Overlapping in two cellular ensembles in the hippocampal CA1 region is important for behavioral tagging

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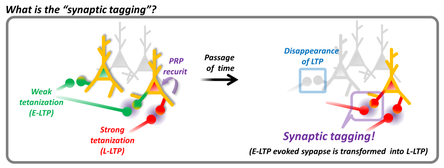
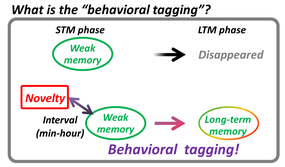
1. Abstract

Behavioral tagging is one form of interactions between two independent memories in which short-term memory (STM) is transformed into a long-term memory (LTM) when the STM-training and another novel experience occur at short time interval. Previous studies showed that behavioral tagging resembles synaptic tagging in the interactions between short-term and long-term events, their requirement for de novo protein synthesis, and time windows between two events (~hours), suggesting that synaptic tagging mechanism may underlie the behavioral tagging.
We previously showed that the number of neurons activated at both novel object recognition (NOR) and novel experience, denoted here overlapping neurons, increased in the CA1 region when the behavioral tagging was successfully achieved. In this study, we examined the role of the overlapping cell ensemble in behavioral tagging. Arc catFISH analysis showed that the increase in the number of overlapping neurons, which in this case were activated during both the retrieval of NOR and the exposure to the initial context, was also observed. Importantly, optogenetic experiment with lentiviral vector expressing ArchT3.0 and cfos-tTA transgenic mice showed that optical silencing of the cell ensemble in the hippocampal CA1 region related to the novel experience, but not different experience, led to an impairment in the NOR memory retrieval. These results suggest that inputs from two different pieces of information, NOR and novel experience, converged on the same neuronal ensemble in the hippocampal CA1 region after achievement of behavioral tagging. Together, our results suggest that overlapping in two cellular ensembles in the hippocampal CA1 region is required for behavioral tagging and synaptic tagging mechanism may be involved in the behavioral tagging.

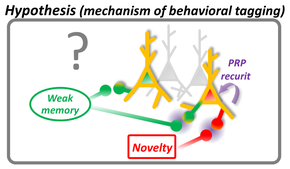
What is the "synaptic tagging"?



What is the "behavioral tagging"?



Hypothesis (mechanism of behavioral tagging)



Behavioral tagging resembles synaptic tagging in the interactions between short-term and long-term events.

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Synaptic tagging mechanism may underlie the behavioral tagging.

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It is expected that these two units of information may be processed in the same neuronal circuit.

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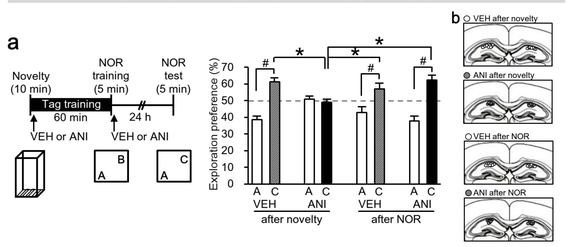
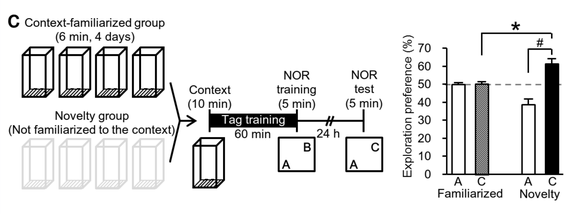
However, there is no evidence about the dynamics of cell assemblies that correspond to these two independent information when the behavioral tagging is achieved.

3. Achievement of behavioral tagging required novelty and hippocampal de novo protein synthesis.

a-b. Anisomycin microinjection into hippocampus immediately after novelty impaired NOR-LTM.

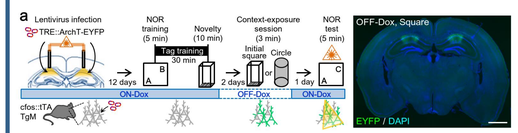


c. Familiarization to novel context inhibited behavioral tagging.

