Using Hierarchical Clustering and Dendrograms to Quantify the Clustering of Membrane Proteins

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***Short Abstract* — Biologists commonly label membrane proteins with gold nanoparticles and then, with high accuracy, locate the center of the gold particles using transmission electron microscopy and image processing software. Important biological information is contained in the clustering of the proteins. Here, we introduce a number, the intrinsic clustering distance, that characterizes the maximum number of clusters in an image. This distance is computed from a hierarchical clustering and dendrogram (or dendogram) algorithm. Previously, the analysis of clustering was based on the Hopkins and Ripley statistics, for which the user must compare statistical plots.**

***Keywords* — dendrogram, dendogram, hierarchical cluster analysis, dose response.**

I. Introduction

Cells communicate with the outside world using signals that consist of a chemical cascade that starts at the cell membrane and proceeds to some internal organelle such as the nucleus or endoplasmic reticulum. Biologist want to know how the dynamic organization of the membrane receptors that start the signaling cascade plays a role in signaling [1]. Briefly, the data we use to study the organization of the membrane receptors is obtained by stimulating the cells, then fixing the cells, removing part of the cell membrane, and then labeling the cytoplasmic part of the receptors with a gold nanoparticles [4]. A digital image of the of the gold particles is created using a transmission electron microscopy (TEM). The positions of the particles are points in the plane generated using image processing software [5]. We are interested in the clustering of the receptors, where clusters are defined by choosing a distance d and then putting two points into the same cluster if they are less than the distance d apart. Our main result shows how to use the data to compute an appropriate d. We applied our ideas to a large set of images generated to understand dose-response dependence of clustering, calcium release and degranulation on the strength of the stimulation of the cells.

II. Summary of Results and Conclusions

In [3] it was proven that small clusters are signaling competent. Therefore rather than fix a distance between particles to find the number of clusters, we define the intrinsic clustering distance dI to be smallest value of d at which there is a maximum number of clusters. From our analysis we concluded that dI is a function of the stimulus. It decreases as the stimulus increases. We use the intrinsic distance to quantify aggregation. The biological data consist of nine data sets, five for one minute and four for two minutes. We focus on the data for one minute with stimulus ranging from 0.000 to 1.000 ug/ml. Our results show that about 70% of the particles are in clusters and the mean of the biggest cluster size is 9. For comparison purposes we pick a cluster distance of 70nm and observe that cluster increases from 71% to 96%, and the biggest clusters size goes from 6 to 60. Bigger clusters are less mobile and don't have a strong relationship with signaling or degranulation [3]. Since the biological data is very noisy, the number of particles per image varies between 72 and 654, we define the cluster ratio rho, which measures how much the biological data clusters compared to the random data. The cluster ratio increases as the stimulus increases. Our results can be use to better quantify the relationship between clustering, calcium release and degranulation.

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