ISSN 2436-5173 (Online)

Metallomics Research

Vol.3 Supplement 1

Abstracts of The 34th Annual Meeting of the Japan Society for Biomedical Research on Trace Elements (BRTE-2023)

JSBRTE
Japan Society for
Biomedical Research on Trace Elements

Metallomics Research

Vol.3 Supplement 1

ISSN 2436-5173 (Online)

Japan Society for Biomedical Research on Trace Elements

Abstracts of The 34th Annual Meeting of the Japan Society for Biomedical Research on Trace Elements (BRTE-2023)

"Trace element researches in Industry-academia-government collaboration by Cross-Disciplinary Researchers"

Tokyo Mitaka City Industrial Plaza, Tokyo, Japan

15-16 September 2023

Organizer: The Japan Society for Biomedical Research on Trace Elements

Co-sponsored by

- The Pharmaceutical Society of Japan
- The Japan Society for Analytical Chemistry (JSAC)
- Japanese Society for Zinc Nutritional Therapy (JZNT)
- Scientific Research on Innovative Areas: Integrated Bio-Metal Science

Organizing committee

[Congress Chair]
Masahiro KAWAHARA (Musashino University)
[Secretariat]
Dr. Ken'ichiro TANAKA, Dr. Midori NEGISHI
[Office]
Department of Bio-Analytical Chemistry,
Faculty of Pharamaceutocal Sciences, Musashino University
1-1-20 Shinmachi, Nishi-tokyo shi, Tokyo 202-8585, Japan

Editorial Office

Seishinsha, co. Ltd.,
Japan society for Biomedical Research on Trace Elements
2-8-13 Fukashi, Matsumoto-shi, Nagano 390-0815, Japan
Tel: +81-263-32-2301
Fax: +81-263-36-4691
Editorial Office: brte-post@seisin.cc
URL: https://www.brte.org/
https://metallomicsresearch.brte.org/

Abstracts of the 34th Annual Meeting of the Japan Society for Biomedical Research on Trace Elements (BRTE-2023)

"Trace Element Researches in Industry-Academia-Government Collaboration by Cross-Disciplinary Researchers"

Preface

It is our great pleasure to publish here the Abstracts of the 34th annual meeting of the Japan Society for Biomedical Research on Trace Elements (JSBRTE) as a supplement issue of *Metallomics Research.*

JSBRTE was established at 1990 to activate a broad range of research fields about metal/metalloids and to provide a platform of researchers. JSBRTE has organized annual meetings in every year from 1990. At 2023, the 34th annual meeting of JSBRTE (BRTE-2023) was held at Tokyo Mitaka City Industrial Plaza from 15 Sep to 16 Sep, 2023. About 100 researchers and students of metals/metalloids research gathers and discussed. In annual meetings, the Japanese abstracts has been published as an issue of Biomedical Research on Trace Elements (BRTE), the official journal of JSBRTE. In this meeting, we decided to publish English abstracts as a supplement issue of *Metallomics Research.*

Metallomics Research is an official journal of JSBRTE, and established at 2021. *Metallomics Research* is an international, peer-reviewed, open access journal publishing significant and novel contributions on the roles of metals, metalloids, and other trace elements in biological function.The journal welcomes investigations from a broad range of research fields including analytical chemistry, biochemistry, toxicology, epidemiology, nutrition, pharmacology, medical sciences, environmental sciences, health sciences, agriculture sciences, and plant biology. We apologize that not all abstracts were included in this issue owing to our a lack of preparedness. However, we believe that this issue will be effective for the international presentation for our research results. We also hope that all participants of BRTE-2023 will be happy and their researches will be successful.

Masahiro KAWAHARA, Prof.
Congress Chair of BRTE-2023
Department of Bio-Analytical Chemistry, Faculty of Pharamaceutical Sciences,
Musashino University, Japan

Contents (digest)

Educational lecture:

Special Lecture:

Symposium 1: collaborated symposium with Japanese Society for Zinc Nutritional Therapy（JZNT）

Symposium 2: collaborated symposium with Scientific Research on Innovative Areas: Integrated Bio-Metal Science

S-05: **Kazuhiro Nishiyama**

Laboratory of Prophylactic Pharmacology, Osaka Metropolitan University Graduate School of Veterinary Science, 1-58 Rinku-ohraikita, Izumisano, Osaka 598-8531, Japan.

"Study on metals and functional modification of G protein-coupled receptors"

S-06: **Yuta AMAGAI[1] and Kenji INABA[1, 2]**

[1] Institute of Multidisciplinary Research for Advanced Materials, Tohoku University, Sendai, Miyagi 980-8577, Japan. [2]Medical Institute of Bioregulation, Kyushu University, Fukuoka 812−8582, Japan.

"Protein quality control systems in the early secretory pathway regulated by zinc ions"

S-07: **Takanori KOBAYASHI**

Research Institute for Bioresources and Biotechnology, Ishikawa Prefectural University.

