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OPTIMIZING PRECISION NANOPARTICLE DELIVERY FOR MAGNETO-MECHANICALLY-BASED CALCIUM MODULATION

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Abstract:
Modulating calcium influx events through mechanically actuated magnetic nanoparticles (MNPs) at the cell membrane or in the cytosol of neurons has been trending in the past few years. The mechanical force load is a critical parameter that can be fine-tuned through MNPs concentration and the incubation time. Commonly, MNPs get incubated for 24 h to improve the probability of neuronal uptake; however, critical cellular processes such as protein turnover also coincide during that time frame. Hence, we optimized the incubation time for MNPs to be below < 24 h while showing similar calcium response events in neurons. To achieve this goal, we recorded transient calcium fluorescence (Fluo-4 AM, 10x Leica DMI-8, 4 fps) in dissociated primary rat cortical neurons (at 14 days *in vitro*) with starch-amine functionalized MNPs (incubated for 4 h and 24 h) over 12 min, where a magnetic field actuated 40-80 pN forces on the MNPs from 4 min to 8 min. We decomposed the mean somatic calcium signals into a resting concentration (lower envelope) and calcium events (magnitude and frequency) and correlated them with the Sørenson-Dice. All data was compared with the Mann-Whitney U test (N = 4 cultures, Single neuron cells: control = 2262, 4 h = 2424, 24 = 1062). Our results showed that both incubation times provided increases in resting concentration and calcium event frequency and decreased event magnitude and synchronous correlation in comparison to the control. Although similar, we observed that the 4 h incubation time increased the calcium event response with higher frequency and decreased event magnitudes and synchronous correlation when compared to the 24 h incubation. Altogether, our results show that shorter MNPs incubation times can make calcium influx more precise and potentially overcome adverse effects stemming from protein turnover.

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