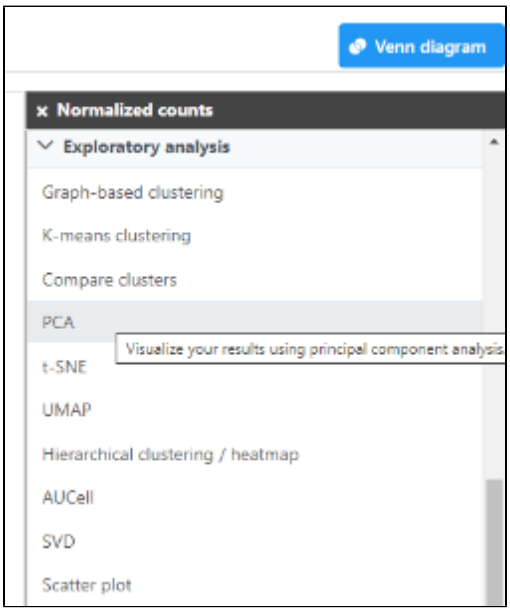
Perform Exploratory analysis

- Use Principle Components Analysis (PCA) to reduce dimensions
- Classify cells based on a marker for expression

Use Principle Components Analysis (PCA) to reduce dimensions

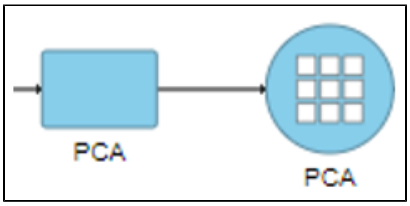
- Click the **Normalized counts** data node
- Expand the **Exploratory analysis** section of the task menu
- Click **PCA**



In this tutorial we will modify the PCA task parameters, to not split by sample, to keep the cells from both samples on the PCA output.

- Uncheck (de-select) the **Split by sample** checkbox under *Grouping*
- Click **Finish**

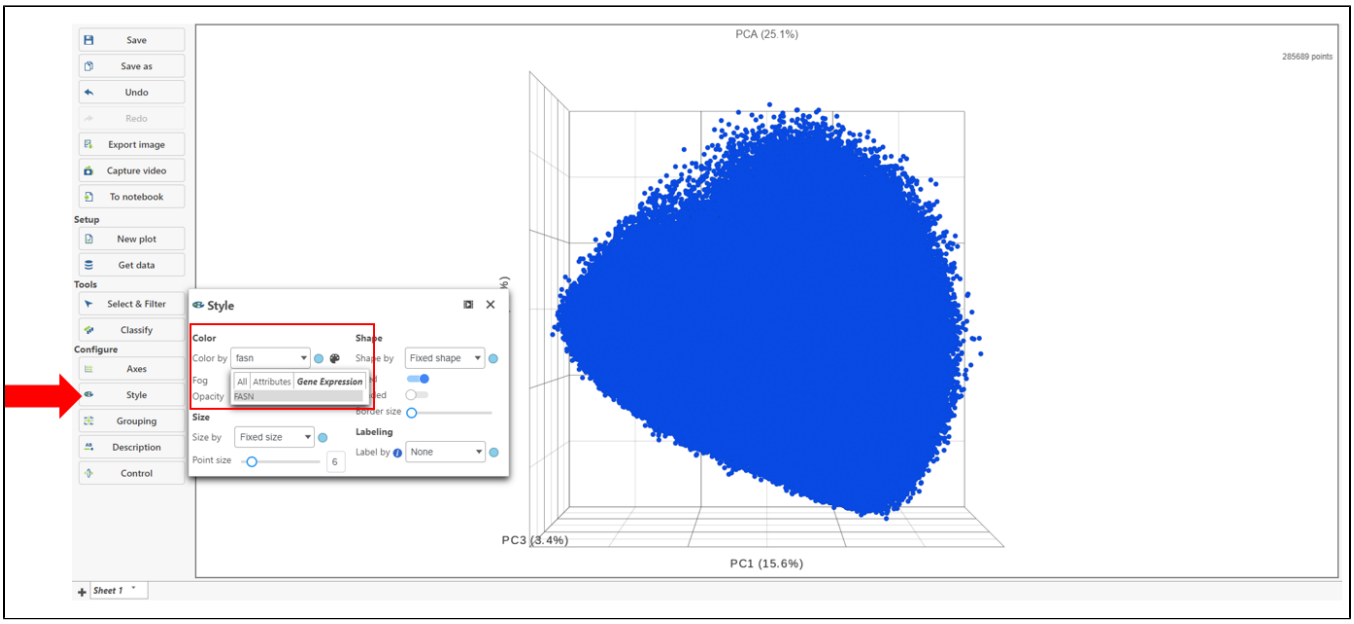
- Double-click the circular **PCA** node to view the results