"Plant iron signaling by iron- and zinc-binding ubiquitin ligase HRZ"

Award Lectures

JSBRTE Young Investigator Award

A-01

Takamasa KIDO

Department of Public Health and Environmental Medicine The Jikei University School of Medicine, Tokyo 105-8461, Japan.

"Mechanism of thymic and splenic immune system impairment by zinc deficiency"

A-02

Yu-ki TANAKA

Graduate School of Pharmaceutical Sciences, Chiba University

"Development of applied analytical methods for trace elements by ICP mass spectrometer"

Hamari Chemicals Award

A-03

Ayako Fukunaka

Institute for Molecular & Cellular Regulation, Gunma University

"The elucidation of the role of trace elements in lifestyle associated disease"

JSBRTE Award

A-04

Masahiro Kawahara

Department of Bio-Analytical Chemistry, Musashino University

"Involvements of trace elements in the pathogenesis of neurodegenerative diseases"

Educational Lecture

EL-1:

Trace Element Research from the Viewpoint of Nutrition: Reexamination of Common Theories, Changes in Mineral Formulations, and Preparation of Trace Element-Deficient Animal

Munehiro YOSHIDA
Faculty of Chemistry, Materials and Bioengineering, Kansai University

In the field of trace element nutrition, several textbooks have arisen that need to be reexamined. On the other hand, there have been historical changes in mineral formulations used in animal nutritional studies, and there are still cases where inappropriate formulations are used. In addition, in studies in which trace element deficient animals have been prepared, the level of deficiency may differ among investigators. In this presentation, the common knowledge that needs to be reviewed, the cases that lead to the use of inappropriate mineral formulations, and the preparation of trace element deficiencies in laboratory animals are presented.

Special Lecture

SL-1:

Applications using synchrotron X-rays in biology and medicine: a point of view in atoms

Mari SHIMURA[1,2]
[1]RIKEN, SPring-8 Center, Koto 679-5148, Japan. [2]National Center for Global health and Medicine, Tokyo 162-8655, Japan.

A case of disease that antibody treatments works is likely caused by one specific molecule; however, there are still many diseases for unknown mechanism. Since they may be caused by multiple molecules, it is essential to examine them with a different point of view. Since molecules are consist of atoms, we have focused on elemental imaging and a relationship between isotope ratio and cancer survival rate. X-ray imaging at SPring-8 have revealed new findings of elemental distribution with biological kinetics. Higher zinc isotope ratio found a mortality high-risk group, which contained cases showing normal ranges in laboratory test values. These data suggested that analyzing atoms (elements) may help to understand a life phenomenon for unknown mechanism.
References: Shimamura T, Takeo Y, Moriya F, et al.: Ultracompact mirror device for forming 20-nm achromatic soft-X-ray focus toward multimodal and multicolor nanoanalyses, Nature Communications, in press. Fukunaka A, Shimura M, Ichinose T, et al.: Zinc and iron dynamics in human islet amyloid polypeptide-induced diabetes mouse model, Scientific Report, 2023; doi: 10.1038/s41598-023-30498-y. Hastuti AAMB; Costas-Rodríguez M, Matsunaga A, et al.: Cu and Zn isotope ratio variations in plasma for survival prediction in haematological malignancy cases, Scientific Report 2020. Matsuyama S, Maeshina K, Shimura M: Development of X-ray imaging of intracellular elements and structure, JAAS. 2020. 35, 1279-1294.

Symposium 1: collaborated symposium with Japanese Society for Zinc Nutritional Therapy（JZNT）

S-01:

Effect of zinc administration in patients with chronic liver diseases
-From prevention of sarcopenia to suppression of cancer development-

Atsushi HOSUI
Department of Gastroenterology and Hepatology, Osaka Rosai Hospital

[Aim] It has been reported that zinc deficiency occurs in patients with chronic liver diseases (CLD) and that administration of zinc preparations maintains and improves liver function and inhibits cancer development [1]. In this study, we aimed to clarify the frequency of sarcopenia, its incidence over time, and its risk factors in patients with CLD.
[Conclusion] The prognosis of sarcopenia in patients with CLD is poor, and careful follow-up is necessary, especially in elderly patients with hypozincemia. The effectiveness of zinc administration in preventing sarcopenia was studied in a small number of patients, and further accumulation of cases is warranted.

[1] Hosui A, Kimura E, Abe S, et al. Nutrients. 2018. 10: 1955.

S-02:

We have to learn many things from both correlation and causation between serum low zinc level and critical illness of COVID-19. −Reducing health disparity and design for primordial prevention for Japan in the 21st century

Hiroyuki YASUI
Department of Analytical and Bioinorganic Chemistry, Division of Analytical and Physical Sciences, Kyoto Pharmaceutical University.

