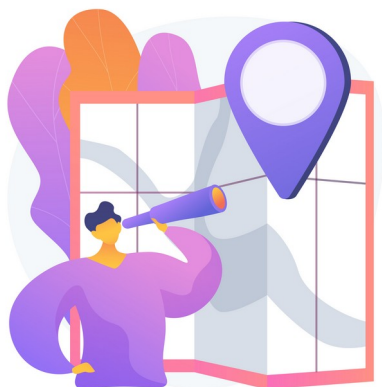


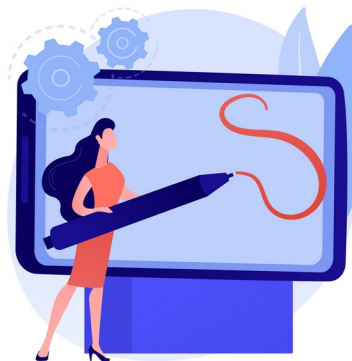


## Chapter 2

# Navigation, Annotations, Measurements



Designed by Freepik



Designed by Freepik

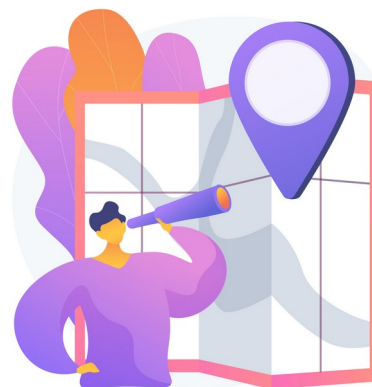


Designed by Freepik



## Chapter 2

# 2.1 Navigation

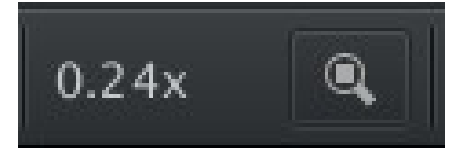


Designed by Freepik

## II. Navigation, Annotation, Measurements

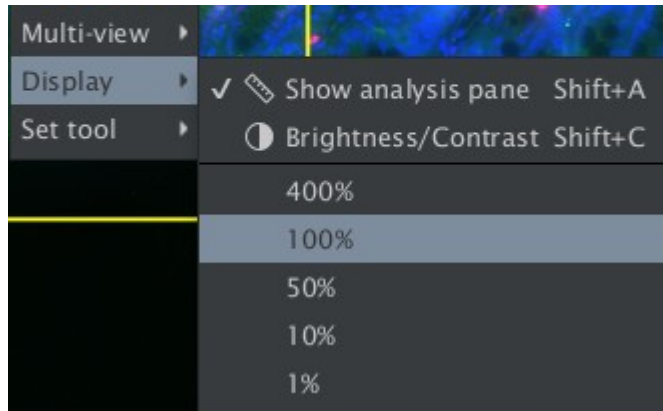
### The Image Viewer

Open image	Double click
Pan	Hold left button and move the mouse
Shift half a field of view	Arrow keys
Zoom in and out	Mouse wheel or shift+up / shift + down
Move to location	Click into the overview

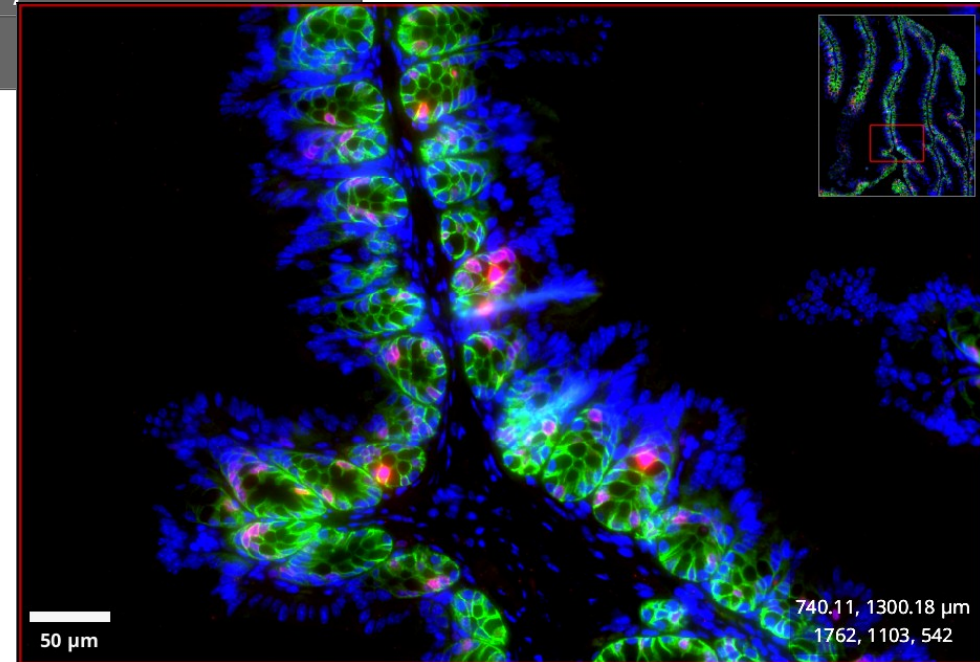


Set zoom manually  
Fit image to window

overview



Set zoom from the viewer's context menu



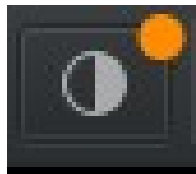
scale-bar

position / intensity

## II. Navigation, Annotations

# Adjust Brightness and Contrast

## B&C



1, 2, 3...

- Toggle channel

Shift+c

- Show B&C

Apply to similar images

### Brightness & contrast

Channel	Show <input checked="" type="checkbox"/>
<span style="color: blue;">■</span> Channel 1	<input checked="" type="checkbox"/>
<span style="color: green;">■</span> Channel 2	<input checked="" type="checkbox"/>
<span style="color: red;">■</span> Channel 3	<input checked="" type="checkbox"/>

Filter channels by name  \*

Settings  Save

Show grayscale  Invert background

Channel 1  Log histogram

Channel min  460...

