Towards visualizing mRNA transport

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Synaptic plasticity is thought to be important for learning and memory processes, and is ensured by local translation of a selection of mRNAs that have been previously targeted to the synapse.
The aim of this project is to visualize mRNA transport in live *Drosophila* embryos and larvae. Towards this aim we are adapting a yeast system developed by Bertrand et al. (Molecular Cell, vol.2, 437-445, 1998) for use in *Drosophila*. This system uses two different constructs: a Green Fluorescent Protein (GFP) fused to the capsid protein of the MS2 bacteriophage and an artificial reporter mRNA containing 3 modular components: (1) a *LacZ* gene to control for mRNA translation, (2) stem-loop structures that are recognized by the capsid protein present in the GFP-fusion protein. (3) RNA-targeting sequences that direct the reporter mRNA to the synapse. To date the targeting sequences that direct mRNAs to the synapse in *Drosophila* embryos and larvae have not been elucidated. However in most transported mRNAs this sorting process relies on targeting signals that are located in the 3'UTR of these mRNAs. We therefore made 3 different reporter mRNA constructs containing the 3'UTRs from mRNAs encoding the *Drosophila* homologues of either Ca2+/calmodulin dependent protein kinase, the fragile X metal retardation protein, the glutamate receptor IIa and a control construct containing a 3'UTR that lacks targeting information. We will shortly be investigating whether our reporter constructs are transported to the processes of ML-DmBG2-c2 cells (a CNS derived *Drosophila* cell-line) and to the synapses of the transgenic fly lines expressing these constructs. In addition we are generating 3 separate fly-lines that express three slightly different GFP-fusion proteins. In larvae and embryos expressing both a targeted reporter mRNA and the GFP-fusion protein we hope to be able to visualize the trafficking of GFP labeled reporter mRNAs.

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