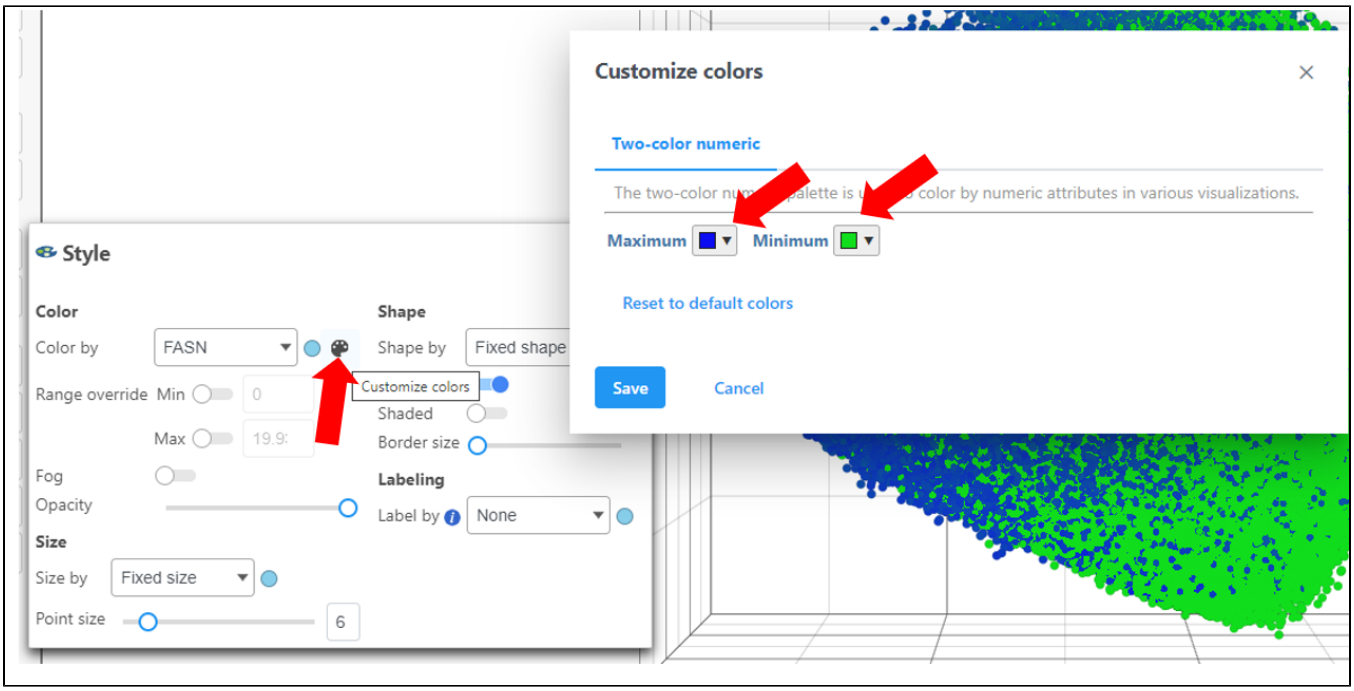
From this PCA node, further exploratory tasks can be performed (e.g. t-SNE, UMAP, and Graph-based clustering).

