

# Kappa 1.0

## User Manual



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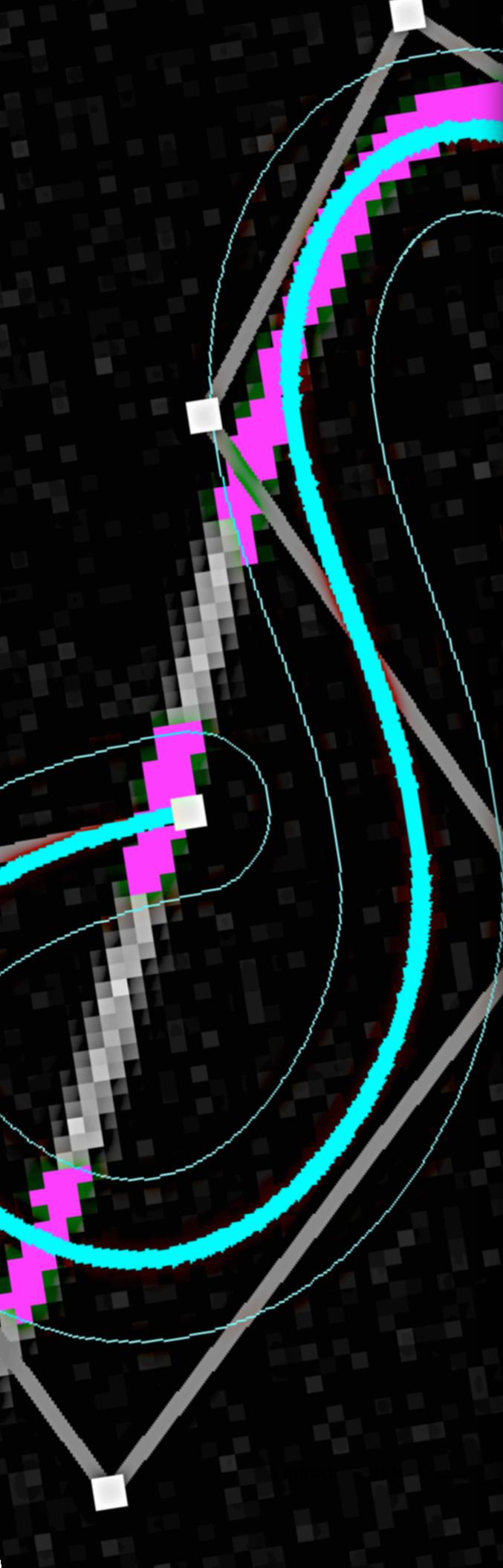
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# 1

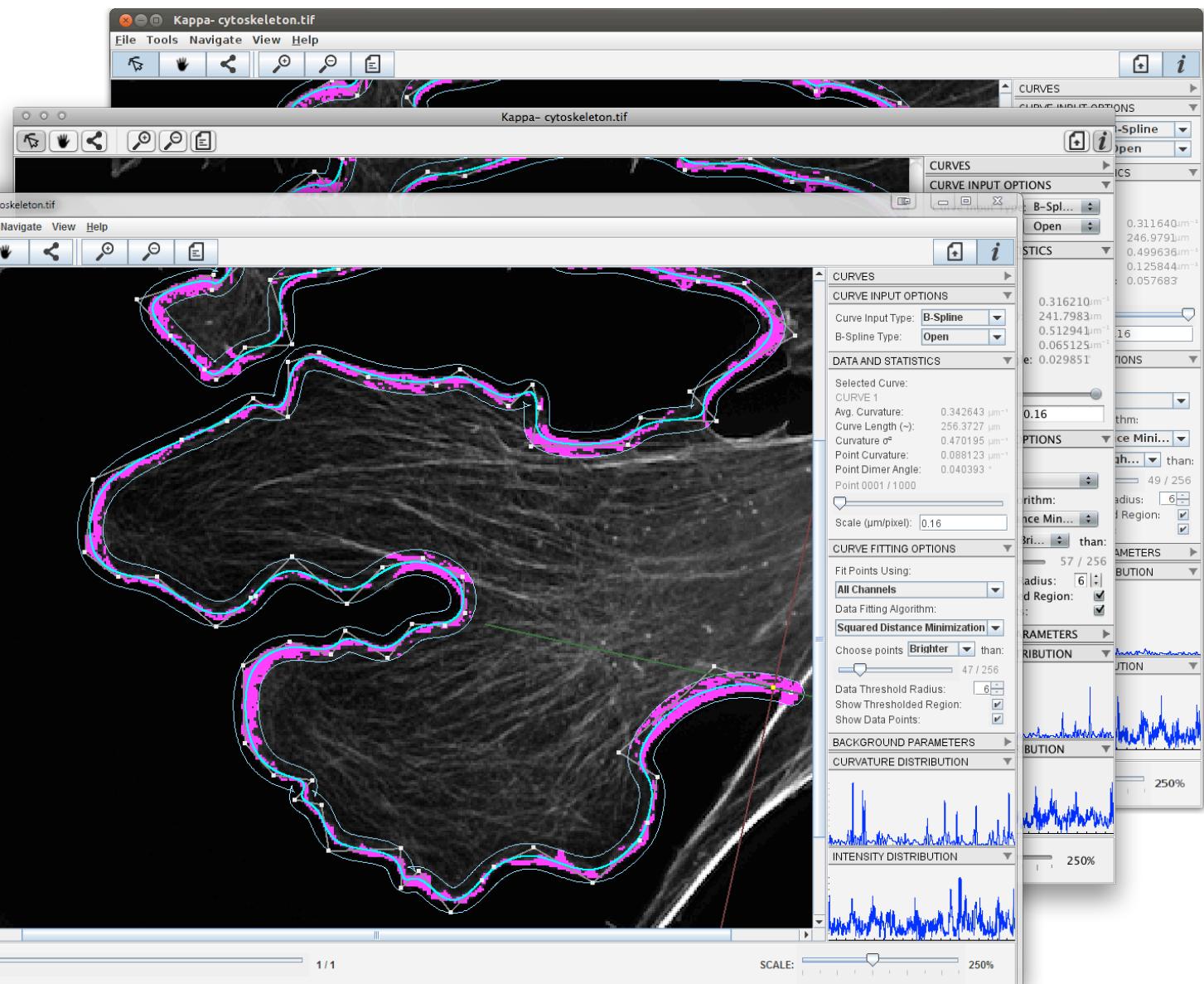
## Section

An Introduction to  
Kappa

# Introduction

Kappa is a Curvature Analysis Program developed in Java. It allows a user to measure curvature in images in a convenient way. You can trace an initial shape with a B-Spline curve in **just a few clicks** and then fit that curve to image data with a minimization algorithm. It's fast and robust.

Since it's developed in Java, it's also **cross-platform**. Kappa has been tested on Windows, Mac OS X, and Linux. It will work on any operating system with a recent version of Java installed.

