Protein Metric Calculator

**Overview:** Referring to our Touchstone paper, it is important to benchmark spatial samples using these statistical metrics that we have developed and tailored to Spatial OMICS. However, the main scope of the previous project is the RNA samples; this is a translated script that operates on protein samples instead.

**Input:** The input for this script is the default expression matrix flat file that you can get from AtoMx.

**Functions:**

*getNcells*: This function simply returns the number of cells in the sample.

*getFlPerCell*: This function sums up all the fluorescence value from the matrix and divide it by the number of cells such that we can find the average fluorescence per cell.

*getComplexity*: This function finds the number of genes that is required to contribute to 50% of the total fluorescence in the sample. It then is normalized by the size of the gene panel.

*getEntropy*: This function returns the Shannon Entropy value given the matrix.

*getSparsity*: We define matrix values that are lower than 2 standard deviations from the mean as empty values. This function finds the number of empty values over the total size of the matrix.

*getMeanSNR*: This function finds the mean of the top 20% expressing cells and divide it by the mean of the bottom 10% expressing cells to find the signal-to-noise ratio.

**Tutorial:**

A comparison of a graph

Description automatically generated with medium confidence

The script returns a box plot like the one above in which each box represents a different tissue. This is a quick way to compare the metrics across different tissue samples. Other statistics can be viewed by generating html files of the summary statistics. Furthermore, you can also generate summary statistics for comparisons between disease states. Feel free to include other areas for comparison by altering the code.