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| **Feature Category** | **CellProfiler Module** | **Feature Examples** |
| Image Processing | ColorToGray | Split CH2 and CH3-CH2 channels for parallel image processing |
| Image Processing | ImageMath | CH2 and CH3-CH2 post-processing to enhance image feature contrast |
| Image Processing | ApplyThreshold | Remove pixels intensities below set threshold to eliminate noise and image granularity |
| Image Processing | EnhanceOrSuppressFeatures | Suppress speckles, ring shapes, neurites to improve object identification |
| Object Processing | IdentifyPrimaryObjects | Axonal segmentation using CH2 channel |
| Image Processing | FilterObjects | Generate axon filter to remove from subsequent CH2 channel image processing |
| Object Processing | MaskImages | Mask axons from CH2 images |
| Object Processing | IdentifyPrimaryObjects | Identify and count TAMs using masked CH2 images |
| Object Processing | IdentifyPrimaryObjects | Segment and count nuclei using CH3-CH2 images |
| Measurement | MeasureObjectSizeShape | Measure segmented nuclei area and shape features |
| File Processing | ExportToSpreadsheet | Export results for machine learning input |

**Supplementary Table 1. CellProfiler pipeline for quantitative SRH**