IMARS
HIGHLIGHTS

Research Commentaries for the Members of The International Maillard Reaction Society

A Non-profit Research and Education Organization in Biomedicine and Food Science

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IMARS
Established in 2005, the International Maillard Reaction Society gathers researchers and clinicians involved in the field of carbonyl reactions in foods, biology and medicine. It promotes research on Maillard Reaction and protein glycation and their numerous applications. It also organizes regular international congresses on the same theme, in addition to those that have been taken place since 1979.
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Editorial comments

In this issue, I am delighted to publish two articles submitted by the Japanese Maillard Reaction Society (JMARS) members.

We had the 29th JMARS meeting in Sendai, Miyagi, Japan, on Oct 27-28, 2019. The first article entitled "Dicarbonyl compounds are generated from auto-oxidation of glucose" is written by **Dr. Akari Sugawa** at the Department of Life Sciences, Graduate School of Bioscience, Tokai University, Kumamoto, Japan, who got the Young Investigator Award of the 29th JMARS meeting. Under the supervision of Prof. Ryoji Nagai, she proposes that glucosone generated from the oxidation of glucose modifies proteins, and thereby forms the AGEs.

The second article entitled "The role of vitamin B6 in schizophrenia" is written by **Dr. Kazuya Toriumi** at Project for Schizophrenia Research, Department of Psychiatry and Behavioral Sciences, Tokyo Metropolitan Institute of Medical Science, Tokyo, Japan, who got the Poster Presentation Award of the 29th JMARS meeting. Under the supervision of Prof. Makoto Arai, he demonstrates the pathogenic impact of vitamin B6 deficiency in schizophrenia and the possibility of a therapeutic approach based on vitamin B6 supplements.

I sincerely thank their significant contributions to the IMARS Highlights and expect their further development on glycation research. The IMARS Highlights editors always look forward to the submission of your articles related to glycation research in the field of food and medical sciences, any comment to the articles published in IMARS Highlights.

Lastly, we are trying to head off the coronavirus crisis. I hope all of us overcome this difficult time very soon.

Please stay safe, and with best wishes,

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Dicarbonyl compounds are generated from auto-oxidation of glucose

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The non-enzymatic reaction between proteins and reducing sugars, called the Maillard reaction, was first reported in the field of food chemistry more than 100 years ago. This reaction also progresses *in vivo* and results in the modification of protein functions **(1)**. The modification of amino residues with glycation induces the conformational change of 3D structures of proteins **(2)**. In addition, the Maillard reaction forms advanced glycation end-products (AGEs) by means of several steps such as oxidation, dehydration, condensation or fragmentation. AGEs such as N^{ε}-(carboxymethyl) lysine (CML) **(3)** and N^{δ}-(5-hydro-5-methyl-4-imidazolon-2-yl) ornithine (MG-H1) **(4)** are also generated not only from glucose but also from intermediate carbonyls such as glyoxal (GO) **(5)** and methylglyoxal (MGO) **(6)**. Thornalley *et al*. **(7)** reported that GO is major product formed by incubating glucose in 100 mM phosphate buffer. Similarly, Baynes *et al*. **(8)** reported that GO as the major carbonyl compounds when glucose was incubated in 200 mM phosphate buffer. In particular, α-dicarbonyls (α-DCs) such as GO and MGO are highly reactive compounds that were generated during the Maillard reaction. However, Baker *et al*. **(9)** reported that glucosone (GLN) is an oxidative product of Amadori compounds which is formed in the early stages of the Maillard reaction. We also previously demonstrated that GLN was generated from the oxidation of glucose **(10) (Fig. 1)**.

