X ISTERH CONFERENCE

TRACE ELEMENT RESEARCH ON HEALTH AND DISEASES

KEIO PLAZA HOTEL TOKYO, TOKYO, JAPAN 18-22 NOVEMBER 2013

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CONTENTS

Foreword			3
Plenary Lecture			4
Symposium	1	Zinc Deficiency in the Aged	8
	2	New approaches to metallothionein research and the recent clinical findings	10
	3	Is Dietary Exposure to Arsenic a Cause for Concern?	11
	4	Populations at Risk for Trace Element Deficiencies	12
	5	Advanced Analytical Techniques	13
	6	Molecular Aspects of Metalloid Toxicity	15
	7	Copper Balance and the Related Factors	16
	8		17
	9	Trace Elements and Disease Burden	18
	10	Recent Advance in Menkes Disease	19
	11	Trace element problems in Developing countries : malnutrition, infection and immunity	20
	12	Frontier of Neurometals	22
	13	Update on the Health Benefits of Bioactive Trace Elements Not Generally Recognized as Essential	23
	14	New Insights into the Role of Manganese in Health and Disease	24
	15	Selenium in Nutrition and Health	26
	16	Review and prospect of radioactive materials before and after Fukushima nuclear accident in Japan. - environment, food and radiation exposure -	27
Mini Symposium	1	Toxicity of Metals: Arsenic	29
	2	Toxicity of Metals: Cadmium	29
	3	Selenium	30
	4	Wilson disease	32
Poster			33

Foreword

TRACE ELEMENT RESEARCH ON HEALTH and DISEASES

The present issue of Journal of Trace Element in Medicine and Biology contains abstracts submitted to the 10th Conference of the International Society for Trace Element Research in Humans (ISTERH) that is an international forum for clinicians and researchers to exchange information regarding advances in trace element research. The conference was organized in collaboration with Japanese Society for Biomedical Research on Trace Elements (JSBRTE) and Japanese Society of Clinical Nutrition (JSCN), and is held at Keio Plaza Hotel in Tokyo on 18-22, November, 2013. The theme of the 10th ISTERH is TRACE ELEMENT RESEARCH ON HEALTH and DISEASES.

ISTERH is a scientific society on various problems caused by trace elements that was established by Drs. Prasad, Brewer, Okada and Tomita and other big professors working on trace elements in 1984 and has been contributing to human problems caused by trace elements. It is a badge of honor for us to organize the memorable 10th Conference.
The 10th ISTERH will cover the recent advances in analysis of trace elements, metallomics, environmental problems of trace elements, genetic diseases and various problems caused by trace element disorders.

The program accommodated eight plenary lectures presenting distinguished scientists of the world. Sixteen symposia were organized/chaired by Toshiyuki Fukada and Taiho Kambe (symposium 1), Masao Sato and Katsuyuki Nakajima (sym.2), Paevez Harris and Jun Yoshinaga (sym.3), Jeanne Freeland-Graves and James McClung (sym.4), Diago Iwahata and Kazumi Inagaki (sym.5), Seiichiro Himeno and Yasumitsu Ogra (sym. 6), Noboru Saito and Mamoru Nishimuta (sym.7), Sangeeta Shukla (sym.8), S.K. Roy (sym.9), Stephen G Kaler and Haruo Shintaku (sym.10), Ingrid S Surono and Rizky Abdulah (sym.11), Asushi Takeda (sym 12), Forrest H Nielsen (sym.13), Michael Aschner and Wei Zhang (sym. 14), Thomas Ong and Shinobu Ida (sym.15), and Hideo Sugiyama and Satoshi Yoshida (sym. 16). Three mini-symposia and poster presentations are also in the program. We have 173 abstracts from presenters coming from many countries all over the world.

A large number of sponsors have supported individual speakers or symposia. Four Luncheon Seminars are supported by Nobelphama Co. Ltd, Novo Nordisk Pharm. Ltd, Nestlé Health Science, and Eli Lilly Japan K.K.. Without support, it would not have possible to organize the symposium. We hereby express our gratitude for this indispensable support and thank all our sponsors.

Tokyo is a capital city where modern and traditional cultures of Japan are mixed together. Until around 100 years ago, we had closed our country and stopped the communication with foreign countries for 300 years. During the time we had incubated our own culture. Thus you can see our characteristic culture in Tokyo as well as in other districts. As you know, Sky Tower in Tokyo is now the highest tower in the world, and this year Mount Fuji was declared a World Heritage Site. I hope that you will enjoy sightseeing of Tokyo area.

Hiroko Kodama
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PL-1
Zinc in Human Health: It's discovery 50 years ago and the current impact

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The essentiality of zinc in humans was established in 1963. During the past 50 y, tremendous advances in both clinical and basic sciences of zinc metabolism in humans have been observed. The major factor contributing to zinc deficiency is high phytate-containing cereal protein intake in the developing world, and nearly 2 billion subjects may be zinc deficient. Conditioned deficiency of zinc has been observed in patients with malabsorption syndrome, liver disease, chronic renal disease, sickle cell disease, and other chronic illnesses. Major clinical problems resulting from zinc deficiency in humans include growth retardation; cell-mediated immune dysfunction, and cognitive impairment. In the Middle East, zinc deficient dwarfs did not live beyond the age of 25 y, and they died because of intercurrent infections. In 1963, we knew of only 3 enzymes that required zinc for their activities, but now we know of >300 enzymes and >1000 transcription factors that are known to require zinc for their activities. Zinc is a second messenger of immune cells, and intracellular free zinc in these cells participate in signaling events. Zinc has been very successfully used as a therapeutic modality for the management of acute diarrhea in children, Wilson's disease, the common cold and for the prevention of blindness in patients with age-related dry type of macular degeneration and is very effective in decreasing the incidence of infection in the elderly. Zinc not only modulates cell-mediated immunity but is also an antioxidant and anti-inflammatory agent.

PL-2
Monitoring of body iron metabolism in health and disease

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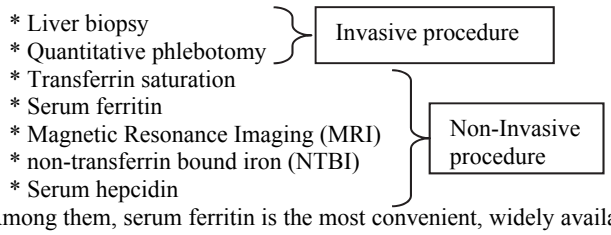
Iron is an essential metal for haemoglobin homeostasis. However, excessive accumulation of iron causes organ dysfunction through the production of reactive oxygen species (ROS). Iron metabolism in the body involves a delicate balance of storage and transport which is regulated by several factors. One of the factors is hepcidin, the newly identified peptide. Accumulation of iron occurs as there is no passive excretory mechanism. Furthermore, hereditary factors, repeated transfusions and many other disease conditions contribute to exogenous iron overload and its accumulation. Notably, liver is the most important organ for iron storage and metabolism. It has the largest capacity to sequester excess iron by depositing iron as ferritin and hemosiderin, and regulating body iron metabolism through hepcidin production. Several mechanisms have been postulated for hepatocyte iron uptake such as transferrin- mediated pathway and non-transferrin bound iron mediated pathway. However, the precise mechanism of hepatocyte iron uptake is not fully elucidated. In chronic liver diseases such as chronic hepatitis C, alcoholic liver disease and non-alcoholic fatty liver disease, iron is accumulated in the liver. It has also been postulated that iron could enhance the progression of fibrosis in such conditions. Iron reduction therapies such as phlebotomy, iron reduction diet therapy and iron chelation may be effective in improving liver function and delaying the progression of fibrosis. The preventive effects of iron reduction in the development of hepatocellular carcinoma have also been postulated. The factors responsible for iron toxicity are the free irons, non-transferrin-bound iron (NTBI), labile plasma iron (LPI) in the circulation, and the labile iron pool within the cells. Direct and indirect methodologies have been adopted for the measurement of body iron.



Among them, serum ferritin is the most convenient, widely available and cost effective modality, although its specificity is occasionally problematic. However, a direct physicochemical determination of iron in liver biopsied specimen remains the gold standard for the evaluation of body iron status. Recently, new detection methods for monitoring body iron such as MRI, non-transferrin bound iron, and hepcidin have gained importance in the estimation of iron metabolism in health and iron-related diseases.

PL-3
Elemental speciation analysis in human health assessment

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Trace elements have both positive and negative impacts on human health. Although some elements (e.g. Hg, Pb, Cd) are considered to have only toxic effects, many trace elements provide beneficial effects up to certain exposures before exhibiting detrimental effects, and in some cases (e.g. Se) the concentration window of beneficial effects is extremely narrow. Adding an extra level of complexity is the influence of the chemical form of the element. The bioaccessability, bioavailability and transformation of an element in humans depend on which elemental species is ingested. The importance of this type of information for environmental and human health has driven the area of research known as elemental speciation analysis, whereby methods are developed to identify and quantify the various species of an element. The methods can be applied to environmental situations to determine the elements' behaviour and fate in nature, and also in human metabolic studies to determine the elements' fate and effects in humans. Increasingly, speciation analysis is being used to link environmental and human health studies.

One of the main methods of speciation analysis couples chromatography with mass spectrometry. In this way the various species in a sample can be separated, either by gas chromatography or, more commonly, by high performance liquid chromatography (HPLC), and then selectively detected by a combination of atomic mass spectrometry (such as inductively couple plasma mass spectrometry, ICPMS) and molecular mass spectrometry (such as electrospray mass spectrometry, ESMS). The two mass spectrometric methods perfectly complement each other, and together allow both quantification and identification of elemental species. The methods are used to investigate a range of topics relevant to human health assessment, such as metal contaminants in food, and the uptake, transformation and excretion of toxic and essential elements in humans. Quantitative data can be obtained by analysis of human biological fluids such as urine and blood. The presentation will provide examples of the application of elemental speciation analysis to human health assessment.

PL-4
The Promise of Copper Lowering Therapy with Tetrathiomolybdate in the Cure of Advanced Previously Incurable Cancer and Treatment of Inflammatory Diseases

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Tetrathiomolybdate (TM) complexes copper with protein in a strong tripartite complex. This complex with food protein is excreted in the stool causing a negative copper balance. In the blood this complex, TM, free copper, and albumin, renders free copper unavailable and nontoxic. TM, developed for Wilson's disease, is very effective for the neurologic presentation, for which no other drug is optimal.

Lowering copper to intermediate levels with TM, levels high enough to avoid clinical deficiency, inhibits many cytokines, such as those promoting fibrotic, inflammatory, autoimmune diseases, and cancer. TM is efficacious in mouse models of pulmonary fibrosis and cirrhosis, inhibiting transforming growth factor beta (TGFbeta) and tumor necrosis factor alpha (TNFalpha). TM is efficacious in mouse models of inflammatory disease involving liver and heart, and four mouse models of immune modulated disease. In the human, TM reached both primary endpoints in a one year randomized controlled trial in primary biliary cirrhosis, an autoimmune attack on bile ducts. TM has great promise in the disease areas of fibrosis,inflammation, and autoimmunity. Wherever steroids are useful, TM will be better and safer.

Most excitingly, recent developments show that TM can cure advanced, previously incurable, metastatic cancer. Tumor growth requires angiogenesis (Folkman). Many angiogenic promoters are copper dependent, particularly those active at the micrometastatic cancer level. MAJOR CLUE: TM COMPLETELY INHIBITED CANCER GROWTH IN THE HER/2neu MOUSE GENETIC MAMMARY CANCER MODEL. Micro clusters of cancer cells, still there after a year of TM therapy, couldn't grow because of a lack of angiogenesis, while all controls had large cancers. Human trials ignored this clue, and tried TM only only against advanced cancers. These can recruit many promoters, many not copper dependent, and only modest effects in these trials led to little interest. Now, two US groups are curing metastatic cancer with TM by harking back to the mouse clue. One group requires conventional suppression to no evidence of disease (NED) , although the disease is considered incurable because of micrometastases. After three years of TM the cancer is cured, with 10 different advanced metastatic, heretofore incurable cancers. The second group uses TM from the beginning, and conventional therapy plus certain therapies aimed at cancer metabolic vulnerabilities, curing many patients with different metastatic cancers. We have begun a randomized controlled trial of TM in micrometastatic osteosarcoma in dogs. It appears TM can cure many previously hopeless case of cancer.