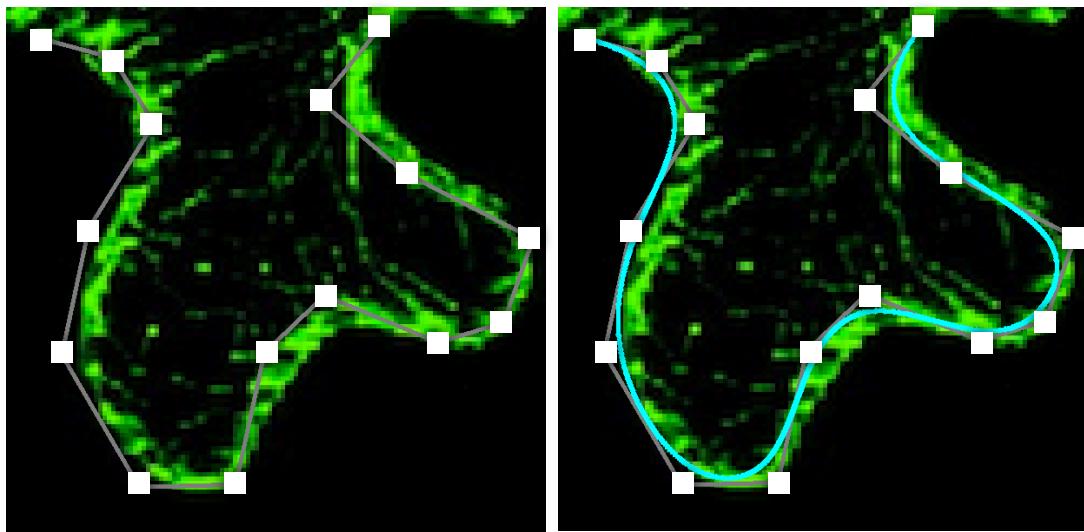


# An Overview: 3 Easy Steps

Kappa is very easy to use! You can track the curvature of an object on an image in just a few steps:

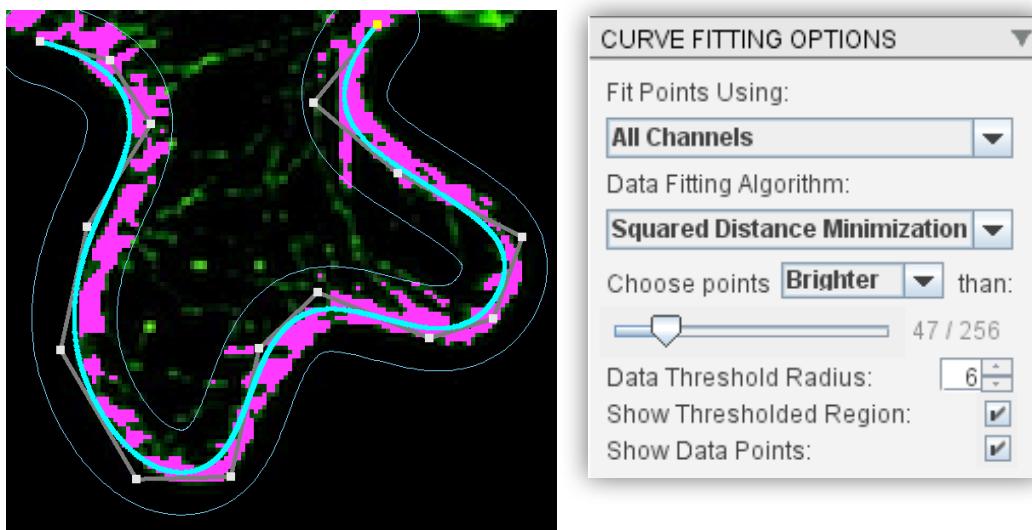
## Step 1: Define an Initial Curve

The user can click to define control points that loosely track the shape of an object. Then, by pressing **enter**, the corresponding B-Spline curve will appear.

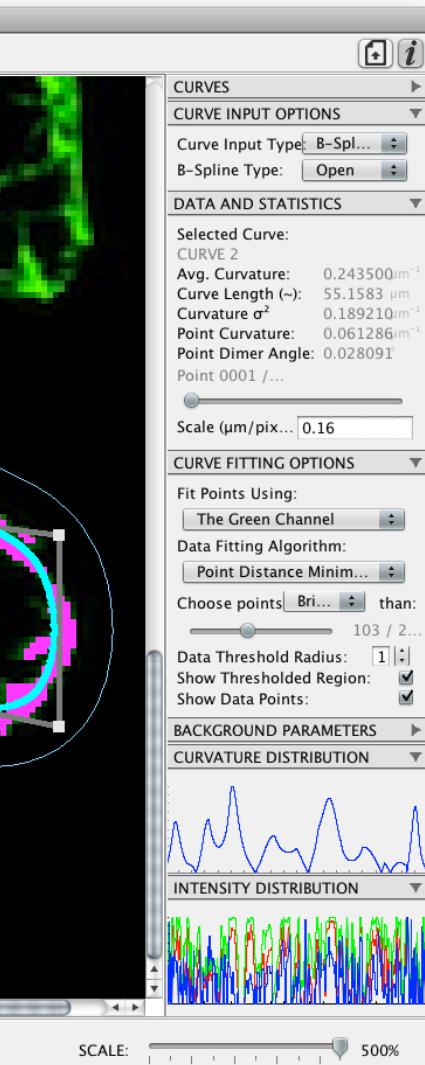


## Step 2: Choose what data you want to fit to

The user can then adjust the data points that the curve will be fit to. Parameters such as the colour channel, the brightness threshold, and the distance from the initial curve can be modified. Any data within these thresholds will be fit.

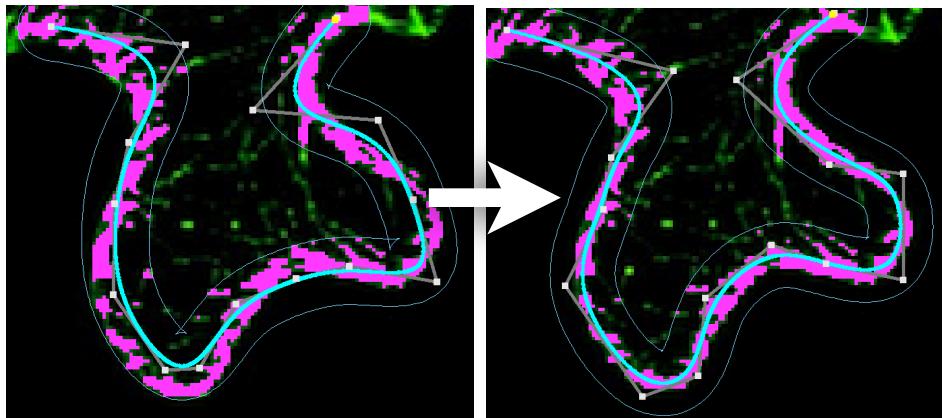


## Step 3: Fit the Curve to the Data



Kappa will fit the curve to the chosen data points using a least-squares based minimization algorithm. This reduces the problem of human bias in the initial curve, and improves the accuracy of the results.

You can then look at the resulting measurements in the sidebar, or export your data into an Excel Compatible Format.



**Top Left:** A B-Spline curve, before fitting is performed.

**Top Right:** The same B-Spline curve, after the fitting algorithm has been run.

**Left:** The Info Panel, showing at-a-glance information from the curve.

## Demo Video

As the old adage goes, a picture is sometimes worth a thousand words. In that case, a 2 minute, 30 FPS video is worth about 3,600,000 words! No seriously, check out the following video to get a great overview of our software:



**An Introduction To Kappa**  
Tracking Curvature in Biological Images with B-Splines  
Duration: 2:42  
<http://www.youtube.com/watch?v=6nyHhhvLGms>

# Our Paper

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Manuscript Under Review. To be updated upon acceptance.