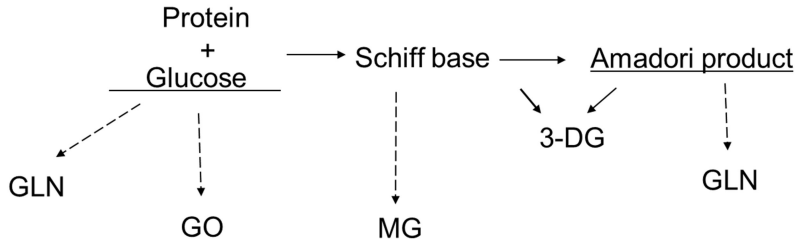

Fig. 1 *Formation pathway of α-dicarbonyl compounds in the Maillard reaction*

Although GO and MG are reported to generate AGEs, GLN-derived AGEs have not been reported yet. Our preliminary study demonstrated that GLN also reacts with proteins to form an adduct, in which the structure has not been identified yet due to its instability.

GLN could play a potential role in the destabilization of protein conformation by non-enzymatic modification *in vivo*. The structure of the GLN-derived AGEs will be published elsewhere in near future.

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The role of vitamin B6 in schizophrenia

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Vitamin B_6 (VB6) is a generic name for a family of six interconvertible pyridine compounds, pyridoxal (PL), pyridoxine (PN), pyridoxamine (PM), and their respective 5′-phosphorylated forms (PLP, PNP, and PMP, respectively). Of these forms, it is well known that PM can potentially inhibit advanced glycation end product (AGE) formation by blocking oxidative degradation of the Amadori intermediate of the Maillard reaction [1, 2]. Additionally, PM can also directly scavenge reactive carbonyl compounds and potentially trap reactive oxygen species [3-6]. By combining these multiple activities, PM is believed to be a potential inhibitor of AGE formation and is a promising drug candidate for the treatment of diabetic complications and an anti-aging agent [7]. PLP is a cofactor for > 150 enzymes (including representatives of every major enzyme class), which account for approximately 4% of known enzymes [8]. PLP-dependent enzymes are involved in the metabolism of neurotransmitters including dopamine (DA), noradrenaline (NA), serotonin (i.e., 5-hydroxytryptamine), glycine, D-serine, glutamate, γ-aminobutyrate, and histamine [9]. Thus, it is speculated that VB6 deficiency causes an accumulation of AGEs and affects the metabolism of many neurotransmitters, leading to impairment in brain function.

Schizophrenia is a heterogeneous psychiatric disorder characterized by positive symptoms, such as hallucinations and delusions, negative symptoms, such as anhedonia and lack of emotion, and cognitive impairment. We have reported that approximately 20% of patients with schizophrenia exhibit accumulation of pentosidine and lower levels of VB6 in the peripheral blood [10-12]. Patients with enhanced carbonyl stress (CS) experienced a more severe clinical course relative to those without CS [12]. The proportion of cases classified as inpatients was 80.8% in those with CS, whereas it was 23.9% in non-CS patients ($P < .0001$). In addition, the mean (± SD) cumulative duration of hospitalization in patients with CS was markedly longer than that of those without CS (17.4 ± 16.9 years versus 4.2 ± 9.2 years; $P = .0002$). Furthermore, we noted higher daily doses of antipsychotics in CS patients compared with non-CS patients (1143.9 ± 743.6 mg/day versus 773.8 ± 652.4 mg/day, respectively; $P = .02$). It should be noted that severe clinical features observed in patients with CS, such as higher inpatient status, longer duration of hospitalization and larger prescribed doses of antipsychotic medication, are very similar to those in treatment-resistant schizophrenia as defined by Kane et al. [13]. If we limit the discussion to VB6 deficiency in patients with schizophrenia, > 35% of patients with schizophrenia exhibit low levels of PL (clinically defined as < 6 ng/mL in males and < 4 ng/mL in females). These results have been replicated by other groups [14, 15].

Furthermore, we found that PL level is inversely proportional to severity score on the Positive and Negative Syndrome Scale (PANSS) [12]. These findings suggest that VB6 deficiency may contribute to the development of schizophrenia symptoms.

