

### 1. Main axes:

- Click and drag to zoom/pan view
- Scatter shows all nuclei colored by spot count
- Use DAPI+FISH overlay to mask spots & create/delete cells

#### 2. Thumbnail Axes:

- Shows nuclei density across the entire scan
- Rectangle corresponds to the position displayed in the main axes
- Drag rectangle or double click to change positions

#### 3. Centroid List:

- Spot counts for all nuclei (ranked high to low)
- Double click on value to move to corresponding cell
- Use up/down arrow keys to move through list

#### 4. Sliders:

- Adjust contrast of the FISH image in main axes
- 5. Checkbox:
  - Toggle plot settings for main axes
- 6. Channel drop-down:
  - Change current FISH channels
- 7. Colormap drop-down:
  - Change colormap for scatter plot in main axes
- 8. Shuffle colors:
  - Change spot and centroid colors in main axes
- 9. Save and export:
  - Save spot, nuclei, and mask tables
  - Export spot summary table (spots per nucleus)

# 10. Zoom and pan view:

- While in zoom mode, click and drag on main axes to zoom-in
- While in pan view mode, click and drag to move through scan
- Right click to zoom-out by 2X
- Double click to return to home view

## 11. Add/delete masks and nuclei:

- Mask spots in current channel or mask nuclei for all channels
- Delete erroneous nuclei and add missing nuclei
- If nucleus is masked/added/deleted, spots in view will be reassigned

#### 12. Threshold axes:

- Drag vertical blue line to set spot intensity threshold
- Or use text box in upper right corner