## Check out our Source Code

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To download the source code or file a bug, please visit:

<https://github.com/kevanlu/Kappa>

## Contact Us

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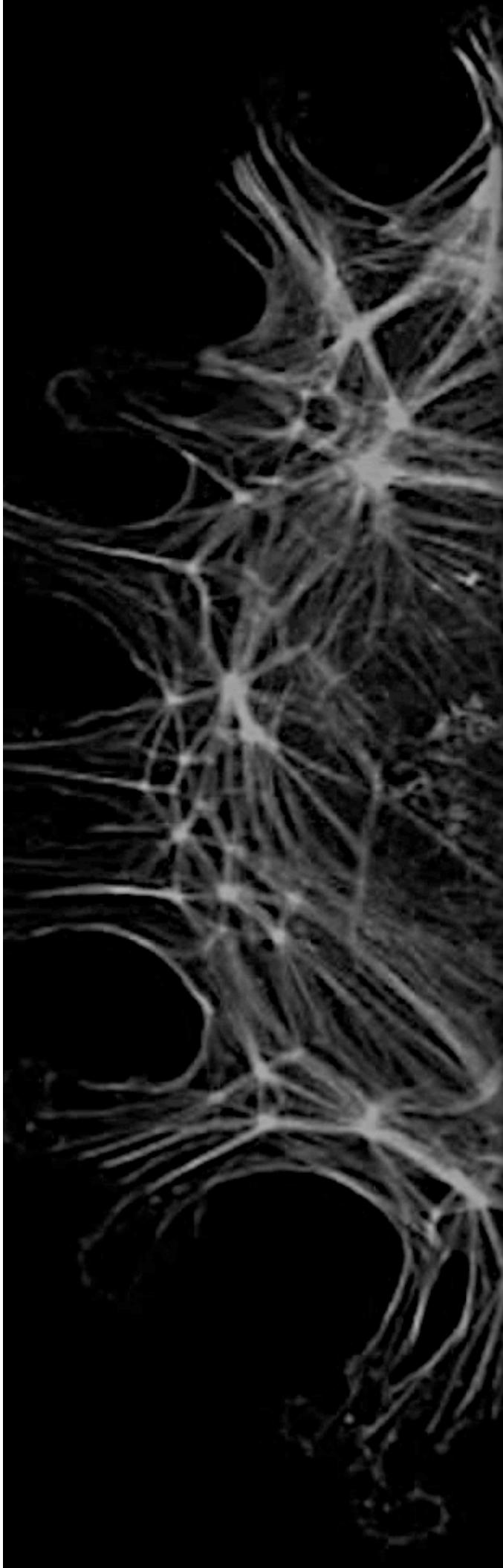
To get in touch with the developer of this software, please email:

**Developer:** Kevan Lu

[lord.cookies@gmail.com](mailto:lord.cookies@gmail.com) (Don't judge!)

**Supervised By:** Gary Brouhard

[gary.brouhard@mcgill.ca](mailto:gary.brouhard@mcgill.ca)



# Section 2

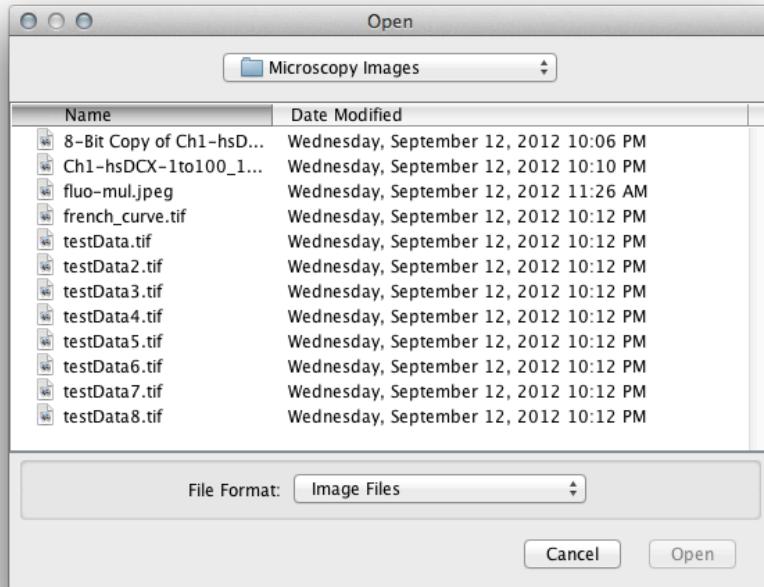
Working with Kappa

# Workflow

## Opening Files

Kappa uses ImageJ libraries for its file support. Consequently, Kappa supports the following file types: **TIFF, GIF, JPEG, PNG, DICOM, BMP, PGM and FITS**. Uncompressed **AVIs** are also supported.

Opening files is straightforward: Go to **File → Open**, and choose a file.

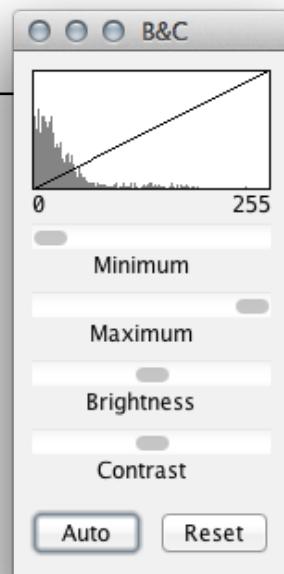


## Manipulating the Image

To zoom in or zoom out of the image, the user can simply click the zoom-in (⊕) or zoom-out buttons (⊖) (See **Section 3** for details on the User Interface).

The user can also use the panning tool (✋) to drag the image around.

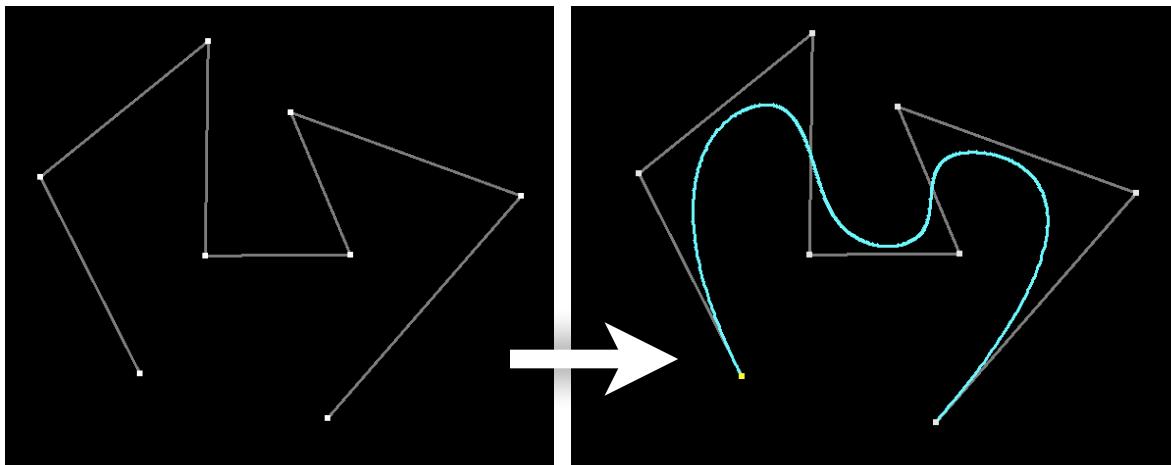
To adjust the brightness or contrast of the image, go to **Image → Adjust Brightness/Contrast**. Kappa uses the Brightness/Contrast panel from ImageJ.



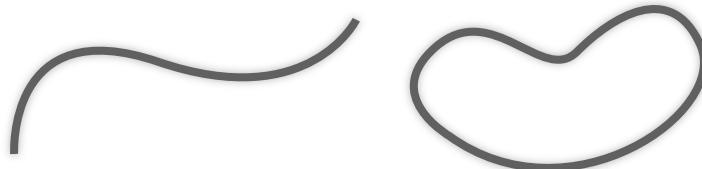
# Entering and Adjusting Curves

## Entering a Curve

Switch to the **Spline Input Tool** () , and click to enter control points. When you are done entering control points, press enter and a curve will be formed.

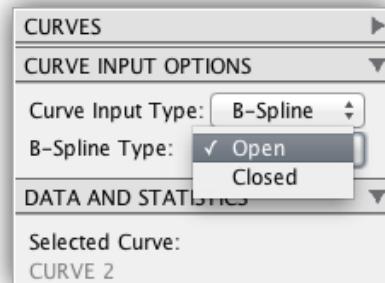


You do not have to enter an *open* B-Spline (with free ends). You can also enter a *closed* B-Spline, which will close on itself. To do this, just change the setting in the curve input panel.



