Neuroepigenetics 2 (2015) 1–8

Contents lists available at ScienceDirect

Neuroepigenetics

journal homepage: www.elsevier.com/locate/nepig

Original research

DNA methylation regulates neurophysiological spatial representation in memory formation

Eric D. Roth [a,b], Tania L. Roth [a,b], Kelli M. Money [b], Sonda SenGupta [b], Dawn E. Eason [b], J. David Sweatt [b,*]

[a] Department of Psychological and Brian Sciences, University of Delaware, Newark, DE 19716
[b] Department of Neurobiology and Evelyn F. McKnight Brain Institute, University of Alabama at Birmingham, Birmingham, AL, 35294

ARTICLE INFO

Article history:
Received 29 December 2014
Received in revised form 9 March 2015
Accepted 10 March 2015

Keywords:
Memory
Spatial memory
Place cells
Epigenetic
Neuroepigenetics
DNA methylation
Bdnf
DNMT
Hippocampus

ABSTRACT

Epigenetic mechanisms including altered DNA methylation are critical for altered gene transcription subserving synaptic plasticity and the retention of learned behavior. Here, we tested the idea that one role for activity-dependent altered DNA methylation is stabilization of cognition-associated hippocampal place cell firing in response to novel place learning. We observed that a behavioral protocol (spatial exploration of a novel environment) known to induce hippocampal place cell remapping resulted in alterations of hippocampal *Bdnf* DNA methylation. Further studies using neurophysiological in vivo single-unit recordings revealed that pharmacological manipulations of DNA methylation decreased long-term but not short-term place field stability. Together, our data highlight a role for DNA methylation in regulating neurophysiological spatial representation and memory formation.

Introduction

De novo gene expression within the hippocampus has long been recognized for its necessary role in synaptic plasticity and the long-term retention of learned behavior. Given that memory formation requires experience-driven patterns of gene expression, this has led to the search for molecular mechanisms both adequately sensitive to environmental stimuli and capable of driving and maintaining transcription-dependent cellular changes. As originally proposed by Crick nearly 3 decades ago (Crick, 1984), studies over the past few years have implicated epigenetic mechanisms including DNA methylation as key mediators of memory formation and stabilization. For example, several studies indicate that histone acetylation, an epigenetic change that alters chromatin structure in a manner that promotes gene transcription, plays an important role in mediating hippocampal gene expression associated with contextual fear (Barrett et al., 2011; Miller et al., 2008; Oliveira et al., 2011), object recognition (Barrett et al., 2011; Oliveira et al., 2011), and spatial (Bousiges et al., 2010) memory formation. Methylation of histones in patterns permissive to gene transcription also supports the formation of contextual fear memory (Gupta et al., 2010; Gupta-Agarwal et al., 2012). Furthermore, consistent with the role of histone modifications in memory, disturbances in hippocampal histone acetylation have been associated with disruptions in hippocampal plasticity and memory capacity in aged rats and in animals with neuronal loss (Fischer et al., 2007; Peleg et al., 2010).

Similarly, studies have shown that active regulation of DNA methylation and demethylation within the hippocampus supports neural plasticity and memory formation. DNA methylation is an epigenetic mechanism typically associated with gene silencing, but studies suggest that it may also be associated with active gene transcription (Chahrour et al., 2008; Uchida et al., 2011; Yasui et al., 2007). Experience-driven changes in gene expression following contextual fear conditioning are associated with methylation of the *PP1* gene (Miller and Sweatt, 2007) and demethylation of the *reelin* (Miller and Sweatt, 2007) and *Bdnf* (Lubin et al., 2008; Mizuno et al., 2012) genes. Changes in DNA methylation and expression of hippocampal *Bdnf* are also associated with object recognition memory (Munoz et al., 2010) and the memory of traumatic experiences (Roth et al., 2011). As further evidence for the role of DNA methylation in memory processes, mice with disruptions in proteins associated with DNA methylation (including MeCP2 and DNMT1) show impairments in long-term potentiation and fear memory formation (Feng et al.,

* Corresponding author at: University of Alabama at Birmingham, Department of Neurobiology, 1825 University Blvd, SHEL 1010, Birmingham, AL 35294-2182. Tel.: +1 205 975 5196; fax: +1 205 975 5097.
E-mail address: dsweatt@uab.edu (J.D. Sweatt).

