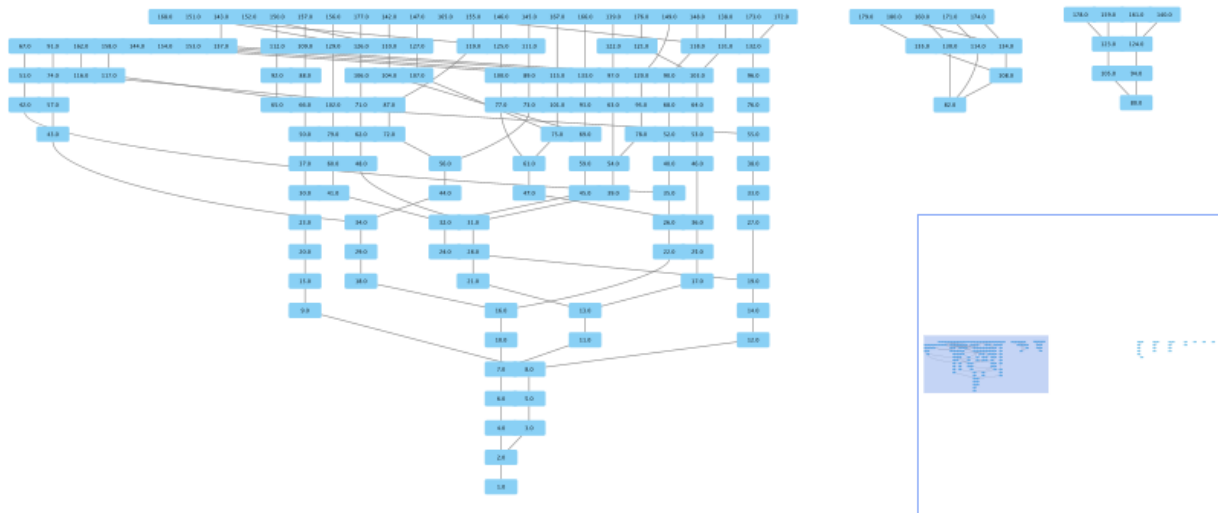
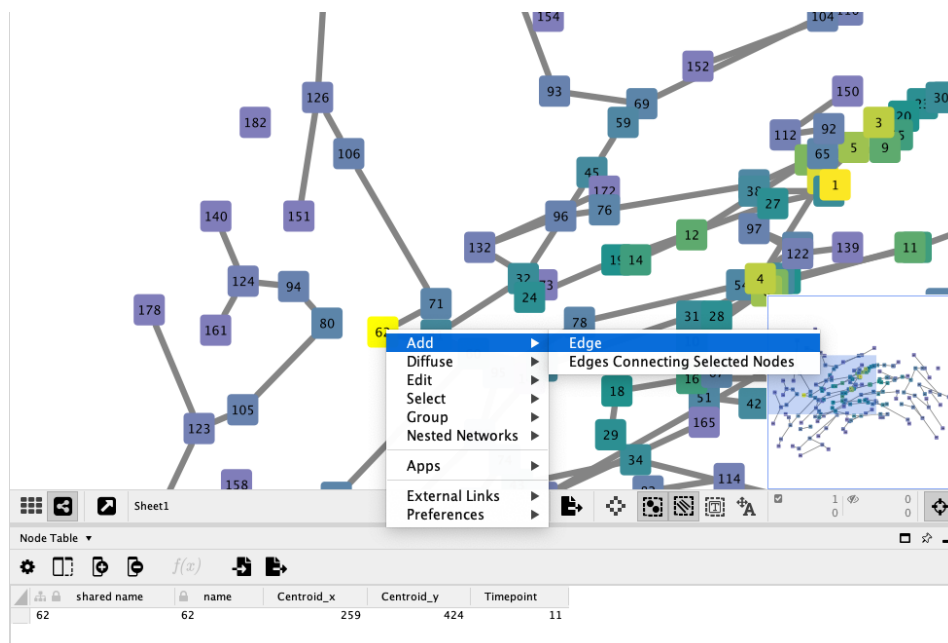


This tutorial contains step-by-step instructions to post-process STrack results using Cytoscape. This provides an interactive framework to:

- Display Strack results and modify the network's appearance to your liking:



- Edit STrack results by removing/ adding tracks



1. Install Cytoscape on your computer:

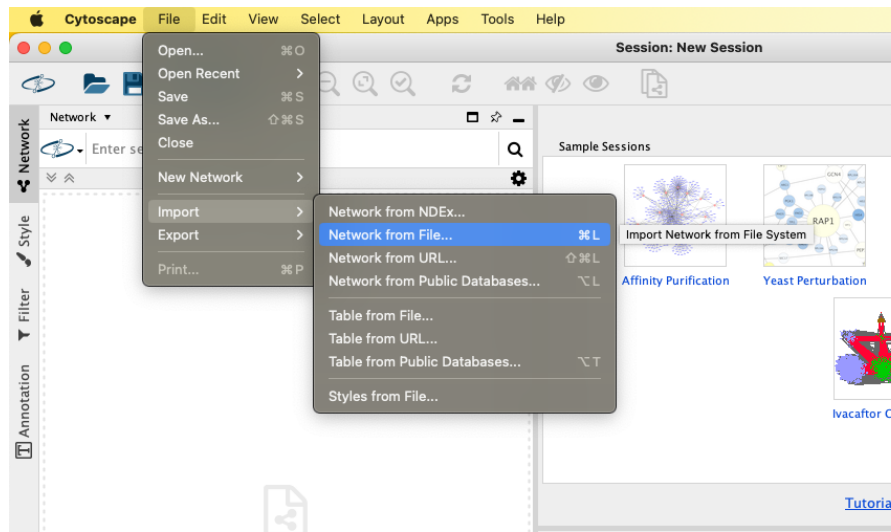
You will find information on how to download and install Cytoscape on this web page: <https://cytoscape.org/>

The web page also contains exhaustive documentation, tutorials and videos on Cytoscape. Feel free to explore the contents if you need any information that you lack in this short and condensed tutorial.

2. Import the “complete\_tracking\_table.csv”, that contains tracks that were identified by STrack for all timepoints in your timelapse, into Cytoscape:

Click on File > Import > Network from File

Browse to the “complete\_tracking\_table.xlsx” and click on “Open”



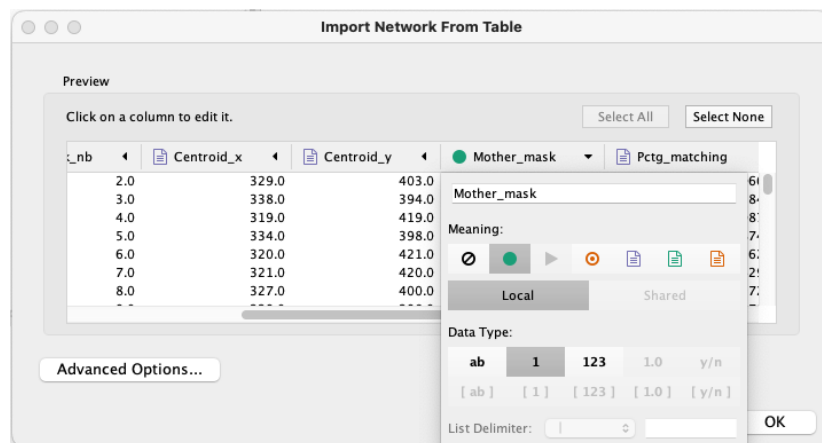
3. Define the columns to use as source node and target node (in this step, we define the mother cells as source nodes and the daughter cells as target nodes to build a network of all links).

Click on the “Mother\_mask” column title > select “1” as Data Type > Select ● (Source node) as Meaning

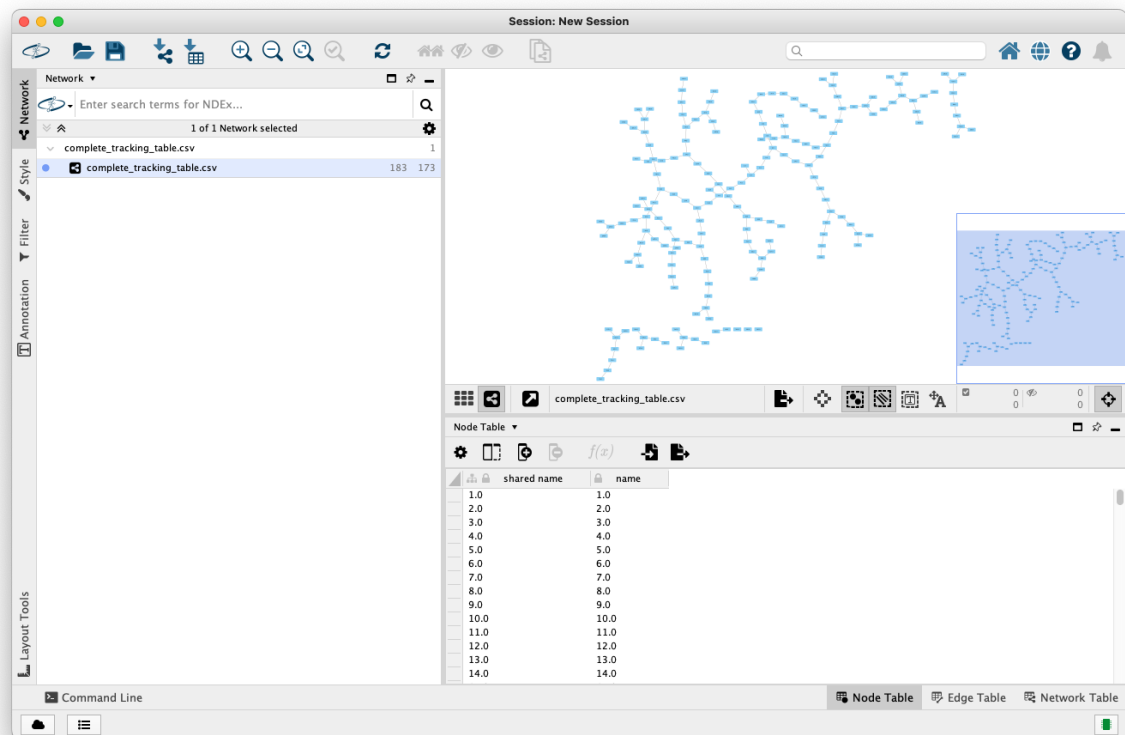
Click on the “Mask\_nb” column title > select “1” as Data Type > Select the orange target (Target node) as Meaning