PL-5
Copper dysfunction as a risk for Alzheimer's disease: predictive value of MCI conversion into dementia, meta-analytic evidence of increased copper and Non-ceruloplasmin copper in AD and new studies on ATP7B gene variants

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Alzheimer's disease (AD) is the most common form of dementia. Diverse and independent pathogenetic pathways have been proposed to explain the basis of the disease, which can be disrupted at the same time, and each contributes to the disease etiology.
A number of studies since late '90 have shown that metal dyshomeostasis may enhance metal-related toxicity and beta-amyloid oligomer formation and precipitation in plaques within the AD brain. More recently, alterations in copper metabolism have been reported to associate with AD onset and progression. Studies performed *in vivo, in vitro*, in living patients and *in silico* studies have demonstrated that brain local and systemic defects in copper metabolism are associated with typical deficits of AD.
Rather than total copper, the relationship between copper and ceruloplasmin (Cp) seems the key issue in interpreting studies of copper in AD. Specifically, Non-ceruloplasmin (Non-Cp) copper originates from a defect of the ATP7B pump to mounting copper into nascent Cp in the hepatocyte, and is released in serum as an exchangeable and toxic pool of copper loosely bound to small molecular weight nutrients, available for oxidative stress reactions via Fenton and Haber-Weiss chemistry also in the brain, crossing the brain blood barrier.
Our group has collected a bulk of evidence sustaining a role of copper derangement in the rise of the risk for AD. Specifically, Non-Cp copper was associated with cognitive decline, with markers of AD, namely beta-amyloid and Tau proteins in the cerebrospinal fluid, with clinical worsening in an AD cohort clinically followed for a year; with Mild Cognitive Impairment (MCI), considered an incipient AD condition; with a faster rate of conversion from MCI to AD full dementia, and recently this evidence has been supported by meta-analytic studies analyzing all the data published from 1983 to 2013 on this topic which demonstrated that serum copper and Non-Cp copper are higher in AD than in controls.
We extended our study dissecting the pathways of copper involvement in AD starting from the key copper-regulating gene, ATP7B, also known as the Wilson's disease gene: 4 Single Nucleotide Polymorphisms (SNPs) informative of the gene structure and a rare Wilson's disease mutation, were associated with the risk of having AD and with higher levels of Non-Cp copper. A new patented devise to measuring directly Non-Cp copper is now available to assess the risk of a copper dysfunction.

PL-6
A century of progress for the diagnosis of Wilson disease

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When in 1912 Kinnier Wilson published his work *Progressive Lenticular Degeneration: a Familial Nervous Disease Associated with Cirrhosis of the Liver* as part of his dissertation for the Edinburgh MD degree, he correctly speculated that his patient's brain disease was caused by their liver disease. He postulated a "morbid toxin" produced by the cirrhotic liver was etiopathogenic. It was 33 years later when Glazebrook detected excess copper in the basal ganglia of a brain of a patient deceased from their Wilson disease and correctly postulated that the accumulation of copper in the liver was due to the inability to properly excrete copper.
Specific diagnostic signs and biochemical findings to help diagnose Wilson disease followed this recognition that excess copper was the cause of Wilson disease. The corneal pigmented rings now known as Kayser-Fleischer rings were described separately in 1902 and 1903, but years passed before it was accepted that the corneal pigmentation was caused by the copper deposition, and even more years before the high frequency of the presence of Kayser-Fleischer rings for patients with neurologic presentations of their Wilson disease compared to the lower frequency in those with hepatic presentations of their disease was recognized. Recognition that serum ceruloplasmin concentration could aid in the diagnosis of Wilson disease came from the observations of Scheinberg and Gitlin in 1952, who were the first to report that a low serum level is found in 96% of Wilson disease homozygotes. However, it was Sternlieb and Scheinberg who later found that 20% of heterozygotes also have low ceruloplasmin concentrations without clinical manifestations of Wilson disease. Further diagnostic advances came with the ability to perform percutaneous liver biopsy. Liver biopsy diagnoses of Wilson disease by determining the hepatic copper content, with values above the threshold of 250 mcg/g dry weight liver (later a lower threshold was demonstrated to have more sensitivity but less specificity) being highly specific for Wilson disease in the absence of prolonged cholestasis. The liver biopsy also permitted the identification of the histologic and ultrastructural effects of copper accumulation (including the unique effects of copper on mitochondria), on the liver. The ability to perform transjugular biopsy on even coagulopathic patients expanded the use of this diagnostic test even further.
Perhaps the most exciting advances have followed genetic and molecular genetic studies of Wilson disease. It was not until 1953 that Wilson disease was clearly recognized as an autosomal recessive disorder. Following the recognition that the disease was localized to chromosome 13 by linkage analysis, further study of the regions of chromosome 13 as well as parallel work on finding genes homologous to the Menkes disease gene, *ATP7A*, the gene for Wilson disease was identified as *ATP7B*. The identification of disease specific mutations on each allele has enhanced our diagnostic capabilities, and has moved this technology as first line testing in siblings of established patients.
Taking into consideration the clinical, biochemical and molecular testing results for patients with Wilson disease, a scoring system was developed at an International Meeting on Wilson's and Menkes disease in Leipzig Germany that has since been validated and included as part of the EASL guidelines for diagnosis and treatment of Wilson disease. This scoring system is useful for clinicians to help determine when to further pursue a diagnosis of Wilson disease.
In summary, there are many advances in the past century since the description of Wilson disease by Kinnear Wilson that allow us to accurately diagnose this fascinating disorder that presents with such varied phenotype. However, outside of screening programs for genetic diseases, it still requires the clinician to be cognizant of Wilson disease in the right patient to request the necessary testing and help avoid delays in diagnosis.

PL-7
Metallostasis in Alzheimer's disease and Parkinson's disease

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The brain houses high concentrations of transition metals Zn, Cu and Fe. Zn and Cu are released during glutamatergic neurotransmission, and their reuptake fatigues with age. This increases the average concentration of extracellular Zn and Cu (a phenomenon called "metallostasis") leading to Aβ aggregation in Alzheimer's disease (AD) and other downstream pathologies. Intraneuronal cortical Fe metallostasis is a feature of aging, and is exaggerated in AD where Fe is trapped by neurofibrillary tangles. A marked rise in Fe in the substantia nigra is a feature of both mutation-associated and sporadic Parkinson's disease (PD). We hypothesized that the main proteins implicated in the pathology of AD and PD are in proximity to these metals because they function to regulate neuronal metal homeostasis.

1. APP is the neuronal chaperone of ferroportin and is needed for Fe efflux. It is decreased in the SN in PD.
2. PBT2, a Zn/Cu ionophore that has induced cognitive improvement in phase 2 testing in AD, reverses metallostasis.
3. Tau knockout mice accumulate Fe in the cortex and nigra, causing neuronal loss, parkinsonism and cognitive loss with advancing age. The neurodegeneration is inhibited by Fe-chelator treatment. Soluble (functional) tau levels are decreased in AD and PD.
4. Loss of Zn flux in the glutamatergic synapse causes accelerated age-dependent cognitive decline in ZnT3 ko mice, a phenocopy of AD.
5. Presenilins 1 and 2 play major roles in the uptake and turnover of Zn and Cu. The SOD1 activation pathway is sensitive to PS loss.

Metallostasis may be the upstream factor that leads to proteostasis in AD and PD and is an upstream target for new pharmacological approaches.

References:
Duce, *et al*. *Cell* **142**, 857–67 (2010).
Lei, P. *et al. Nat Med* **18**, 291–295 (2012).
Adlard *et al*. *J Neurosci* **30**, 1631–6 (2010).
Greenough. *et al. JBC* **286**, 9776–86 (2011).

PL-8
Zn acts as a signaling molecule.

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Zinc (Zn) is an essential nutrient that acts as a second messenger to regulate a large number of physiological functions in at least two ways. Late Zn signaling depends on transcriptional changes in Zn transporters (1). We have shown roles for a number of Zn transporters including Slc39a6/Zip6/Liv1, Slc39a13/Zip13, Slc30a5/Znt5, and Slc39a14/Zip14 (2-6). All these studies indicate that the regulation of Zn transporters affects intracellular Zn status, Zn content and Zn distribution in various intracellular signaling pathways. The expression of such transporters is sensitive to various extracellular stimuli, which can therefore have indirect effects on intracellular signaling molecules via Zn-binding. In early Zn signaling, extracellular stimuli have direct effects by modifying intracellular Zn-homeostasis. For example, FcεR1-stimulation induces an increase in intracellular free Zn without any intermediary Zn-transporter, a phenomenon we named the "Zn wave"(7). Zn waves seem to originate around the ER several minutes after stimulation in a manner independent of changes in the expression of Zn transporters. In fact, we showed that a pore-forming Zn permeable channel on the ER membrane is involved in the Zn wave (8). It has been reported that Zn is present in mast cell vesicles, but its roles are unknown. We showed that the Slc30a2/ZnT2 is required for the retention of Zn in mast cell vesicles. Furthermore, using ZnT2KO mice we showed that Zn released from mast cells upon FcεR1-stimulation plays roles in wound healing (8). In this talk, I will elaborate on the role of Zn and Zn signaling in various cellular homeostasis systems.

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1) Hirano et al, **Adv. Immunol**. 97:149, 2008; 2) Yamashita et al, **Nature** 429: 298, 2004; 3) Fukada et al, **PLoS ONE** 3(11):e3642, 2008; 4) Kitamura et al, **Nature Immunol**.7: 971, 2006; 5) Nishida et al, **J. Exp. Med.** 206:1351, 2009; 6) Hojyo et al, **PLoS ONE** 6(3):e18059, 2011; 7) Yamasaki et al, **J. Cell Biol.** 177: 637, 2007; 8) Yamasaki et al., **PLoS ONE** 7(6):e39654, 2012; 9) Nishida et al., manuscript submitted

S-1-1
Zinc and Aging: An introduction

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Aging, biological responses followed by time axis, takes place in numbers of cells, therefore, influence health and disease conditions in which zinc homeostasis is also affected. Although the mode of physiological and pathological status is frequently affected by the status of zinc, the relation between zinc and aging are not fully understood. This symposium will aim to share the updated information about the significant link of zinc and aged phenomenon, and to discuss the future directions.
Recent studies have highlighted that the zinc ion acts as a signaling mediator: zinc signal. Series of studies demonstrated that zinc signal participates in physiology and diseases. I will address zinc signal contributes to our health and involves in diseases, by highlighting bone homeostasis and immune regulation, and discuss that zinc signal selectively controls intracellular signal transduction and the following cellular events, called zinc signal axis which may provide with understanding the role of zinc in aged phenomenon.

Reviews:
J. Biol. Inorg. Chem. (2011) 16:1123–1134
Metallomics (2011) 3: 662-674
J. Bone. Miner. Meta. (2013) 31:129-135, 2013

S-1-2
Basic and Clinical Aspects in Zinc and Degenerative Diseases of Ageing

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Essentiality of zinc for humans was only established fifty years ago. Zinc deficiency is now known to affect nearly two billion subjects in the developing world. It affects adversely cell mediated immunity, increases oxidative stress and inflammatory cytokines. In the developed countries nearly 30% of the elderly are zinc deficient. Many chronic diseases seen in the elderly such as atherosclerosis, diabetes, neuro-degenerative disorders, Parkinson's disease and age related macular degeneration (AMD) may be due to chronic inflammation and increased oxidative stress. Zinc supplementation studies in the elderly have shown decreased incidence of infections, decreased oxidative stress, and decreased generation of inflammatory cytokines. Decreased incidences of blindness in patients with AMD and increased atheroprotective effect have been observed in the zinc supplemented elderly. Zinc is a molecular signal for immune cells and many transcription factors involved in gene expression of inflammatory cytokines and adhesion molecules are regulated by zinc.

S-1-3
Zinc Deficiency Anemia in Patients with Chronic Kidney Disease (CKD)

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Background: Recently, we reported Zinc (Zn) deficiency anemia in dialysis patients and effectiveness of Zn supplementation. We evaluate here the correlation between serum Zn concentration (s-Zn) and renal anemia in non-dialysis CKD patients.
Materials & Methods: The correlation between s-Zn and anemia was evaluated in 123 non-dialysis CKD patients. Of these, 10 cases with normal iron metabolism and a low s-Zn were selected to receive administration of Polaprezinc (containing 34mg of Zn per daily) for three months.
Result: It has been shown that s-Zn become significantly lower with the progression of CKD. There is a significant correlation between e-GFR and s-Zn ($p<0.01$). It has also been found that the patients with low s-Zn had a significantly lower hemoglobin level (Hb) ($p<0.01$). Patients with Zn supplementation showed improvement in Hb from 9.5 to 10.4g/dl.
Discussion: It has been reported that the Zn absorption is inhibited in patients with chronic renal failure. It is suggested that highly advanced renal impairment leads to lower Zn absorption which results in aggravated anemia in CKD patients. For such patients, the administration of Polaprezinc has been effective.