Since most severely ill hospital patients with COVID-19 in Sakai City Medical Center exhibited zinc deficiency, we aimed to examine the correlation and causation between the patient's serum zinc levels and severe cases of COVID-19. Serum zinc < 60 μg/dL was defined as the criterion for hypozincemia, and patients continuously with these zinc levels were classified in the hypozincemia cohort. To evaluate the cross-sectional and longitudinal data, we performed multivariate analyses. In this symposium, we would like to talk the present results and examples of international mechanism studies, and additionally bring a place for arguments about health education and health promotion for Japan in the 21st century which we learned, to the table of BRTE2023.

S-03:
A New Strategy for Breast Cancer Therapy by Using Zinc

Tomoka TAKATANI-NAKASE[1,2]
[1]*Department of Pharmaceutics, School of Pharmacy and Pharmaceutical Sciences, Mukogawa Women's University,* [2]*Institute for Bioscience, Mukogawa Women's University, 11-68, Koshien Kyuban-cho, Nishinomiya, Hyogo 663-8179, Japan*

Breast cancer is the most common malignant tumor and the leading cause of cancer deaths in women. Effective molecular targeted drugs for the treatment of breast cancer have been developed, and have led anticancer therapy among cancers. However, breast cancer cells frequently acquire malignant phenotypes at an early stage, resulting in recurrence, metastasis, and resistance to treatment. Therefore, it is critical to develop novel therapeutic approaches and fully elucidate the mechanisms of breast cancer. We have been shown that zinc transported by zinc transporters selectively regulates target molecules and play an important key role in the sensitivity of anticancer drugs as well as the process of breast cancer pathogenesis. In this symposium, we summarize the zinc network and its molecular mechanisms in breast cancer, and discuss the possibility and future challenges of innovative breast cancer therapies by using the function of zinc.

Symposium2: collaborated symposium with Scientific Research on Innovative Areas: Integrated Bio-Metal Science

S-05:

Study on metals and functional modification of G protein-coupled receptors.

Kazuhiro Nishiyama[1]
[1] *Laboratory of Prophylactic Pharmacology, Osaka Metropolitan University Graduate School of Veterinary Science, 1-58 Rinku-ohraikita, Izumisano, Osaka 598-8531, Japan.*

β-adrenergic receptors (βARs) are G-protein-coupled receptors (GPCRs) that control sympathetic stimulation responses and are also attracting attention as targets for therapeutic drugs for heart failure. Previously, we found that transient receptor potential canonical (TRPC) 6 channels, which are permeable to Zn^{2+} and Fe^{2+}, increase βAR-stimulated cardiac contractility (positive inotropy) in mice. Therefore, we generated a Zn^{2+}-nonpermeable TRPC6 mutant knock-in mouse (TRPC6 KYD). Cardiomyocytes isolated from TRPC6 KYD mice were stimulated with βAR, and the isolated myocardium from TRPC6 KYD mice produced less cAMP in response to βAR stimulation than those from wild-type mice.
These data suggest that TRPC6 channel activation in ventricular myocytes enhances cardiac contractility through Zn^{2+} influx. In the future, it is expected that the TRPC6 activator will contribute to the development of new treatments for heart failure.

S-06:

Protein quality control systems in the early secretory pathway regulated by zinc ions

Yuta AMAGAI[1] and Kenji INABA[1, 2]
[1] *Institute of Multidisciplinary Research for Advanced Materials, Tohoku University, Sendai, Miyagi 980-8577, Japan.* [2] *Medical Institute of Bioregulation, Kyushu University, Fukuoka 812−8582, Japan.*

The early secretion pathway comprising the endoplasmic reticulum and Golgi apparatus is the space where newly synthesized secretory and membranous proteins acquire their native structures. Thus, the protein quality control system is highly developed in these organelles. Previously, we have found that the chaperone protein ERp44 binds Zn^{2+} in the Golgi to enhance its function. However, the labile Zn^{2+} concentration in the Golgi lumen and its regulatory mechanisms are largely unknown. Here, we measured labile Zn^{2+} concentrations in the Golgi apparatus of HeLa Kyoto cells, using a novel zinc probe: ZnDA-1H. By quantitative Zn^{2+} imaging in combination with systematic knockdown experiments, we found that the Golgi-resident zinc transporters (ZnTs) differently regulate Zn^{2+} homeostasis at the Golgi cisternae and thereby tune the traffic and functions of ERp44.

S-07:
Plant iron signaling by iron- and zinc-binding ubiquitin ligase HRZ

Takanori KOBAYASHI[1]
[1]*Research Institute for Bioresources and Biotechnology, Ishikawa Prefectural University.*

Iron is essential for various vital processes, including photosynthesis and respiration. To avoid deficit and toxicity of iron, plants transcriptionally induce the expression of various genes involved in iron uptake and translocation in response to iron deficiency. We previously identified a promising candidate for intracellular iron sensor in plants: hemerythrin motif-containing RING- and zinc-finger protein (HRZ)[1]. HRZ negatively regulates major plant responses to iron deficiency[1]. Physiological and biochemical evidence suggests that the stability and function of HRZ are affected by iron nutritional conditions, possibly through iron and/or zinc binding to its five characteristic domains. We propose that HRZ proteins sense and transmit cellular iron status by iron-dependent degradation of their ubiquitination targets.