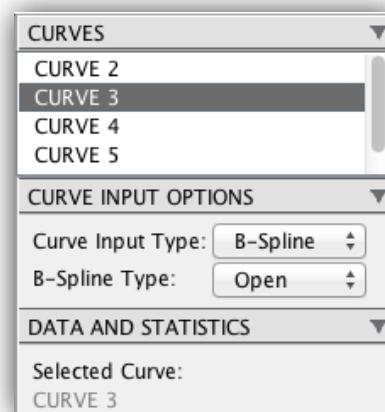
An Open B-Spline

A Closed B-Spline



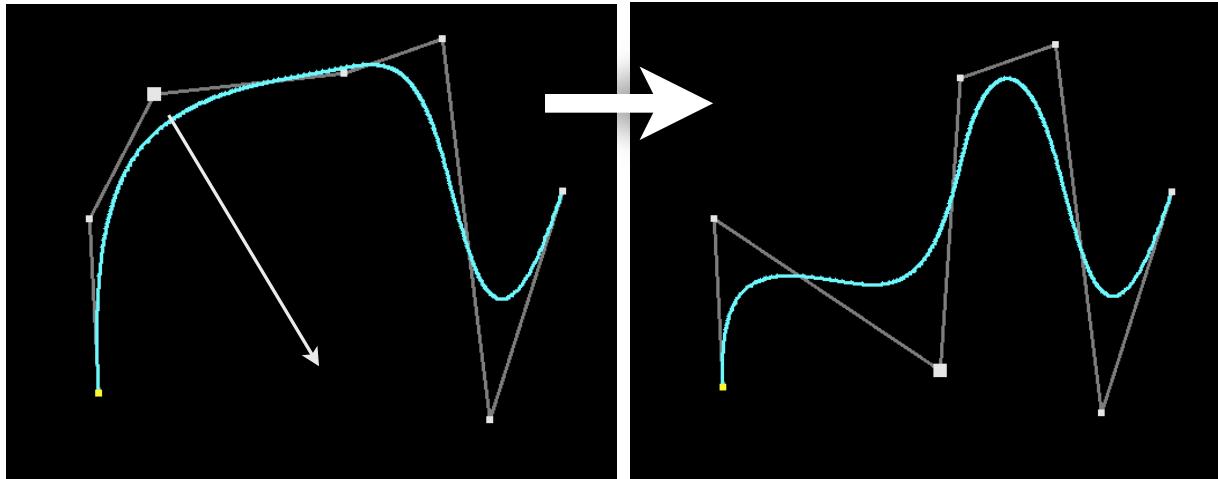
## Selecting Curves

Switch to the **Selection Tool** () , and click on a curve to select it. Hold SHIFT and click to select multiple curves. You can also select curves by clicking their name on the list of curves displayed in the side panel.



## Adjusting Curves

If you want to adjust a curve that you have already entered, select it with the selection tool, and then click and drag on the control points.

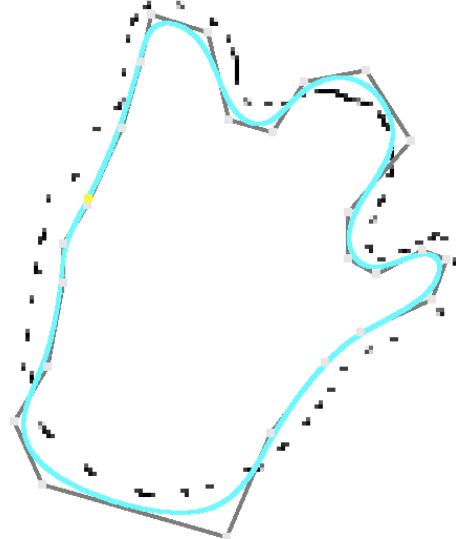


## Using the Curve Fitting Tools

Now that you have an initial curve, Kappa has built-in curve fitting tools that let you fit it to a set of data points. Using these tools is straightforward.

### The Initial Curve

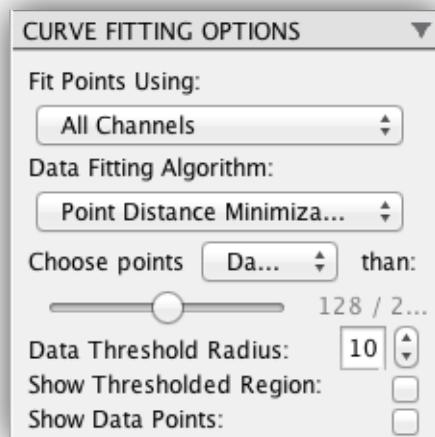
Your initial curve should loosely follow the shape of the data points. An example B-Spline is shown to the right.



### Selecting Data Points

Kappa will look near the initial curve for any pixels that are brighter or darker than a threshold. You can adjust this threshold level, as well as how far from the curve you will look, in the sidebar (under **Curve Fitting Options**).

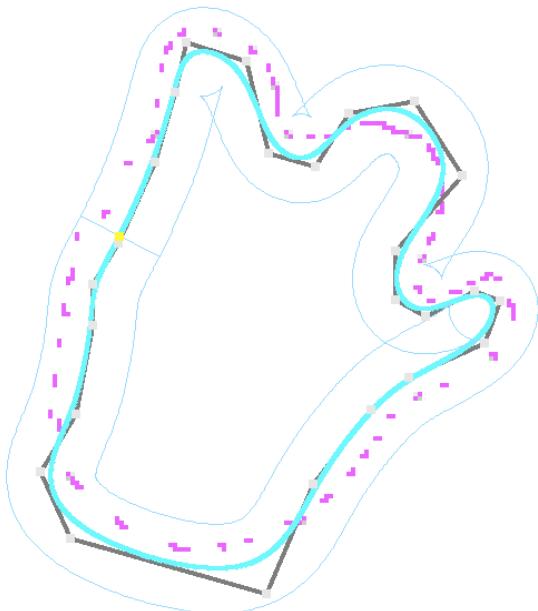
You can also choose to look only on the Red, Green, or Blue Channels, if you have an image with fluorescent labeling.



Clicking “Show Thresholded Region” will let you see the selection range, and clicking “Show Data Points” highlights any included data points in magenta.

The same initial curve, with selected data points highlighted, is shown here.

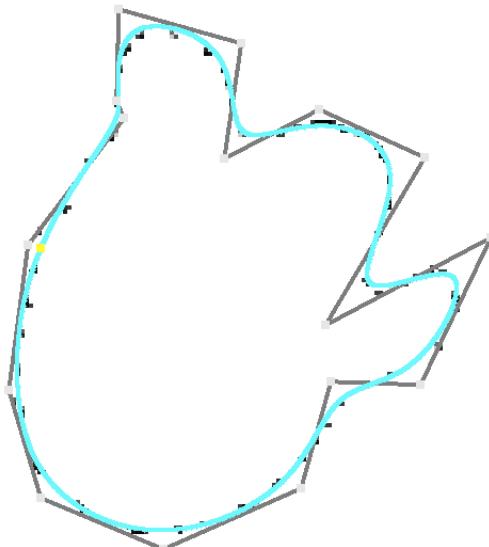
Before Curve Fitting



## Fitting the Curve

To fit the initial curve to the data points, go to **Tools → Fit Curve**. Alternatively, pressing **F** will do the same thing.

