Neuroscience

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Quantification of Spine Alterations with Imaris

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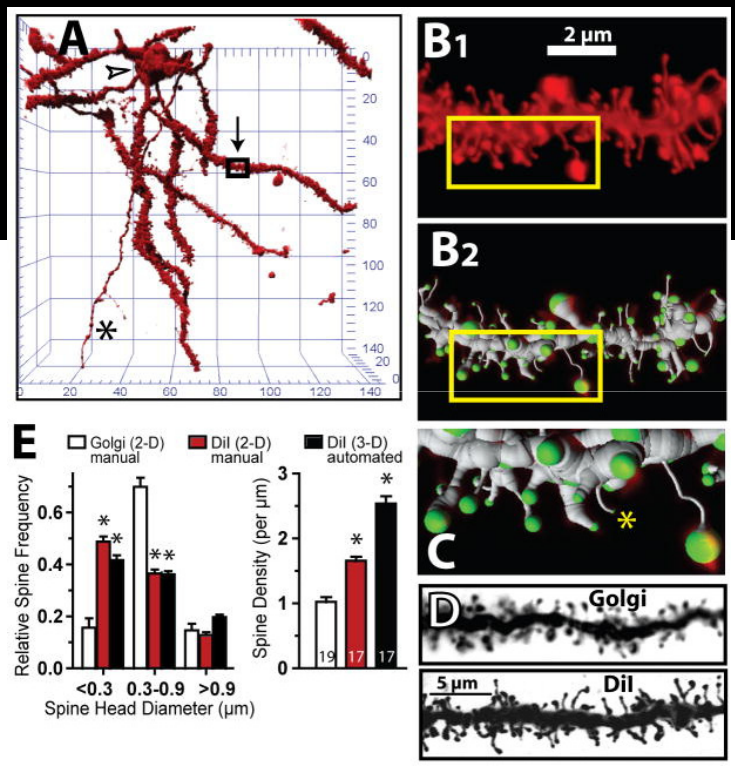
Effects of cocaine withdrawal on spine plasticity



Rendering and quantification of a confocal image of a diolistically-labeled, DiI filled dendrite segment from an MSN. **A)** DiI-filled neuron in the NAcore. Arrow and box identify segment of dendrite rendered in panels B-D. Grid= µm. *= axon; open arrowhead= soma. **B)** Panel B1 shows a confocal image of the dendrite segment indicated in panel A, and B2 shows filling of the dendrite using the Filament module in the IMARIS software package. Green identifies the end segment of each spine where the estimate of d_h was made. **C)** Illustrates higher magnification of the boxed region in panel B. *= thin filapodia-like spines ($d_h \leq 0.2$ µm). D) Representative light micrograph of Golgi-stained dendrite and confocal image of DiI-labeled dendrite. **E)** Comparison of spine density and d_h between Golgi impregnated and DiI-labeled dendritic spines. The number of neurons quantified is shown in the bars in the right panel. *p< 0.05, comparing DiI with Golgi staining using a Bonferroni post hoc.

Changes in cellular neuroplasticity may underlie behavioral changes after chronic and acute cocaine exposure. Dendrite spine morphology is thought to regulate synaptic strength therefore playing a critical role in synaptic plasticity.

Shen and colleagues have developed a method utilizing confocal imaging techniques and Imaris FilamentTracer software to detect changes in spine head diameter and spine density. Together with changes in post-synaptic density protein levels and field amplitude measurements, Shen et al used automated spine classification results to show differences in synaptic strength in cocaine treated animals versus saline treated animals. They also used EM results to validate that the automated method using FilamentTracer is more accurate then 2D golgi-staining results.

To measure changes in spine density and spine head diameter, medium spiny neurons from the nucleus accumbens core were labeled with DiI and the entire profile of the neuron was acquired with a Zeiss LSM 510 confocal. FilamentTracer was used to first reconstruct the dendrites, and then to identify the spines. Once Spines were identified MeasurementPro was used to quantify spine density and head diameter. These measurements were taken at various time points in cocaine treated and saline treated animals to determine if there were changes in synaptic strength. The automated FilamentTracer calculations were compared to manual calculations to verify that the automated measurements were successful.

Overall, Shen and his colleagues showed that by using the automated spine detection in FilmentTracer, changes in synaptic plasticity induced by cocaine treatment could be quantified and the effects of treatment and withdrawal were identified. The changes demonstrated here may have implications for changes in behavior associated with chronic cocaine exposure and withdrawal.

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Altered Dendritic Spine Plasticity in Cocaine Withdrawn Rats. Shen, Toda, Moussawi, Bouknight, Zahm and Kalivas.