To uncover the relationship between lower VB6 levels and schizophrenia, we developed a VB6-deficient mouse model by feeding male C57BL/6J mice a VB6-deficient diet containing low levels of VB6 (5 μg/100 g pellets) from 8 to 12 weeks of age, while control mice were fed a normal diet, with VB6 at 1.4 mg/100 g (Toriumi K *et al.,* submitted). After 4 weeks of feeding, the plasma levels of VB6 in VB6-deficient mice decreased to approximately 3% of that in control mice. To date, accumulation of AGEs due to VB6 deficiency has not been observed. However, the body weight of mice fed the VB6-deficient diet did not increase, leading to a significant difference in body weight between VB6-deficient and control mice. Next, to evaluate the effect of low VB6 levels on mouse behavior, we performed behavioral tests using VB6-deficient mice. In the social interaction test, VB6-deficient mice exhibited less interaction compared with control mice, corresponding to increased negative symptom-like behavior. These behavioral data suggest that VB6 deficiency may be associated with negative symptoms. To investigate whether VB6 deficiency affected the function of monoaminergic neuronal systems, the tissue content of monoamines and their metabolites in various regions of the brain were measured. A marked increase in 3-methoxy-4-hydroxyphenylglycol (MHPG) was observed in the prefrontal cortex, the striatum, the nucleus accumbens and the hippocampus compared to that in controls, which is consistent with many clinical reports that have described MHPG levels in patients with schizophrenia. Furthermore, due to the increased levels of MHPG, the ratio of MHPG to NA was significantly increased in VB6-deficient mice, suggesting that the activities of noradrenergic neuronal systems were increased in VB6-deficient mice. Finally, we found increased MHPG levels in the peripheral blood of patients with schizophrenia compared with healthy controls, which demonstrates an increase in noradrenergic signaling in humans. Moreover, we demonstrate that MHPG concentration is negatively correlated with peripheral blood levels of VB6. These results strongly suggest that VB6 deficiency is linked to enhanced NA signaling in humans, and that VB6-deficiency may be involved in schizophrenia symptoms via hyperactivation of the noradrenergic system.

In contrast, to test whether VB6 supplementation could improve clinical symptoms in patients with schizophrenia, we conducted a 24-week, open-label clinical trial with high-dose PM supplementation in ten CS patients with schizophrenia [16]. PM was initiated at a dose of 1200 mg/day, supplementing the existing antipsychotic regimen, with the daily dose (1200, 1800, or 2400 mg/day) set at the discretion of the attending physician during the trial. Serum pyridoxal concentrations reached levels up to 345.3-fold higher than baseline with PM administration and plasma levels of pentosidine decreased by 26.8%. In the clinical assessment, the overall reduction rates for the PANSS positive, negative, general, and total subscales, and the Brief Psychiatric Rating Scale (BPRS) scores, were 6.9%, 5.8%, 9.9%, 8.1%, and 10.8%, respectively, which did not reach statistical significance, probably due to the small number of participants. Two patients experienced marked improvement in clinical symptoms, and one exhibited considerable improvement in psychological symptoms accompanied by a moderate decrease in plasma pentosidine levels. Additionally, four participants experienced

improvement in the side effects of antipsychotics, such as Parkinsonism. Unfortunately, severe adverse drug reactions occurred in two patients administered a dose of 1800 mg/day, in whom clinical symptoms and course were very similar to those of Wernicke's encephalopathy. However, the patients recovered completely following thiamine supplementation. After observing these adverse reactions, we measured serum thiamine levels and performed head magnetic resonance imaging in the other participants but observed no abnormal findings.

In summary, our findings suggest that VB6 deficiency may be involved in the pathophysiology of schizophrenia. Further study, therefore, of VB6 deficiency to fully understand the molecular mechanisms underlying schizophrenia development is warranted. Accordingly, we are currently conducting a phase IIb clinical trial with high-dose PM supplementation in patients with schizophrenia. We anticipate a day in which PM will become widely used as a novel therapeutic drug in those with treatment-resistant schizophrenia and increased CS.

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Highlights of the glycation literature
(January 2020 - March 2020)

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