After Curve Fitting



## Saving and Loading Curves

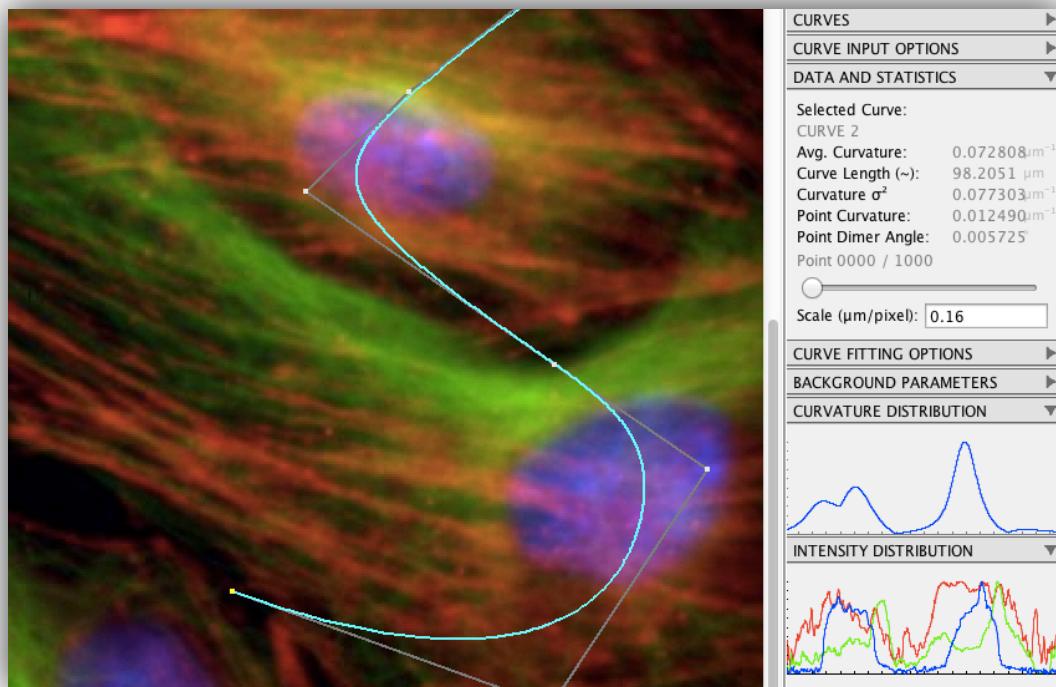
You can save the curves that you have entered into a separate **.kapp** file, which can be opened at a later time. To save your currently entered curves, go to **File → Save Curve Data**, or alternatively press **⌘S** (CTRL-S on Windows). Similarly, to load previously saved curves, go to **File → Load Curve Data**, or press **⌘L** (CTRL-L on Windows)



fitted curves.kapp

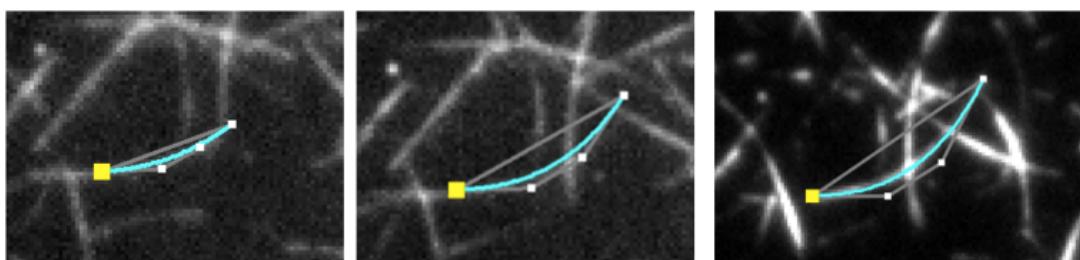
## Viewing Curve Properties

If a curve is selected, measurements will appear on the sidebar. In addition, a plot of the curve's **curvature distribution**, as well as the corresponding **pixel intensity distribution**, will be displayed. These measurements are based on a scale factor (in  $\mu\text{m}/\text{pixel}$ ). The default setting is  $0.16 \mu\text{m}/\text{pixel}$ , or  $160 \text{ nm}/\text{pixel}$ . You can change this scale factor in the sidebar.



## Working with Image Stacks

If you have a stack of images, you can adjust the shape of a curve on any frame you want. Then, for any intermediate frames, Kappa will *automatically* translate the control points. This lets you conveniently track **changes in shape** over an image stack. To change frames, you can drag the frame slider in the bottom left corner.



An example of tracking shape changes across multiple frames. By moving the control points, the curve will adjust as well.

# Exporting Data from Kappa

The **Export Panel** (↗) lets you export your curve data. Here you have options allowing you to specify what values you want to export.

The data will be exported as a comma delimited file (.csv), which can then be opened by most other applications.



**EXPORT OPTIONS:**

**PER DATA POINT OPTIONS:**

- Export X Coordinate
- Export Y Coordinate
- Export Curvature ( $\kappa$ )
- Export MT Dimer Angle