S-1-4
Zinc, diabetes mellitus and cardiovascular disease

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Impaired zinc homeostasis features prominently in Type 2 diabetes mellitus (DM) and cardiovascular disease (CVD). The possibility that replenishment with zinc could provide an adjunct therapy for these diseases is compelling.
Our aim was to evaluate the effects of zinc supplementation on risk factors for CVD and DM. A systematic review and meta-analysis were undertaken.
In relation to CVD risk factors, the findings from clinical trials showed an overall lack of effect of zinc supplementation on plasma lipids. When the trials were assessed based on health status of the participants, zinc resulted in a significant decrease in high-density lipoprotein (HDL) in healthy subjects (an undesirable response), but a significant rise in HDL in those with DM, indicating a decreased risk of CVD. In DM patients, zinc supplementation decreased glucose concentrations and %HbA1c. These observations were supported by results from cross-sectional surveys and from cohort studies.
Improved zinc status affects favourably the cardio- metabolic profile in people with DM. Further understanding of the cellular homeostasis of zinc will help to explain the effects of zinc in humans.

S-1-5
Serum Zinc Concentration and C-viral Chronic Liver Disease in the Aged

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Zinc plays an important role as a component of numerous enzymes, and its deficiency results in various disorders, including liver diseases. Patients with chronic hepatitis C, particularly those with liver cirrhosis (LC) and liver failure, have low serum zinc concentrations.
The relationship between hepatocellular carcinoma (HCC) and serum concentrations of zinc and low serum concentrations of zinc in patients with HCC has been frequently examined in the literature.
In our previous study on patients with C-viral chronic liver disease, serum zinc concentration decreased with the progression of liver disease. Serum zinc concentration was correlated with levels of albumin and platelet counts. Furthermore, the concentration of serum zinc was lower in patients aged above 60 years than in those aged below 60 years. Serum zinc concentration is also related with the occurrence of HCC.
Supplementation therapy of zinc is necessary for patients with C-viral chronic liver disease and low zinc concentration to lower the risk of HCC occurrence and improve outcomes.

S-1-6
Zinc Supplementation in the Geriatric Hospital

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Serum zinc level decreased with advancing age. Although the normal range of serum Zn is 84-159μg/dL, hypozincemia below 65μg/dL is observed in 20% of inhabitants in the epidemiological study in USA and in Japan. Serum zinc level in the hospitalized patients at the Geriatric word is significantly lower than that in the healthy old people.
In our study, the mean serum zinc concentration and the standard deviation (m ± SD) did not differ between young and old if the multiple blood analysis is in normal range.
It means that the decreased serum zinc level in the advanced age indicates merely based on malnutrition. Serum zinc correlates well to the serum albumin and hemoglobin.
In the geriatric hospital hypozincemia below 30 is often seen in the bed-ridden patients . After 6 months Zn supplementation with 34mg daily, serum Zn concentration elevated to 100μg/dL in the effective cases. Incidence of pneumonia decreased and the decubitus ulcers showed the healing tendency with Zn supplementation. In these cases serum Cu decreased prominently. Further studies are necessary about the copper and zinc metabolisms in the supplementation therapy for the zinc deficiency.

S-1-7
Zinc in Alzheimer's disease

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Fatigue of extracellular Zn^{2+} homeostasis may underlie amyloid pathology in Alzheimer's disease (AD). Zn released upon glutamatergic neurotransmission is concentrated in presynaptic terminals by ZnT3, whose ablation rescues amyloid pathology (in a transgenic AD model), but also accelerates age-dependent cognitive decline[1]. Presenilins, whose mutations cause familiar AD, mediate a large fraction of zinc uptake into all cells[2], and the impact of mutations on zinc uptake as a factor driving amyloid pathology is being evaluated. PBT2 is a zinc ionophore that restores cognition and decreases brain Aβ in APP transgenic mice within days[3]. Phase IIa data for mild AD demonstrated a reduction in CSF Aβ42 with significant improvement in cognitive performance within 12 weeks. The speed of these improvements suggested that PBT2 may also be correcting other aspects of synaptic dysfunction in early stage disease, such as the trans-synaptic movement of zinc. Indeed, oral PBT2 treatment of aged and cognitively impaired wildtype mice (C57Bl/6) with no amyloid burden caused almost complete normalization of Morris water maze performance within 11 days (ANOVA $p < 0.001$), and selectively elevated zinc levels in the hippocampal CA1. PBT2 both decreases amyloid load, and restores zinc homeostasis, leading to improved synaptic function.

S-1-8
Zinc transporters and Aging: Closing Remarks

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Coordinated zinc mobilization across the cellular membrane by zinc transporter proteins is indispensable for controlling a number of physiological events. In human, over 20 zinc transporters play pivotal roles for maintenance of zinc homeostasis (Fukada T and Kambe T, Metallomics, 2011, 3, 662-674). Expression changes of many zinc transporters, including increased or decreased expression, ectopic expression and mislocalization, have been suggested to be involved in a variety of diseases processes, and actually, loss-of-function mutations of five zinc transporters genes have been shown to result in genetic disorders. Therefore, changes of their expression pattern, which are associated with aging, may be involved in the development of many chronic diseases seen in the elderly, such as presented in this symposium. I will review zinc transporters functions in human, and briefly discuss how zinc contributes to good health in the elderly, from the perspective of zinc transporters and aging.

Fukada and Kambe, "Molecular and genetic features of zinc transporters in physiology and pathogenesis" *Metallomics* (2011) 3: 662-674

S-2-1
Mechanisms of metallothionein gene transcription and epigenetic changes in the promoter

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Mammalian metallothionein (MT) genes are transcriptionally activated by the essential metal zinc as well as by environmental stresses, including toxic metal overload and redox fluctuations. In addition to playing a key role in zinc homeostasis, MT proteins can protect against metal- and oxidant-induced cellular damage, and may participate in other fundamental physiologic and pathologic processes such as cell survival, proliferation, and neoplasia. Metal response element (MRE)-binding transcription factor-1 (MTF-1) plays an essential role in the activation of MT gene transcription. Previously, zinc has been shown to induce the formation of a co-activator complex containing MTF-1 and the histone acetyltransferase p300 which plays an essential role in the activation of MT1 gene transcription. We showed that zinc induced epigenetic changes in chromatin of the MT1 promoter. It is possible that the epigenetic changes modulate of MT gene transcription. In my talk, I will discuss the mechanisms of MT gene transcription and the epigenetic changes.

S-2-2
Possible inhibitory function of Metallothionein in Obesity

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Adipose tissue is a primary site of energy storage in the body and an important endocrine organ. Changes in adiposity, an excess obesity, have been known to lead obesity-related comorbidities such as type 2 diabetes and cardiovascular diseases. Thus factors to regulate adiposity would be useful tool to protect obesity-related metabolic diseases.
We have previously reported that female metallothionein–null (MT-/-) mice showed higher sensitivity to high fat diet (HFD)-induced obesity as compared with wild type (WT) mice (1). In order to find out the new strategy for manipulation of obesity-related diseases, we have investigated the potential function of MT to regulate adipose tissue generation and adipocyte differentiation.
The results suggest that both MT and androgens have an ability to inhibit the HFD-induced obesity. Also MT has an inhibitory effect on adipogenic differentiation in preadipocytes (ADCs).
In conclusion, those results revealed that the modulators of MT function might regulate the fat tissue homeostasis.
(1) Sato M, et al, Faseb. J. 2010; 24, 2375-2384.

S-2-3
Enzyme-linked immunosorbent assay for metallothionein- III in humans and experimental animals.

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We have developed an easy and specific enzyme-linked immunoassay (ELISA) for the determination of metallothinein-3 (MT-3) in human and experimental animals for the research of heavy metal, using a specific monoclonal antibody raised against human recombinant MT-3 (rMT-3). The epitope mapping of the antibody was conducted using mouse, rat, and human MT-3s and peptide fragments of human MT-3. MT-1/2, MT-3 knock-out (KO) mice and human brain and liver were used for the evaluation of the ELISA. The human MT- 3 NH2 terminal peptide (Fr. 1-17) was the demonstrated epitope of this antibody. The reactivity of this ELISA in brain homogenate of MT-3 KO mouse was significantly low compared with the wild type and MT-1/2 KO mice. An effective pretreatment procedure of the tissue homogenate was also established for this MT-3 ELISA. In conclusion, this competitive ELISA is an easy and specific method for measuring the brain and other tissue MT-3 levels in experimental animals and in humans.

S-2-4
Significance of urinary metallothionein in Japanese farmers exposed to cadmium through consumption of home-harvested rice

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There are mine-derived cadmium (Cd)-polluted areas in Akita prefecture, Japan, where farmers have been exposed to Cd through home-harvested rice consumption. We measured urinary metallothionein (MT) levels in these farmers and investigated their significance in Cd toxicity. After obtaining informed consents, we conducted health examinations on them in 2010–2011 and obtained blood and urinary samples (39–93 years old, 336 males and 417 females). Urinary MT had a significant positive relationship with urinary Cd, but not with age, blood Cd, blood lead, and urinary arsenic in both males and females. In addition, urinary alpha 1-microglobulin and beta 2-microglobulin, indicators for renal tubular function, generally showed closer relationships with urinary MT than with blood or urinary Cd. These results indicate that urinary MT increases along with both Cd-induced MT production and renal tubular dysfunction. Furthermore, cases of advanced renal tubular dysfunction were only observed among farmers with low MT producers, suggesting that they are at high risk of Cd-induced renal injury.

S-2-5
Plasma metallothionein I/II in liver diseases

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We have previously developed an easy and specific competitive ELISA for determining metallothionein I and -II (MT I/II) simultaneously in serum or plasma and other biological specimens of human and experimental animals (1).
MT I/II levels in chronic active hepatitis C patients were significantly lower compared to normal controls. Among the chronic hepatitis cases, MT I/II levels increased gradually followed by the progression of the disease such as liver cirrhosis, hepatocarcinoma. In particular, we have found significantly higher MT I/II levels in Wilson disease patients which were very similar with the plasma levels of LEC rat (model animal of Wilson Disease).
Further we have found the positive correlation between MT I/II and the following plasma parameters; bilirubin, γ-GT, ZTT, blood glucose, HbA1c in liver disease cases.
In particular, MT I/II was found to be a significant diagnostic marker of Wilson Disease.

(1) Nakajima et al . Clinica Chimica Acta 411 (2010) 758–761

S-3-1
Dietary intake of inorganic arsenic in the Japanese

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We estimated dietary intake of inorganic arsenic, a naturally occurring carcinogen, in the Japanese using a market basket survey and probabilistic estimation.
An LC-hydride generation-ICP-MS system was used to measure inorganic arsenic concentrations in 19 food composites prepared from 159 food items purchased in Shizuoka, Japan to estimate inorganic arsenic intake from 19 food categories. Cereals and seaweeds, in particular, rice and *hijiki*, comprised the major sources of inorganic arsenic, at a combined 90% of daily intake (21 μg As/day).
The above measurements were used to estimate the distribution profile of inorganic arsenic intake in Japanese people using a probabilistic approach, which considers variability in inorganic arsenic concentrations and daily consumption amounts for rice and *hijiki* in the Japanese. The 50th and 95th percentile estimates for inorganic arsenic intake were 19 and 65 μg As/day, respectively. We confirmed that inorganic arsenic intake levels in Japanese people represent a significant cancer risk.

S-3-2
Arsenic exposure as associated with carotid intima-media thickness and cardiovascular disease risk: the effects of dietary pattern modification

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No previous studies have evaluated the effects of dietary pattern modification with regard to the association between arsenic exposure from drinking water and subclinical or clinical endpoints of cardiovascular disease (CVD). The present study was a case-cohort study nested in the Health Effects of Arsenic Longitudinal Study in Bangladesh, and assessed the combined effects of well arsenic and dietary patterns with regard to the risks of CVD and heart disease. We conducted cross-sectional analyses in 959 subjects to evaluate whether the association between arsenic exposure and carotid intima-media thickness (cIMT) varies by dietary pattern. Urinary arsenic and cIMT showed stronger correlations in those who adhered less to a balanced diet, i.e., those who adhered more closely to a gourd vegetable diet or a Western diet. Moreover, the effects of well arsenic and the Western diet were additive in terms of increasing the risk of CVD and heart disease. Dietary patterns may influence associations between arsenic exposure and subclinical and clinical endpoints of CVD.