Channel max  14916.5

Viewer gamma  1

Auto Reset

Apply to similar images

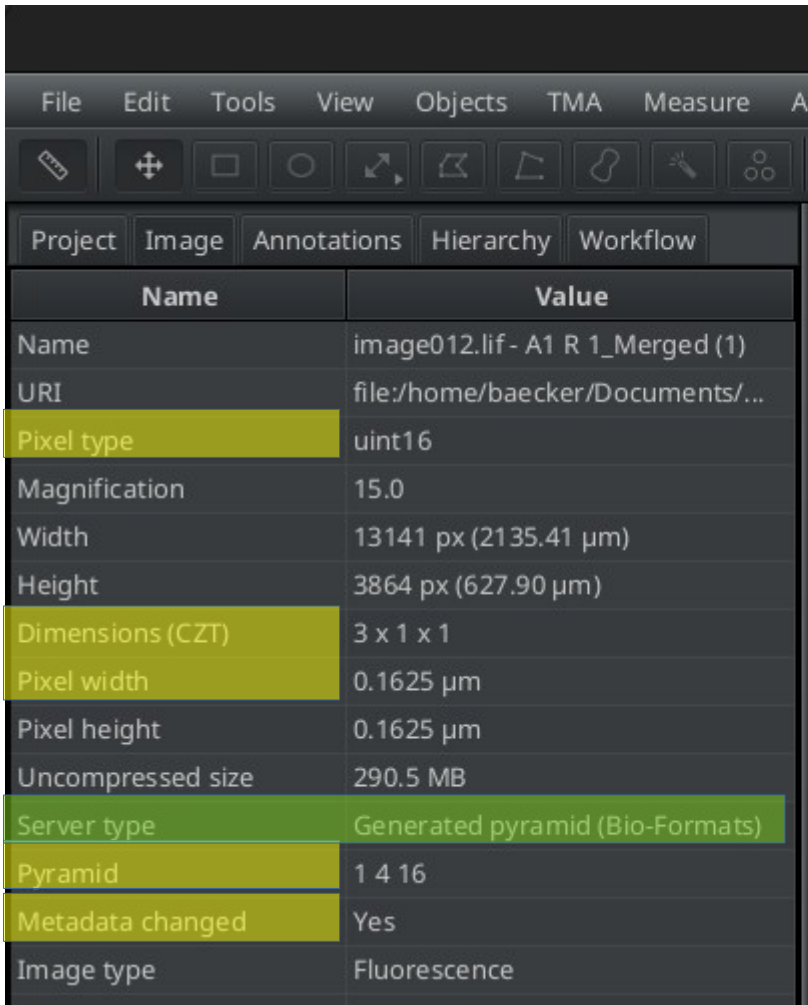
*Inverted background - interpret colors cautiously!*

### R 1\_Merged

654.59, 459.16  $\mu\text{m}$   
2778, 2412, 677

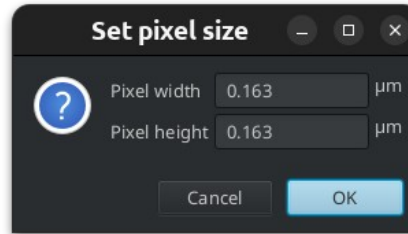
## II. Navigation, Annotation, Measurements

### The image-tab

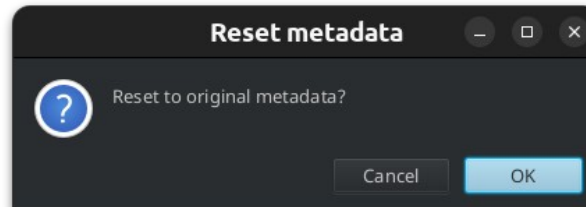
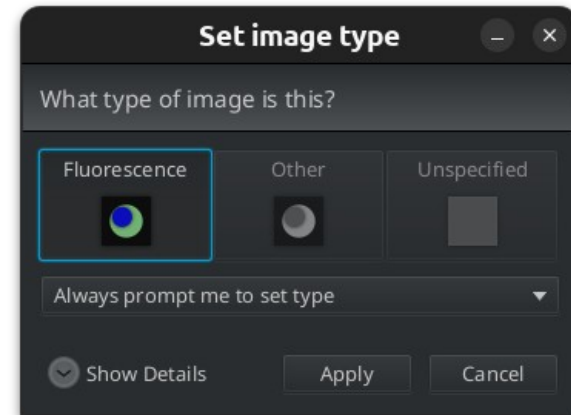


The screenshot shows the 'Image' tab selected in the software interface. Below the menu and toolbar, there is a table with the following data:

Name	Value
Name	image012.tif - A1 R 1_Merged (1)
URI	file:/home/baecker/Documents/...
Pixel type	uint16
Magnification	15.0
Width	13141 px (2135.41 $\mu\text{m}$ )
Height	3864 px (627.90 $\mu\text{m}$ )
Dimensions (CZT)	3 x 1 x 1
Pixel width	0.1625 $\mu\text{m}$
Pixel height	0.1625 $\mu\text{m}$
Uncompressed size	290.5 MB
Server type	Generated pyramid (Bio-Formats)
Pyramid	1 4 16
Metadata changed	Yes
Image type	Fluorescence



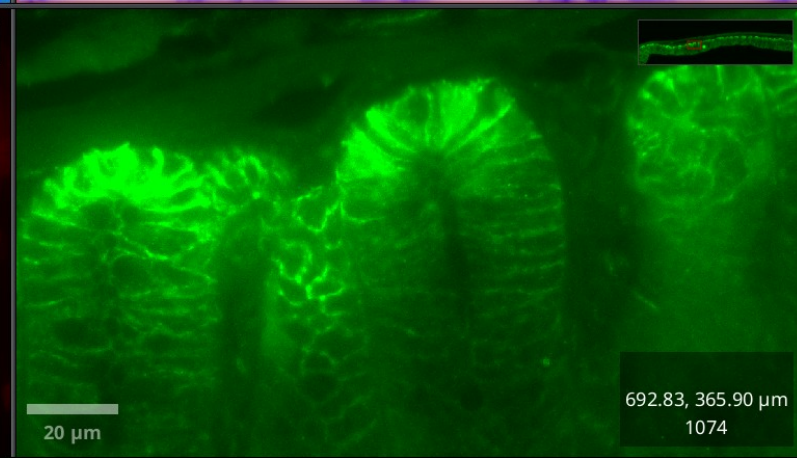
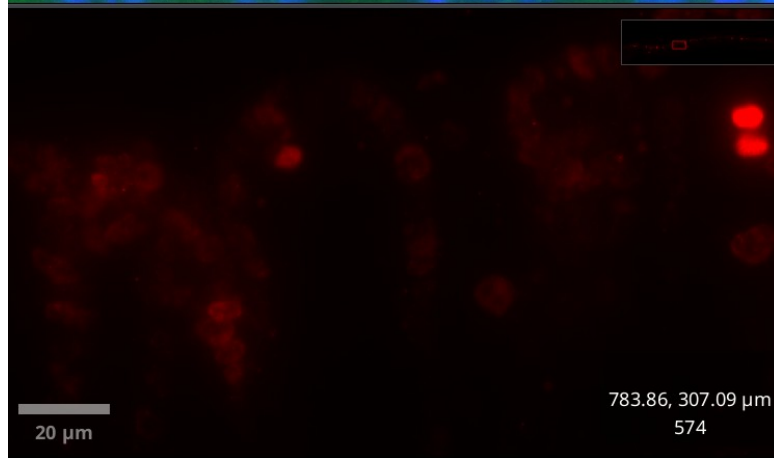
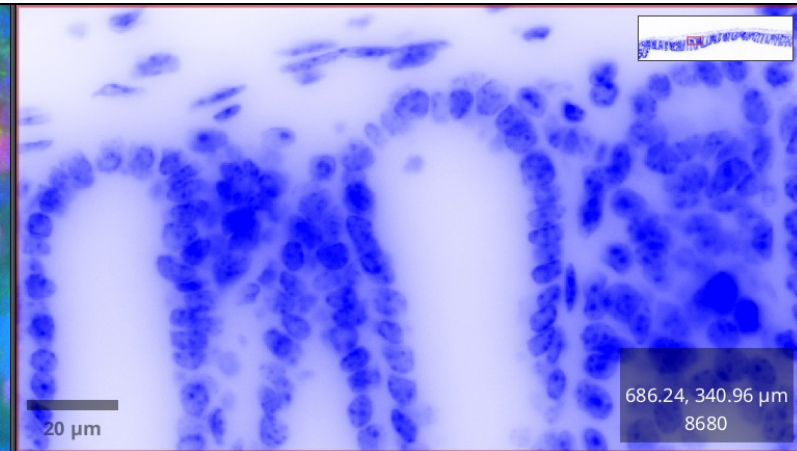
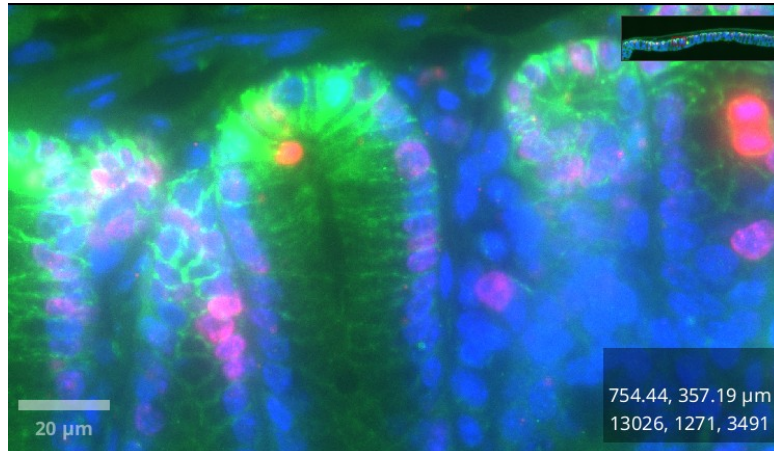
You can also use  
line or area annotations