Classify cells based on a marker for expression

- Choose **Style** under *Configure*
- **Color by** and search for *fasn* by typing the name
- Select *FASN* from the drop-down

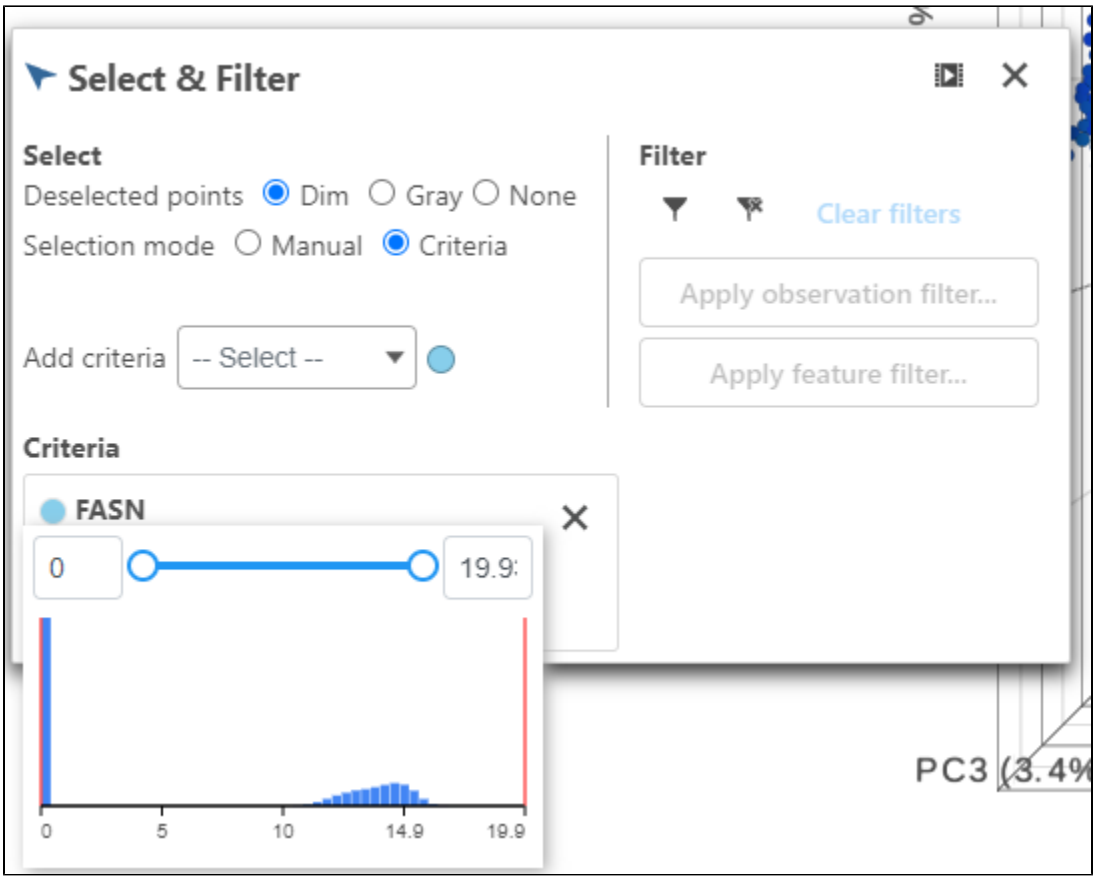


The colors can be customized by selecting the **color palette** then using the color drop-downs as shown below.