Click on all the other column titles > Select ○ (Not imported) as Meaning

Finally, Click on “ok”



#### 4. Your network was now imported into Cytoscape!



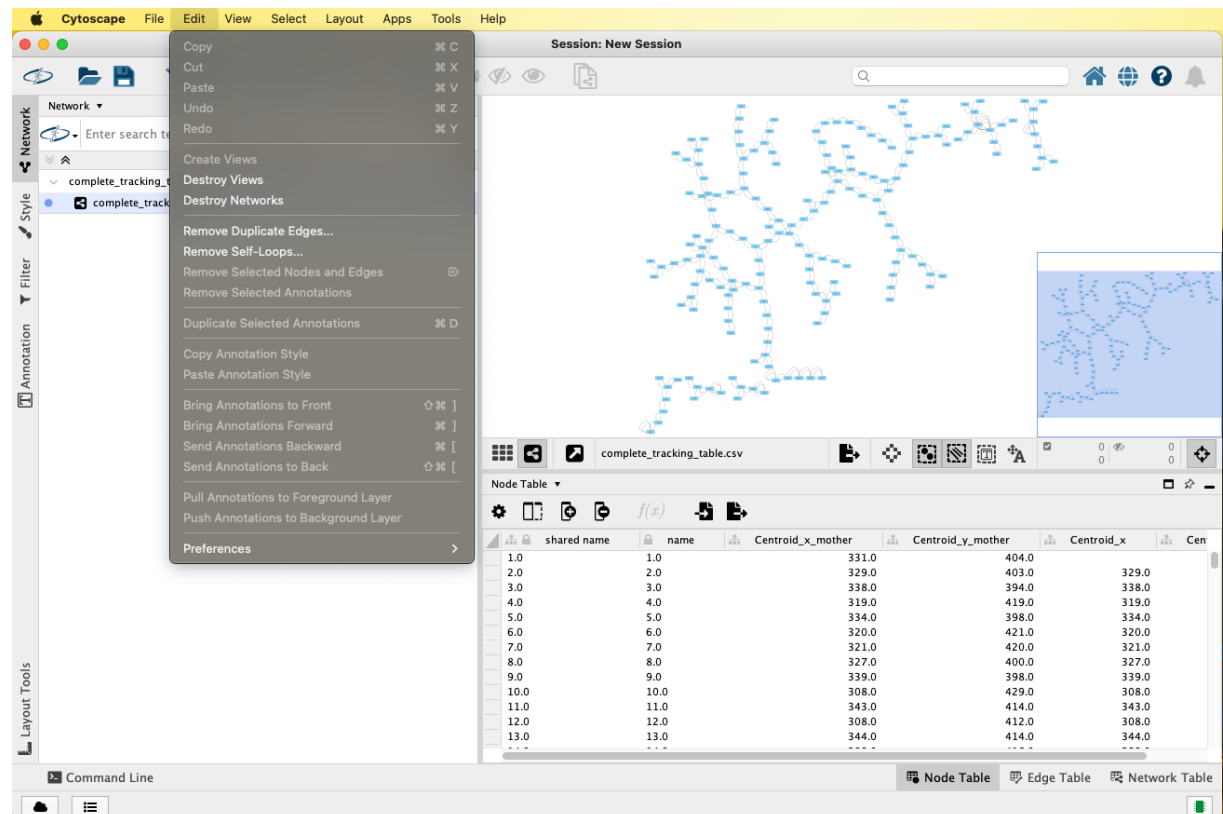
Duplicate edges or self-loops might have appeared in the graph by mistake.

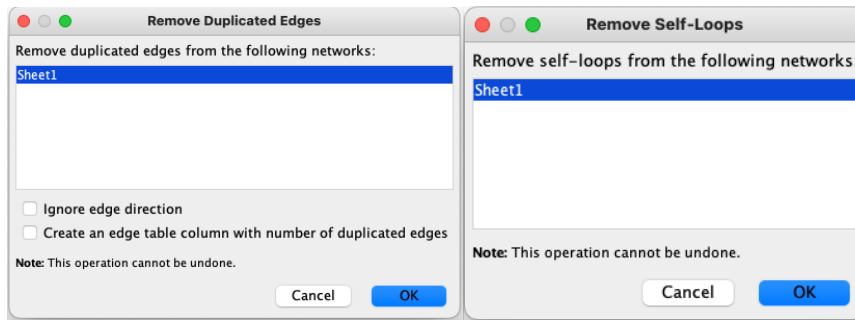
To remove duplicate edges:

Click on Edit > Remove Duplicate Edges > Select the “Sheet1” file > Click on ok

To remove self loops:

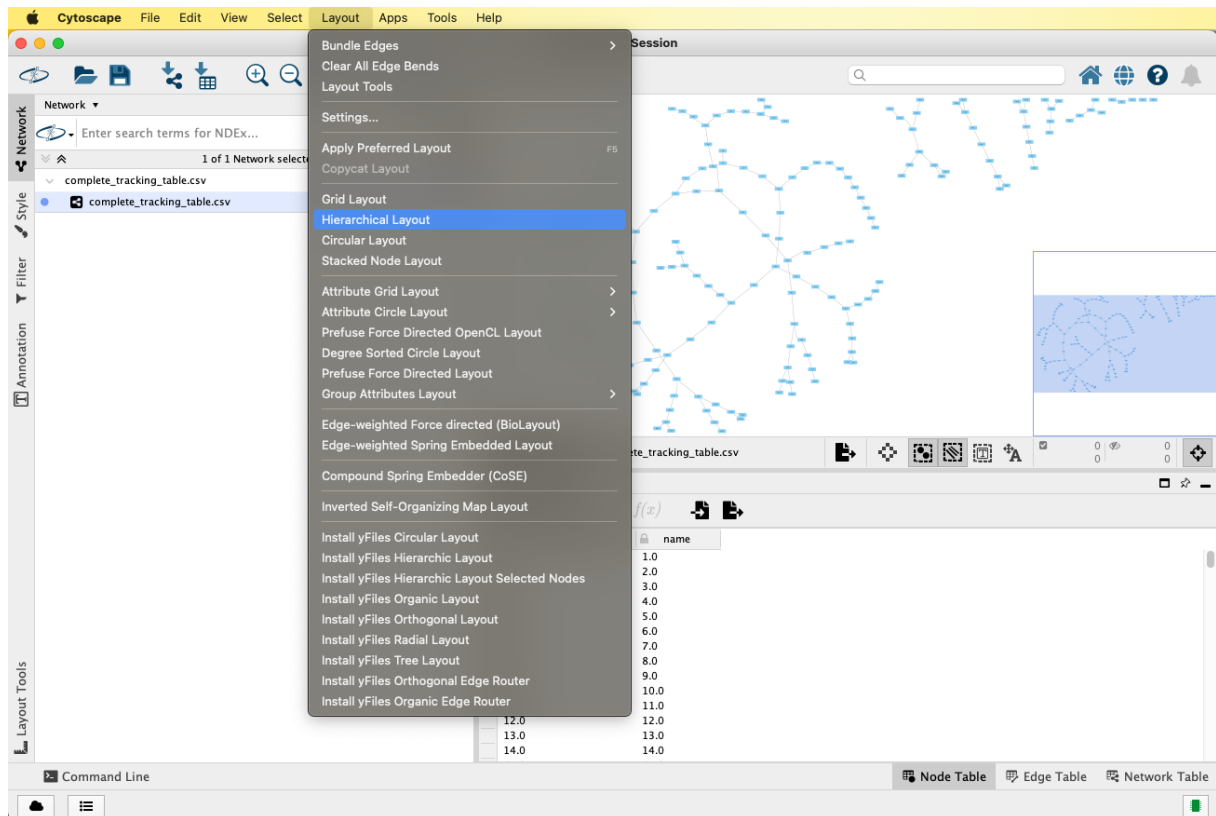
Click on Edit > Remove Self Loops > Select the “Sheet1” file > Click on ok