## II. Navigation, Annotation, Measurements

### Multi-View

- Duplicate image
- Synchronize viewers
- Allows to
  - See channels separately



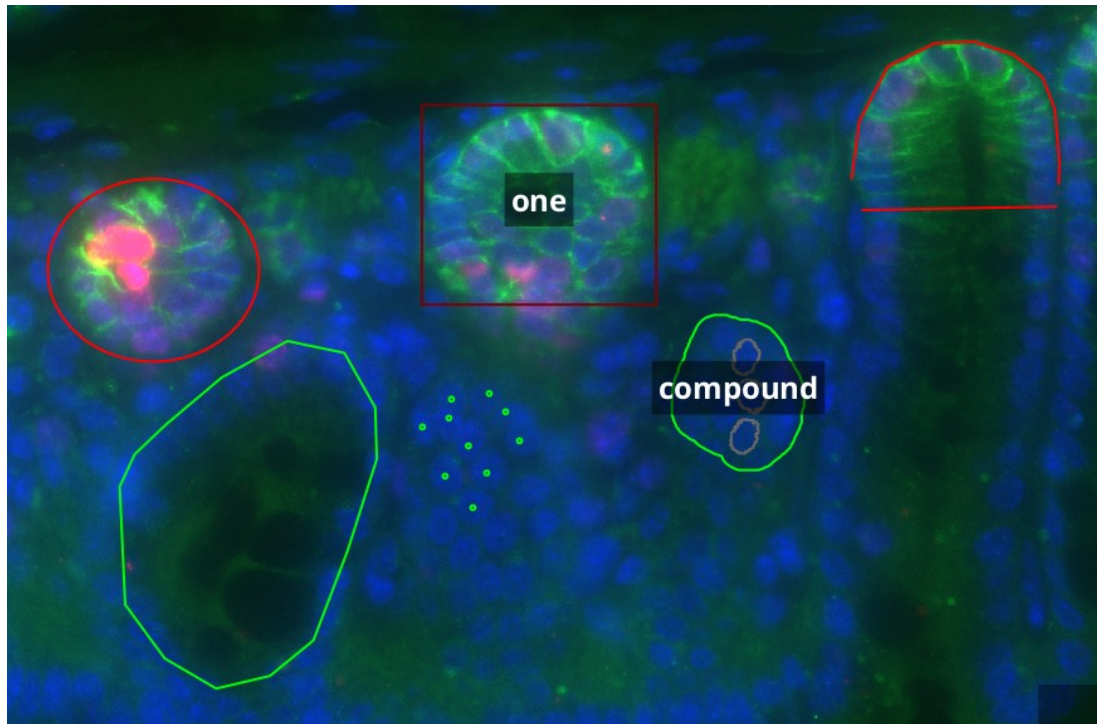


## Chapter 2

# 2.2 Annotations



### Annotations

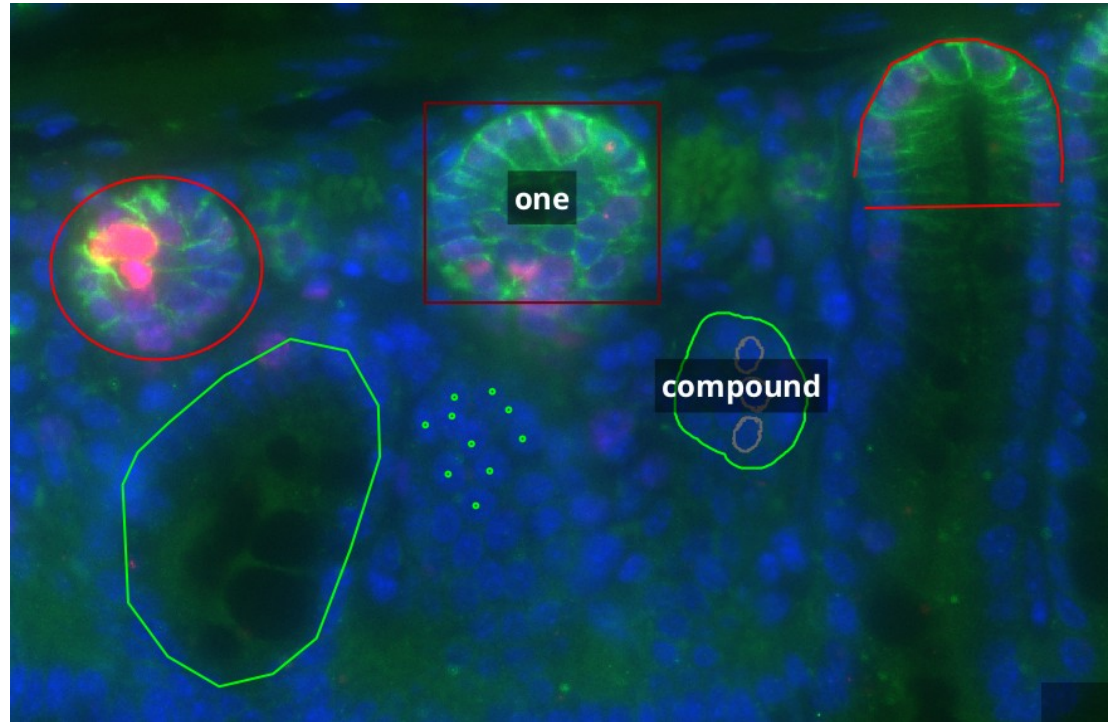


- Highlight a zone
  - Diagnostic for a zone
  - Name + description
- Classify zones
- Measure
  - Area, perimeter, diameter, length, number, intensities
- Select
  - Region of interest for further analysis
  - Work on small zone to find parameters
- Result of an analysis
  - Tissue segmentation