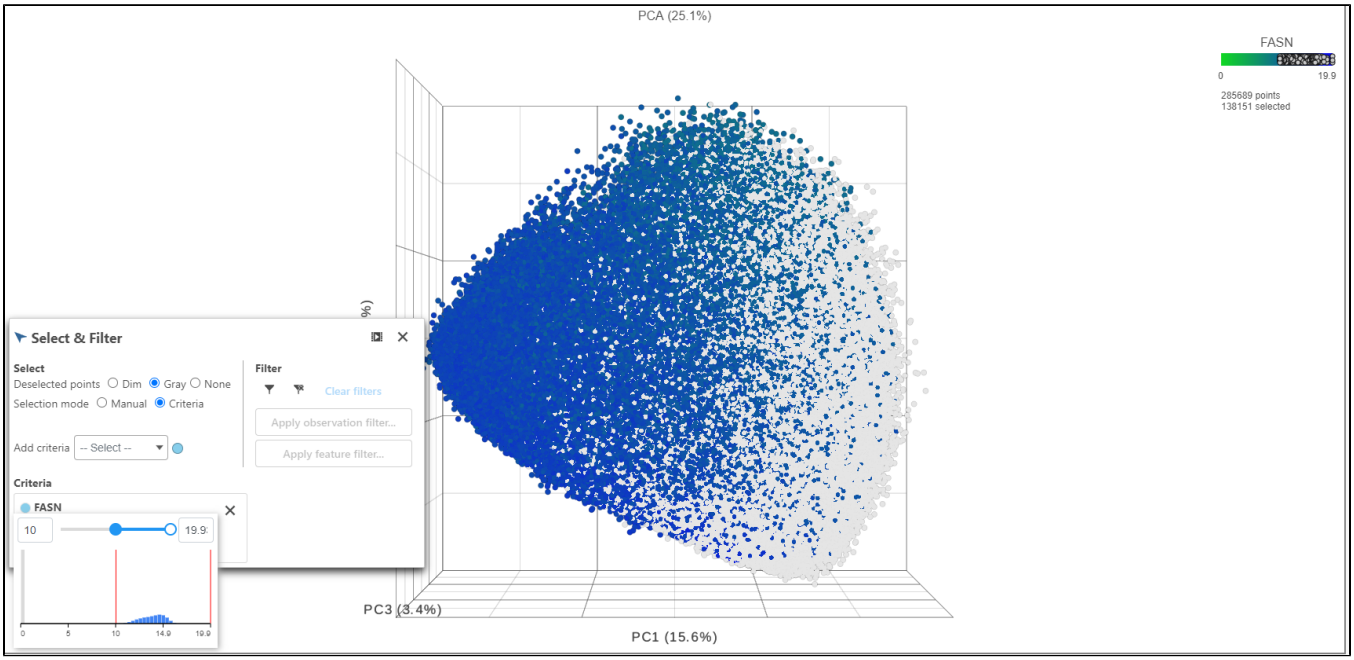
Ensure the colors are distinguishable such as in the image above using a blue and green scale for **Maximum** and **Minimum**, respectively.

- Click **FASN** in the legend to make it draggable (pale green background) and continue to drag and drop FASN to **Add criteria** within the **Select & Filter** *Tool*
- Hover over the slider to see the distribution of FASN expression