Reference:
[1] Kobayashi T, Nagasaka S, Senoura T, Itai RN, Nakanishi H, Nishizawa NK: Iron-binding haemerythrin RING ubiquitin ligases regulate plant iron responses and accumulation. *Nat. Commun.* 2013. 4: 2792.

Award Lectures

JSBRTE Young Investigator Award

A-01:

Mechanism of thymic and splenic immune system impairment by zinc deficiency

Takamasa KIDO
[1] *Department of Public Health and Environmental Medicine The Jikei University School of Medicine, Tokyo 105-8461, Japan.*

Zinc deficiency causes dysfunction of the immune system. We have demonstrated that zinc deficiency causes thymic atrophy/fatty degeneration and a splenic-derived inflammatory response. Also, we investigated the beneficial effects of IL-4 administration or zinc supplementation on zinc deficiency. It was suggested that atrophy / fatty degeneration in the thymic cortex region of zinc deficiency rats impairs the maturation process of T cells. In the spleen, the activity of the anti-inflammatory 'Th2 lymphocyte-M2 macrophage' pathway was reduced and the inflammatory response was enhanced. Importantly, IL-4 injection or zinc supplementation can reverse the effects of zinc deficiency on immune function.

A-02:

Development of applied analytical methods for trace elements by ICP mass spectrometer

Yu-ki TANAKA[1]
[1]*Graduate School of Pharmaceutical Sciences, Chiba University*

Inductively Coupled Plasma-Mass Spectrometer (ICP-MS) is extensively employed for the precise measurement of trace elements within living organisms. Various analytical techniques utilizing ICP-MS, including laser ablation analysis, speciation analysis, isotope ratio measurement, and single particle/single cell analysis, have been applied to evaluate the metabolism, functions, and toxicity of trace elements, particularly biometals. In this presentation, I will discuss two main topics:
(1) Evaluation of calcium metabolism through isotopic composition analysis: The isotopic composition of calcium in serum and bone samples was analyzed by ICP-MS and used as an indicator to assess calcium metabolism in animals suffering from metabolic bone diseases.
(2) Development of analytical methods for single-cell elemental analysis: I developed the analytical methods to analyze elements at a single-cell level using bacteria, fungi, algae, and mammalian cells, highlighting their potential use in biomedical research.

JSBRTE Award

A-04:

Involvements of trace elements in the pathogenesis of neurodegenerative diseases

Masahiro Kawahara
Department of Bio-Analytical Chemistry, Musashino University.

Increasing evidence suggests that the dyshomeostasis of trace elements is implicated in the pathogenesis of various neurodegenerative diseases including Alzheimer's disease, prion diseases, Parkinson's disease, and vascular type of senile dementia. Disease-related proteins such as β amyloid protein, prion protein, α-syunuclein commonly form β-pleated sheet structures and exhibit neurotoxicity. Metals can bind to these proteins and cause their conformational changes, and enhance neurotoxicity. Moreover, these proteins or its precursor co-localize in the synapses and regulate metal homeostasis. Therefore, the disorders of the disease-related proteins can induce the dyshomeostasis of metals in the synapses, triggers synaptic degeneration and neuronal death. rupt the metal homeostasis and Meanwhile, excess Zn and Cu in the synapses are involved in the pathogenesis of vascular type dementia. Considering these results together, it is possible that trace elements in the synapses and the interactions between disease-related amyloid proteins are involved in the pathogenesis of these neurodegenerative diseases. Possible treatment based on metals are also discussed.

Oral Presentations

O-01:

Elucidation of the activation and physiological functions of *S*-selanylated glyceraldehyde-3-phosphate dehydrogenase.

Yuko MINAMI[1], Koki SHINODA[2], Shuhei MICHIGAMI[2] Mitsuka KOZAKI[1], Takayuki OHNUMA[1,2,3], Toru TAKEDA[1,2]
[1]Department of Advanced Bioscience, Graduate School of Agriculture, Kindai University, [2]Department of Bioscience, Kindai University, [3]Agricultural Technology and Innovation Research Institute, Kindai University, 3327-204, Nakamachi, Nara 631-8505, Japan

In this study, we investigated the effects of *S*-selanylation on the kinetics of glyceraldehyde-3-phosphate dehydrogenase (GAPC), and also discussed the physiological significance of *S*-selanylated GAPC in plant cells. It became clear that the increased specificity constant ($k$cat/K_m) of *S*-selanylated GAPC is due to the formation of selenol groups, which are more nucleophilic than thiol groups, on the catalytic Cys residues. Selenol groups reacted more rapidly with glyceraldehyde3-phosphate (G3P) than thiol groups, resulting in increased affinity and reactivity with G3P. Intracellularly, GSSeSG essential for *S*-selanylation was found to exist stably at pH 7.0 or below. In the cytosol that have taken up selenate or selenite, GAPC is *S*-selanylated via GSSeSG produced by the action of GSH and selenite, thereby avoided the toxicity of selenium oxyanions in the cytosol.