S-3-3
Distribution of arsenic species in brown, polished and milled rice

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The concentration of total arsenic (As) and As species, namely arsenite [As(III)], arsenate [As(V)], monomethylarsonic acid (MMAA) and dimethylarsinic acid (DMAA), in 10 rice flour samples and the distribution in the rice grains from which the flour were made were determined by ICP-MS and HPLC-ICP-MS.
Portions of the brown rice were polished / milled to different degrees to yield milled off samples and polished rice samples. All samples were used for the determination of total As and As species. Recoveries and mass balances in all samples showed good agreements with the starting materials. As(III), As(V), MMAA and DMAA were detected in the samples, and the sums of the concentrations of all As species in the extract were 86 %–105 % of the total As concentration in each case.
The concentrations of the As species in polished rice decreased with increased milling away of the outer layers of the rice grains. There was a tendency for As(III) and As(V) to be present in the outer layers of the grains with DMAA compared with the inside.

S-3-4
Dietary exposure to arsenic in different ethnic communities in the United Kingdom

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Exposure to arsenic, including inorganic arsenic, can be significant in populations that consume large quantities of rice. We investigated the relationship between arsenic exposure and diet in different groups in the United Kingdom. We monitored urinary arsenic concentrations in different ethnic groups including Asians, African and Europeans. The Bangladeshi s, who are the largest consumers of rice, show higher concentration of urinary inorganic arsenic. To reduce arsenic exposure from rice consumption, we analysed different types of rice from Bangladesh and other regions of the world to identify varieties that have low arsenic content. These studies revealed that certain varieties of aromatic rice from Bangladesh contained much lower concentration of arsenic and relatively higher concentration of selenium and zinc compared to non-aromatic rice. Our studies suggest that exposure to inorganic arsenic from rice is a problem in some communities and this can be reduced by lowering their intake of rice and/or consuming certain types of aromatic rice that are not only low in arsenic but have higher concentrations of selenium and zinc.

S-3-5
Implication of relationship between arsenic toxicity and arsenic metabolites

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Although some organoarsenicals, such as dimethylarsinic acid (DMA^V), arsenobetaine (AB), arsenocholine (AC), and trimethylarsine oxide (TMAO) are found in seafood, information on the metabolism of organoarsenicals in mammals is limited as compared to inorganic arsenicals. In the present study, we investigated tissue distribution, metabolism, and excretion of organoarsenic compounds in rats. The rats were fed on arsenic-depleted rodent chow to decrease the basal level of tissue arsenic contents. Arsenic concentration in RBCs increased remarkably only in DMA^v-treated group. Approximately 35%, 72%, and 61% of the dose was excreted in urine and feces in 2 days in DMA^V-, AB-, and AC-treated groups, respectively, while virtually the entire dose of TMOA was excreted in urine in 1 day. Reduction of organoarsenicals is implicated in the deposition of arsenic in tissues.

S-4-1
Dietary Zinc and Zinc Deficiency

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Humans digest animal flesh and absorb it's zinc. Red meat is a common source. White meat is relatively low in zinc. Plant seeds (grains, legumes, nuts) contain more zinc than other plant parts, and are rich phytate. At alkaline pH phytate, some dietary fibers, and Maillard browning products form complexes that humans are unable to digest. Milling and/or fermentation of remove phytate and dietary fibers. Milling also removes zinc, but fermentation does not. Populations that consume a wide variety of foods have a lower risk of dietary zinc deficiency. Risk increases when food variety is limited, and grains, legumes and nuts are primary sources of protein. Zinc deficiency impairs functions (e.g., growth, immunity, neuropsychological performance) before common laboratory tests such as plasma zinc become abnormal. Diagnosis of zinc deficiency before clinical signs become obvious requires an understanding of habitual food frequencies, iron status, and the likely presence of other deficiencies. These relationships became evident from randomized controlled zinc treatment trials.

S-4-2
Patients at risk for trace element deficiencies: bariatric surgery

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Obesity is a worldwide epidemic associated with diseases such as diabetes mellitus and cardiovascular disease. Current methods for weight loss are not very effective, particularly for those with morbid obesity. Surgical therapy may be recommended for those with a BMI >40, or >35 with co-morbidities. It can produce significant weight loss and improve/ resolve co-morbidities including diabetes, hypertension and hyperlipidemia. Yet successes are tempered by adverse effects on trace element absorption and status. A PubMed literature search will identify studies for inclusion in a meta-analysis. Publications from January 1980 to February 2013 that contain key words 'bariatric surgery or gastric bypass,' 'trace element or mineral or zinc or iron or copper or iodine or manganese', and 'absorption or status or rate or level' will be used. Inclusion criteria will be human markers that reflect changes in trace element status before/after bariatric surgery. Appropriate trace elements supplements that should be recommended after specific types of bariatric surgery will be addressed.

S-4-3
Female athletes: A population at risk of mineral deficiencies and associated performance decrements

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Adequate mineral status is essential for optimal human performance. Female athletes are at risk for mineral insufficiency due to inadequate dietary intake, menstruation, and inflammatory responses to heavy physical activity. Recent studies from our laboratory and others have documented poor iron status and associated declines in both cognitive and physical performance in female athletes. Similarly, poor vitamin D and calcium status have been observed in female athletes. Insufficient vitamin D and calcium status may be associated with injuries, such as stress fracture, which may limit a female athlete's ability to participate in regular physical activity. This presentation will focus on recent studies detailing the prevalence of poor mineral status in female athletes, as well as associated decrements in human performance. Factors associated with declines in mineral status during physical training, including the inflammatory response, will be reviewed. Finally, countermeasures for the prevention of inadequate mineral status will be described.

S-4-4
Estimation of iron requirements by numerical analysis of population-based indicators

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Iron deficiency is one of the most prevalent nutritional diseases worldwide. Recommendation of dietary references for iron is crucial to protect people at risk of iron deficiency. A numerical method for estimating iron requirements has been developed using the population-based nutritional indicators: a prevalence of inadequate iron status and a distribution of iron intake in a certain population. Inadequate iron status is prevalent in Japan. Thus, the iron indicators obtained from the National Health and Nutrition Surveys 2003 to 2007 held annually by the Japanese government were analyzed. Less than 30 ng/mL of serum ferritin was considered as indicating inadequate iron status (iron depletion). For 18-29 years old women, median iron requirements were from 7.4 to 8.5 mg, iron intakes covering up to 80 mL menstrual blood loss per cycle were 10.9 to 12.6 mg and iron intakes covering 97.7% of the population were 13.5 to 15.6 mg. For 30-49 years old women, median iron requirements were from 8.4 to 9.1 mg, iron intakes covering up to 80 mL menstrual blood loss per cycle were 12.4 to 13.4 mg and iron intakes covering 97.7% of the population were 15.3 to 16.5 mg.

S-4-5
Physiological roles of copper in mammalian iron homeostasis

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Body iron levels are controlled by modulation of iron absorption in the proximal small bowel, as no regulated means to excrete iron exist. Moreover, iron deficiency results in adverse physiological outcomes, and as such, iron absorption is enhanced during states of deficiency. Upregulation of iron absorption occurs via transactivation of genes encoding iron transporters by a hypoxia-inducible transcription factor (Hif2α). Coincident with increased iron absorption, copper levels are elevated in the duodenal mucosa, liver and serum. Consistent with this, copper homeostasis-related genes are activated in duodenal enterocytes during iron deficiency, including those encoding intracellular copper binding proteins (metallothionein 1/2) and a copper export protein (Menke's copper-transporting ATPase [Atp7a]). It was further noted that *Atp7a* gene transcription is co-regulated with iron transport-related genes by Hif2α, providing a mechanistic explanation for alterations in copper transport during iron deficiency. These discoveries illustrate novel iron-copper interactions, supporting the hypothesis that copper is critical for proper control of body iron metabolism.

S-5-1
Inductively Coupled Plasma Time-of-Flight Mass Spectrometry for Single Cell Analysis for Immunological and Functional Assay

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Flow cytometry is an essential tool for dissecting the functional complexity of hematopoiesis, and this technique uses fluorescence from fluorophore and requires compensation due to spectral interference when more than 5 antigens are simultaneously measured.

A next-generation single-cell cytometry based on inductively coupled plasma time of flight mass spectrometry, ICP-TOF-MS, has been developed recently, which detects metal ions instead of fluorophore. Because of better spectral resolution and abundance sensitivity, ICP-TOF-MS doesn't require compensation. A special polymer for "mass cytometry" has also been developed to tag metallic elements to antibodies. More than 30 different enriched isotopes of lanthanide can be tagged to almost any antibody, including both cell surface and intracellular probes.

We report here the use of mass cytometry for the simultaneous sub-population and functional (response to agonist) assay of PBMC samples comparing conventional data analysis and multi-dimensional data analysis.

S-5-2
Demonstration of high selectivity/sensitivity of ICP-QQQ in challenging applications

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ICP-MS is widely used for trace element analysis due to its excellent analytical performance characteristics including wide dynamic range, low detection limits and multi-element analysis capability. However, in real sample analysis, spectral interferences often make it difficult to achieve the full potential of low detection limits. Most modern ICP-MS instruments use collision/reaction cell technology to remove or alleviate these interferences; collision cell mode discriminates analyte ions from interfering ions by virtue of their different sizes; and reaction cell mode removes interferences using chemical reaction. To date, reaction cell mode has been used in quite limited applications because the reaction chemistry in the cell of conventional, single quadrupole ICP-MS is not well controlled, often leading to erroneous results.

This problem is now solved with the new triple quadrupole ICP-MS (ICP-QQQ), which operates in MS/MS and provides a new reaction cell capability that enables higher selectivity and sensitivity compared to conventional ICP-QMS. I will present some results from the analysis of complex matrices that were acquired using reaction cell mode of the ICP-QQQ.

S-5-3
High Sensitive Elemental Analysis of Single Yeast Cells by Time-Resolved Inductively-Coupled Plasma Mass Spectrometry using a High Efficiency Cell Introduction System

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Single-cell elemental analysis has gained substantial interest in the biochemical and metallomics fields, because elemental composition of single cells and their distribution can also reveal information about their health and growth state, for example Zn and Cu imbalances in brain tissues.
A time-resolved inductively coupled plasma mass spectrometry (ICP-MS) is one of the powerful tools for single cell element analysis. If a suspension of cells is directly introduced into the plasma and the moment that cell decomposed inside the plasma is measured with a low integration time, a high density ions plume corresponding to a single cell event can be detected.
We developed a high efficiency cell introduction system (HECIS) for the time resolved ICP-MS measurement. We will show the analytical results for single yeast cells.

S-5-4
Elemental Distribution and Chemical Speciation by Synchrotron Radiation X-ray Fluorescence Analysis

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Some plants are known to contain high amount of heavy metals such as Pb, Cd, and Cu in their bodies. This trait has the potential to be exploited in phytoremediation, a soft method in which plants are used for the cleanup of heavy metal-polluted soils. Since heavy elements such as Cd is a highly toxic element for plants, it is interesting to reveal the transport and accumulation mechanisms of toxic elements in plants. We are engaged in applying the synchrotron radiation X-ray fluorescence analysis to several kinds of plants accumulating toxic elements. X-ray absorption fine structure (XAFS) analysis can directly elucidate the elemental oxidation states in plants with minimal sample preparation. X-ray fluorescence (XRF) spectroscopy using a synchrotron radiation (SR) microbeam is powerful tool to analyze the cellular scale of elemental distribution. We here introduce the accumulation of Cd in rice plants and tobacco BY-2 cultivated cell, and also that of Cr in fern of *Pteris vittata* L.

S-5-5
New metal tag for highly selective and sensitive analyses by HPLC/ICP-MS

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Amino acids are the important components of proteins and are essential for life. They are obtained from the diet and are utilized throughout the body. In addition, amino acids work as hub compounds in metabolic pathways, cell signaling, and other cellular phenomena. Therefore, the analytical method for amino acids is a fundamental requirement. However, the volumes of amino acids in biological samples are usually very small.
Then, we have established a highly selective and sensitive amino acid analytical method, using reversed phase HPLC/ICP-MS. We derivatized amino acids with the metal tag reagent. The derivatized amino acids were detected by ICP-MS. Our metal tag reagents have the different property to the major metal tag for analysis of protein; ex. DOTA = 1, 4, 7, 10-tetraazacyclododecane –N, N', N'', N'''-tetraacetic acid. They are more hydrophobic. They are retained in the reverse phase column and suitable to analyze low molecular weight compounds.
We will present our new metal tag and application data for biological samples.

S-6-1
Effects of arsenic on immunological functions

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Arsenic is an environmental pollutant derived from the Earth's crust. Many people around the world are exposed to arsenic-polluted underground water from wells and suffer from chronic arsenic poisoning, which causes cancer in multiple organs. This is considered to be the most critical problem in arsenic-contaminated areas. On the other hand, dysfunctions of immune systems lead to the survival and progression of tumor cells. Among the immune systems, NK cells and T cells play an important role in tumor immunity; NK cells have cytotoxic activities to tumor cells, which are activated by the cytokines released from T cells. In order to investigate the mechanisms underlying the carcinogenic actions of arsenic, we examined the effects of arsenite on the functions of NK cells and T cells. In this symposium, we will introduce the influence of arsenite on the functions of the cultured cell lines of NK and T cells. We also prepared primary NK and T cells from the spleen of arsenite-exposed or control mice, and examined the functions of these cells.