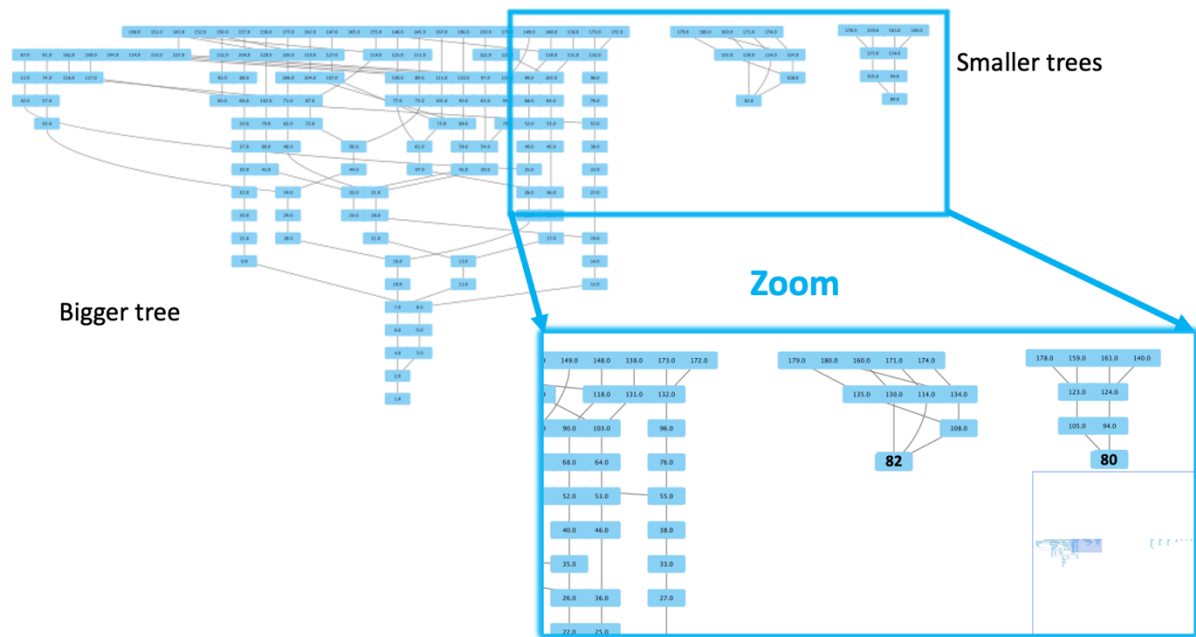


## 5. Change the networks layout

Click on Layout > Hierarchical Layout



This layout shows the network similarly to a lineage tree. In our example, this allows us to see that the results of STrack seem to contain one big tree + several smaller trees. This might be the result of a tracking error, and we might want to re-attach the smaller trees to the bigger one.



For instance, we see that the nodes 82 and 80 might have been wrongly considered as lonely cells, and we might want to re-attach them to the bigger tree.

## 6. Import information on the cells into Cytoscape

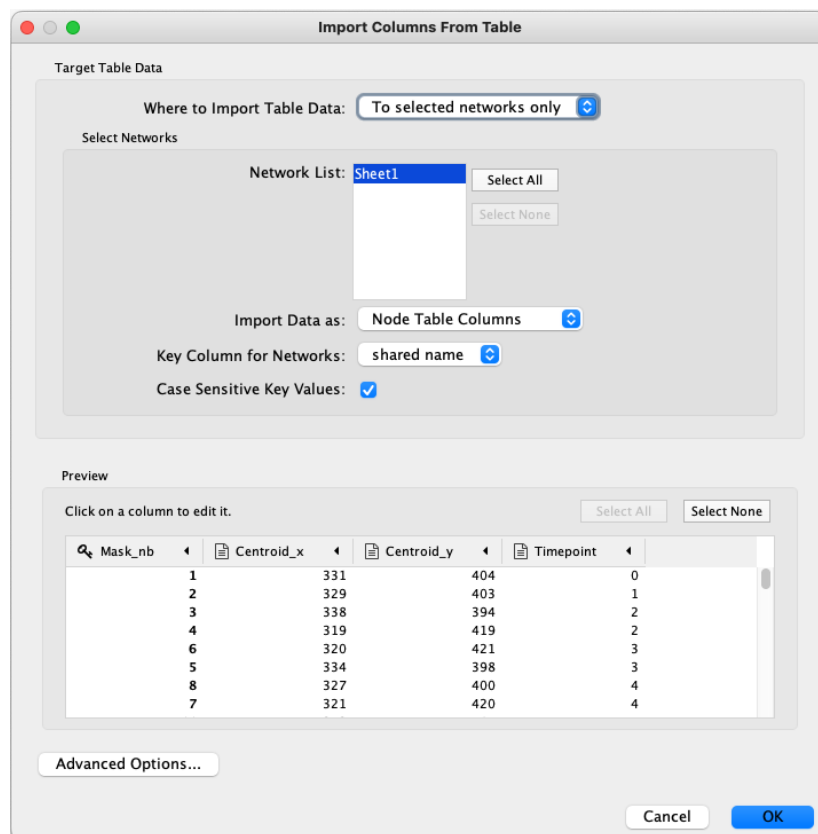
We will now import information on the cells (such as their x-y coordinates in your timelapse images, their timepoints, ...). We import this information from the “tracked\_cells\_table.xlsx” file, that was exported by STrack.

Click on File > Import > Table from File

Browse to the “tracked\_cells\_table.xlsx” and click on “Open”

In the “Where to Import Table Data” field, select “To selected networks only”