O-02:

Effects of selenite on the expression of genes involved in selenoprotein biosynthesis in the methanogenic archaeon *Methanococcus maripaludis*

Mikihisa ONODA[1], Riku AONO[1], Masao INOUE[1,2], Anna OCHI[1] and Hisaaki MIHARA[1]
[1]College of Life Sciences, Ritsumeikan University, [2]R-GIRO, Ritsumeikan University

In a methanogenic archaeon, *Methanococcus maripaludis*, the methanogenic pathway comprises two types of proteins: Sec-type proteins containing selenocysteine (Sec), such as FruA, and Cys-type proteins containing cysteine (Cys) instead of the Sec residue, such as FrcA. Here, we investigated the effects of selenite on the growth and the expression of genes involved in selenoprotein biosynthesis of this microorganism. We observed no differences in the growth phenotype of *M. maripaludis* with or without selenite. In the presence of selenite, the intracellular mRNA levels of *fruA* increased, whereas those of *frcA* decreased, suggesting that the expression of Sec-type proteins is induced by selenium. On the other hand, we observed no changes in the mRNA levels of *selB* and *selD*, which are involved in selenoprotein biosynthesis. These results suggest that the selenoprotein biosynthetic genes are expressed even in the absence of selenium, possibly to enable immediate synthesis of selenoprotein upon the availability of external selenium.

O-06:

Burning mouth syndrome (glossodynia) cotreated with zinc and L-carnosine

Kensaku SAKAE[1,2], Machi SUKA[2], and Hiroyuki YANAGISAWA[2]
[1]Department of Psychiatry, Keieikai Yashio Hospital, [2]Department of Public Health and Environmental Medicine, The Jikei University School of Medicine.

Burning mouth syndrome (BMS), also known as glossodynia, frequently co-occurs with taste disturbance (TD) and xerostomia (XS)—symptoms of zinc (Zn) deficiency. Therefore, Zn deficiency may be associated with BMS. However, owing to the lack of a sensitive biomarker, Zn deficiency may be overlooked in BMS patients. We report 2 cases of BMS secondary to Zn deficiency that were successfully cotreated with 150 mg/d of polaprezinc (PLZ, including 34 mg of Zn and 116 mg of L-carnosine [CAR]) and 2 g/d of CAR. In both cases, TD and XS improved after treating with PLZ. Exploring Zn deficiency in BMS patients is important. In addition, both cases suggested that 2 g/d of CAR may relieve burning mouth symptoms and that Zn may augment this effect and/or prevent relapse.

Reference:
Sakae K, Suka M, Yanagisawa H. Burning mouth syndrome cotreated with zinc and L-carnosine: two case reports. *J Clin Psychopharmacol*. 2023; 43: 387-389.

O-07:

Dehydroeffusol from *Juncus effusus* prevents amyloid $\beta_{1\text{-}42}$-mediated hippocampal neurodegeneration via metallothionein synthesis

Atsushi TAKEDA[1], Mako TAKIGUCHI[1], Haruna TAMANO[1,2]
[1]School of Pharmaceutical Sciences, University of Shizuoka. [2] Shizuoka Tohto Medical College, Shizuoka.

Here we examined the effect of dehydroeffusol, from *Juncus effusus* on amyloid $\beta_{1\text{-}42}$ ($A\beta_{1\text{-}42}$)-mediated hippocampal neurodegeneration. Dehydroeffusol (15 mg/kg body weight) was orally administered to mice once a day for 6 days and then human $A\beta_{1\text{-}42}$ was injected intracerebroventricularly followed by oral administration for 12 days. Neurodegeneration in the dentate granule cell layer was rescued by dehydroeffusol. Pre-administration of dehydroeffusol for 6 days rescued $A\beta_{1\text{-}42}$-mediated neurodegeneration. Interestingly, pre-administration of dehydroeffusol increased synthesis of metallothioneins, intracellular Zn^{2+}-binding proteins, in the dentate granule cell layer, which can capture Zn^{2+} from Zn-$A\beta_{1\text{-}42}$ complexes. The present study indicates that pre-administration of dehydroeffusol protects $A\beta_{1\text{-}42}$-mediated neurodegeneration in the hippocampus by reducing intracellular Zn^{2+} toxicity, which is linked with induced synthesis of metallothioneins. Dehydroeffusol, a novel inducer of metallothioneins, may protect $A\beta_{1\text{-}42}$-induced pathogenesis in Alzheimer's disease.