S-6-2
Structure activity relationships in organotin-induced toxicity via retinoid X receptor signaling pathway

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We previously reported that tributyltin and triphenyltin are potential endocrine disruptors to act as retinoid X receptor (RXR) agonist. However, the structure of these organotin compounds is entirely distinct from established RXR ligands such as 9-cis retinoic acid and rexinoids. To extend our knowledge of the correlation between the structures of organotins and their RXR agonist activity, we here assessed RXR agonist activity of various organotin compounds using human RXRα (hRXRα) mutants with altered ligand binding potential. As a result; 1) the trialkyl or triaryl substituents are required for organotins to act as RXR agonists; 2) R316 of hRXRα, which is critical amino acid residues for rexinoids to interact with hRXRα, is not required for agonist activity of trioganotins; 3) C432 of hRXRα is critical for transactivation by triorganotins. Our results suggest that triorganotins and rexinoids have distinct ligand-binding properties of RXR. In the symposium, I will discuss the relationship between potential toxicity of organotin compounds as endocrine disruptors and their ligand-binding properties of RXR.

S-6-3
Novel lipid peroxidation dependent cell death by deficiency of selenoprotein GPx4

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Selenium is an essential trace element for cell growth and human health. Severe selenium deficiency in human and cultured cells induces heart disease (Keshan disease) and cell death. GPx4 is one of selenoproteins and an intracellular antioxidant enzyme that directly reduces phospholipid hydroperoxide produced by oxidative stress. To clarify the physiological and pathological role of GPx4 and its substrate phospholipid hydroperoxide, we made several tissue specific GPx4 KO mice. GPx4 deficient tissue cells (late spermatocyte, retina and cardiac cells) induce cell death and each GPx4 deficient mice display male infertility, retina degradation and cardio sudden death. To investigate the mechanism of cell death induced by depletion of GPx4, we used tamoxifen inducible GPx4 depleted MEF cells. Tamoxifen treatment induces generation of phospholipid hydroperoxide until 24hr and cell death at 72hr. Vitamin E suppressed this cell death. Our detail experiments demonstrated that cell death induced by depletion of GPx4 is not apoptosis, necrosis and autophagic cell death. These results demonstrated that deficiency of GPx4 induce novel cell death caused by lipid peroxidation.

S-6-4
Identification of novel tellurium metabolites in selenium-accumulating plants

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Tellurium (Te) is a widely used metalloid in industry because of its unique chemical and physical properties. However, information about the biological and toxicological effects of Te in plants and animals is limited. To reveal the metabolic pathway of Te in plants, garlic, a well-known selenium (Se) accumulator, was chosen as the model plant. Garlic was hydroponically cultivated and exposed to sodium tellurate, and Te-containing metabolites in the water extract of garlic leaves were identified. At least three Te-containing metabolites were detected by HPLC-ICP-MS, and two of them were subjected to HPLC-ESI-MS-MS for identification. The MS spectra obtained by ESI-MS-MS indicated that one of the metabolites was *Te*-methyltellurocysteine oxide (MeTeCysO). The other was assigned as cysteine *S*-methytellurosulfide. These results suggest that garlic can transform inorganic tellurate into a Te-containing amino acid, the so-called telluroamino acid. This is the first report addressing that telluroamino acid is *de novo* synthesized in a higher plant.

S-7-1
The relationships between Cu intakes and serum Cu levels in inpatients

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In this study 293 inpatients (103 males, 190 females) were recruited, whose biochemical data were obtained at fast early in the morning.
In 85 inpatients below 30 µg/dl of serum Cu there was a significantly positive correlation between serum Cu levels and leukocyte counts.
The selection of enteral liquid foods was limited to ones with low Cu contents 15 years ago in Japan. From about 0.1 mg to 0.6 mg of daily Cu intakes, serum Cu increased parallel to Cu intakes, and then leveled off until 1.2mg of daily Cu intakes. Serum ceruloplasmin(Cp) showed the same tendency. From these cross point adequate intake was calculated, being 0.68mg/day. Nomocupremia in either 18 males or 30 females during 4.3-4.7 months, showing daily Cu intakes of 0.686 mg in males and of 0.584 mg in females.
Severe Cu deficiencies were influenced by daily Cu intakes, indicating the importance to ingest adequate amount of Cu in enteral liquid foods.

S-7-2
Equilibrium copper intake(ECuI) estimated by human mineral balance study.

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To determine the copper (Cu) intake required to maintain Cu balance, we investigated the relationship between Cu intake and excretion for young female Japanese in 13 balance studies (n=131, 20±1 y, 160 ±5 cm, 51.9±5.0 kg) conducted at NIHN. Durations of balance periods ranged between 8 and 12 ds. Dietary Cu intake ranged between 1.05 and 2.54 mg/d. Urine output of Cu was low and relatively consistent among the first three experiments, and thereafter, urine output of Cu was presumed to be 0.05 mg/d. Balance distribution medians for Cu was positive, so the data were adjusted to set the medians of the balance to zero. Intake and balance for Cu were divided by body weight (BW), lean body mass (LBM), and standard body weight (SBW), which was calculated using height and standard body mass index measurements (BMI=22), and ECuI was calculated as the intercept of a simple regression equation. ECuI was 30.9 µg/kgBW/d, 42.6 µg/kgLBM/d, and 29.7 µg/kgSBW/d, respectively.

S-7-3
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If liquid food with poor copper is used for long period in enteral nutrition, anemia and white corpuscle reduction appears as copper deficiency. When we supplemented with copper using pure cocoa, copper deficiency improved. As copper one-day dose using pure cocoa, we presumed 0.6mg. Because copper may rival zinc in intestinal absorption, we consider 1 to 10 for copper zinc ratio to be standard. When inflammatory reaction serves as backdrop, serum copper becomes high and serum zinc becomes low. We tried two studies. First, they were cases of enteral nutrition sent to hospital for infection. When they were treated with antimicrobial drugs, and inflammatory reaction became low, serum copper fell and serum zinc rose. The next was the same result in oral diets. When using serum copper and zinc levels for the nutritive evaluation of acute period, it is necessary to take this phenomenon into consideration.. By carrying out zinc supplement with zinc-rich gastric ulcer medicine, copper deficiency was shown, reducing the dose of the medicine and noticing the disorder of absorption in copper deficiency.

S-7-4
Influence of serum concentrations of copper, content ratio of zinc and copper in products of liquid nutrient under long term enteral nutrition.

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Copper and zinc are known to be antagonistic in absorption. In Dietary Reference for Japanese(enacted in 2000), the ratio of copper and zinc was 1:6. Contents of liquid nutrient were adjusted by this reference. There were many reports with high serum copper in user of liquid nutrient confirming this reference, without elevated serum zinc. High ratio of copper/zinc was assumed. Absorptions of copper and zinc in digestive tract are conflict. The ratio of copper and zinc was modified 1:10 in Dietary Reference, 2005. By readjusting by the reference, high serum copper depleted in enteral nutrition. Serum trace elements are influenced by serum protein. Serum trace elements have to be considered about malnutrition, inflammation and BMI. Recently, liquid nutrients with enough trace elements are developed to prevent trace elements deficiency. When suffering from pressure sores, zinc-rich supplemental foods act effectively cure. Excessive zinc intakes lead to copper deficiency because of antagonistic absorptions.

S-8-1
A Biochemical and Molecular Approach in the Reversal of antituberculosis drugs Induced hepatotoxicity

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Hepatotoxicity is one of severe implications caused by antituberculosis drugs (ATD). This investigation focused on the evaluation of defensive role of *Nigella sativa* (NS) against liver injury caused by ATD. Albino rats were divided into six groups of six each. Group 1 served as control. Groups 2-6 were administered ATD for 8 weeks and group 2 served as experimental control. Groups 3, 4 and 5 were administered NS at the doses of 125, 250 and 500 mg/kg, *p.o.* respectively for 8 weeks. Group 6 was administered silymarin (50 mg/kg). Serum LFTs and KFTs were elevated in serum after ATD administration. ATD significantly increased LPO, TNF-α, IL6, DNA damage and decreased the activities of antioxidants and GSH cycle enzymes, i.e., SOD, CAT, GR, GP_X and G6PDH in liver. CYP 2E1 showed sharp depletion as assessed by estimating AH activity after ATD exposure. NS recouped enzymatical activities and DNA damage. NS could inhibit oxidative stress by suppressing LPO and augmenting antioxidant enzymes towards control. Thus, results of this study indicated excellent hepatoprotective activity of NS.

S-8-2
Therapeutic potential of trace elements supplemented with Spirulina against Beryllium toxicity

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Trace elements iron (Fe) and magnesium (Mg) are the key nutrients in dietary sources to treat a wide variety of diseases. Beryllium (Be) is one of the most toxic elements causes tissue damage. This study focused on hepatoprotective activity of *Spirulina* Extract (SE) monotherapy and in combination with Fe and Mg. Be was administered to albino rats at a dose of 1 mg/kg (*i.p.*) for 28 days, followed by monotherapy of SE at 200 mg/kg (*p.o.*) and in combination as SE and Fe, SE and Mg, SE with Mg and Fe (7mg/kg, *p.o.*) for 7 consecutive days. Be significantly depleted Hb, blood sugar and uric acid whereas it enhanced serum protein, bilirubin, urea and creatinine. Administration of Be also caused a significant rise in LPO level and a fall in GSH, SOD, catalase, ALP and G6pase in the liver and kidney depicting failure of the antioxidant defense mechanism it was also supported by histopathological studies. AAS also revealed high concentration of Be in liver and kidney. Thus, it can be concluded that combined treatment along with trace elements could be a better treatment for Be than monotherapy.

S-8-3
Amelioration of gold nanoparticles against acetaminophen induced hepatorenal toxicity in rats

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The therapeutic benefits of gold preparations have been reported from as early as 2500 BC. The aim of this study was to evaluate the therapeutic efficacy of gold nanoparticles (AuNP) against acetaminophen (APAP) induced toxicity. Albino rats were administered APAP at a dose of 2 g/kg *p.o.* once only. After 24 hours of APAP administration, animals were administered AuNP at a dose of 100 µg/kg *p.o.* and silymarin at a dose of 50 mg/kg *p.o.* once only. APAP administered group showed significant increase in AST, ALT, LDH, cholesterol, bilirubin, albumin, urea, uric acid and creatinine in serum which indicated liver and kidney damage. A significantly increased LPO and a decreased level of GSH, SOD, catalase and G6pase after APAP exposure indicated oxidative stress in liver and kidney. Administration of AuNP reversed above variables significantly towards normal level. Thus it can be concluded that AuNP played beneficial role in reducing APAP induced toxicity and after an adequate amount of research it can be used in the development of drug against hepatorenal damage.

S-8-4
A study on acrylamide induced biochemical alterations

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Acrylamide (ACR) is found in various deep-fried and oven-baked foods and has become a worldwide concern because of its generation in a variety crisps, bread and biscuits. ACR is associated with disturbances in the oxidative stress and hepatotoxicity. The present study was carried out to investigate the biochemical and histopathological effects of ACR on female albino rats. Animals were divided into two groups of six animals each. Group I was served as control group that received a daily oral administration of distilled water while Group II received ACR at the dose of 41mg/kg b.w., intragastrically for ten days regime. Levels of AST, ALT, urea, uric acid, creatinine, LPO were significantly increased whereas SOD, CAT and GSH were depleted due to toxic effect of ACR indicated oxidative stress. It can be concluded that ACR when given orally produced significant toxicity. This is also evident by histopathological examinations which showed hydropic degeneration of hepatic cells, hypotrophied Kupffer cells, mild necrotic changes in the hepatic cells.

S-8-5
SILICA induced occupational hazard in India

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Silicosis is occupational disease caused by mining, tunnelling, sandstone grinding etc. Epidemiological study was conducted to investigate the occupational health manifestations among the stone crushing workers of Gwalior region,(India). Exposed workers had significantly higher rate of radiographic abnormalities.Workers had higher prevalence of cough, wheezing and shortness of breath. Significant fall in pulmonary function test's Forced vital capacity (FVC), Forced expiratory volume (FEV_1), FEV_1/FVC%, Peak Expiratory Flow (PEF), FEF between 25% and 75% of FVC ($FEF_{25-75\%}$), abnormalities in chest radiographs and raised serum ACE level were noted in silica exposed group. Chest radiography revealed prominent bronchovascular markings with tiny nodular opacities in lung zones. Thus exposure to SiO_2 dust in subjects confirms the presence of pulmonary fibrosis. Further,study was also designed to examine the ability of Glycyrrhiza glabra(GG), to attenuate sub chronic SiO_2 induced injury in the liver, kidney and lung of rats. The treatment with alcoholic extract of GG at 500 and 1000mg/kg significantly improved antioxidant status and lessened the DNA damage.