http://dx.doi.org/10.1016/j.nepig.2015.03.001

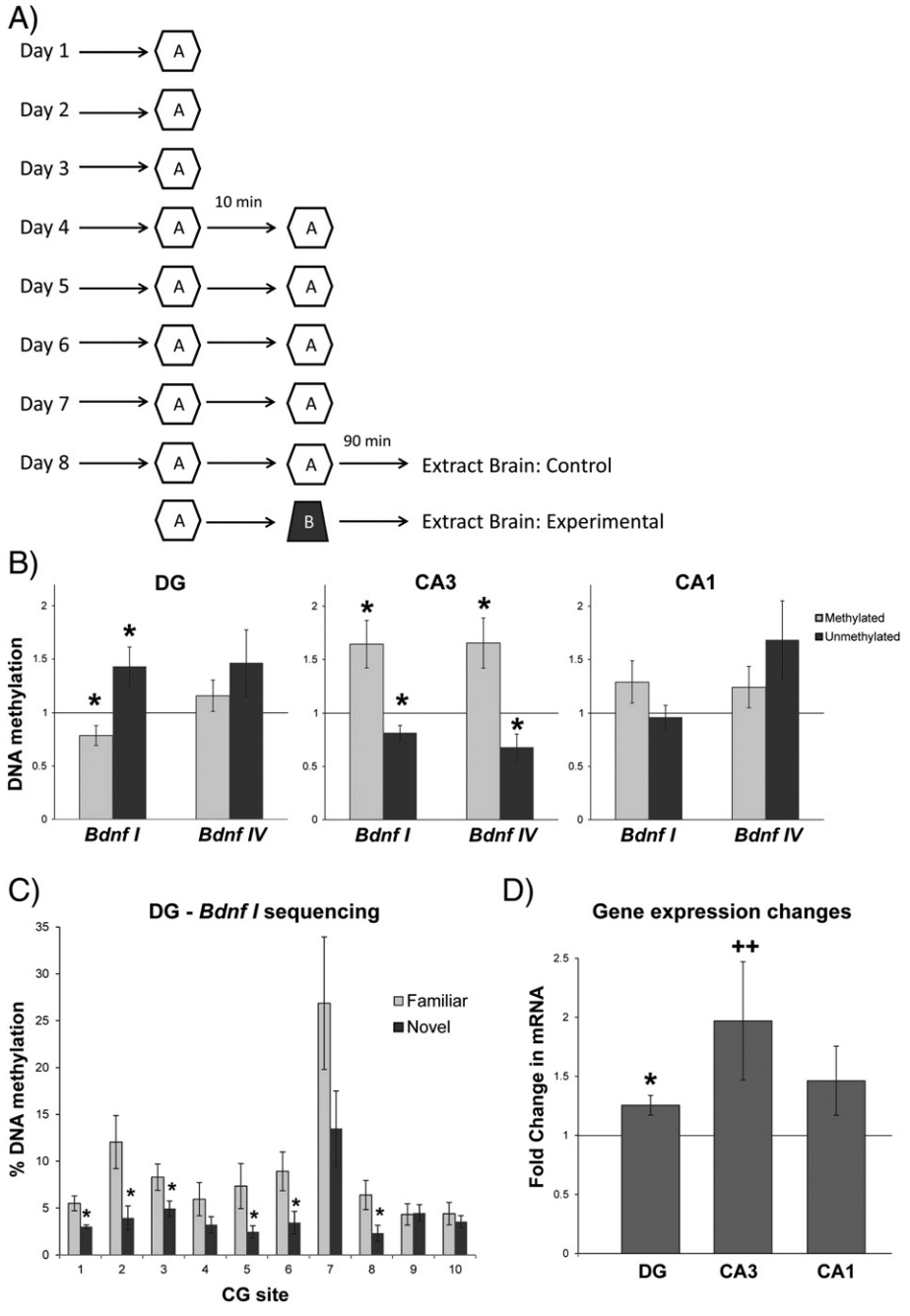

Fig. 1. (A) Graphic depicting the 8-day protocol used in experiment 1. For 8 days, rats performed a foraging task in a familiar environment (environment A). On days 4–8, rats performed the task in two 20-lap sessions separated by 10 minutes. On days 1–7, protocols were identical for the experimental and control group. On day 8, the second 20-lap session for the experimental group was conducted in a novel environment (environment B). Brains were extracted for biochemical analyses 90 minutes after the second foraging session on day 8. (B) Experience-induced alterations of hippocampal *Bdnf* gene DNA methylation. Levels of methylated and unmethylated DNA associated with the *Bdnf* gene in dentate (*Bdnf* I n = 10, *Bdnf* IV n = 11), CA3 (*Bdnf* I n = 10, *Bdnf* IV n = 11), and CA1 (*Bdnf* I n = 11, *Bdnf* IV n = 11) in novel-exposed versus control (familiar-exposed) rats. (C) Methylation analysis of individual CG dinucleotides associated with exon I in the dentate of control and novel-exposed rats (n = 10/group). (D) *Bdnf* mRNA (exon IX) levels in novel-exposed rats relative to familiar-exposed controls (DG n = 8, CA3 n = 9, CA1 n = 9). Error bars represent SEM; $^{*}P < .05$, $^{++}P = .0885$.

2010; Moretti et al., 2006; Nelson and Monteggia, 2011). Memory dysfunction in aged rats has also been linked to aberrant changes in hippocampal DNA methylation (Penner et al., 2011).

Despite this broad molecular and behavioral background, the means by which chemical modification of DNA might control cognition-associated neuronal circuit firing patterns has been left unaddressed. The general goal of the present study was to investigate the potential contribution of DNA methylation in spatial memory formation and the maintenance of neurophysiological spatial representations within the hippocampal neural circuit (Moser et al., 2008). A place cell is a hippocampal pyramidal neuron that encodes space by selectively increasing activity (firing rate) in a specific environmental location (place field). A collection of place cells with place fields in different locations may encode large environmental areas, providing the animal with a neurophysiological spatial representation of the environment (O'Keefe and Dostrovsky, 1971), which is commonly termed a *cognitive map* (Tolman, 1948) and believed to play a key role in navigation and spatial learning and memory. To