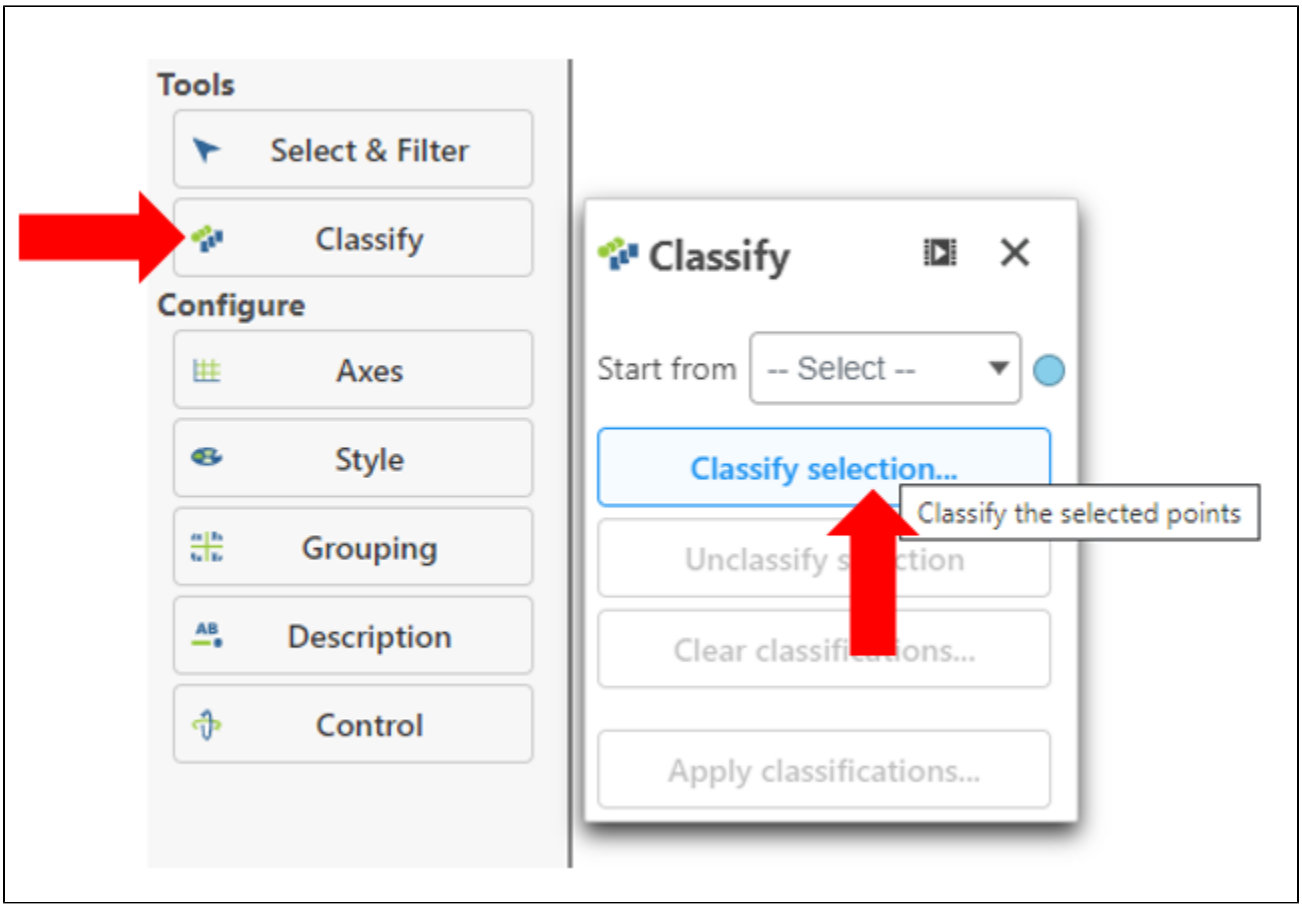


Multiple gene thresholds can be used in this type of classification by performing this step with multiple markers.

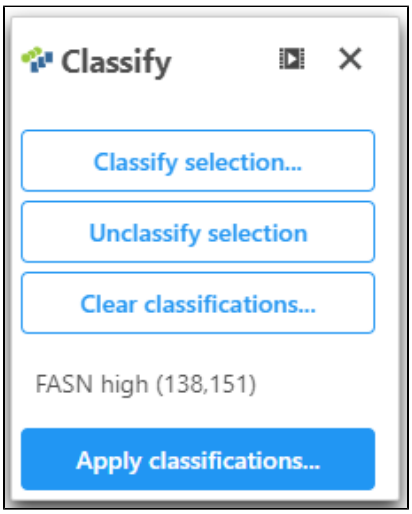
- Drag the slider to select the population of cells expressing high FASN (the cutoff here is 10 or the middle of the distribution).