S-8-6
Influence of chelating agents with supplementation against Aluminum toxicity

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Aluminium is the most abundant metal ion in the biosphere and we are continuously exposed to it in our everyday life through food, beverages, pharmaceutical products etc This study aimed at evaluating the improvement of chelators therapy on histopathological and biochemical parameters in rats. Al (NO3)3 (5mg/Kg/day, i.p.) was administered to rats for 90 days, followed by therapy with NAC/HEDTA+Ca+Fe for 7 days. Significant rise were observed in the activities of serum ALPase, AST, ALT, creatinine and urea whereas serum protein was found to be decline after toxicant exposure. Results indicate that TBARS levels, was significantly higher and total glutathione content were significantly lower in liver and kidney toxicity. The decrease in activity of SDH, ATPase and G6Pase. Therapy showed over all improvement in all the biochemical changes. In the group treated with Al, degenerative changes were provoked cytoplasmic vacuoles, disorganized endoplasmic reticulum, pleomorphic shape of nuclei, swallowed mitochondria with degenerated outer membrane and vacuolization in liver and kidney. Treatment with HEDTA+Ca+Fe was more effective.

S-9-1
Disease Burden due to Trace elements Deficiency

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The possibility that some trace element deficiencies are associated with a reduced anti-oxidant potential in organisms (onset of cancer and atherosclerosis), accelerated aging, developmental retardation in children, an increased incidence of abnormal pregnancies, immunological abnormalities, and lifestyle-related diseases. Reproductive failure may be induced by deficiencies of single or combined trace elements and by imbalances. Selenium deficiency, like copper, iron and zinc deficiency, is also more likely to occur in premature birth infants. Copper deficiency is related to vascular defects such as aneurysms, heart enlargement, heart failure, and A deficiency of copper relative to zinc produces a decrease in HDL (high density lipoproteins) and an increase in LDL (low density lipoproteins). Arsenic, chromium, and nickel are said to contribute to the development of cancer based on the epidemiologic evidence, and beryllium, cadmium, chromium, cobalt, lead, nickel, zinc, and iron have increased risk of disease burden.

S-9-2
Dietary risk exposure to heavy metals among poor and non-poor households in Dhaka city, Bangladesh

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The cadmium, lead, arsenic, mercury, antimony, aluminium and lithium content of foods and beverages consumed by poor and non-poor households in Dhaka were analyzed by ICPMS. Eighty food samples were each collected from three markets that were commonly accessed by poor and non-poor households. Ninety four percent males from poor households and 78% males from non-poor households had an estimated dietary exposure to Cd above MPL. Eight percent males from poor households and 83% males from non-poor households had an estimated dietary exposure to Al above MPL, and were considered at risk from Al contamination. All males of poor and non-poor households likewise had dietary risk exposure to As and Pb. The dietary risk exposure to mercury, antimony and lithium by males from both poor and non-poor households was below MPL, thus noted to be within safe limits.

S-9-3
Zinc Deficiency among Children below Five in India

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The serum zinc deficiency in the children was studied for those of 6-60 months old in five states, Uttar Pradesh(Northern region), Karnataka (Southern), Orissa (Eastern), Gujarat (Western region) and Madhya Pradesh (Central) in Indea. In each state, all the districts with ICDS scheme were enlisted. One district was selected with the help of Random number table (RNT). In each district, all the ICDS projects were selected and one ICDS project was selected with help of RNT. In the selected ICDS project, a cluster 5 Anganwadi centres (AWC) were selected. Among these, 300 children of 6- 60 months were selected for the detailed study. Total number of the subjects was 1,655 (836 males, 819 females). The blood samples were collected from each child. The serum zinc level was analyzed by atomic absorption spectrophotometer. The overall prevalence of zinc deficiency in 5 states was 43.8% . The prevalence of zinc deficiency was highest in Orissa (51.3%), followed by Uttar Pradesh (48.1%), Gujarat (44.2%), Madhya Pradesh (38.9%) and Karnataka (36.2%). These results revealed that zinc deficiency highly prevails among the children belonging to Low-Socio-economic Index (LSI) in India.

S-9-4
Trace element content of foods and estimated intake by poor and non poor households in Dhaka

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Eighty food samples and beverages commonly consumed by poor and non-poor households in Dhaka were analyzed for selected trace elements. The intake of Fe, Mn, Cu, Zn, Mo and Ni by adults in poor and non-poor households was estimated using the Household Income Expenditure Survey 2010. Cereals were the primary contributors of dietary intake of Fe (55.45%), Mn (76.34%), Cu (71.28%), Zn (60.0%), Mo (53.85%) and Ni (76.95%). Vegetables were in second position to supply dietary intake of Fe (17.55%), Mn (7.94%), Cu (9.14%), Zn (9.16%) and Ni (13.62%). Pulses provided with 9.93% Fe, 9.81% Mn, 9.77% Cu, 9.88% Zn and 1.76% Ni of the dietary intake of trace elements. Diets of males from poor household are estimated to be more deficient in trace elements (Fe, Cu, Zn, Co and Se) than non-poor household males. The intake of Mn and Mo was higher than the Recommended Dietary Intake for both poor and non-poor households.

S-10-1
ATP7A-related copper transport diseases: emerging concepts and future trends.

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Involvement of ATP7A in axonal outgrowth, synapse integrity and neuronal activation underscores the fundamental importance of copper metabolism to neurological function. Severe defects in ATP7A cause Menkes disease, an infantile-onset, lethal condition. Neonatal diagnosis and early treatment with copper injections enhance survival in patients with this disease, and can normalize clinical outcomes if mutant ATP7A molecules retain small amounts of residual activity. Adeno-associated virus (AAV)-mediated gene addition plus copper rescues a mouse model of Menkes disease, suggesting a potential therapeutic supplement for patients with complete loss-of-function ATP7A mutations. Remarkably, a newly discovered ATP7A disorder, an isolated adult-onset distal motor neuropathy, has none of the characteristic clinical or biochemical abnormalities of Menkes disease or its milder allelic variant occipital horn syndrome (OHS), instead resembling Charcot–Marie–Tooth disease type 2. These findings indicate that ATP7A has a crucial but previously unappreciated role in motor neuron maintenance, and that the mechanism underlying ATP7A-related distal motor neuropathy is distinct from Menkes disease and OHS pathophysiology. Collectively, these insights refine our knowledge of the neurology of ATP7A-related copper transport diseases and pave the way for further progress in understanding ATP7A function.

S-10-2
Standard value of urine HVA/VMA ratio in neonates to screen for Menkes disease

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Menkes disease (MD) is a lethal disorder of copper metabolism. Early treatment with copper injections can improve outcomes. However, early diagnosis is difficult because clinical features are subtle in neonates. Previously, it was reported that low activity of dopamine β-hydroxylase, a copper-dependent enzyme, led to increase urine homovanillic acid/vanillylmandelic acid (HVA/VMA) ratio in MD patients, and the urine HVA/VMA ratio (cutoff: 4) was useful to screen for MD. However, there was no data in neonates. We examined the standard value of urine HVA/VMA ratio in neonates. 112 cases (age at 1-6 days) were enrolled and classified into the high(>4) and low(≤4) urine HVA/VMA ratio groups. Multivariate logistic analysis revealed ventilation was an independent risk factor of high urine HVA/VMA ratio (odds ratio 21.94; 95%CI: 2.82-247.03; p=0.004). Among 92 cases without ventilation, a mean of urine HVA/VMA ratios was 2.5±0.7. We established the standard value of urine HVA/VMA ratio in neonates which can be useful for early screening for MD.

S-10-3
PET imaging for copper bio-distribution and developing novel treatment for Menkes disease.

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Menkes disease is a lethal X-linked neurodegenerative disorder caused by dysfunctional ATP7A, a copper transporting protein. Copper deficiency displays characteristic clinical features such as kinky hair. Although subcutaneous injection of histidine-copper is a mainstay of treatment, clinical improvement remains limited. With the development of dedicated PET scanners such as microPET, it is now possible to perform functional imaging in small animals at high resolutions. By using this system, we quantified the abnormal distribution of copper in the Menkes disease mouse model. We postulated that copper chelating agents could correct the abnormal bio-distribution in the disease. Indeed, disulfiram, a lipophilic chelator, altered the copper distribution in the entire body and enhanced copper delivery to the brain. Furthermore, disulfiram corrected aberrant copper accumulation kinetics in the kidney. Our work strongly suggested copper chelating agents potentially act as a non-enzymatic transporter of copper to treat Menkes disease patients. Further development strategies to hunt more effective drugs will be discussed.

S-10-4
Changes in body weight and height in survivors of Menkes disease

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Changes in body weight (BW), height (BH), were examined in 40 male patients with Menkes disease (MD) from birth to the age of 1.6-4.7 years. All patients have been treated with copper-histidine by subcutaneous injection. Data were obtained from medical recorders. Comparing to those from the Nationwide Survey on BW and BH of normal children in Japan (2010), birth BW and BH values divided by gestational week in patients with MD did not differ significantly from those values in the 2010 survey data ($P > 0.05$). At the age of start of treatment, BW and BH were below the 3rd percentiles in 42.9% and 66.7% of patients, respectively. During the treatment period, BW and BH were below the 3rd percentile in 14/14 (100.0%) and 11/13 (84.6%) patients (included two patients with earlier start treatment), respectively. These results show that copper-histidine treatment could not improve BW and BH.

S-11-1
The Effect of Zinc on serum, breast milk retinol and zinc level in Maternal Postpartum

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A randomized double blind placebo controlled trial of a single dose of 200,000 I.U of vitamin A on maternal postpartum with daily Zinc supplementation was done in third semester of pregnant women. Third semester of pregnant women were randomized to receive zinc sulfate for six days a week and a single dose of 200,000 I.U. of vitamin A (n=12), and (n=12) placebo Third semester of pregnant women were evaluated weekly for nutrient intake, and serum zinc and retinol, breast milk zinc and retinol at three months. At the end of the study there were a significant increase in serum zinc level (p=0,000), breast milk zinc level (p=0,024), but no significant increase in serum retinol level (p=0,624) and breast milk retinol level (p=0,172) on maternal postpartum. These results suggest that vitamin A supplementation for maternal postpartum could not influence breast milk and serum retinol level. Zinc added vitamin A for post partum did not influence the breast milk retinol level but increased breast milk zinc level. Suggesting adding zinc distribution of high-dose vitamin A on maternal postpartum for retinol breast milk babies first 6 months.

S-11-2
Zinc and probiotic supplementation improve Zinc and Selenium status of Indonesian young children

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Zinc deficiency is quite common in developing countries, especially in Asia with the Eastern diet rich of phytate. Probiotic, zinc and their combination were supplemented to 12-24 months Indonesian children for 90 days; nutritional status (serum zinc, serum selenium and bodyweight) as well as immune response were assessed and compared with placebo. Microencapsulated *L. plantarum* IS-10506 was supplemented at 10^{10} cfu/day as probiotic.; 20 mg zinc sulfate monohydrate (8 mg zinc elemental) was supplemented as zinc. The results showed that supplementation of probiotic and zinc tend to increase the serum zinc and significantly increased serum selenium ($p<0.05$) of young children after 90 days supplementation as compared to placebo and also significantly increased bodyweight of the young children (($p<0.05$). Taken together, a combination of probiotic *L. plantarum* IS-10506 at 10^{10} cfu/day and 8 mg zinc elemental supplementation showed potential ability in improving nutritional status of pre-school children.

Keywords : zinc, probiotic *L. plantarum* IS-10506, dadih, pre-school children

S-11-3
The Effects of Iron/Folic Acid/Zinc Supplementation on Insulin-Like Growth Factor-1 Levels in Anemic Pregnant Women

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Zinc, a micronutrient associated with anemia and growth, is thought to potentially alleviate anemia in pregnant women and increase fetal growth. The present study aimed to clarify the mechanism responsible for this improvement by administering iron/folic acid/zinc supplements in pregnant women with anemia and documenting levels of Hb and Insulin-like Growth Factor-1 (IGF-1). This was a quasi experimental study with a pre-test and post-test non-randomized control group design. The treatment group comprised pregnant women with anemia who were administered supplements containing iron, folic acid and zinc (n=18; 60 mg, 0.25 mg and 50 mg, respectively). Subjects in the control group received supplements with only iron and folic acid (n=18; 60 mg and 0.25 mg, respectively). A significant difference in IGF-1 levels were found between groups ($p<0.05$). The control group showed improvement in anemic status and fetal growth through increases in Hb and IGF-1, while this was not observed for the treatment group. We conclude that increases in Hb contribute to increases in IGF-1 by as much as 36.5%.

