Bolding K, Biedenkapp J (2006) What Can Immediate-Early Gene Expression Tell Us about Spatial Memory Retrieval? J. Neurosci 26:1659-1660

Gusev PA, Cui C, Alkon DL, Gubin AN (2005) Topography of Arc/Arg3.1 mRNA expression in the dorsal and ventral hippocampus induced by recent and remote spatial memory recall: dissociation of CA3 and CA1 activation. J Neurosci 25:9384-9397.

Dear Journal of Neuroscience,

We very appreciate the interesting and careful review of our article. However, we disagree with some key statements of the review. Kevin Bolding and Joseph Biedenkapp claim that retention of the platform location reported in our article "was quite poor during both the recent and remote probe trials" based on search time in the target quadrant, and that the behavioral data make more difficult to interpret the meaning of Arc gene activation. In addition, the authors note that there is evidence suggesting that systems consolidation does not occur in Morris water maze task.

We did not use the dwell time as a measurement of memory and made it clear in the *Methods*, p.9386: "Navigation skills were evaluated based on the number of "target area" (12.5x12.5 cm) crossings and latency to the first target crossing". Based on our experience and the water maze literature, we reasoned that the number of crossings over platform location and latency for the first platform crossing during the probe test are the most appropriate and the most sensitive parameters to assess rats' spatial navigation in the water maze protocol that we use. Dwell time in quadrant analysis may not be such a sensitive and specific parameter for spatial learning because it has recently been shown to be associated with the procedural aspect of the water maze task not with navigation ability *per se* while the latency for the first platform crossing would be a more appropriate parameter to evaluate navigation (Micheau et al., 2004).

The distribution of quadrant center crossings between the target and non-target centers indicated a significant increase in the number of crossings for the target location in our study. This

analysis has been adopted from the original works that introduced the water maze learning paradigm (Morris, 1984), and from recent publications (Ramirez-Amaya et al., 2001). Swimming controls clearly lack such a spatial bias in quadrant center crossings. Moreover, the number of crossings over the target center was significantly higher in water-maze trained rats as compared to swimming controls both at 24-hr and 1-mo delays; and first crossing latency was not different for rats performed on recently and remotely acquired tasks. These data indicate robust spatial learning and no spatial memory decline over a 1-mo period. Our behavioral study of a reminding effect on long-term memory performance indicated that during a probe test, the number of platform location crossings in the tank was a more precise and sensitive parameter as compared to quadrant analysis. We did not see any effect of reminding on dwell time and distance while the number of platform crossings had significantly increased after previous reminding trials that were given a day before a remote memory test (Gusev, unpublished observation). Thus, despite the fact that dwell time in the target quadrant in some probe tests was not significantly increased in our article, the data demonstrate that rats' performance become spatially biased due to training as precisely shown by the number of platform location crossings and first latency crossing.

We also disagree with authors on the water maze memory consolidation issue. Although the effects of partial and complete hippocampal lesions on rats' performance on remote water maze task do not favor predictions that follow from both the standard and multiple trace memory models (Martin et al., 2005), a gene knockout study of long-term water maze memory does support the idea of some sort of systems level consolidation for this task (Frankland et al., 2001). Accordingly, as we suggested in the *Discussion* p.9394, "an analysis of Arc mRNA expression in the neocortex in necessary for a more definite conclusion." We did not analyze the number of activated neurons because we believe that a density analysis should better reflect the gradation of neuronal activity including synaptic. Once we

complete the analysis for the entire brain, the dynamics of memory underlying activity may not favor predictions of either model as well.

In conclusion, the goal of our article was to decipher the activity patterns of the hippocampus that could help to identify sparse enduring neuronal alterations underlying long-term spatial memory in the future studies.

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