Essential Roles of Nutrient Factors in Learning and Memory

Satoshi KIDA
Graduate School of Agriculture and Life Sciences, The University of Tokyo, Tokyo, Japan

Summary Essential nutrient factors, including water- and fat-soluble vitamins, minerals, and essential amino acids, play an important role in brain function. We have investigated the roles of these nutrients in learning and memory in mice. Interestingly, we found that dietary deficiency of vitamin B1 or magnesium, inhibition of the vitamin A signaling pathway, and restricted intake of tryptophan impair hippocampus-dependent memory. Furthermore, magnesium deficiency causes neuroinflammation in the hippocampus. Conversely, dietary heat-killed Lactobacillus species enhance hippocampus-dependent memory. These results suggest that the nutrient factors investigated in our studies have strong influences on hippocampus-dependent memory performance.

Key Words learning, memory, hippocampus, vitamin B1, vitamin A, magnesium

We have investigated the roles of vitamin B1 and A, magnesium, gut bacteria, and tryptophan in learning and memory through deficient and restricted diets, mouse genetics, and dietary supplementation. I review our findings below.

1. Vitamin B1
Vitamin B1 (B1) was discovered by Umetaro Suzuki in 1913 (1). B1 functions as a coenzyme in the glucose metabolism system to generate ATP. Importantly, B1 is essential for the maintenance of nervous system function, especially in the brain, where glucose is used to produce ATP. Beriberi is a well-known B1-deficiency disease with motor deficits in the peripheral nervous system. Wernicke-Korsakoff syndrome (W-K syndrome) is also caused by B1 deficiency, and is associated with alcoholism, the prominent pathophysiology of which is memory impairment (2). Although the symptoms of beriberi are quickly rescued by B1 intake, the memory impairment observed in W-K syndrome is not. Interestingly, W-K syndrome is observed in alcoholism. Furthermore, B1 deficiency has also been proposed to be associated with the onset of Alzheimer’s disease (3). Unfortunately, the mechanism underlying the memory impairment caused by B1 deficiency remains unclear.

To address this issue, we have investigated the mechanism of memory impairment caused by B1 deficiency in mice (4). Mice were fed a B1-deficient diet for 10 d with daily administration of pyrithiamine, a B1 antagonist, and then fed a normal diet for 3 wk of recovery treatment (B1-deficient recovered mice). B1-deficient recovered mice showed normal motor coordination; however, they demonstrated impaired hippocampus-dependent memory formation, including spatial memory in the Morris water maze, social recognition memory, and contextual fear memory. These memory deficits persisted for at least 6 mo after recovery from B1 deficiency, indicating that, as in W-K syndrome, B1 deficiency induces memory deficits that cannot be recovered by feeding a normal diet in mice. In contrast, the amygdala-dependent memory formation of B1-deficient recovered mice was normal, suggesting that B1 deficiency impairs hippocampus-dependent memory. These findings indicate that B1-deficient recovered mice can serve as a model of W-K syndrome, with chronic memory impairment that is associated with impaired hippocampal function.

The hippocampus is a central region for episodic memory formation. Therefore, we analyzed the morphological damage caused by B1 deficiency to the hippocampus. Interestingly, a decrease in the number of neurons was observed in the hippocampus of mice recovering from B1 deficiency, suggesting that B1 deficiency reduces the number of hippocampal neurons. Subsequently, we analyzed the morphology of hippocampal neurons in Thy1-GFP mice, which express Green Fluorescence Protein (GFP) specifically in neurons, by applying B1 deficiency treatment, and observed a decrease in the dendritic spine density of neurons in the hippocampal dentate gyrus. These results suggest that B1 deficiency causes the degeneration of hippocampal neurons, thereby leading to the impairment of hippocampus-dependent memory (4).

2. Vitamin A
Vitamin A (retinol) is metabolized into all-trans retinoic acid (ATRA) and its isomer 9-cis retinoic acid (9-cis RA). ATRA is a ligand of retinoic acid receptors (RARs), while 9-cis RA is a ligand of RARs and retinoid X receptors (RXRs). RARs and RXRs exert their physiological effects through transcriptional regulation of target genes after binding with ATRA and 9cis-RA.

In the adult brain, most of the subtypes of RARs and RXRs are expressed in most of the regions of the brain, suggesting that the vitamin A-retinoic acid signaling pathway plays an important role in brain function. Unfortunately, the role of vitamin A in higher brain functions has not been clarified using a nutritional

E-mail: akida@g.ecc.u-tokyo.ac.jp
approach since vitamin A deficiency renders animals morbid, making it difficult to clarify the roles of this signaling pathway in brain function. However, studies in mice targeting genes for RARs and RXRs have enabled analysis of this pathway (5).

RARβ knockout or RARβ/RXR double gene-knockout mice showed impaired motor coordination, which makes it difficult to perform behavioral paradigms for learning and memory since these tests require normal basal motor skills at the behavioral level (5). Therefore, to clarify the role of the vitamin A signaling pathway in learning and memory, we induced a pseudo-vitamin A deficiency condition in a brain region-specific manner by generating conditional RAR mutant mice. We generated transgenic mice that inducibly express a dominant-negative mutant of RARα (dnRAR mice) only in the forebrain including the hippocampus using the tetracycline system, which allows for the time-specific induction of gene expression. In these transgenic mice, the inducible expression of dnRAR blocked the expression of a target gene of RAR in the hippocampus. Importantly, dnRAR mice showed impaired hippocampus-dependent memory formation, including spatial memory in the Morris water maze and social recognition memory. These observations suggest that the vitamin A signaling pathway plays an essential role in learning and memory.

