博士論文 (要約)

Dentate granule cell activity during fear memory extinction in freely moving mice

（自由行動下における恐怖記憶消去課題中のマウス海馬歯状回顆粒細胞の活動）

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The hippocampal dentate gyrus (DG) is known to be important for the formation, recall and extinction of episodic memories. The DG of mouse consists of 500 thousand principal cells called granule cells (GCs), which is about 20 times larger than the number of presynaptic neurons in the entorhinal cortex. Because of this large divergence, GCs are considered to assign large amount of information into different neuronal populations with a small overlap between them, which could underlie the accurate recall and modulation of a specific episode among large number of stored memories.

An indispensable feature contributing to the memory processing in the DG is the persistence of neurogenesis in adult animals. After the discovery, substantial evidence demonstrates that neurogenesis persists throughout adulthood in the subgranular zone (SGZ) of the DG in various mammals including humans. About one thousand adult-born GCs (abGCs), which corresponds to 0.2 % of the total number of GCs, are born and integrated every day in the DG circuit of the mouse. Newly generated GCs are reported to exhibit higher synaptic plasticity compared to surrounding mature GCs (mGCs) between four and six weeks after their generation.

Inclusion of highly plastic neurons into neural circuits can provide new substrates for memory. Consistent with this notion, increase of hippocampal neurogenesis has been shown to improve memory formation, whereas its suppression disrupts new memory formation, social behavior and promotes depressive-like responses. Furthermore, the specific time window of

increased plasticity in abGC populations may help to “time-stamp” memories with different timings, supporting the process of differentiating episodic memories by the DG. It has been demonstrated that the blockade of synaptic transmission between abGCs and other neurons disrupts the separation of several episodic memories. The rewiring of existing neural circuits by adult neurogenesis also controls the long-term stability of acquired memories. This notion is based on the results that increasing hippocampal neurogenesis promotes forgetting of hippocampus-dependent memories including fear memory. Moreover, inhibiting adult neurogenesis in the hippocampus induced severe deficits in spatial memory extinction and fear memory extinction.

Studies of monitoring activities of mGCs and abGCs in behaving animals are indispensable for understanding how neurons encode information in real-time. It is known that dentate GCs exhibit spatial specific activation, called place cell activity. In addition, dentate GC activity and the local field potential of the dentate gyrus are modulated by various external stimuli such as reward consumption, and tone discrimination, suggesting the possibility that accurate processing of episodic memories by GCs could involve various external information. However, in spite of all the studies that recorded the real-time activity of mGCs and abGCs in order to elucidate their role in episodic fear memory processes, currently there are no reports that monitor the dynamics of mGCs as well as abGC during retrieval and extinction of episodic fear memories and determine the qualitative differences in activity between them. Furthermore, there is no report identifying the

changes in neuronal activity of both neuronal groups that correspond to the behavioral adaptations seen through the experience of episodic fear memories.

In order to address whether the abGCs act differently from mGCs, and if so, how they act throughout the behavioral changes, I studied the activity of mature and adult-born GCs in freely moving mice for the first time. I recorded their Ca^{2+} transients with a miniaturized microendoscope during cue-dependent retrieval and extinction of fear memory, and related the activities of mGCs and abGCs with the changes in behavior mice experienced through sessions.

I found that only mice that were fear conditioned showed an increase in the number of active mGCs during cued fear retrieval-extinction. Moreover, the activity-dependent induction of Fos immediate early gene by cued fear retrieval-extinction was higher in conditioned mice than unconditioned controls. Regarding the activity rate of mGCs, I found that neurons from unconditioned mice showed a decay in the Ca^{2+} event rate through the retrieval-extinction sessions, while neurons from conditioned mice exhibited a transient increase in the event rate and a change in the active population of mGCs along with extinction. I also observed that mGCs in conditioned mice fired more selectively to the tone than those in unconditioned mice.

In contrast, I saw opposite phenomena when analyzing the activity of abGCs. While in unconditioned mice the size of the active abGCs ensemble was constant through all experimental days, in conditioned mice there was a decrease in the number of active abGCs after fear

conditioning that persisted through the retrieval-extinction sessions. Additionally, a part of abGC population showed Ca^{2+} transients more frequently and periodically. Those active and periodic abGCs in conditioned mice showed similar activity to those in unconditioned mice. In contrast, abGCs with low firing frequency showed a switch of active population during the first memory extinction session. Despite those fear conditioning dependent changes, there were no clear tone cue-related effects in the firing rate and cue selectivity of abGCs.

Finally, I found that chemogenetic inhibition of mGCs not only weakened the cue-dependent retrieval of fear memory but also compromised the effectiveness of the extinction process, whereas generalized ablation of GCs increased cue-dependent freezing and disrupted cued extinction. Taken together, these results suggest that GCs are necessary for the retrieval and extinction of a cued fear memory and that mGCs and abGCs utilize distinct mechanisms for the processing of episodic memories.