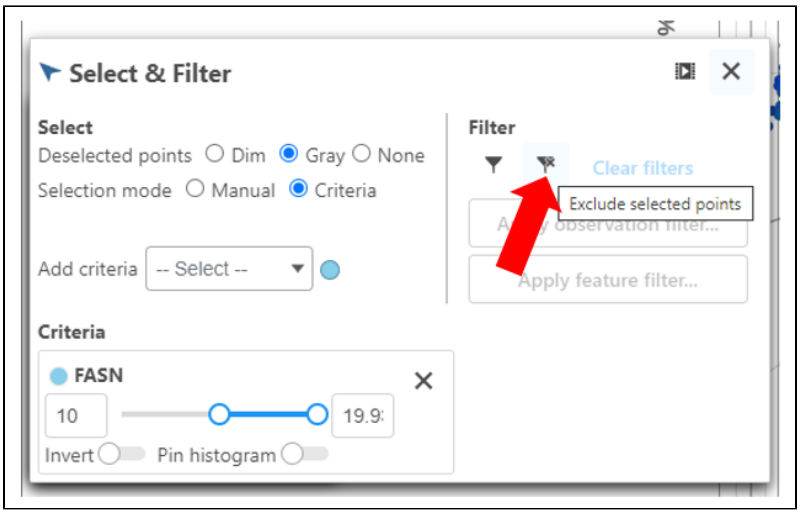
- Click **Classify** under *Tools*
- Click **Classify selection**



- Give the classification a name "FASN high"

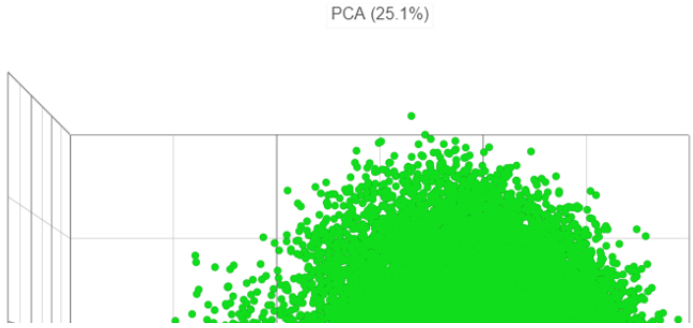


- Under the Select & Filter tool, choose **Filter** to **exclude** the selected cells

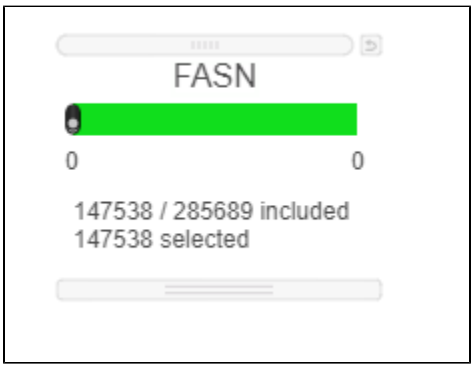


Exit all Tools and Configure options

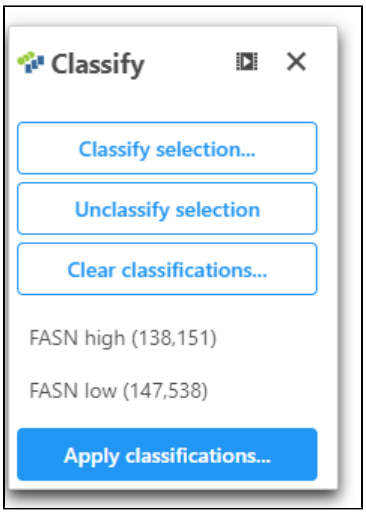
- Click the "X" in the right corner
- Use the **rectangle** selection mode on the PCA to select all of the points on the image



This results in 147538 cells selected.



- Open **Classify**
- Click **Classify selection** and name this population of cells "FASN low"
- Click **Apply classifications** and give the classification a name "FASN expression"



Now we will be able to use this classification in downstream applications (e.g. differential analysis).

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Additional Assistance

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