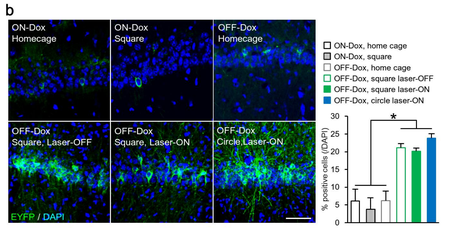


5. Optical silencing of the CA1 cell ensemble related to the original place experience, but not to different experience, led to an impairment in the NOR memory retrieval and a decrease in the ratio of Egr1/zif268 expressing cells in ArchT3.0+light ON cells.

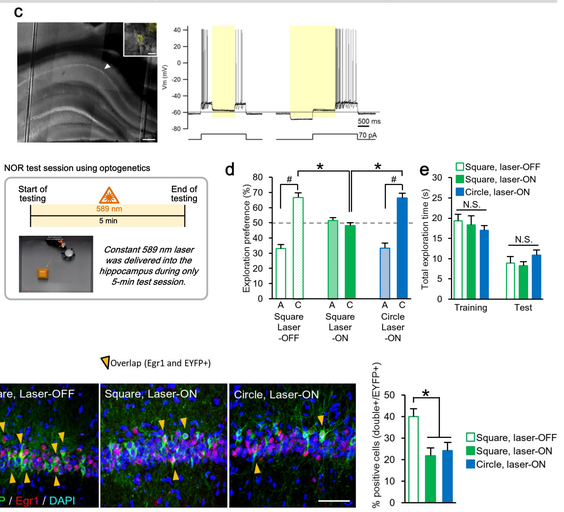
a. Experimental design using Tettag(cfos-tTA) mice, lentivirus vector and optogenetics



b. ArchT3.0-EYFP expression was induced in OFF-Dox- and context-exposure-dependent manners



c-f. Optogenetic experiment followed by immunohistochemistry

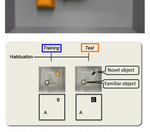


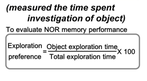
2. NOR-STM was transformed into LTM by exposing mice to a novel place at a short interval of ~1 hour.

Experimental design of novel object recognition (NOR) test

Following objects are used to perform NOR test.

Stamp Elephant Ochoko



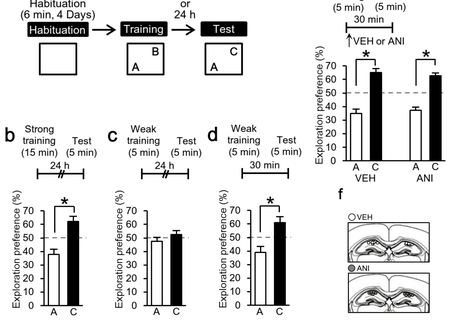


(measured the time spent investigation of object)

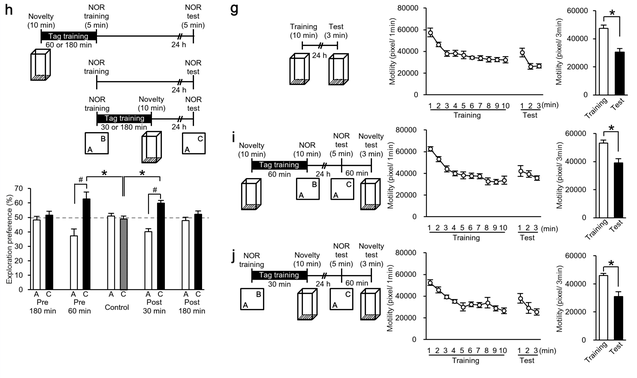
To evaluate NOR memory performance

Exploration preference = Object exploration time / Total exploration time × 100

a-f. Fifteen min but not five min NOR-training formed NOR-LTM, and five min training formed NOR-STM in a hippocampal protein synthesis independent manner.

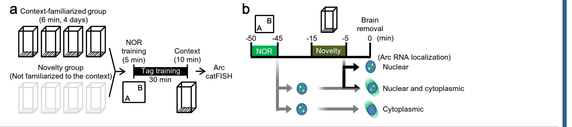


h-j. Establishment of behavioral tagging paradigm using NOR and place memory test which has time window

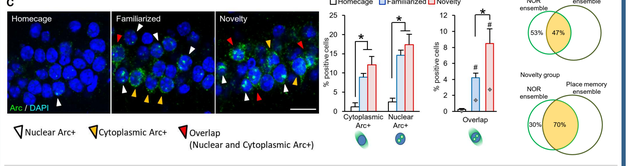


4. Novelty group showed an increase in the number of overlapping neurons in the hippocampal CA1 region compared with the control groups.