**PER CURVE OPTIONS:**

- Export All Data Points
- Export Averages Only

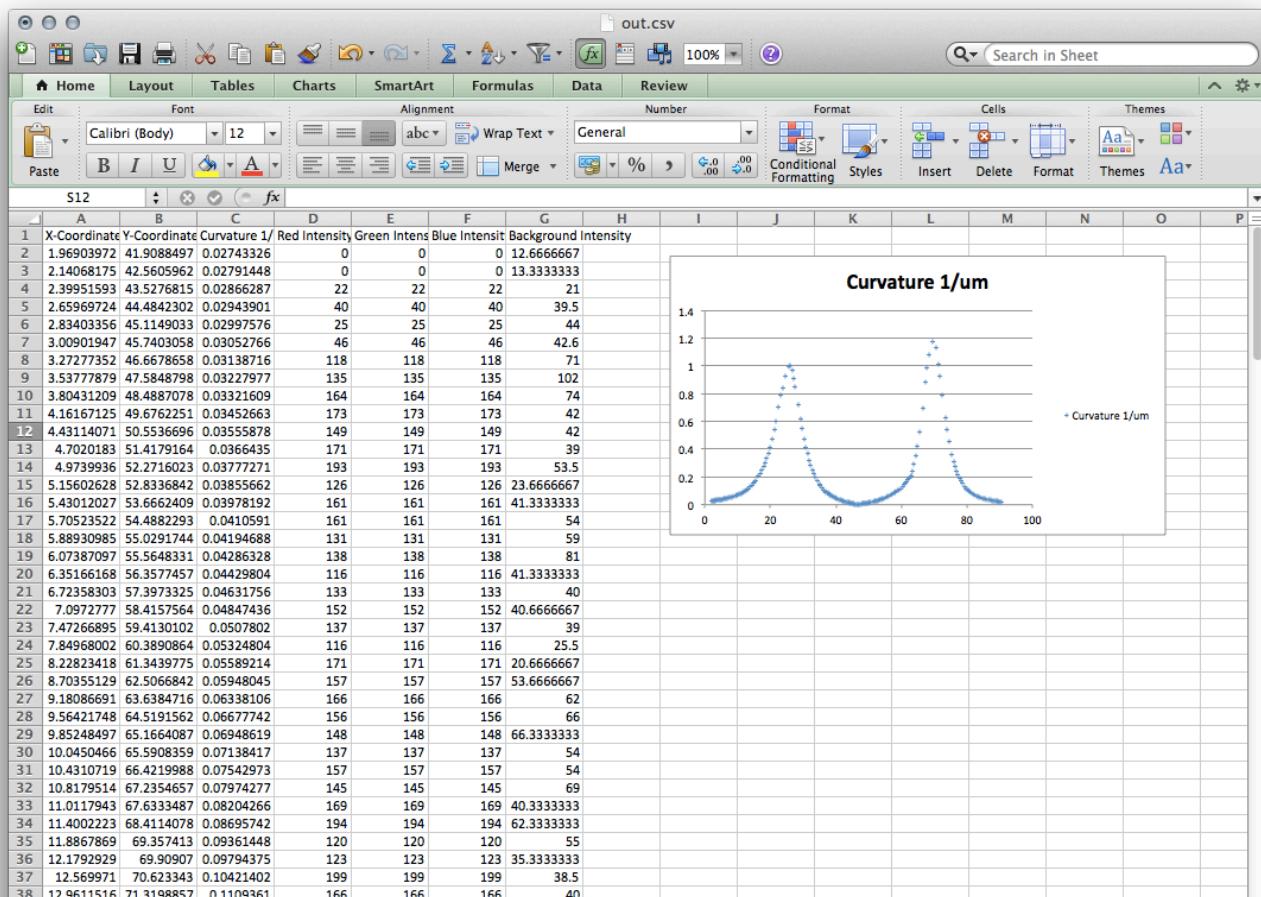
**PER FRAME OPTIONS:**

- Export All Curves
- Export Selected Curves Only

**PER STACK OPTIONS:**

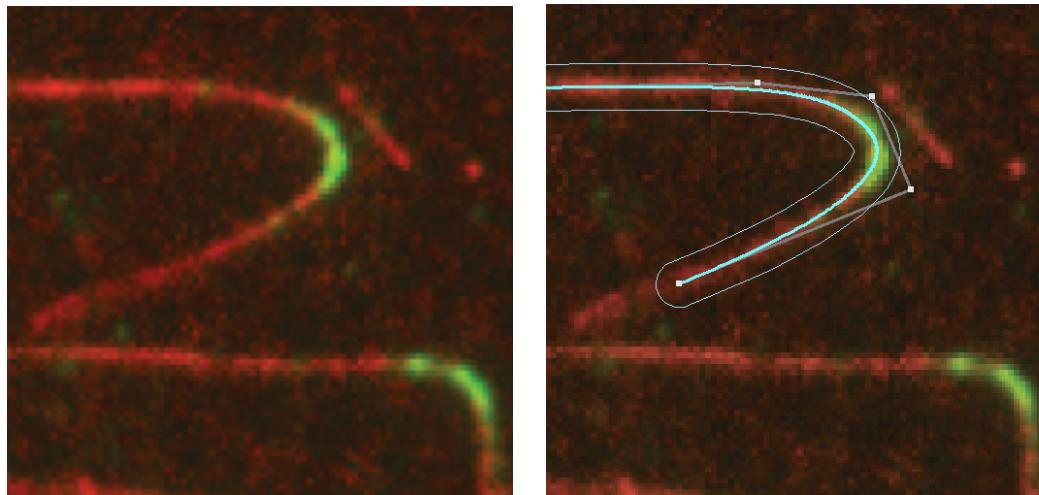
- Export Data in Each Frame
- Export Data in Keyframes Only

**EXPORT**

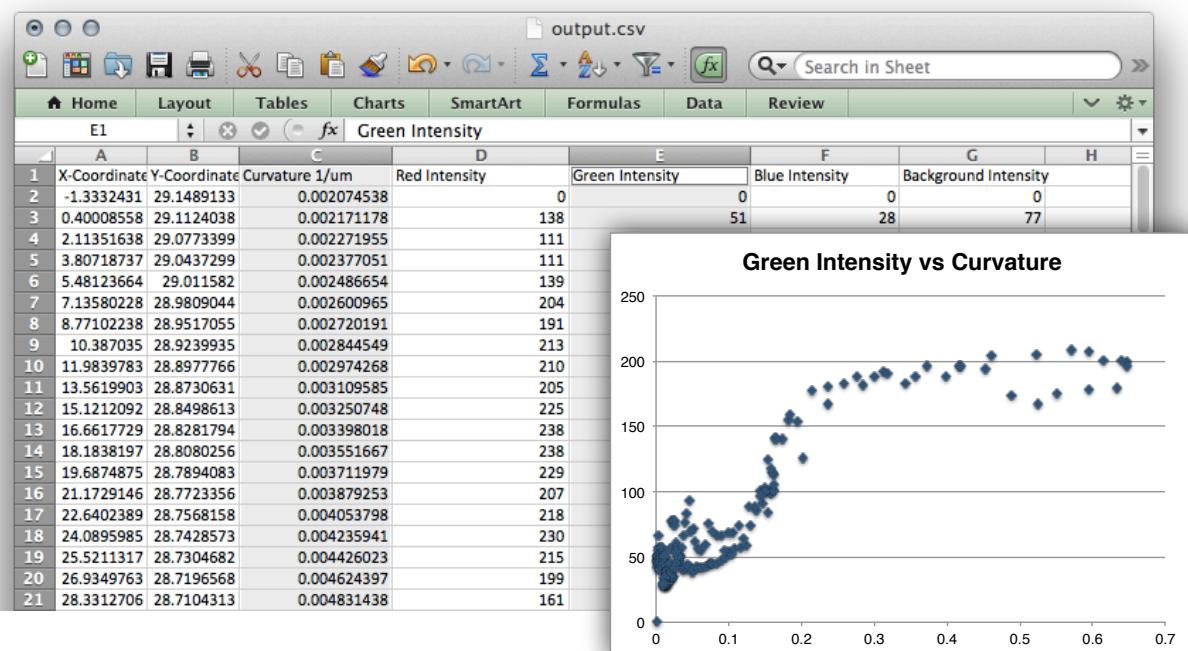


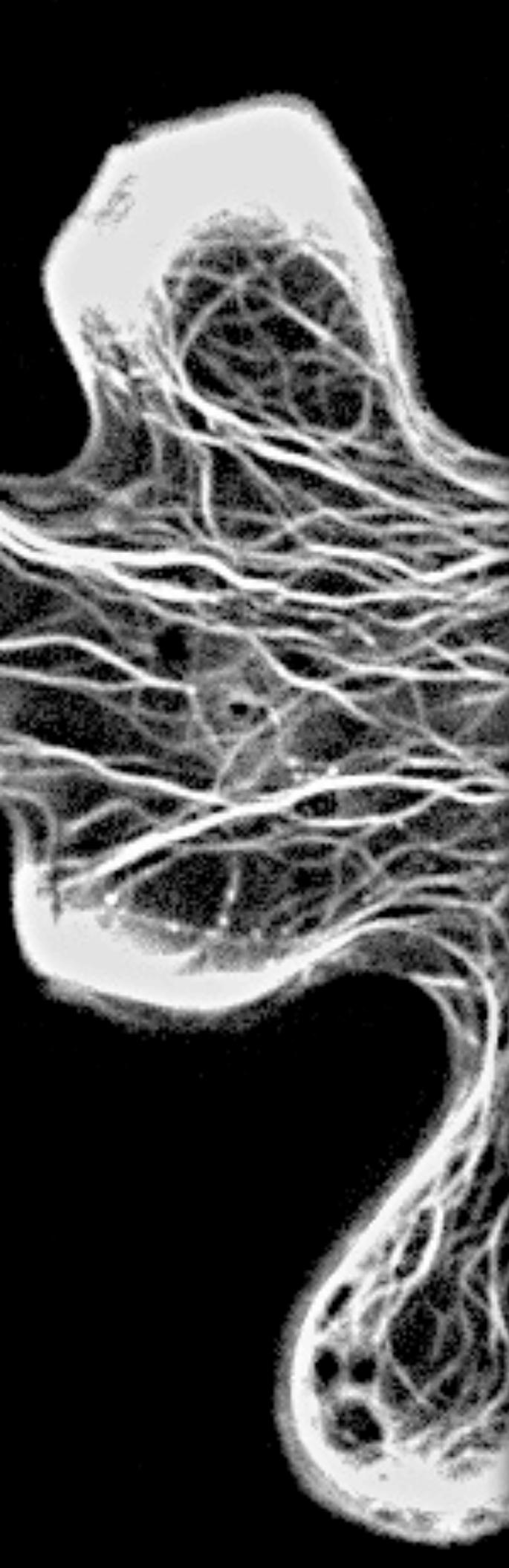
# Correlating Brightness with Curvature

In many cases, it is highly desirable to know whether a fluorescently-tagged protein preferentially binds to a curved surface. In this situation, it would be useful to correlate the **image intensity** on a specific channel with the **curvature**. In Kappa, this is very easy to do.