Click on OK

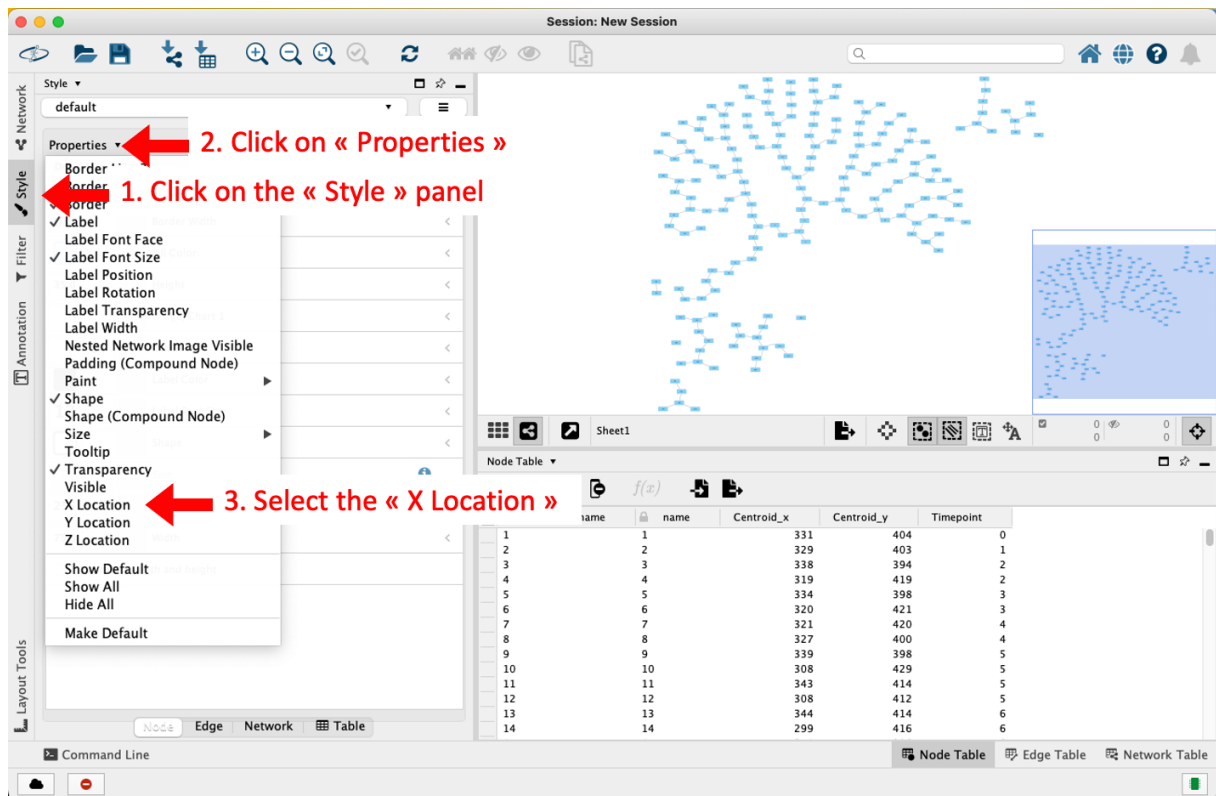


## 7. Visualise the information on cells in Cytoscape

Now that you have imported information on cells into Cytoscape, you can use this information to:

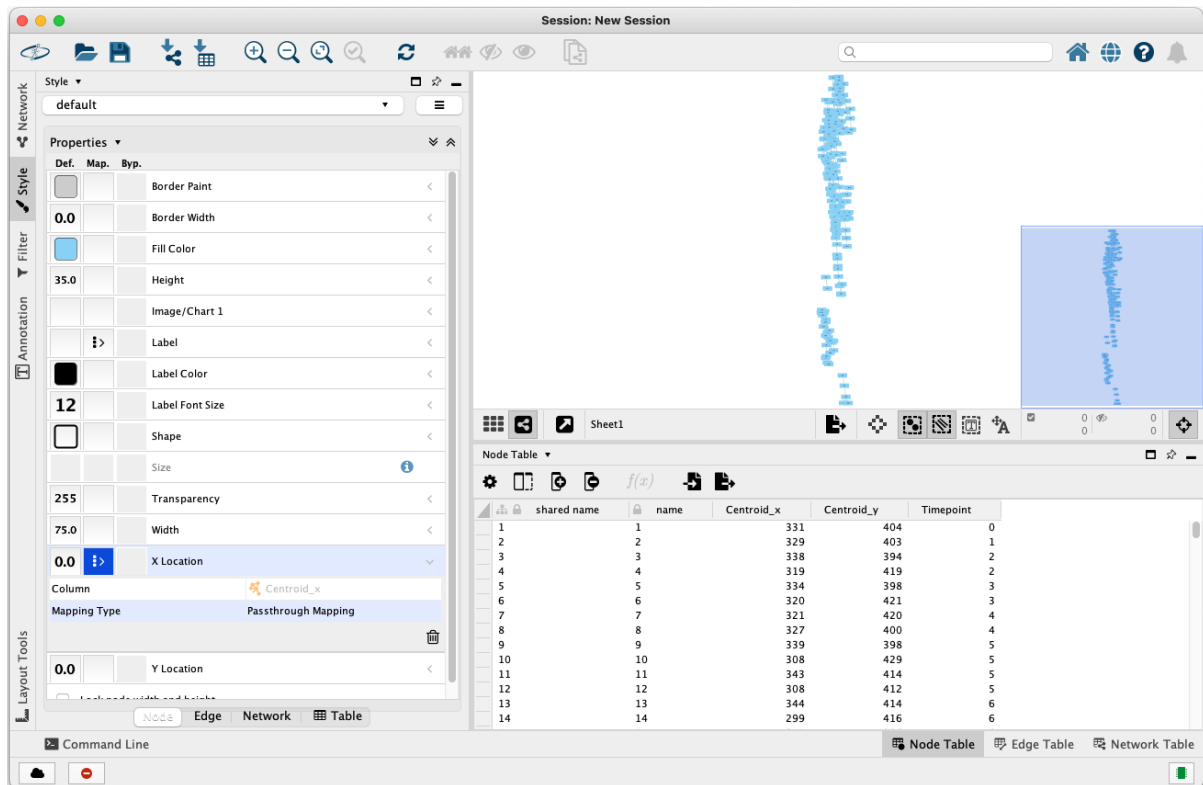
- Change the layout, such that cells are located at their true x-y coordinates
- Color cells according to their timepoint