### Annotations

- Area
  - Rectangle / Square (shift)
  - Ellipse / Circle (shift)
  - Polygon
  - Brush
  - Wand
- Line
  - Straight
  - Polyline
- Points
  - Count

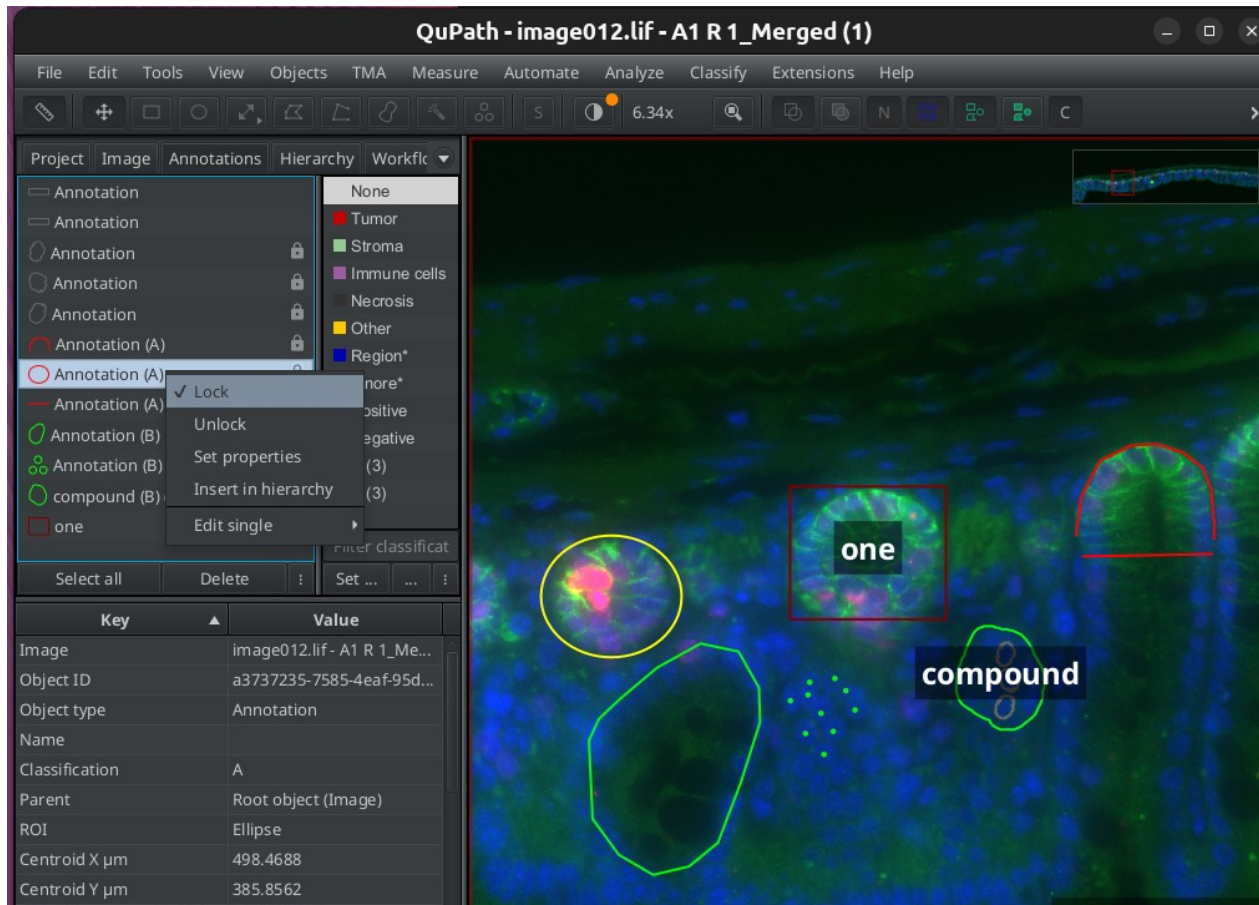
M



## II. Navigation, Annotation, Measurements

# Annotations Tab

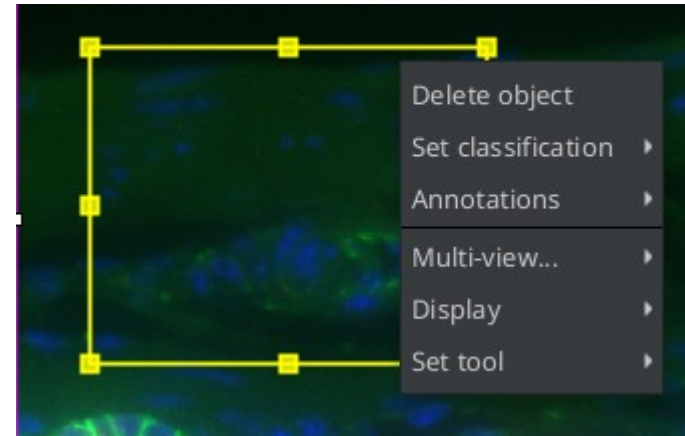
- Lock finished annotations
  - protected from modification
  - “selected” for actions
- Unlock
  - to modify again
- Select annotation
  - click In list
    - select
  - double click in list
    - select and show
  - or double click on it in the image
    - alt or ctrl + click



### Selecting Annotations

- Select all annotations
  - ctrl+alt+a
    - ctrl+shift+a makes an annotation of the whole slide
- Add annotation to selection
  - ctrl+left-click on unselected
- Remove annotation from selection
  - ctrl+left-click on selected