Consistent with the observations in RARβ knockout or RARβ/RXR double knockout mice, inducible dnRAR-expressing mice showed impaired long-term potentiation (LTP) in the hippocampal CA1 region after a single tetanus stimulation. Interestingly, this impairment of LTP in dnRAR mice was rescued by four tetanus stimulations; dnRAR mice showed LTP levels comparable to those of wild-type mice. As mentioned above, dnRAR mice failed to form social recognition memory in massed training conditions; however, similar to the observations of LTP, they showed normal social recognition memory in spaced training conditions. Thus, these findings that the impaired hippocampal LTP and memory in dnRAR mice could be rescued by strong stimulation or training conditions, respectively, suggest a correlation between hippocampal neural plasticity and hippocampus-dependent memory, raising the possibility that the vitamin A signaling pathway regulates memory performance by controlling hippocampal neural plasticity (6).

3. Magnesium

Magnesium acts as a co-factor for more than 300 enzymes and is an essential mineral in a variety of tissues. Minerals are required for the regulation of ion channels and therefore have a significant impact on brain function. N-methyl-D-aspartic (NMDA)-type glutamate receptors act as voltage-gated calcium channels and activate intracellular calcium signaling pathways. Mg²⁺ contributes to the “Mg²⁺ block” of NMDA-type glutamate receptors and allows calcium ion influx in response to depolarization in a voltage-dependent manner. Importantly, previous studies suggested that elevated concentrations of magnesium ions in the brain enhance learning and memory (7), while magnesium deficiency leads to memory impairment (8).

We analyzed the effects of dietary magnesium deficiency in mice using a behavioral battery similar to that of B1-deficient recovered mice (9). No abnormalities in amygdala-dependent memory or locomotor activity were observed in mice with dietary magnesium deficiency for 5 wk. However, these mice showed deficits in hippocampus-dependent spatial, contextual fear, and social recognition memories. However, no abnormalities in spine density or morphology were observed in the hippocampus, amygdala, or prefrontal cortex of these Mg²⁺-deficient mice. Thus, although magnesium deficiency does not affect the morphology of hippocampal neurons, unlike B1 deficiency, it leads to impaired hippocampus-dependent memory, as in the case of B1 deficiency.

To understand the mechanisms of memory impairment caused by magnesium deficiency, we performed RNA-sequencing analysis using next-generation sequencing followed by gene ontology analysis and found that magnesium deficiency increased the expression of a set of inflammation-related genes. These observations suggest that magnesium deficiency causes neuroinflammation in the brain, which may cause memory impairments (10).

4. Lactobacillus species

Recently, gut microbes have been considered to be important for the maintenance and/or enhancement of brain function including memory performance (11). We examined the effects of feeding dead Lactobacillus to mice as a diet for 1 mo (12), and found that mice fed heat-killed Lactobacillus brevis SBC8803 showed an improvement in hippocampus-dependent social recognition memory. Additionally, adult neurogenesis in the hippocampal dentate gyrus, which is continuously generated by the intake of Lactobacillus. There was also an improvement in hippocampus-dependent memory when neurogenesis was enhanced artificially by the administration of memantine, an antagonist of NMDA-type glutamate receptors (13). Therefore, dead L. brevis SBC8803 may enhance memory performance by promoting adult hippocampal neurogenesis and could function as a dietary memory enhancer.

5. Tryptophan

The essential amino acid tryptophan (TRP) is a precursor of the neurotransmitter serotonin. TRP is metabolized to serotonin via 5-hydroxytryptophan in a two-step enzymatic reaction by tryptophan hydroxylase and aromatic L-amino acid decarboxylase. Serotonin is involved in the regulation of a wide range of brain functions, including learning and memory, appetite, sleep, and especially emotional behaviors such as anger, aggression, and anxiety. TRP intake. TRP concentrations in the brain, and serotonin levels positively correlated since TRP concentrations in the brain do not
reach saturation in the metabolic enzyme reactions (14). Thus, TRP intake determines brain serotonin levels and indirectly influences serotonin action.

To clarify the effects of insufficient TRP intake on brain functions, we examined mice fed a mildly restricted diet (low-TRP diet) with 35% less TRP (15, 16). These mice showed normal weight even after 6 mo of restricted intake. Analysis of a battery of behavioral tests showed that TRP-restricted mice exhibited normal social recognition and spatial memories. In contrast, they demonstrated impaired hippocampus-dependent contextual fear memory (15). Additionally, these mice showed increased locomotor activity in an open field test (novel environment) and increased mobility in a forced swimming test, suggesting that restricted TRP intake leads to manic-like behavior; that is, these mice displayed abnormalities in emotional behavior (16). Therefore, the impairment of contextual fear memory induced by restricted TRP intake may reflect abnormal emotional behaviors.

In summary, vitamin B1 and magnesium deficiency, inhibition of the vitamin A signaling pathway, and restricted tryptophan intake impaired hippocampus-dependent memory. Conversely, Lactobacillus intake enhanced hippocampus-dependent memory. Thus, we found that essential nutrient factors show strong influences on hippocampus-dependent memory performance.

Disclosure of state of COI

No conflicts of interest to be declared.

Acknowledgments

SK was supported by Grant-in-Aids for Scientific Research (A) (15H02488, 18H03944, 19H01047) and (B) (23300120 and 20380078), and Challenging Exploratory Research (20K21265); Grant-in-Aid for Scientific Research on Innovative Areas (Research in a Proposital Research area) (24116008, 24116001, 15H02488, 20380078), and Challenging Exploratory Research (A) (15H02488, 18H03944, 19H01047).

REFERENCES