Click on the “Style” panel > Click on Properties > Select the “X location” and “Y location”



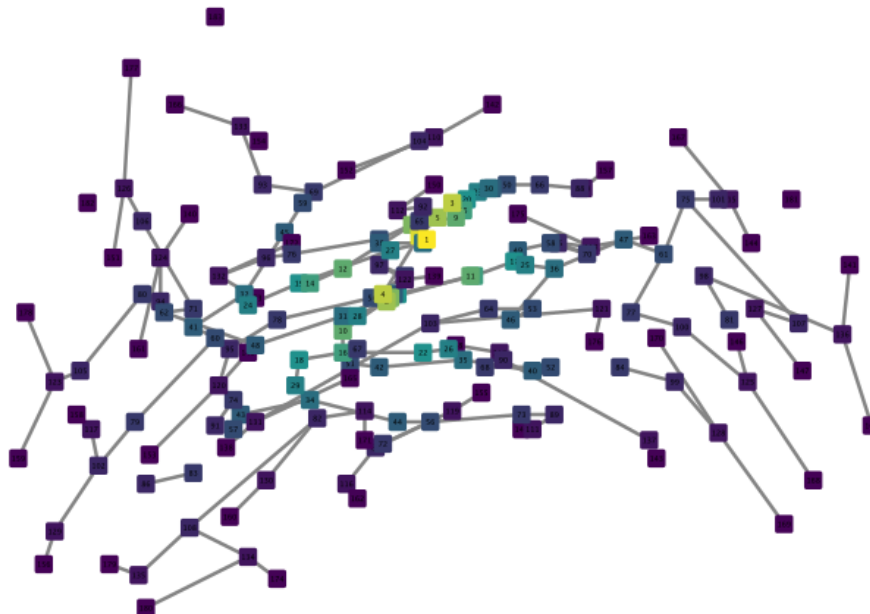
In this menu, you can change the color, size, shape (...) of the nodes. We will only change the nodes X and Y location, and color them according to timepoint.

Click on the X Location > Select “Centroid\_x” as Column > Select “Passthrough Mapping” as Mapping Type



Click on the Y Location > Select “Centroid\_y” as Column > Select “Passthrough Mapping” as Mapping Type  
 Click on “Lock node width and height” under the Y Location  
 Change the “Node Size” and “Label font size” if necessary  
 Click on “Fill Color” > Select “Timepoint” as Column > Select “Continuous Mapping” as Mapping Type

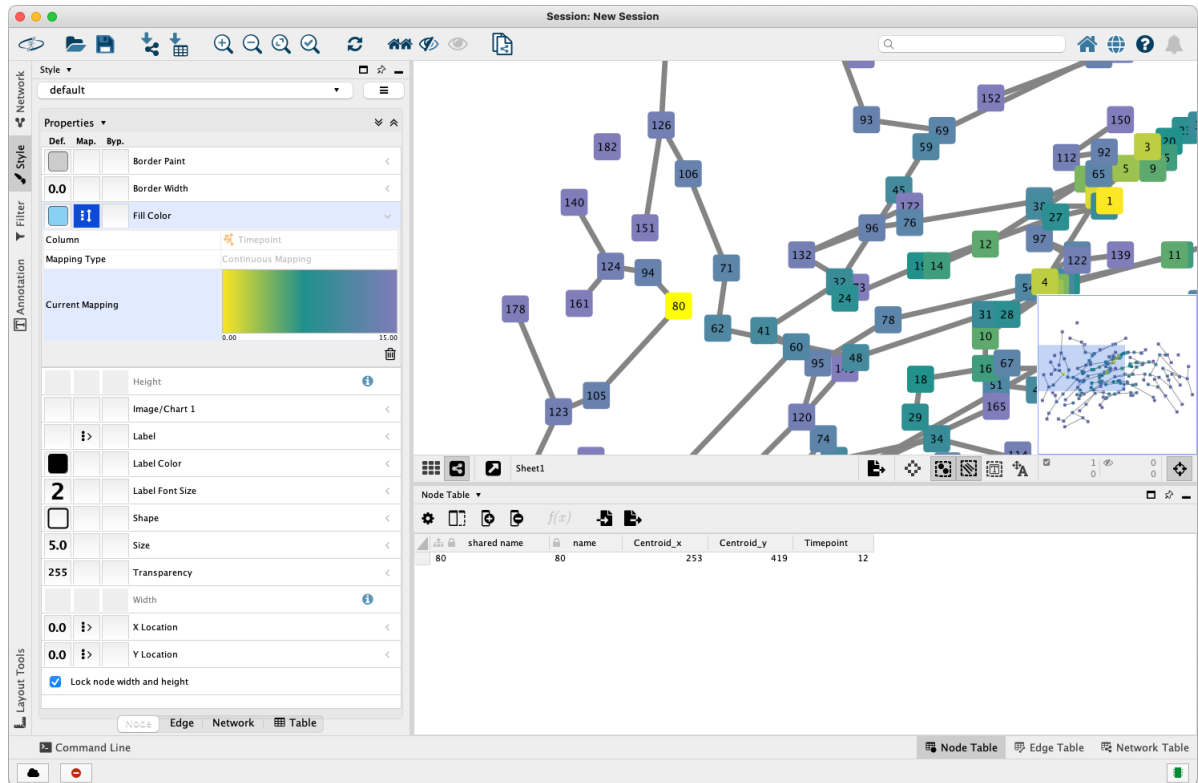
The nodes in your graph should now be located at their correct x-y coordinates, and colored according to time.