When your data is exported, the intensity on each color channel is exported as well. This gives you a measure of the curvature as well as the image intensity, for each point on your curve. To plot the brightness versus the curvature, it is as simple as plotting the two data columns.



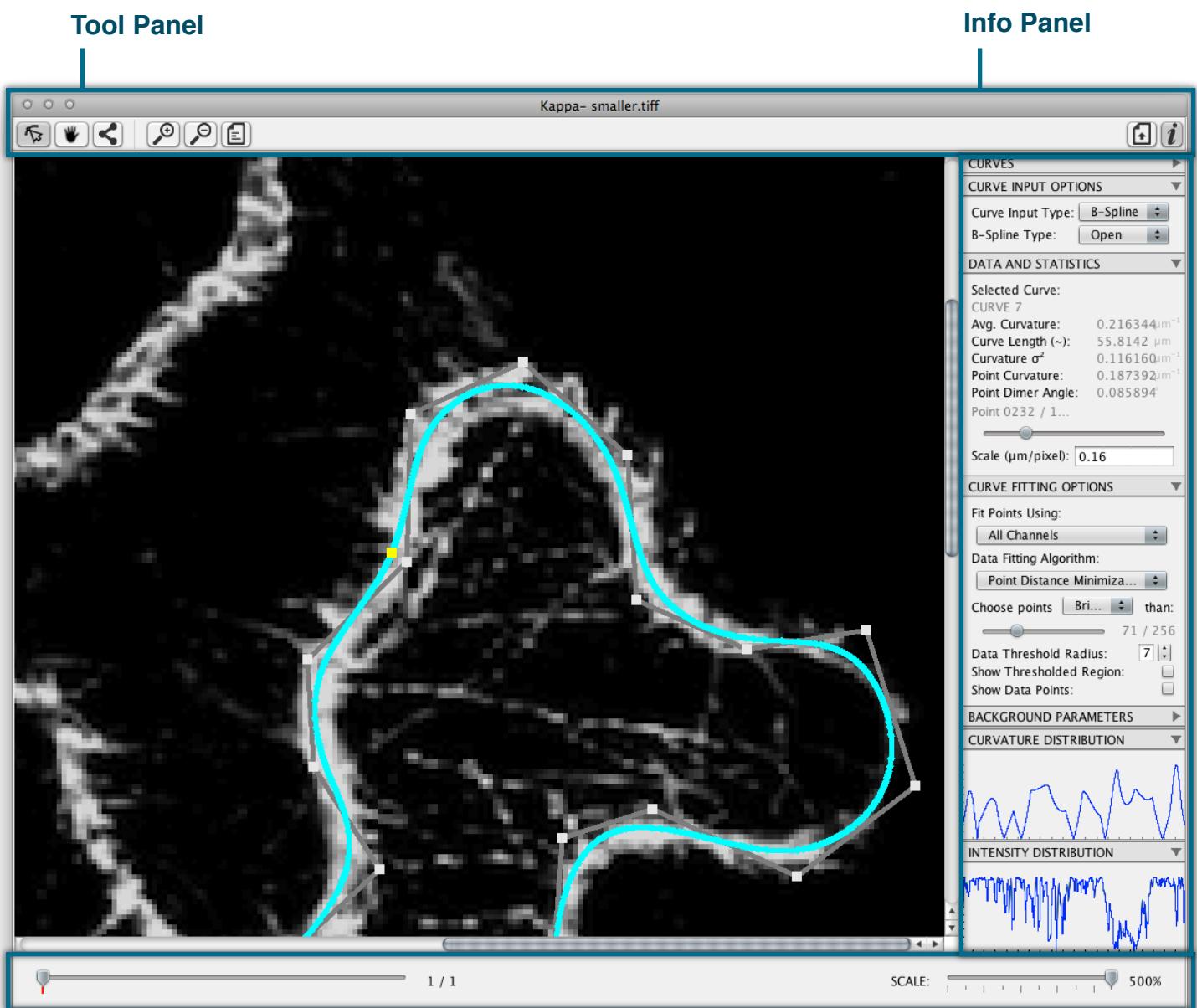


# Section 3

## The User Interface

# The Main Window

The main window of Kappa has 3 panels. The top panel is the **tool panel**. It contains all the tools used to manipulate the image. On the right is the **info panel**. This displays information about selected B-Splines, and allows the user to configure how they want each curve to be fit. Finally, on the bottom is the **control panel**. Here, there are scrubbers that can be used to zoom in or out of an image, or to navigate between frames in an image stack. Clicking the Export button will switch to an **export panel**, where export options can be configured.



# The Tool Panel



## The Selection Tool

Select points and curves that have been entered.



## The Panning Tool

Pan around the image. Press the spacebar to temporarily switch to this tool.



## The Spline Input Tool

Enter curves. When this tool is selected, click to enter control points, and press enter to input your curve.