a. Experimental design of Arc mRNA catFISH and dynamics of Arc mRNA after its transcription

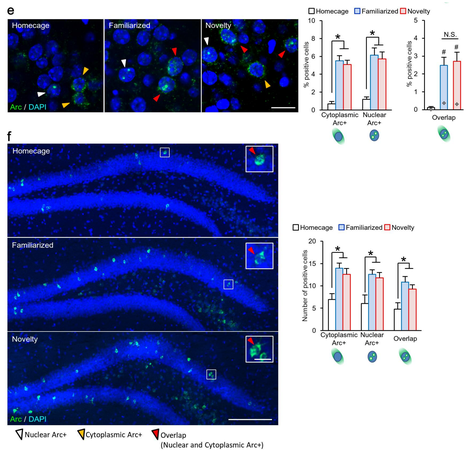


c-d. Results of CA1 region and Venn diagram of ratio of overlapping



Nuclear Arc+ Cytoplasmic Arc+ Overlap (Nuclear and Cytoplasmic Arc+)

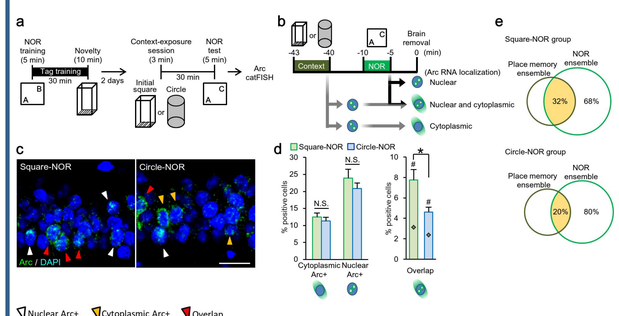
e-f. Results of CA3 and DG regions: the number of overlapping neurons were comparable between familiarized and non-familiarized groups.



Nuclear Arc+ Cytoplasmic Arc+ Overlap (Nuclear and Cytoplasmic Arc+)

6. Arc catFISH analysis showed that the ratio of overlapping neurons were also larger compared with an control group during both NOR and context exposure.

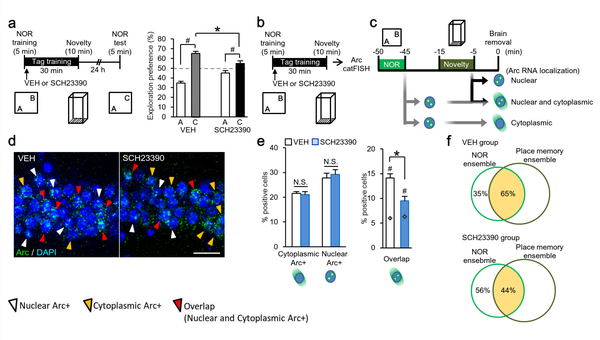
a-d. Experimental design and results of Arc mRNA catFISH



Nuclear Arc+ Cytoplasmic Arc+ Overlap (Nuclear and Cytoplasmic Arc+)

7. Blockade of dopamine D1/D5 receptor during behavioral tagging training affected NOR-LTM and ratio of overlapping neurons.

a-f. Experimental design using dopamine receptor antagonist SCH23390 and results of Arc mRNA catFISH



Nuclear Arc+ Cytoplasmic Arc+ Overlap (Nuclear and Cytoplasmic Arc+)

7. Conclusion

NOR-STM is transformed into LTM by exposing mice to a novel place (Novelty) at a short interval of ~1h in a protein synthesis-dependent manner.

Arc catFISH analysis showed that... the number of neurons activated at both NOR and novel experience, overlapping neurons, increases in the CA1 region when the behavioral tagging was successfully achieved.

Optogenetic experiment with lentiviral vector expressing ArchT3.0 and cfos-tTA transgenic mice showed that... optical silencing of the cell ensemble in the hippocampal CA1 region related to the novel experience, but not different experience, led to an impairment in the NOR memory retrieval.

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These results suggest that... inputs from two different pieces of information, NOR and novel experience, converged on the same neuronal ensemble in the hippocampal CA1 region during behavioral tagging.

Together, our results suggest that ***synaptic tagging mechanism may be involved in the behavioral tagging.***

Hypothesis (mechanism of behavioral tagging)