S-11-4
The Effect of Zinc, Lysine and Vitamin A Supplementation to increase Cellular Immune Response of Pulmonary Tuberculosis Patients

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Tuberculosis is still found due to malnutrition's condition caused by deficiency among macro and micro nutrient. Thus a nutrient supplementation is necessary to accomplish all the required nutrients. This research was conducted at pulmonary Hospital (BP4) of Surabaya, with amount of sample as much as 30 pulmonary tuberculosis patients acid fast bacilli (AFB) + new. The design of the observation was using Randomized Pre Test Post Test Control Group Design, with supplied Double Blind as a treatment. Significant increase was observed in the T CD4+ cell count $p = 0.040$, IFN gamma levels $p = 0.036$ on pulmonary tuberculosis patient after supplementation zinc, lysine, and vitamin A in every day for two months. Significant increase of cellular immune response of pulmonary tuberculosis patients in the supplementation of zinc, lysine, and vitamin A due to zinc, lysine, and vitamin A was observed.

Keywords : Celluler immune response, pulmonary tuberculosis, zinc, lysine, vitamin A.

S-11-5
Effect of adding Zinc to Vitamin A on IGF-1, Bone Age and Linear Growth(H/A) in Stunted Children

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Stunted linked to zinc deficiency is found during gestation, and also in the newborn and children up to adolescence. Adding zinc to vitamin A supplementation on infection and linear growth, is hypothesized to have synergistic effect in decreasing infection and increase linear growth. A randomized double blind placebo controlled trial of 200,000 I.U of vitamin A with daily Zinc supplementation was done in children aged f48 to 60 months, they were randomized to receive 200,000 I.U. of vitamin A and zinc sulfate six days a week (n=12), and 200,000 of vitamin A (n=12) as placebo for six months. Children were evaluated for IGF-1, Bone Age and index Height for Age at six months. At the end of the study there were a significant increase of IGF-1 hormone ($p<0.04$), Z-Score Height for Age ($p<0.001$), and bone age ($p<0.01$) These results suggest that combining vitamin A with Zinc increased linear growth of children, thus may play a key role in controlling stunted of under five children.

KEY WORDS :
Zinc, vitamin A, IGF-1, Bone Age and z score Hight for Age

S-11-6
Nutritional Status as Risk Factor for Iron Deficiency among Women at Reproductive Age

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Underweight and iron deficiency is very prevalent in Indonesia. The association between underweight with some parameters of iron deficiency is still unclear. Hence, it is interesting to investigate the association between body mass index (BMI) and iron deficiency in young women. About 80 apparently healthy young women were randomly selected in a cross sectional study. Body weight and stature were measured, BMI was calculated and compared with age-specific BMI reference values for adolescent subjects. Underweight was defined as a BMI less than 5th percentile. Iron deficiency is represented by anemia (Hb<12g/dL) and iron depleted (serum ferritin less than 12 µg/L). Subject who are underweight is approximately twice to be anemia (OR:2.15; 95%CI: 0.74-6.17) and three times to be iron depleted (OR: 3.167; 95% CI: 0.7-14.335), as compared to those who are not underweight. Iron deficiency is more prevalent among underweight young women and the risk is approximately twice to three times compare to those who were not underweight.

S-11-7
Selenium content in daily consumed foods and estimation of daily intake by populations living in Bandung City Indonesia

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The information on the daily intake of selenium (Se) will benefit the authorities as an early warning to decide if the populations need any Se fortification program to prevent the Se deficiency-related diseases. With the history of Iodine deficiency, study on the Se status of Indonesian population is never been conducted. Thus, currently we don't know what is the Se content of their daily consumed foods? Their Se daily intake? and do they need Se supplementation program? Daily consumed foods in Bandung City were obtained from the local market and analyzed for Se concentration spectrofluorometrically. Estimation of the daily intake were calculated with the data from the food consumption survey of the populations. This information about Se daily intake is an important database that serve as an early warning for Se deficiency in Indonesia.
Keywords: Selenium, Nutrition, Foods, Daily Intake, Indonesia.

S-12-1
Involvement of intracellular Zn^{2+} signal in the dentate gyrus in cognition

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The hippocampus is required for memory retention for a limited period of time after learning. Hippocampal three synapses, i.e., perforant pathway-dentate granule cell, mossy fiber-CA3 pyramidal cell and Schaffer collateral-CA1 pyramidal cell, are glutamatergic (zincergic). The three synapses are stained by Timm's method, which detects histochemically reactive zinc. The zinc serves as a Zn^{2+} signal, predominantly exists in the presynaptic vesicles, co-released with glutamate, and modulates glutamate signaling at zincergic synapses. The role of Zn^{2+} signal in learning and memory is poorly understood. Synaptic plasticity such as long-term potentiation (LTP) is believed to be a cellular model for memory. On the basis of the evidence that Zn^{2+} signal multi-functionally modulates LTP induction in hippocampal slices, in vivo real-time correlation between LTP induction and memory was examined focusing on the selective loss of synaptic Zn^{2+} signal in the dentate gyrus. The present study indicated that intracellular Zn^{2+} signal in the dentate gyrus is required for object recognition memory, probably via dentate gyrus LTP expression.

S-12-2
Zinc chelation reduces hippocampal neurogenesis after traumatic brain injury

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Several studies have shown that traumatic brain injury (TBI) enhances hippocampal neurogenesis. However, the mechanism underlying increased neurogenesis after TBI remains largely unclear. Neurogenesis occurs in the subgranular zone (SGZ) of the hippocampal dentate gyrus (DG). A high level of vesicular zinc is localized in the presynaptic terminals of the DG. Using a TBI model, we tested our hypothesis that zinc plays a key role in modulating hippocampal neurogenesis after TBI. Zinc chelator, clioquinol (CQ, 30 mg/kg), injected into the intraperitoneal space to reduce brain zinc availability. Neurogenesis was evaluated using BrdU, Ki67 and doublecortin (DCX) immunostaining at 1 week after TBI. The number of BrdU, Ki67 and DCX positive cells was increased after TBI. However, the number of BrdU, Ki67 and DCX positive cells was significantly decreased by CQ administration. The present study shows that zinc chelation reduced TBI-induced neurogenesis. Therefore, this study suggests that zinc has an essential role for modulating hippocampal neurogenesis after TBI.

S-12-3
PBT2: A Novel Neuroprotective Agent that Enhances Cognition in Ageing

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The loss of cognitive function is a pervasive and often debilitating feature that characterizes ageing, and there are currently no therapeutics that effectively target the symptoms, let alone the biological substrates, of age-related cognitive decline (ARCD). Given the potential overlap between ageing and Alzheimer's disease (AD), we hypothesized that a novel metal chaperone (PBT2, Prana Biotechnology) previously shown to benefit synaptic plasticity-related endpoints and/or cognitive function in transgenic mouse models of AD and human clinical trials for mild AD patients, would enhance cognition in aged mice. We show here that PBT2 improves learning and memory in aged wildtype mice, concomitant with alterations to anatomical and biochemical substrates that are critical to normal cognition, and which suggest a specific effect of PBT2 on enhancing glutamatergic signaling pathways at the synapse. These data demonstrate that metal chaperones are a novel therapeutic approach for the treatment of ARCD.

S-12-4
Roles of biological trace elements in neurodegeneration (ALS and Fahr's disease (IBGC))
From molecular mechanisms to therapeutic strategies

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Biological metals have been considered to play very important roles in the neurodegenerative diseases such as Alzheimer's disease (AD), Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS) and idiopathic basal ganglia calcification (IBGC). By ICP-MS, we have measured levels of biological metals in the cerebrospinal fluid of patients. The findings suggest that copper (Cu) and zinc (Zn) play important roles. We also have demonstrated that both metallothionein(MT)-I/II and MT-III immunonoreactivity is diminished in the spinal cord of ALS. Then we have shown that MT-III prevents the loss of motor neurons of ALS model mice and prolongs the life span. FD is characterized by calcification in the basal ganglia. Recently, mutations in *SLC20A2*, encoding the type III sodium-dependent phosphate transporter-2 (PiT-2), are found to be a major cause of familial IBGC all over the world. Inorganic phosphate (Pi) plays an important role in the deposit of metals. The findings suggest that improvement of Pi transport is a target of therapeutic strategy for IBGC.

S-13-1
Should Bioactive Trace Elements Not Recognized as Essential, but with Beneficial Health Effects Have intake Recommendations?

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Today, most nutritionists do not consider a trace element essential unless it has a defined biochemical function in higher animals or humans. As a result, even though some elements have been clearly shown to have beneficial actions in higher animals and humans, they are generally ignored when dietary recommendations are formulated. This lack of attention could be blocking an opportunity that would promote the health and well-being of humans. Recently, it has been questioned whether some bioactives or phytonutrients should receive dietary recommendations. Some of the trace elements not established as essential but with beneficial health effects could be considered bioactives or phytonutrients. This includes boron, which is rich in fruits, leafy vegetables, nuts, legumes and pulses; silicon, which is rich in unrefined grains and some vegetables; and nickel, which is rich in pulses, grains, nuts, and chocolate. An appropriate recommendation may be to have an intake of these foods that would provide an amount of a trace element that has been shown to have beneficial effects for health and well-being.

S-13-2
Human Health Benefits of Boron

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Boron (B) has been found to beneficially affect bone growth, central nervous system and immune function, and associated with reduced risk for some cancers. The diverse effects of B indicate that it influences the formation and/or activity of substances that are involved in numerous biochemical processes. Several findings suggest that this influence is through the formation of boroesters in biomolecules containing *cis*-hydroxyl groups, which include those containing ribose (e.g., S-adenosylmethionine, diadenosine phosphates, and nicotinamide adenine dinucleotide), phosphoinositides, glycoproteins, and glycolipids. Through affecting these biomolecules, B might influence cell membrane function and/or integrity, and signaling ions or compounds release or action. As a result of this bioactivity, B can affect cell differentiation, organogenesis and embryogenesis; facilitate hormone action; and modulate inflammatory and immune responses. An intake of less than 1.0 mg/day apparently suppresses the benefits of B. Dietary surveys indicate such an intake is not rare. Thus, increasing B intake should be recognized as a reasonable dietary recommendation to enhance health and well-being.

S-13-3
Is chromium nutritionally or pharmacologically beneficial for glucose metabolism?

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Of all the "essential" elements, the role of chromium (Cr) is the most controversial. Recently, its status as an essential element, first proposed nearly fifty years ago, has been challenged; this will probably result in the general consensus on the status changing. Since 1989, Cr nutritional supplements have received expanding popular attention, not mirrored by scientific advances in understanding how Cr could work at a molecular level. At nutritionally relevant levels, Cr supplementation has no demonstrated beneficial effects on healthy humans while demonstration on humans with altered carbohydrate and lipid metabolism is ambiguous at best. At the same time, recent research on animal models has convincingly demonstrated potential pharmacological uses for Cr(III) compounds if Cr can be established to lack toxic effects. The financial implications of the pharmaceutical verses nutritional verses toxin debate, which underlies much of the research in the last two decades, have made the debate quite heated. For the field to advance, whether or how Cr can function at a molecular level in beneficial and/or deleterious manners must be determined.

S-13-4
Nickel deprivation affects reproduction, blood pressure control and sensory function

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Recent studies from our laboratory documented impaired male reproductive function in nickel (Ni)-deprived rats, consistent with infertility found in earlier studies. Ni is known to modulate function of cyclic nucleotide-gated cation channels (CNG). Because CNGs are distributed in photoreceptor, gustatory receptor and olfactory receptor cells, and kidney tubules, we determined the effects of Ni deprivation on blood pressure, brightness detection, odor preference, and taste preference in rats. Ni deprivation decreased sodium excretion after an oral NaCl load. Ni-deprived rats had increased systolic blood pressure. In the light/dark maze, Ni-deprivation decreased time spent in the dark arm by rats. The sniffing of estrous female urine was increased only in Ni-supplemented rats. Preference to saccharin solution was decreased by Ni deprivation. These findings suggest that Ni in nutritional amounts is beneficial, and possibly has an essential role, in blood pressure control, male reproductive function, vision, olfaction and taste.