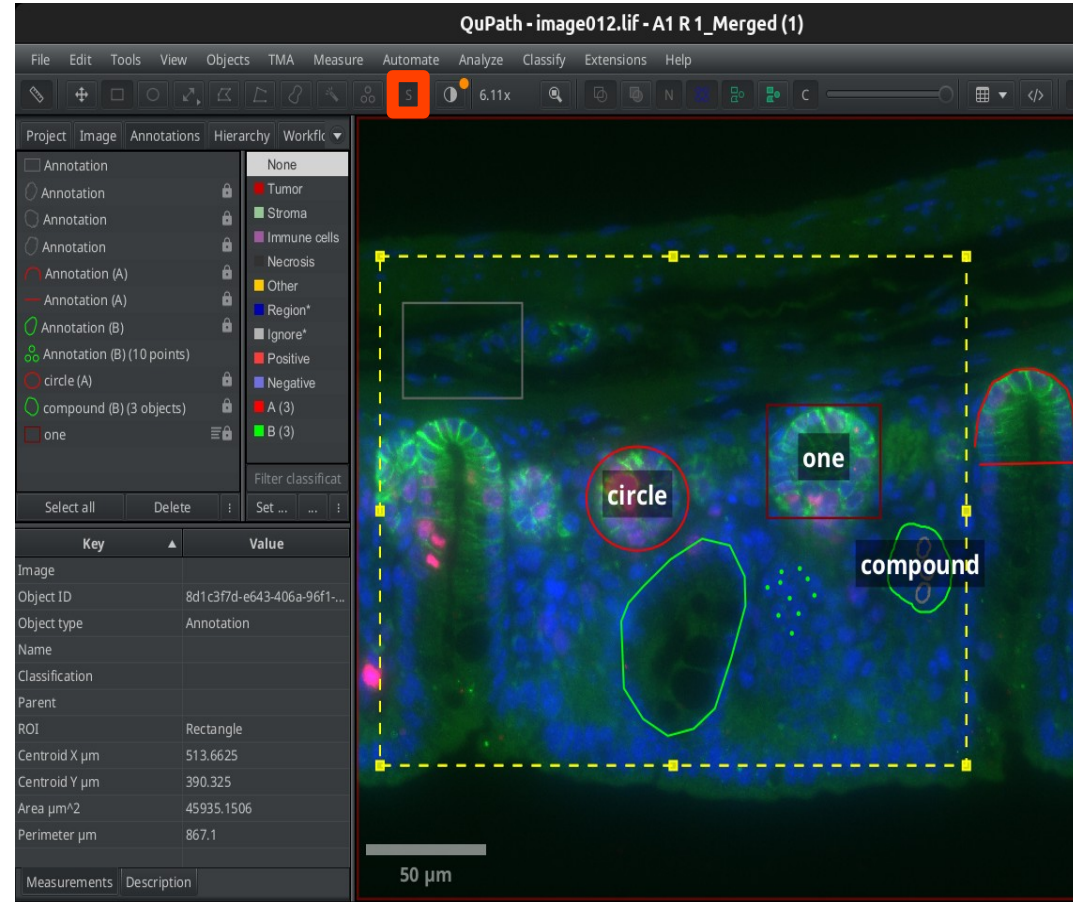
- Delete selected
  - del / backspace
  - menu / context menu



### Selection Mode

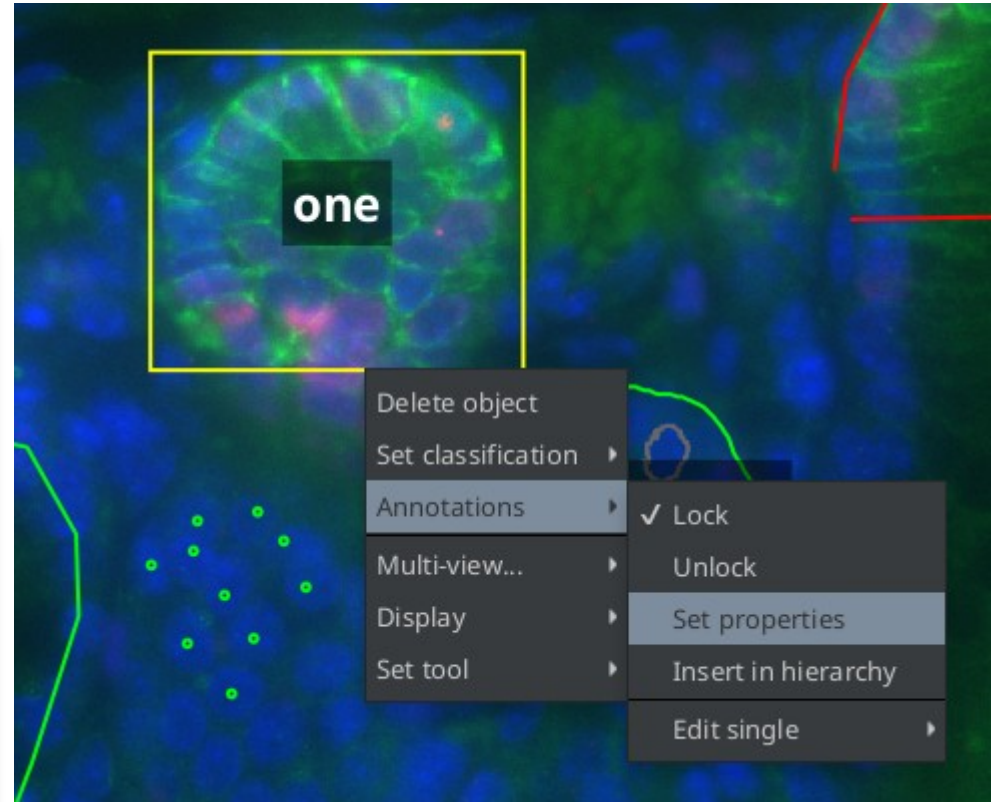
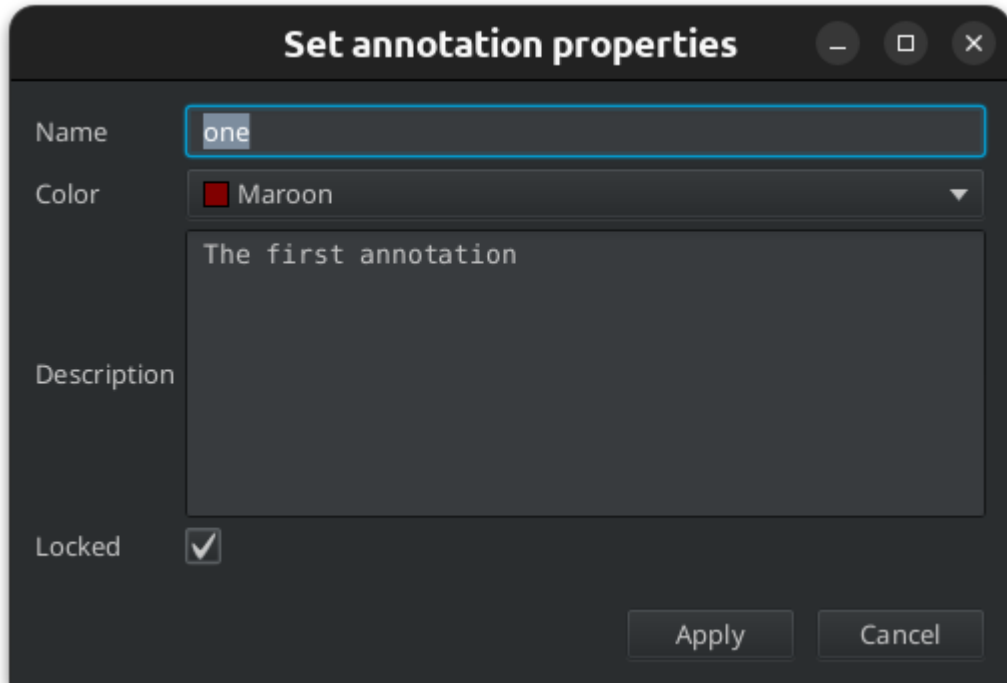
#### Selection Mode

- Toggle with S-button
- When activated
  - Tools select existing annotations
- When deactivated
  - Tools create annotations
- Use shift to add to a selection

