

## IMMUNOFLUORESCENT STAINING REVEALS SYNAPTIC LOCATIONS IN INSECT LARVAE

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**KEY WORDS:** Laser Scanning Microscopy, 3d Reconstruction, Development, Locust, Synapse

In order to reveal the location of synaptic sites in the locust *Schistocerca gregaria*, I labelled specific synaptic proteins. This enabled me to study changes in the location of synapses during postembryonic development.

I used primary antibodies directed against two synaptic proteins of the fruit fly, *Drosophila*: synapsin [1] (a molecule implicated in the formation of the reserve pool of synaptic vesicles), and synaptotagmin [2] (a molecule of the vesicle membrane thought to be a calcium sensor of the synapse). I applied these antibodies to parts of the locust brain that contain the ocellar tracts (parts of the visual system) and visualised them using fluorescent secondary antibodies. Fluorescence was detected using a Leica confocal microscope, and stacks of optical sections were 3d reconstructed using Amira software.

The results (Fig.1) show that both antigens accumulate at synaptic sites. The distribution of these sites within the ocellar tracts changes during postembryonic development. In instars one and two, they are evenly distributed throughout the ocellar tracts, whereas in later instars and adult locusts, most synapses are concentrated in proximal areas of the tracts.

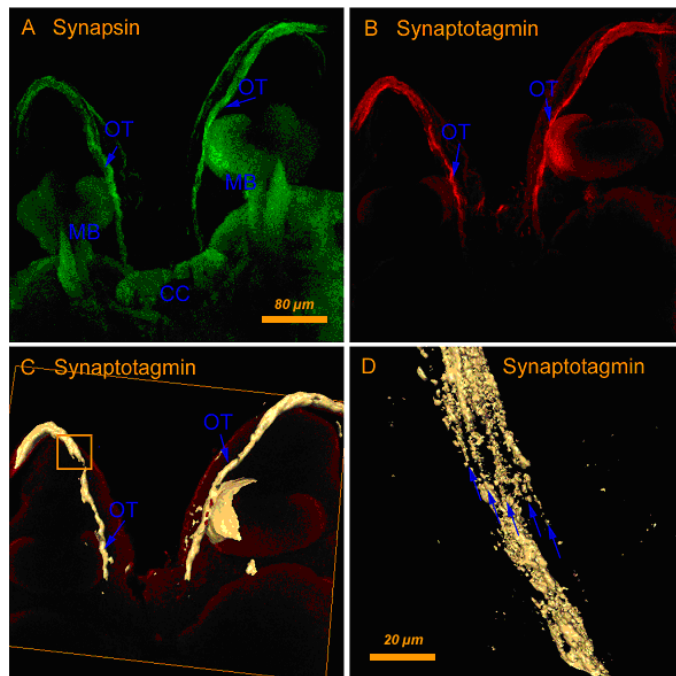


Figure 1: Synaptic proteins in the central nervous system of a second instar locust. A and B stacks of confocal laser scanning micrographs. A: synapsin-immunoreactivity, B: synaptotagmin-immunoreactivity. C: 3d reconstruction of 30 optical sections of synaptotagmin immunoreactivity scanned at low resolution. D: 3d reconstruction of 60 optical sections of synaptotagmin-immunoreactivity at higher resolution. Several strands containing clusters of vesicles are visible (arrows). OT ocellar tract, CC central complex, MB mushroom body.

### REFERENCES:

- [1] BR Klagges, G Heimbeck, TA Godenschwege, A Hofbauer, GO Pflugfelder, R Reifegerste, D Reisch, M Schaupp, S Buchner, and E Buchner, "Invertebrate synapsins: a single gene codes for several isoforms in *Drosophila*". *J Neurosci*, **16**, 3154-65 (1996).
- [2] JT Littleton, HJ Bellen, and MS Perin, "Expression of synaptotagmin in *Drosophila* reveals transport and localization of synaptic vesicles to the synapse", *Development*, **118**, 1077-88 (1993).

Funded by the Austrian Science Foundation (Project P17874-B05)