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## Analysis of Golgi structure by imaging flow cytometry

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The Golgi complex of mammalian cells is a dynamic organelle which regulates the vesicles trafficking in the cell. Although cellular import and export of proteins require active changes of the Golgi membranes these are not accompanied by changes in the general Golgi's structure. On the other hand, other cellular processes require dramatic fragmentation of the Golgi complex. These processes include mitosis in which the Golgi complex is divided between the two daughter cells, in apoptosis in which the Golgi complex undergoes sequential degradation and migration in which partial Golgi fragmentation is required for a proper directionality of the moving cells. Currently, quantification of changes in Golgi structure is performed by counting 100-500 stain cells using basic immunofluorescence. This is followed by a manual classification of the Golgi structure in these cells by subjective assessments. Here we describe the use ofthe unique imaging and analysis abilities of the ImageStream X, a high throughput imaging flow cytometer for a non subjective quantification of Golgi fragmentation. This method provides a way to analyze the changes in Golgi structure in an automated, quantitative and non biased manner. Furthermore, this method enables rapid and accurate way to analyze more than 104 cells per sample. All these above features are exemplified by structural Golgi changes during different mitotic phases. Using these parameters, we provide a robust, non biased statistically powerful analysis of the Golgi structure that can be used in future cell cycle and apoptosis studies.

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