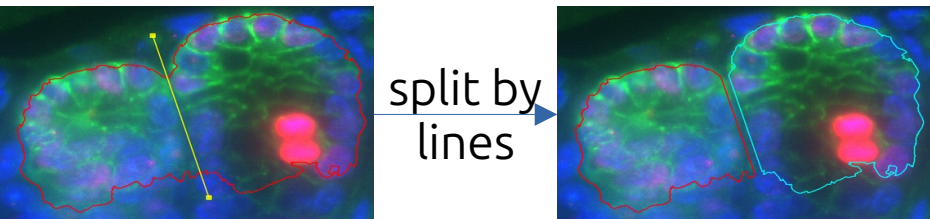
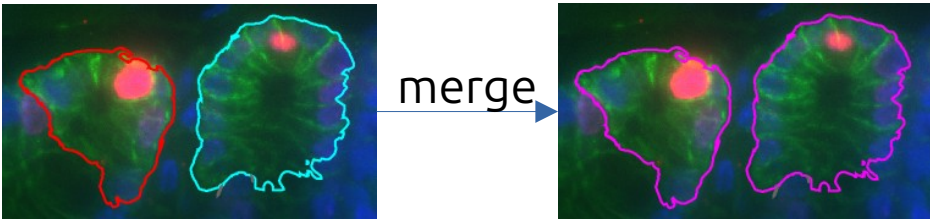
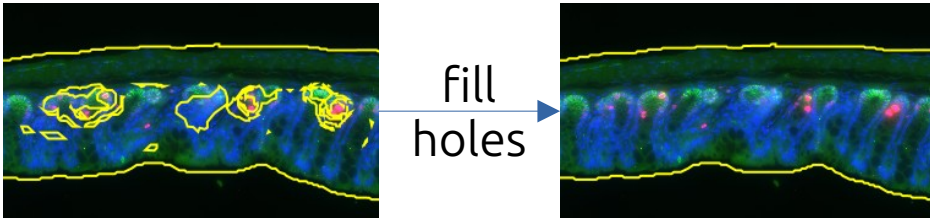
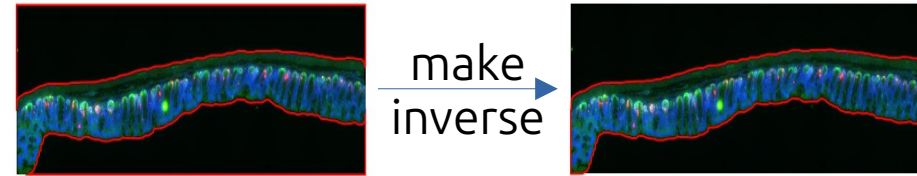


# Annotation Properties

Applied to all selected annotations



### Editing Annotations



- Subtract
- Intersect
- Specify (rectangle, ellipse)
- Transform (rotate)
- Duplicate
- Copy to current plane
- Transfer last annotation (shift+e)

- Annotations context menu
- Objects> Annotations... menu

- Expand annotations
- Remove fragments and holes
- Simplify

## II. Navigation, Annotation, Measurements

### Annotation Hierarchy

Project Image Annotations Hierarchy

- None
- Tumor
- Str
- Im
- Ne
- Oth
- Re
- Ign
- Pos
- Ne
- A
- B

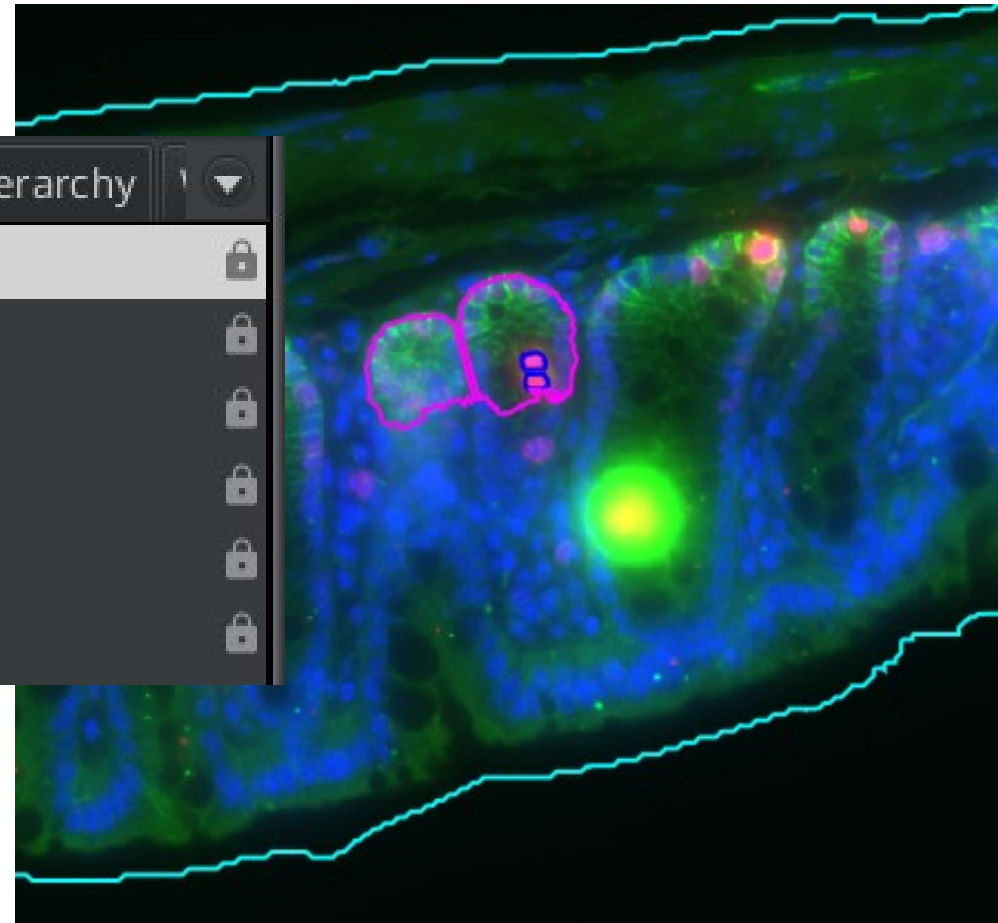
Project Image Annotations Hierarchy

- slide (1/5 objects) (locked)
- section 1 (2/4 objects) (locked)
  - roi 1 (2 objects) (locked)
    - sub-roi 1 (locked)
    - sub-roi 2 (locked)
  - roi 2 (locked)