O-08:

Development and characterization of the *Zip10-EGFP*-KI mouse for the spatiotemporal analysis of ZIP10-expressing cells

Takafumi HARA[1], Misato KIKUCHI[1], Hatsuna MIYAKE[1], Akisa NAKANO[1], Emi YOSHIGAI[1], Takuto OHASHI[1] and Toshiyuki FUKADA[1]
[1]*Molecullar and Cellular Physiology, Faculty of Pharmaceutical Sciences, Tokushima Bunri University*.

We have previously reported that zinc transporter ZIP10 is essential for the development of epidermis and hair follicles. However, the regulatory mechanisms of ZIP10 expression and the lineage fate of ZIP10-expressing cells during the developmental process of skin tissue remain poorly understood. Therefore, we generated *Zip10-EGFP-IRES-CreERT2* knock-in (*Zip10-EGFP*-KI) mice by introducing the EGFP gene downstream of the *Zip10* gene promoter, and analyzed the gene expression and cellular lineage of ZIP10/GFP-expressing cells using the following methods. The results indicated that the spatiotemporal changes occurring in ZIP10/GFP-expressing cells during hair follicle and skin tissue formation. In the future, we plan to isolate ZIP10/GFP-expressing cells and their descendants for RNA-seq analysis to characterize their properties in terms of skin development.

O-09:

Effect of nicotianamine, a plant-derived chelating compound, on recovery of iron deficient mice.

Yoshiko MURATA[1], Makoto FUJISAWA[1], Takehiro WATANABE[1], Hiroyuki KIMURA[2] and Kosuke NAMBA[3]
[1]*Bioorganic Research Institute, Suntory Foundation for Life Sciences, Kyoto 619-0284, Japan.* [2]*Laboratory of Analytical and Bioinorganic Chemistry, Kyoto Pharmaceutical University, Kyoto 607-8414, Japan.* [3]*Department of Pharmaceutical Sciences, Tokushima University, Tokushima 770-8515, Japan.*

Nicotianamine (NA), which is important for the transport of essential elements such as iron, copper, and zinc in plant, is a chelate compound. We administered NA-Fe complexes to iron-deficient mice and examined their effect on restoring iron deficiency compared to Fe only. Mice were fed an iron-deficient diet for 2 weeks and each mouse was orally administered saline, NA-Fe or Fe only. Hemoglobin and serum iron levels recovered better with NA-Fe than with Fe. This study will be conducted with different doses and durations of administration.
Reference:
Murata Y, Murata J, Namba K: *Metallomics Research*. 2022; 2: 1-11.

O-10:

Crucial importance of ferroptosis-resistance in ferric nitrilotriacetate-induced renal carcinogenesis in animals.

Shinya AKATSUKA[1], Zhen CHENG[1], Yingyi KONG[1], Shinya TOYOKUNI[1]
[1]*Department of Pathology and Biological Responses, Nagoya University Graduate School of Medicine.*

Ferric nitrilotriacetate (Fe-NTA) induces Fenton reaction in renal tubules. Mice subjected to repeated Fe-NTA treatment develop oxidative stress induced renal cell carcinoma (RCC). Here we found that susceptibilities to the carcinogenic effects of Fe-NTA vary greatly with the genetic strain of experimental animals. A/J strain mice obtained RCCs in a high incidence, while C57BL/6 mice exhibited quite a low incidence. We then searched for factors involved in the difference in RCC development, focusing on the acute or subacute responses to Fe-NTA treatment. Transferrin receptor was more decreased in A/J mice than in C57BL/6 mice. The expressions of ferritin and GPX4 were higher in A/J mice than in C57BL/6 mice. Based on these results, we consider that A/J mice can suppress lipid peroxidation through decreasing the expression of transferrin receptor and sustaining GPX4 level to prevent massive ferroptosis and that it was the survived cells with DNA damage that developed into the RCC.

O-11:

Underlying factors of hypercupremia in patients with severe motor and intellectual disabilities (SMID) living in a residential facility for SMID

Aya TOKUMITSU and Yuichi KUSUNOKI
Department of Pediatrics, Hokkaido Ryoikuen.

[Objective] Hypercupremia is not rare in patients with severe motor and intellectual disabilities (SMID). We investigated underlying factors of hypercupremia in our SMID patients. [Methods] 1) Forty-five patients with serum copper levels over 150 μg/dL between 2018 and 2022 were selected and investigated. 2) The relationship between serum copper levels and various blood test results in 2020 were conducted using Pearson correlation analysis. [Results] 1) Twenty-nine patients were found to have some disease presumably associated with hypercupremia. Of those, 11, 9, 3, 2 and 4 patients had respiratory problems, urological problems, malignant diseases, autoimmune diseases and other diseases, respectively. 2) Serum copper levels were significantly and positively correlated with erythrocyte sedimentation rate. [Conclusions] SMID patients frequently have chronic respiratory and urological problems. These complications of SMID may cause various inflammations, and then result in hypercupremia.

