Electronic Supporting Information

On the mechanism of electrochemical vesicle cytometry: chromaffin cell vesicles and liposomes

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Abstract

The mechanism of mammalian vesicle rupture onto the surface of a polarized carbon fiber microelectrode during electrochemical vesicle cytometry is investigated. It appears that following adsorption to the surface of the polarized electrode, electroporation leads to formation of a pore at the interface between a vesicle and the electrode and this is shown to be potential dependent. The chemical cargo is then released through this pore to be oxidized at the electrode surface. This makes it possible to quantify contents as it restricts diffusion away from the electrode and coulometric oxidation takes place. Using a bottom up approach, lipid-only transmitter-loaded liposomes were used to mimic native vesicles and the rupture events occured much faster in comparison with native vesicles. Liposomes with added peptide in the membrane result in rupture events with duration slower than that of liposomes and faster in comparison to native vesicles. Diffusional models have been developed and suggest that the trend in pore size is dependent on

soft nanoparticle size and diffusion of the content in the nanometer vesicle. In addition, it appears that proteins form a barrier for the membrane to reach the electrode and need to move out of the way to allow close contact and electroporation. The protein dense core in vesicles matrixes is also important in the dynamics of the events in that it significantly slows diffusion through the vesicle.

Table SI-1. Diameter mean values and size distribution of liposomes and chromaffin vesicles obtained with Nanoparticle Tracking Analysis (NTA)

Sample	Mean (nm)	SD (nm)
Liposome control	328.3 ± 5.1	150.1 ± 3.0
Liposome with peptide	206.5 ± 6.5	78.5 ± 5.1
Chromaffin vesicle	339.8 ± 4.9	118.8 ± 3.5

SD – standard deviation calculated by the NTA software; numbers represents mean values ± standard deviation; *Liposome control*: *n* (measurements) = 4; *n* (sample preparations) = 2; *Liposome with peptide*: *n* (measurements) = 6; *n* (sample preparations) = 3, *Chromaffin vesicles*: *n* (measurements) = 23; (vesicle isolations) = 7.

$t \approx 45$ min $t \approx 75$ min $t \approx 105$ min
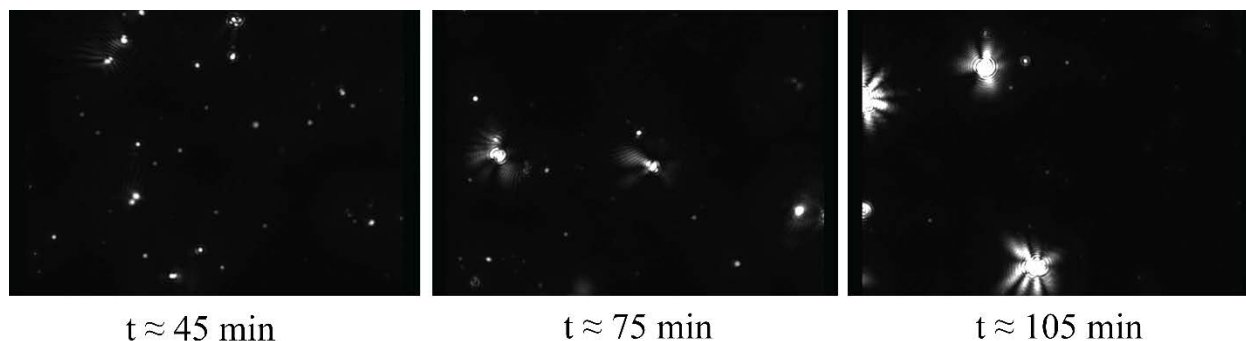
Figure SI-1. NTA video screenshots of liposome with peptide showing the aggregation over the course of time; t=0 is considered the time point when the last stage of the sample preparation was done. In Table SI-1 is shown the mean diameter of liposomes with peptide.