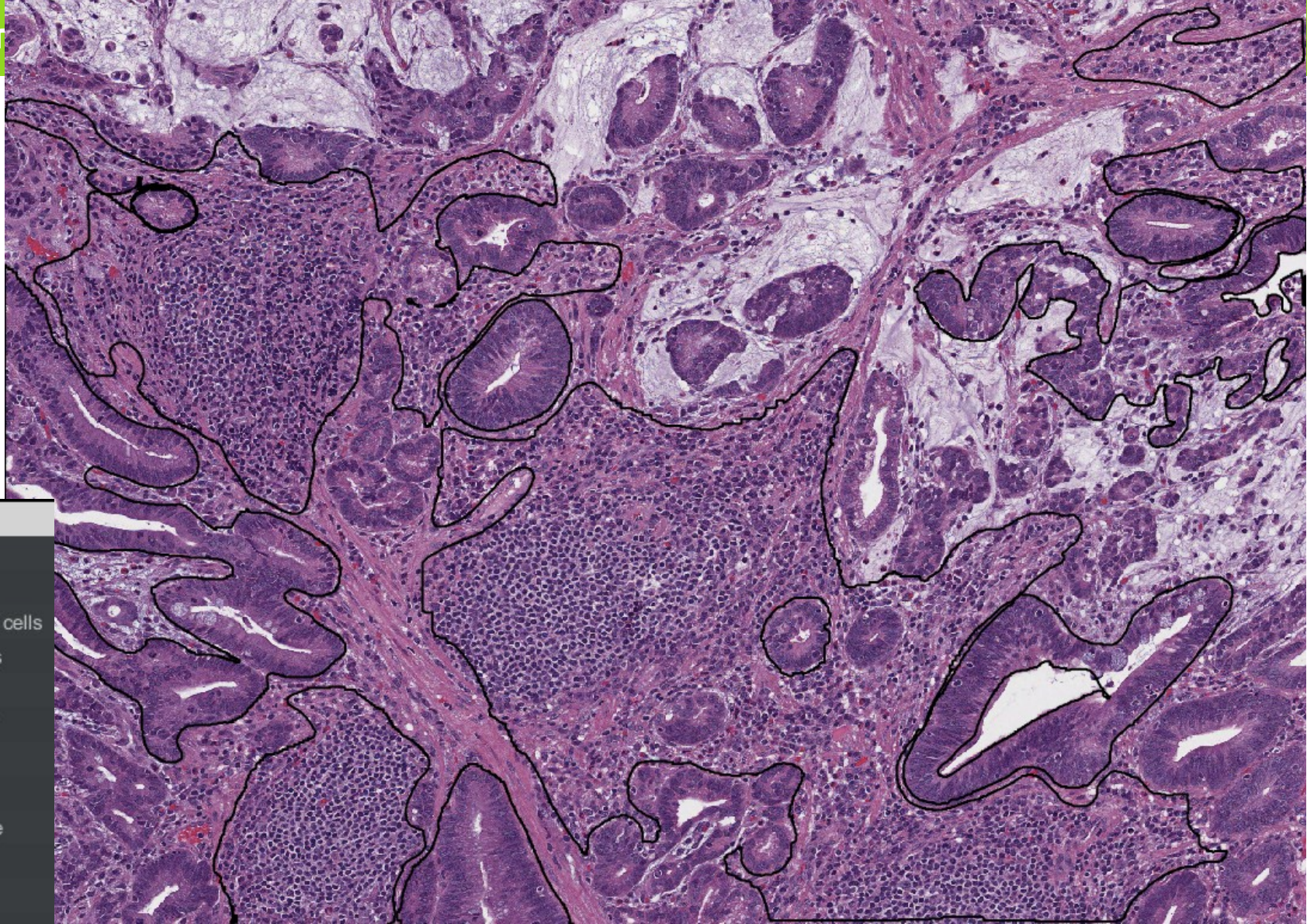
Select all Delete ⋮

Key

- Lock (checked)
- Unlock
- Set properties
- Insert in hierarchy
- Edit single
- Edit multiple



# Annotation Classification



	Annotation	None
	Annotation	Tumor
	Annotation	Stroma
	Annotation	Immune cells
	Annotation	Necrosis
	Annotation	Other
	Annotation	Region*
	Annotation	Ignore*
	Annotation	Positive
	Annotation	Negative
	Annotation	A
	Annotation	B

## II. Navigation, Anno

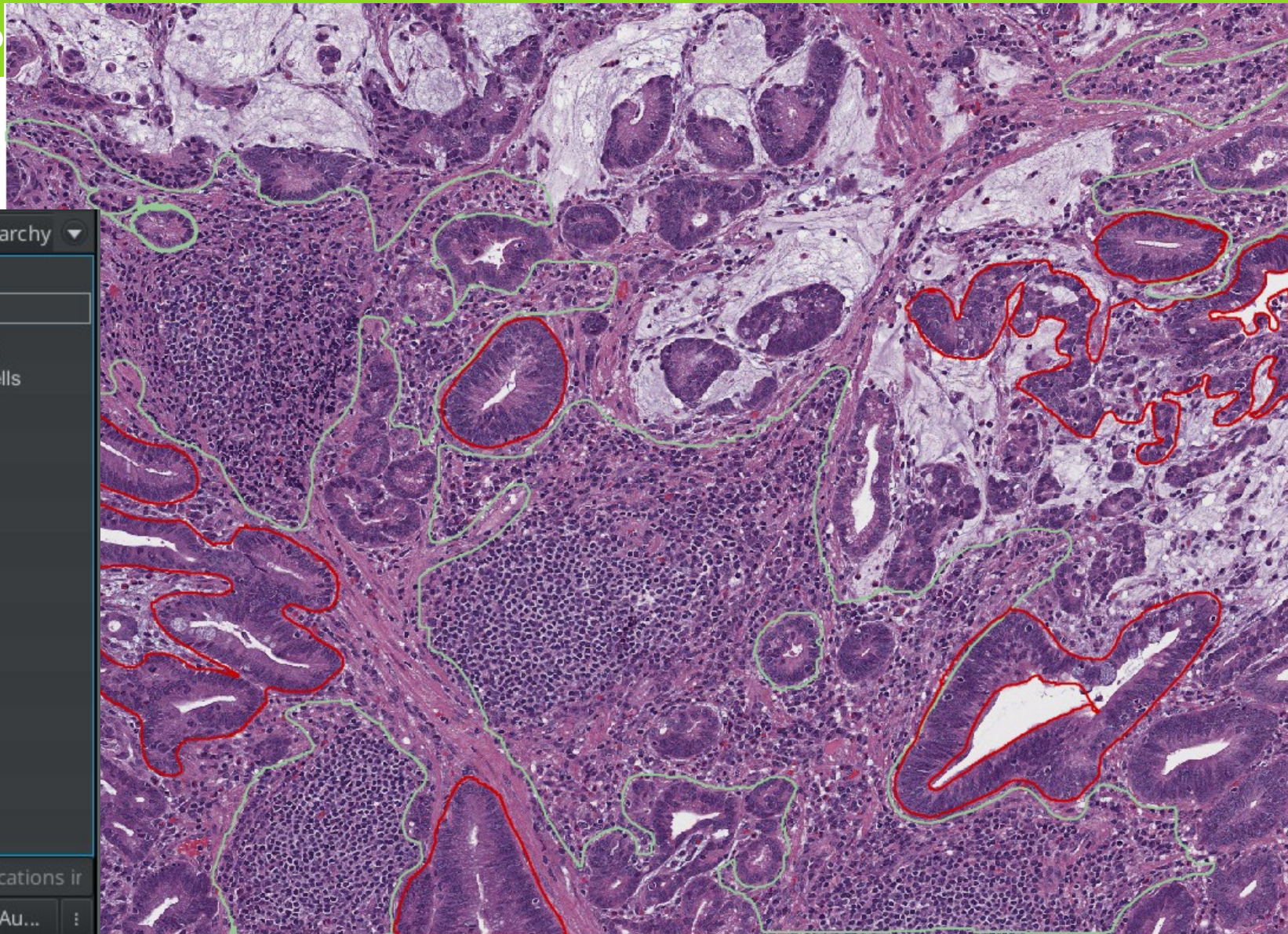
# Annotation Classification

Project Image Annotations Hierarchy ▾

- Annotation (Str... None
- Annotation (Str... Tumor (6)
- Annotation (Str... Stroma (5)
- Annotation (Str... Immune cells
- Annotation (Str... Necrosis
- Annotation (Tu... Other
- Annotation (Tu... Region\*
- Annotation (Tu... Ignore\*
- Annotation (Tu... Positive
- Annotation (Tu... Negative
- Annotation (Tu... A
- Annotation (Tu... B