O-15:

Formation of Selenium-adduct of Hepatic Protein in Rats Administered Selenite above a Nutritional Level

Munehiro YOSHIDA, Tingting WANG and Xin ZHANG
Laboratory of Food and Nutritional Sciences, Faculty of Chemistry, Materials and Bioengineering, Kansai University

To investigate the effects of selenium (Se) administration above the nutritional level on the liver, protease hydrolysates of rat liver treated with Se above the nutritional level were analyzed by HPLC-ICPMS. Se at 50 μg/d/rat was administered intraperitoneally as selenite or selenomethionine (SeM) to Se-deficient rats for 7 days, and the protease hydrolysates of the liver were analyzed by HPLC-ICPMS. Selenocystine (SeC) and SeM were detected in the SeM group, while in the selenite group, in addition to SeC, several Se compounds were found, one of which had the same retention time as that of SeM. Next, protease hydrolysates of liver from rats fed diets containing 0.2 or 3.0 μg/g Se were analyzed by HPLC-ICPMS; only SeC was detected at the 0.2 μg/g dose, but at the 3.0 μg/g dose, in addition to SeC, a Se compound with retention times closed to SeM was detected. These results indicate that Se-adducted proteins occur in the liver of rats treated with selenite above the nutritional level, and that the toxicity of selenomethionine may be lower than that of selenite.

O-16:

Identification of intestinal bacteria metabolizing *Se*-methylselenocysteine and its influence on the selenium metabolism in a host animal

Kazuaki TAKAHASHI[1], Sayano IIJIMA[2], Sakie HORIAI[2] and Yasumitsu OGRA[3]
[1]Graduate School of Horticulture, Chiba University, Chiba 263-8522, Japan. [2]Faculty of Pharmaceutical Sciences, [3]Graduate School of Pharmaceutical Sciences, Chiba University, Chiba 260-8675, Japan.

Selenium (Se) is an essential trace element and exists in various chemical forms. Their bioavailability depends on their chemical forms. We have shown that the intestinal microflora modified the bioavailability of Se in a host animal. In this study, we elucidated metabolites of *Se*-methylselenocysteine (MeSeCys), a major selenocompound in plants, by intestinal bacteria, and evaluated the bioavailability of the intestinal selenometabolites in rats. A strain isolated from rat feces transformed MeSeCys into dimethyldiselenide (DMDSe) and dimethylselenide (DMSe). DMDSe was utilized for biosynthesis of selenoproteins as Se source similar to MeSeCys, but DMSe was not. These results suggested that the intestinal Se conversions to DMDSe and DMSe by intestinal microflora could be one of the key factors for Se metabolism in host animals.

Poster presentations

P-02:

Synthesis of *N,N*-dimethylated selenoneine and its *Se*-methylation

Misuzu HAKODA[1], Noriyuki SUZUKI[2], Yasunori FUKUMOTO[2] and Yasumitsu OGRA[2]
[1]Faculty of Pharmaceutical Science, Chiba University. [2]Graduate School of Pharmaceutical Science, Chiba University.

The methylation and demethylation of selenium (Se) are considered to be the important metabolic processes for utilization or detoxification of Se compounds. From this viewpoint, naturally-occurring selenium compounds are classified into the two groups. Se compounds in the non-methylated group are highly bio-active and chemically unstable, and Se compounds in the methylated group are relatively bio-inactive and chemically inert. A bioselenium compound, selenoneine (2-selenyl-*Nα,Nα,Nα*-trimethyl-L-histidine) exhibits relatively low toxicity, although it has a non-methylated Se in its molecule. In this study, we intended to synthesize *N,N*-dimethylated selenoneine (Me2-selenoneine), and evaluated the *Se*-methylation of selenoneine and Me2-selenoneine. As a result, either selenoneine or Me2-selenoneine was not *Se*-methylated by the typical *Se*-methyltransferases (SeMT1 and SeMT2).

P-06:

The suppressive actions of zinc complexes on *in vitro* AGEs formation and the treatment effects on diabetes by intraperitoneal administration to KK-A^y mice

Masaki KOJIMA[1], Yuki NAITO[1], Katsuyuki TSUKAMOTO[1], Masayuki YAGI[2], Katsuhiko YOSHIZAWA[3], Yutaka YOSHIKAWA[4] and Hiroyuki YASUI[1]
[1]Laboratory of Analytical and Bioinorganic Chemistry, Kyoto Pharmaceutical University, [2]Anti-Aging Medical Research Center and Glycative Stress Research Center, Doshisha University, [3]Laboratory of Environmental Science, Mukogawa Women's University, [4]Department of Health, Sports, and Nutrition, Kobe Women's University.

