Luminol Electrochemiluminescence for the Analysis of Active Cholesterol at Plasma Membrane in Single Mammalian Cells Dechen Jiang, Guangzhong Ma

Key state labortorary of analytical chemistry for life science and School of Chemistry and Chemical Engineering, Nanjing University, Jiangsu, 210093, China

A luminol electrochemiluminescence assay was reported to analyze active cholesterol at plasma membrane in single mammalian cells. The cellular membrane cholesterol was activated by the exposure of the cells to low ionic strength buffer or the inhibition of intracellular acyl-coA:cholesterol acyltransferase (ACAT). The active membrane cholesterol was reacted with cholesterol oxidase in the solution to generate a peak concentration of hydrogen peroxide near the electrode surface, which induced a measurable luminol electrochemiluminescence. Further treatment of the active cells with mevastatin decreased the active membrane cholesterol resulting in a drop in luminance. No change in the intracellular calcium was observed in the presence of luminol and potential, which indicated that our analysis process might not interrupt the intracellular cholesterol trafficking. Single cell analysis was performed by placing a pinhole below the electrode so that only one cell was exposed to the photomultiplier tube (PMT). Twelve cells were analyzed individually and a large derivation on luminance was observed, which exhibited the cell heterogeneity on the membrane active cholesterol. The observation of cell heterogeneity on the membrane active cholesterol might provide a new insight on the study of intracellular cholesterol trafficking.