Filter classifications in

Select... Del... : Set sel... Au... :



### Annotations vs. Detections

#### Annotation

- Zone
- Limited number
- Name, description
- Class
- Measurements
- Can be parent in hierarchy
- Can be child in hierarchy
- Can be modified



#### Detection

- Cell (nucleus + surrounding area)
- Large number
- **No name or description**
- Class
- Measurements
- **Can not be parent in hierarchy**
- Can be child in hierarchy
- **Can not be modified**





## Chapter 2

### 2.3 Measurements



Designed by Freepik

Inserm



### Measurements

Key	Value
Image	101.png
Object ID	63467c99-2b52-4e78-8a0f-85...
Object type	Annotation
Name	
Classification	Tumor
Parent	Root object (Image)
ROI	Polygon
Centroid X px	518.6721
Centroid Y px	623.2351
Num Detections	86
Num A	0
Num B	4
Area px <sup>2</sup>	11959.8043
Perimeter px	448.4494

Within selected annotation

- Number of detections
- Number of detections for all classes

By default

- Area
- Perimeter

### Shape Measurements

Analyze>Calculate features>Add Shape Features

#### Shape features

Add shape features to selected objects.

Note that not all measurements are compatible with all objects.

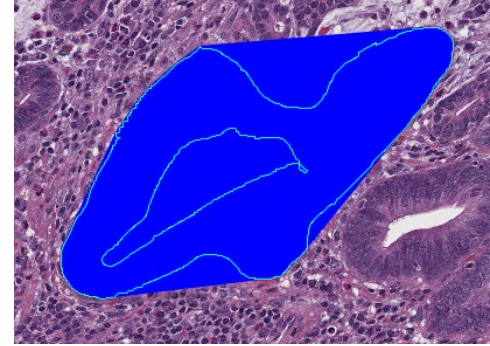
- Area
- Length
- Circularity
- Solidity
- Maximum diameter
- Minimum diameter
- Nucleus/Cell area ratio

Select all

Select none

Apply

Cancel



Solidity = Area / Convex Area

$$c = 4 \cdot \pi \cdot \frac{\text{area}}{\text{perimeter}^2}$$

circularity

## II. Navigation, Annotation, Measurements

### Compute intensity features

#### Resolution

Downsample

1

#### Regions

Region

ROI

Tile diameter

200

px (full resolution image)

#### Channels/Color transforms

- Optical density sum
- Hematoxylin (color deconvolved)
- Eosin (color deconvolved)
- Residual (color deconvolved)
- Red
- Green
- Blue
- Hue (mean only)
- Saturation
- Brightness

#### Basic features

- Mean
- Standard deviation
- Min & Max
- Median

#### Haralick features

- Compute Haralick features

Haralick distance

1

Haralick number of bins

32.0

## Intensity Measurements

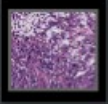

ROI: 1.00 px per pixel: Hematoxylin: Mean	0.4315
ROI: 1.00 px per pixel: Hematoxylin: Std.dev.	0.1847
ROI: 1.00 px per pixel: Hematoxylin: Min	-0.0441
ROI: 1.00 px per pixel: Hematoxylin: Max	1.4258
ROI: 1.00 px per pixel: Hematoxylin: Median	0.4275
ROI: 1.00 px per pixel: Eosin: Mean	0.2711
ROI: 1.00 px per pixel: Eosin: Std.dev.	0.0911
ROI: 1.00 px per pixel: Eosin: Min	-0.2869
ROI: 1.00 px per pixel: Eosin: Max	0.7897
ROI: 1.00 px per pixel: Eosin: Median	0.2755

Analyze>Calculate features>Add Intensity Features

## II. Navigation, Annotation, Measurements

# Show Annotation Measurements of on image

Annotations: 101.png

Thumbnail	Image	Object type	Name	Classification	Parent	ROI	Centroid X px	Centroid Y px	Area px <sup>2</sup>	Length px	Solidity	Max diameter px	Min diameter px
	101.png	Annotation		Tumor	Root object (Image)	Geometry	1158.4	578.33	40152.7	2714.1	0.614	416.58	221.44
	101.png	Annotation			Root object (Image)	Rectangle	665.5	596	1586552	5046	1	1786.7	1192
	101.png	Annotation		Stroma	Root object (Image)	Polygon	1232.2	445.69	10702.1	825.84	0.53	206.5	140.22
	101.png	Annotation		Stroma	Root object (Image)	Polygon	1220.7	305.46	13109	722.66	0.79	238.87	99.06

Column filter

Show histograms Copy to clipboard Save

Display histograms and scatter plots

## II. Navigation, Annotation, Measurements

### Export Measurements

#### Export measurements

Available

Selected

98.png

99.png

102.png

103.png

104.png

105.png

106.png

107.png

100.png

101.png

2 selected

With data file only

Output file

Choose

Export type

Separator

Columns to include (Op...

Populate

Reset

Cancel

Export

A	B	C	D	E	F	G	H	I
Image	Object	Object type	Name	Classification	Parent	RQI	Centroid X px	Centroid Y px
101.png	dd69	Annotation		Stroma	Root object (Image)	Geometry	281.49	
101.png	be29	Annotation		Stroma	Root object (Image)	Geometry	776.09	
101.png	4d64	Annotation		Stroma	Root object (Image)	Geometry	327.22	
101.png	0391	Annotation		Stroma	Root object (Image)	Polygon	1220.7	
101.png	20b5	Annotation		Stroma	Root object (Image)	Polygon	1232.2	
101.png	5841	Annotation		Tumor	Root object (Image)	Geometry	146.19	
101.png	75d5	Annotation		Tumor	Root object (Image)	Geometry	1078.6	
101.png	6348	Annotation		Tumor	Root object (Image)	Polygon	486.25	
101.png	6340	Annotation		Tumor	Root object (Image)	Polygon	518.8	
101.png	35d1	Annotation		Tumor	Root object (Image)	Geometry	1158.4	
101.png	c49c	Annotation		Tumor	Root object (Image)	Geometry	1190.9	
101.png	d1e0	Annotation			Root object (Image)	Rectangle	665.5	
101.png	bb5c	Annotation			Root object (Image)	Rectangle	570.5	

#### Export Type

- Image
- Annotations
- Detections
- Cells
- TMA cores