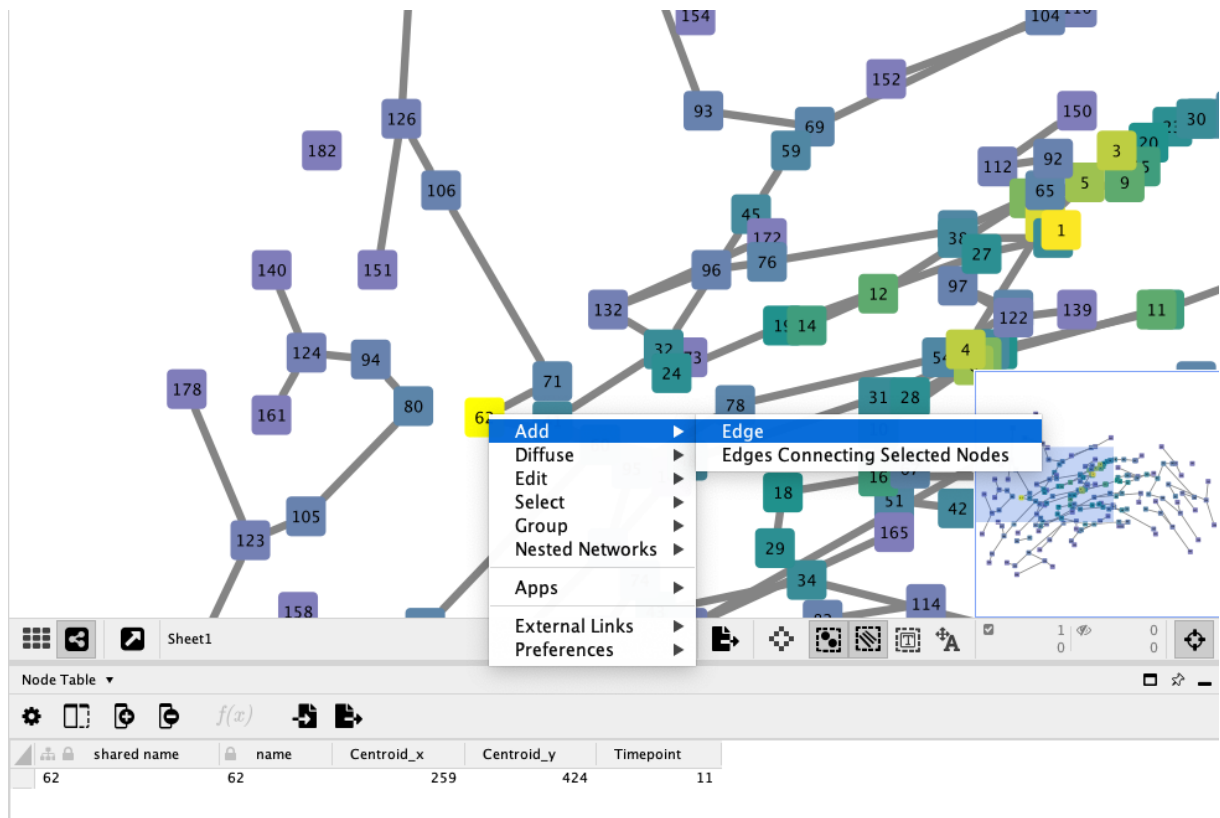


## 8. Correct the tracks by adding or removing links between nodes

By clicking on nodes in the Cytoscape network, we can see their respective node information in the Node table below. As an example, we clicked on node 80, and we see its x and y coordinates in the Node Table below. The Node table also shows us that node 80 came from timepoint 12.



The node 62, that is located very close to node 80, came from timepoint 11 and has only one daughter cell (the node 71). We can thus safely correct STrack's results, by adding an edge between node 80 and node 62. To do so, we select the node 62 and right click > Add > Edge and click on the node 80.



An edge was now created between the nodes 80 and 62.

In order to remove an edge, click on it and right click > Edit > Cut

Cytoscape offers many possibilities, and we've only explored a very few of them in this tutorial. If you're missing any information on how to use Cytoscape, feel free to consult this web page: <https://cytoscape.org/>