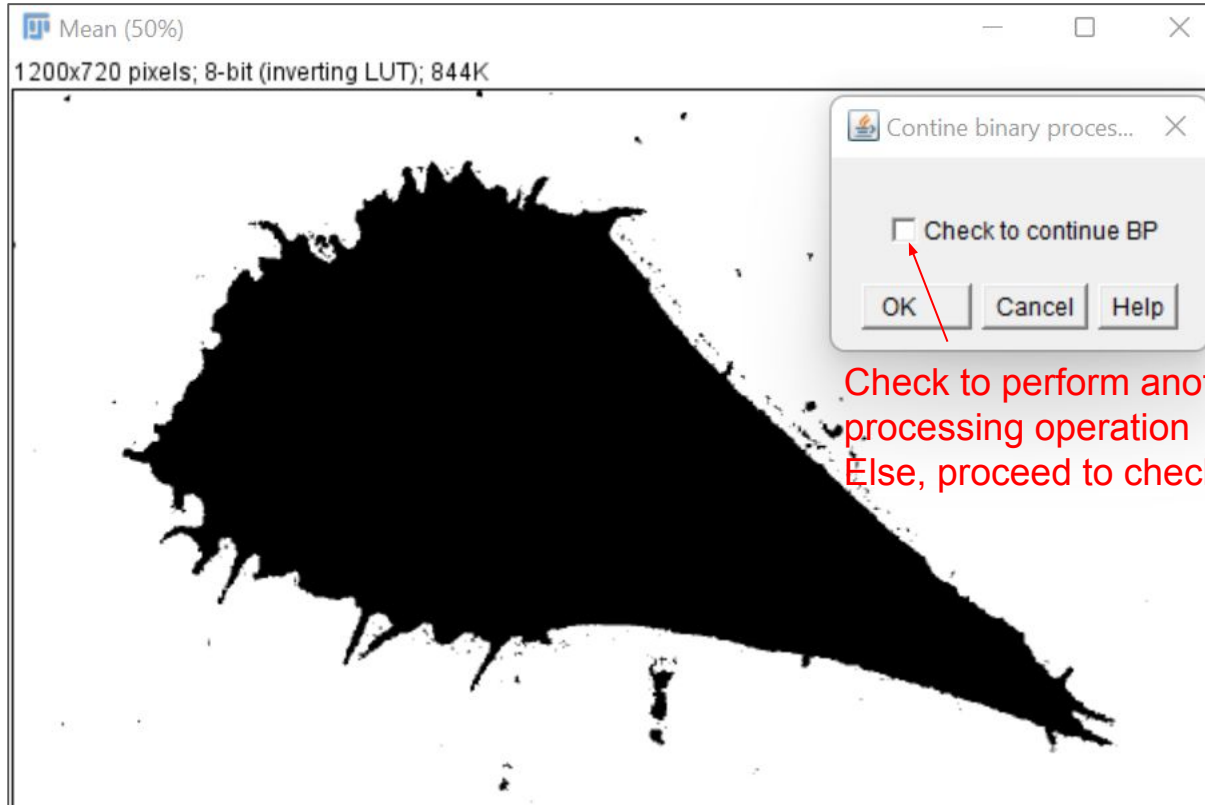
Checkpoint #4: Binary Processing

Record binary processing operation and parameters

Checkpoint #4: Binary Processing

Checkpoint #5: Analyze Particles

Checkpoint #5: Analyze Particles
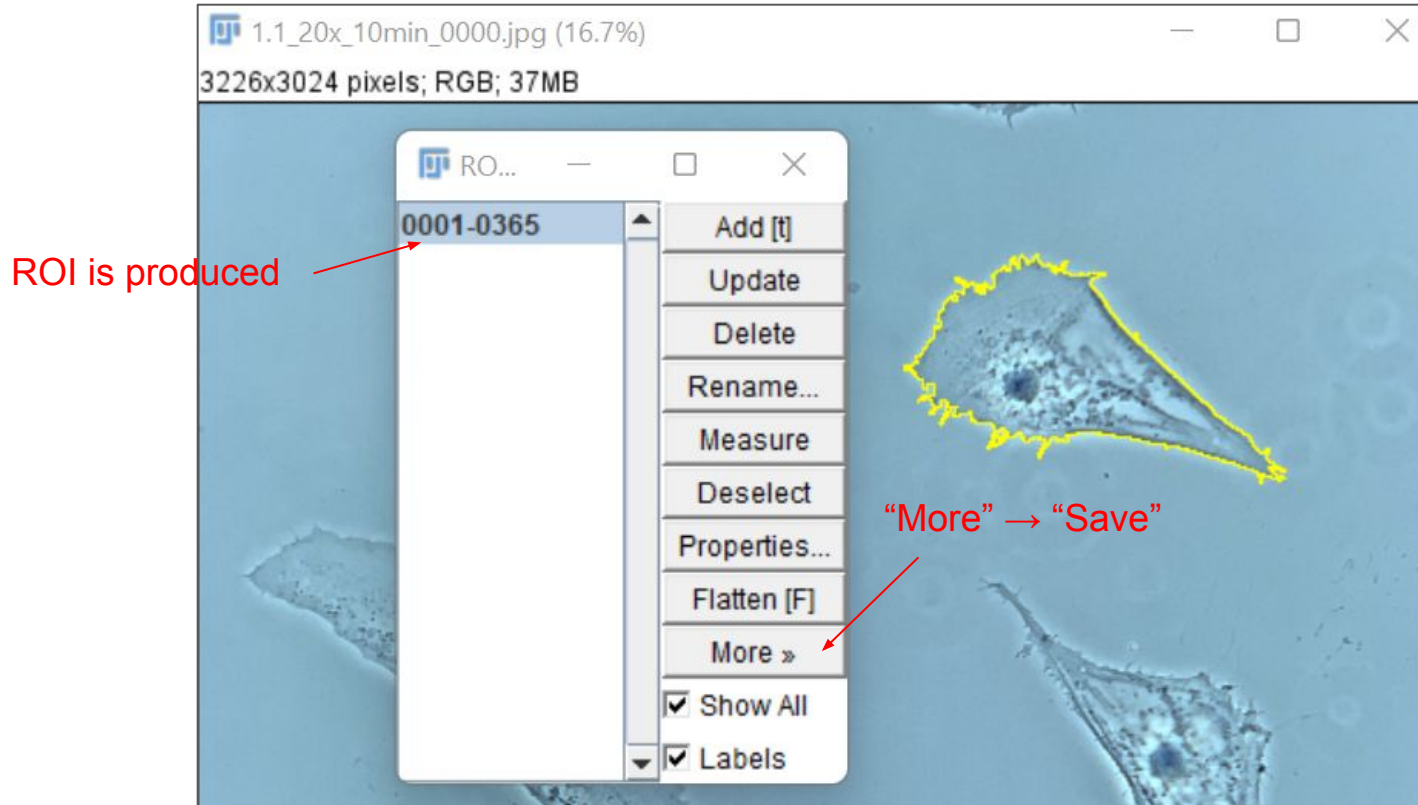
Process another image or STOP