Diabetes mellitus (DM) is a chronic metabolic disease, and the hyperglycemic conditions increase the amount of advanced glycation end products (AGEs) produced by the glycation reaction. In this study, we investigated both *in vitro* and *in vivo* anti-glycation effects of Zn complexes, which have been reported to show insulin-like effects. In an *in vitro* experiment, Zn complexes significantly reduced the fluorescent AGEs and pentosidine. Additionally, we administered [Zn(hkt)$_2$] by *i.p.* injection to KK-A^y mice for 14 days and evaluated its treatment effects on DM and AGEs production. This study is the first report of Zn complexes showing the anti-glycation actions and suppressing pentosidine formation in *in vitro* study.

P-09:

Changes in autophagy function related to abnormalities in lipid metabolism in mature pups induced by high-fat diet intake in pregnant and lactating mother rats and post-weaning pups

Takeshi SAITO[1], Yuki OHNO[2], Shojiro YAMASAKI[3] and Masaaki KURASAKI[4]
[1]Faculty of Health Sciences, Hokkaido University, Sapporo 060-0812, Japan. [2]Graduate School of Health Sciences, Hokkaido University, Sapporo 060-0812, Japan. [3]Sapporo Medical University, Sapporo 060-8556, Japan. [4]Faculty of Environmental Earth Science, Hokkaido University, Sapporo 060-0810, Japan.

The purpose of this study was to clarify whether the alteration of autophagy function is involved in the abnormal lipid metabolism in the liver of mature pups due to high-fat diet intake in pregnant and lactating mother rats and post-weaning pups, and the effect of green tea extract intake in lactating mother rats. High-fat diet intake in pregnant and lactating mother rats and post-weaning pups causes abnormal liver lipid metabolism and oxidative stress, and that one of the causes is a decrease in autophagy function due to defective formation of autophagosomes in the liver. Furthermore, the intake of green tea extract by lactating mother rats during the lactation period was shown to be effective in improving the abnormal lipid metabolism and impaired autophagy function in the liver caused by high-fat diet intake.

P-10:

β-Adrenergic receptor activation by isoproterenol prevents amyloid $\beta_{1\text{-}42}$-mediated hippocampal neurodegeneration via metallothionein synthesis

Haruna TAMANO[1,2], Kotaro TAMURA[1], Mako EGAWA[1], Yuya KAWANO[1], Atsushi TAKEDA[1]
[1]School of Pharmaceutical Sciences, University of Shizuoka. [2] Shizuoka Tohto Medical College, Shizuoka.

We examined whether isoproterenol, a β-adrenergic receptor agonist reduces neurodegeneration caused by amyloid $\beta_{1\text{-}42}$ ($A\beta_{1\text{-}42}$), for which intracellular Zn^{2+} dysregulation is a trigger. Neurodegeneration by $A\beta_{1\text{-}42}$ was canceled after co-injection of isoproterenol. Isoproterenol did not affect $A\beta_{1\text{-}42}$ staining (uptake) in the dentate granule cell layer. In contrast, isoproterenol reduced intracellular Zn^{2+} level increased by $A\beta_{1\text{-}42}$. The synthesis of intracellular metallothioneins (MTs), Zn^{2+}-binding proteins was not increased in the dentate granule cell layer by $A\beta_{1\text{-}42}$ injection, but increased by co-injection of isoproterenol. Phenylephrine, an α_1-adrenergic receptor agonist did not increase MT synthesis. These data indicate that isoproterenol increases MT synthesis and cancels neurodegeneration via intracellular Zn^{2+} toxicity after $A\beta_{1\text{-}42}$ injection. It is likely that MT synthesis induced by β-adrenergic receptor-mediated signaling contributes to prevent the $A\beta_{1\text{-}42}$ pathogenesis.

P-15:

Effects of long-term combined administration of zinc and manganese or zinc and copper on learning and memory in aged mice

Kaoru YOSHIDA[1], Min Gi[2,3], Shugo SUZUKI[2], Masaki FUJIOKA[2], Isao TERAMOTO[4]
[1]Dept. of Food Science and Nutrition, Doshisha Women's College of Liberal Arts [2] Dept. of Molecular Pathology, Osaka Metropolitan University Graduate School of Medicine, [3]Dept. of Environmental Risk Assessment, Osaka Metropolitan University Graduate School of Medicine, [3]Dept. of Parasitology, Osaka Metropolitan University Graduate School of Medicine

Our previous studies showed that long-term Zn administration to aged mice dose-dependently inhibited learning and memory in novel object recognition test (ORT) and step-through passive avoidance test, suggesting that excess Zn administration impaired long-term memory and object recognition memory in aged mice. In the present study, we investigated the effects of long-term combined administration of Zn and Mn or Zn and Cu on learning and memory in aged mice. The combination of Zn and Mn did not cause the ORT-induced memory impairment observed in the Zn alone treatment. On the other hand, the combination of Zn and Cu showed a tendency to decrease memory performance compared to Zn alone. These results indicate that Cu may enhance Zn-induced the impairment in learning and memory, while Mn may conversely suppress the impairment in learning and memory.