## Zoom In



## Zoom Out



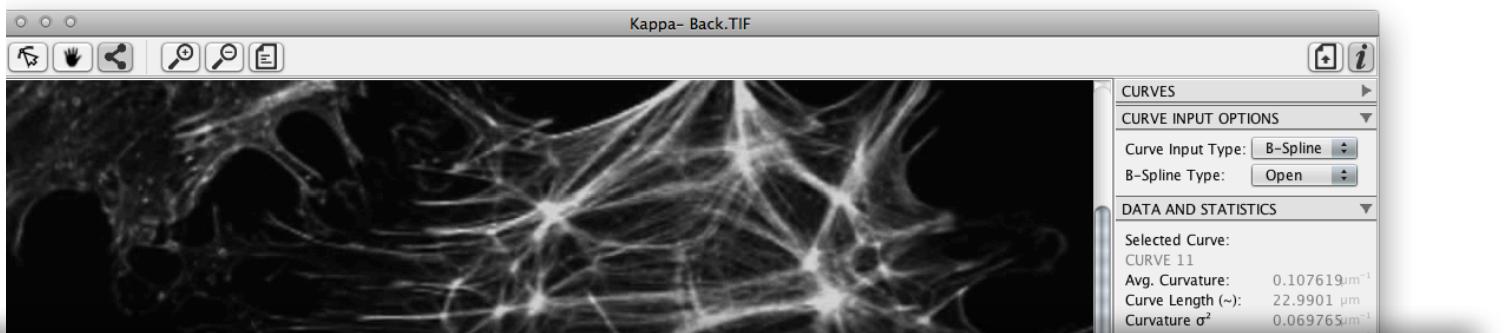
## Switch to the Export Panel

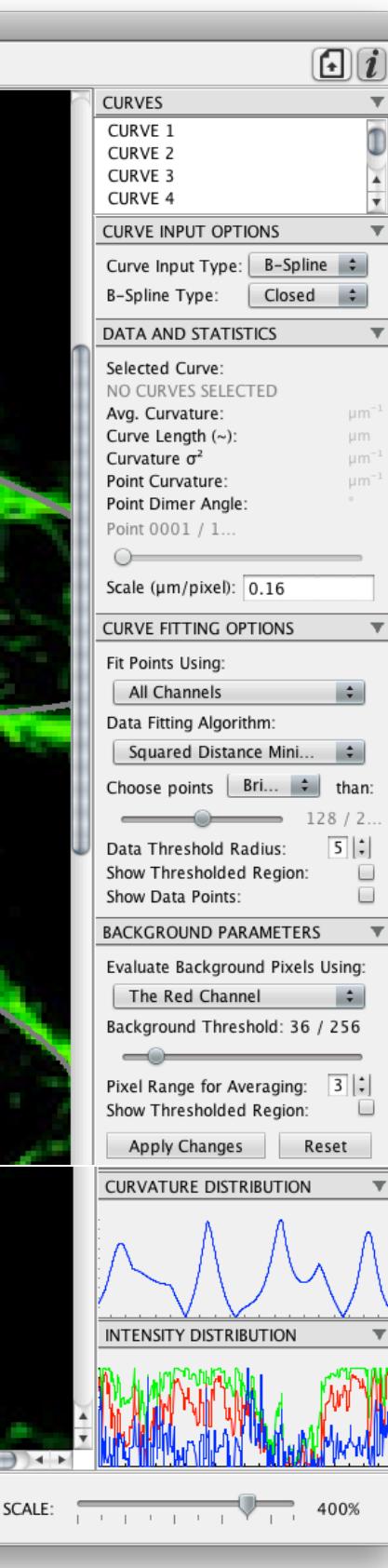
Clicking this button switches the info panel with the export panel. The export panel allows you to specify what data you want to export from the program. If the export panel is already displayed, clicking this button will hide the panel.



## Switch to the Info Panel

Clicking this button switches the export panel with the info panel. If the info panel is already displayed, clicking this button will hide the panel.





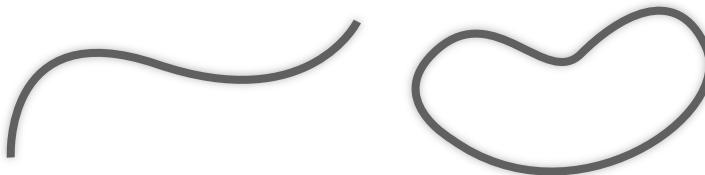
# The Info Panel

## The Curves Pane

Here is where all the entered curves are listed. You can select and deselect different curves by clicking on them.

## The Curve Input Pane

This panel provides options on what type of curve you wish to input. You can either input a **Bézier Curve**, or a cubic **B-Spline**, which is a spline comprised of several cubic Bézier Curves. If you enter a B-Spline, you also have the option of specifying whether the spline will be **open** or **closed**: an open curve will have free ends, while a closed curve will close on itself.



An Open B-Spline

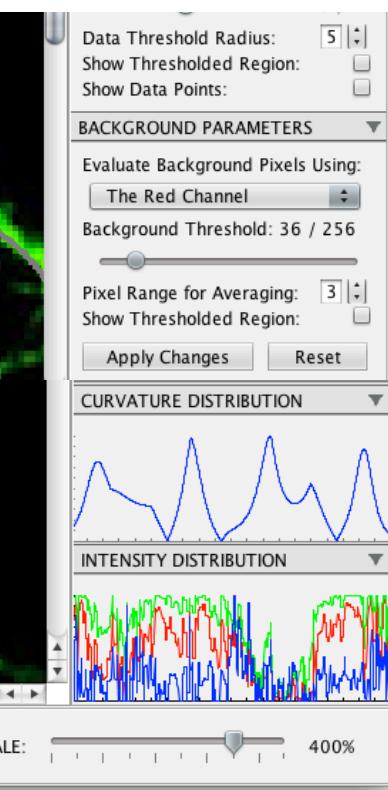
A Closed B-Spline

## The Data and Statistics Pane

This panel gives you measurements from the currently selected curves. Values that are displayed include the **average curvature**, the **curve length**, and the **curvature standard deviation**. Additionally, you can view the curvature at any point by dragging a slider.

## The Curve Fitting Options Pane

Here you have settings for Kappa's curve fitting features. How these features are used are described in more detail in the previous section.



## The Background Parameters Pane

This panel lets you select pixels that belong to the image background. These pixels are then used to estimate background intensities across the entire image. The resultant background values can optionally be used to normalize intensity measurements.

## The Curvature Distribution Pane

This panel shows a plot of the curvature along each point on the currently selected curve.

## The Intensity Distribution Pane

This panel shows a plot of the red, green, and blue intensity along each point on the currently selected curve.

# The Control Panel

## The Layer Slider

The layer slider (**bottom left**) lets you go through multilayer images.

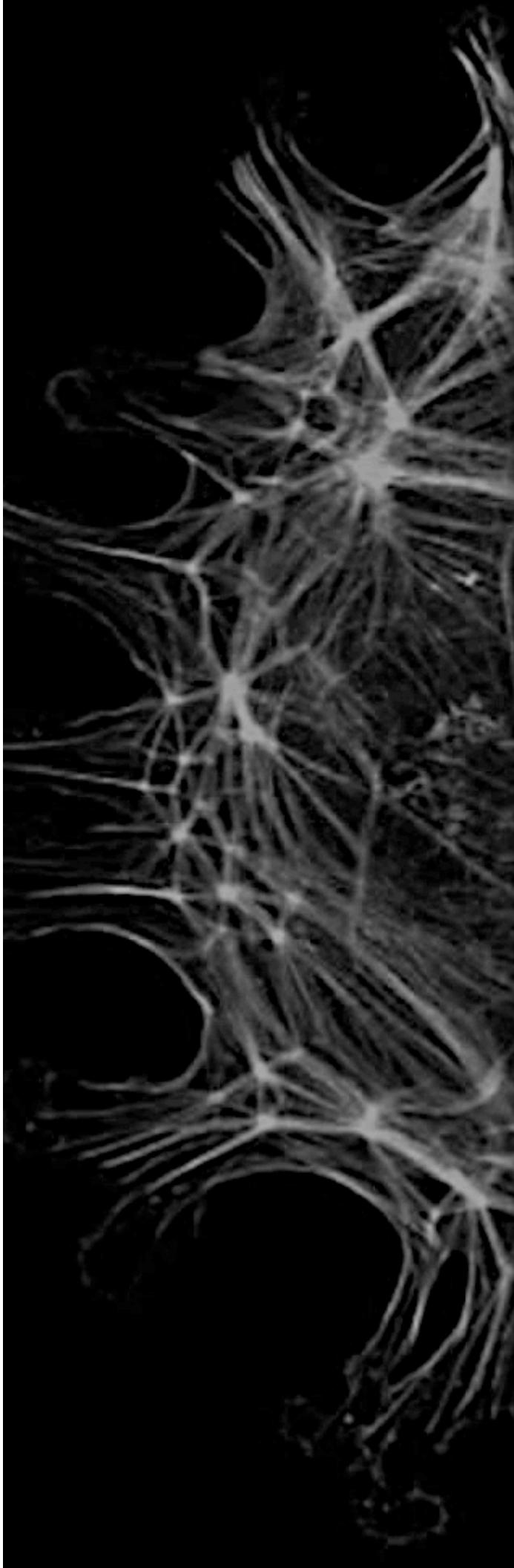
## The Zoom Slider

The zoom slider (**bottom right**) lets you zoom in or out of the image.



Layer Slider

Zoom Slider



# Section 4

## Keyboard Shortcuts

Operation	Keyboard Shortcut
File Handling Shortcuts	
Open an Image	⌘O
Save Curve Data	⌘S
Load Curve Data	⌘L
Navigation Shortcuts	
Switch to the Selection Tool	V
Switch to the Panning (Hand) Tool	H
Switch to the B-Spline Input Tool	B
Zoom-In	⌘= (CTRL=)
Zoom-Out	⌘- (CTRL-)
Skip to Next Frame	↖→
Skip to Previous Frame	↖←
Skip to Next Keyframe	⌘→ (CTRL→)
Skip to Previous Keyframe	⌘← (CTRL←)
Operation Shortcuts	
Fit Curve	F