S-13-5
Dietary Silicon and Connective Tissue Health

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Silicon (Si) is major constituent of the mammalian diet, derived from cereals and cereal-based products, some fruits and vegetables and from drinking/mineral waters. On average at least 40% of ingested Si is absorbed, as orthosilicic acid. Variation exists between foods and between dietary supplements due to differences in Si speciation. Mechanism of uptake is not established but is generally assumed to be by passive diffusion. The majority of absorbed Si is excreted in urine, but an unknown proportion must be retained as fasting tissue Si levels are increased with dietary intervention. Evidence exists for the kidneys as the site of regulation of Si status and Si responsive genes have been identified, including a Si transporter. Dietary Si intake is associated with higher bone mineral density in men, premenopausal women and post-menopausal women taking hormone replacement therapy, suggesting a Si-oestrogen interaction. In rats, dietary Si depletion led to changes in bone mineral composition and inhibition of growth plate closure. Mechanism of action of Si in bone and connective tissue health is unclear but overwhelming data suggests a role in extracellular matrix synthesis and stabilisation.

S-14-1
Bone as the Storage Site of Manganese (Mn) Exposure: Relationship to Mn Levels in Brain

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Limited data in literature suggest that nearly 43% of body Mn accumulates in the skeletal system. This study was designed to examine the time course of Mn accumulation in bone tissues and to seek the association between Mn storage in bone and its concentration in brain. Rats received a daily oral gavage at dose of 50 mg Mn/kg as $MnCl_2$ for 0, 2, 4, 6, 8 or 10 weeks. The data showed that Mn levels in all bone samples increased as the exposure duration increased. Mn was relatively evenly distributed in bone samples collected from leg, arm and skull, although the humerous (arm) bone had the highest Mn levels among all four bones ($p>0.05$). Synchrotron X-ray fluorescence data revealed that the spongy bone appeared to accumulate more Mn than did the compact bone. Linear regression analyses revealed that bone Mn levels were statistically significantly associated with Mn levels in the CSF, striatum, hippocampus and choroid plexus ($p<0.05$). Our results indicate that Mn exposure can lead to a significant accumulation of Mn in bone, particularly in the spongy bone mass; bone is the storage site for body Mn.

S-14-2
Decreased brain volumes in manganese-exposed welders

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This study evaluates morphological changes in brain volume among welders, and investigates the relationship between structural brain abnormalities and subclinical dysfunction in this population. We used voxel-based morphometry (VBM)to assess differences in gray- and white matter brain volumes between 40 welders with chronic Mn (manganese) exposure and 26 age-matched control subjects. Brain volumes in the globus pallidus and cerebellar regions were significantly diminished in welders with chronic Mn exposure compared to controls (FDR-corrected $p < 0.05$). These changes in brain volume were negatively correlated with grooved pegboard scores for both dominant and non-dominant hands. In conclusion, brain volume reductions in the globus pallidus and cerebellum are measurable even in the subclinical manganism, and the amount of volume reduction correlates with neurobehavioral motor deficits. Our findings therefore indicate that volumetric measurement could be a useful indicator of subclinical manganism.

S-14-3
Manganese Neurotoxicity: Lessons From Worms to Neonates

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Manganese is an essential mineral. Exposure to high Mn levels results in lesions in the basal ganglia, referred to as manganism. Combining genetics and biochemical assays, we established in the nematode (*C. elegans*) that dopamine (DA) is responsible for Mn-induced DAergic neurodegeneration and that this process (1) requires functional DA-reuptake transporter (DAT-1) and (2) is associated with oxidative stress and lifespan reduction. Additional studies tested if parenteral nutrition represent risk factors for increased Mn brain deposition. Brain Mn was assesed by T1 relaxation (T1R) times. Brain Mn and its relationship to total dietary Mn, total days on PN, conjugated bilirubin levels and blood Mn concentrations was determined. Dietary Mn exposure was be inversely associated with GP T1R. The results show that T1–weighted MRI can be used to screen infants on prolonged PN for increased brain Mn deposition. Hepatic cholestasis was also found to be a risk factor for increased brain Mn deposition in neonates receiving PN. These data suggest that PN may be a risk for Mn neurotoxicity

Keywords: manganese, parenteral nutrition, MRI, dopamine

S-14-4
^{18}F-FP-(+)-DTBZ (^{18}F-AV-133) brain PET scan in chronic manganese intoxication and idiopathic Parkinson's disease

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Excessive exposure to manganese dust or fume may induce a neurological abnormality called manganism, similar to idiopathic Parkinson's disease (IPD). ^{18}F-9- fluoropropyl-(+)-dihydrotetrabenazine (^{18}F-AV-133) is a novel positron emission tomography tracer for imaging the vesicular monoamine transporter II in dopaminergic neuron degeneration which might be indicative for IPD. Fifteen ^{18}F-AV-133 PET studies were conducted including 4 normal controls (age: 62.3±4.9 years), 9 Parkinson's disease (PD) patients (age: 60.8±6.0 years) and 2 typical manganism patients (age: 62, 69 years) with Hoehn and Yahr stages 2 to 3. Volumes of interest of the bilateral putamen, caudate nuclei and occipital cortex as the reference region were delineated from individual MRI. Standardized uptake value ratio was evaluated in the striatum and compared to OC from 14 dynamic image sets. ^{18}F-AV-133 uptake decreased in PD patients at the bilateral striatum as compared with normal control and manganism groups. In manganism patients, there was no decreased uptake of the radioactivity involving the bilateral striatum. In addition, there was symmetrical distribution of monoaminergic radioactivity in the bilateral striatum. Our findings indicate ^{18}F-AV-133 with PET could provide a useful neuroimage biomarker in the differential diagnosis between manganism and IPD.

Key words: manganese, manganism, brain ^{18}F-AV-133, PET, Parkinson's disease

S-14-5
Metal concentrations in CSF and blood plasma from patients with neurodegenerative disorders

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A slow but steady increase in neurodegenerative disorders has been noted in recent decades. In motor neuron disease (MND) spinal neurons degenerate causing widespread fatal muscle atrophy. What is causing this degeneration? Indications for metallotoxic etiologies exist. Clusters are noted in metal rich regions. Animal exposure experiments show specific spinal cord metal accumulations.

Twenty-two metals were measured in cerebrospinal fluid (CSF) from 17 MND patients and 10 controls. Significantly increased concentrations were found for Mn, Al, Cd, Co, Cu, Zn, Pb, V and U. Manganese showed the most prominent differences between cases (median 5.67 mg/L) and controls (median 2.08 mg/L). CSF Mn concentrations were higher than plasma Mn concentrations (median 0.91 mg/L), suggesting Mn transport into the central nervous system. Most detected metals in CSF are well known neurotoxicants. Properties of barrier systems will be discussed and the possibility of Mn accumulations and multi-metal toxicity as a major factor contributing to the relentless course of MND introduced.

S-14-6
Biomarkers of iron status in welders exposed to manganese

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Iron (Fe) and manganese (Mn) share many transporters in humans, and low serum ferritin (s-fer) is related to higher blood Mn levels. Welders may inhale high amounts of Fe, but little is known if this may affect Mn homeostasis. Fe status biomarkers and Fe, Mn and cobalt (Co) in whole blood (B), serum (S) and urine (U) was studied in 137 welders and 137 referents. Alcohol consumption and systemic inflammation were assessed with carbohydrate deficient transferrin (sCDT) and C-Reactive Protein (sCRP) in serum. The geometric mean (GM) exposure to Mn was 214 µg/m^3 and to Fe 1524 µg/m^3. The GM s-fer was 119 µg/L in welders and 98 µg/L in referents. The GM serum hepcidin were 8.4 and 6.5 µg/L. S-fer was associated with sCDT and sCRP, but not with exposure. The increase in s-fer was 23% among 63 welders and 19% among 65 referents (p=0.84) who had been examined six years previously. Regression analysis showed negative associations between S-fer and U-Co, B-Co and S-Co, and positive associations with U-Mn, S-Mn and B-Mn. In conclusion, exposure to Fe and Mn had no impact on biomarkers of iron status. sCDT and sCRP was positively associated with s-fer. S-fer and S-CDT had impact on several elements in biological fluids.

S-14-7
X-ray fluorescence imaging and spectroscopy in trace element research: focus on mechanisms of Mn neurotoxicity.

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X-ray fluorescence imaging quantifies multiple trace elements (biologically relevant and toxic) in tissues and single cells. Tissue level imaging (20x20 um resolution) of rat brains exposed to Mn (model of Mn induced neurotoxic condition in humans) revealed Mn accumulations in: (globus pallidus (GP)≈substantia nigra compacta (SNc)>CA3 layer of hippocampus (HP)>thalamus>caudate putamen>cortex). Treated rats received intraperitoneal injections of 6 mg Mn/kg for 4 weeks, the control group received saline solution. Single cells images (300x300 nm resolution) from the CA3/HP and SNc showed Mn localization in cytoplasm and nucleus. Mn was not increased in localized intracellular Fe or Cu accumulations. Spatial Mn distribution did not follow the distribution of Fe, Cu or Zn. However, strong Fe/Mn correlation in control ($r=0.84$, $p<0.01$) and treated ($r=0.52$, $p=0.02$) brains suggested common transport mechanisms likely via transferrin receptor. In HP strong Mn/Zn ($r=0.68$, $p<0.01$) correlation suggested additional, unique for HP, Mn transport via Zn pathway.
Keywords: manganese, X-ray fluorescence imaging, Globus Pallidus, Substantia Nigra Compacta.

S-15-1
Brazilian nuts as a promising selenium-containing functional food in health and disease.

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Food, provide nutrients and non-nutrients compounds that, in general, may act modulating several cell signaling pathways, and also gene expression, promoting health benefits. However, the relationship between food and health is still unclear due to many lacks in the food science and nutrition knowledge. So, we will focus on relationships between food quality, it's intake and influence on the nutritional status of groups of population and also their potential to reduce the risk of development of disease in healthy individuals and/or under condition of diseases, with emphasis on selenium (Se) and its relationship with related enzymes polymorphisms. Alterations in Se status may result in suboptimal amounts of selenoproteins, which have been associated with increased oxidative stress. The Pro198Leu polymorphism in the glutathione peroxidase-1 gene is predicted to be functional, so the response of Se status, GPx activity, and levels of DNA damage to a Se supplementation trial between the genotypes related to this polymorphism was investigated. Trials were conducted in Brazil with Brazilian nut supplementation and the results will be presented.

S-15-2
Targeting the epigenome with selenium for cancer prevention

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Increasing evidence shows that nutrients, including selenium, influence epigenetic processes. Selenium has been shown to modulate the activity of enzymes that integrate the epigenetic machinery, including DNA methyltransferases and histone deacetylases and acetyltransferases. Selenium regulation of microRNAs has also been recently reported. Cancer development involves progressive alteration of epigenetic marks. Global DNA hypomethylation, gene promoter hypermethylation and histone onco-modifications are associated with genomic instability and aberrant expression profiles of genes involved with cell proliferation, death and differentiation Selenium is a promising anticancer nutrient that has been shown to exert its protective effects through several mechanisms, including epigenetic modulation. Because epigenetic marks are potentially reversible and early altered during carcinogenesis, the epigenome represents a promising target for cancer prevention with selenium. Data showing the influence of selenium on the epigenome and its consequences for cancer prevention will be presented.

S-15-3
Selenium deficiency in children with intestinal dysfunction treated with parenteral nutrition and/or elemental diet

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We have three males and two females (2 - 20 years old) with selenium deficiency. They needed nutritional support because of hypoxic-ischemic encephalopathy in 4 patients, and short bowel syndrome in one case. Three cases were received enteral nutrition of elemental diet (ED) which contains no or few selenium. Parenteral nutrition (PN) with no selenium was performed for two children. In all five patients the serum selenium levels were lower than 2 μg/dL. Their clinical features were browning hair, whitening nails, macrocytic anemia and low heart function. Four children were received trace element supplement. One patient was administered rice water and fish soup. After selenium replacement therapy, all patients showed elevation of serum selenium level, and their clinical features were improved. It is important to consider about selenium deficiency in patients treated with PN and/or ED.

S-16-1
Environmental Contamination and its Temporal Changes around the Fukushima Site

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The Japan Atomic Energy Agency (JAEA) has conducted large-scale environmental monitoring and mapping projects around the Fukushima Daiichi Nuclear Power Plant, in collaboration with many organizations commissioned by the Ministry of Education, Culture, Sports, Science, and Technology. Thus far, large-scale monitoring activities have been carried out several times, and temporal changes in local contamination conditions have been clarified. This presentation will summarize results from these projects.
By analyzing collected soil samples, as well as in situ measurements made with portable Germanium detectors, soil deposition maps were created for Cs-134, Cs-137, I-131, Te-129m, Ag-110m, Pu-238, Pu-239+240, Sr-89, and Sr-90. Activity ratios of I-131, Te-129m, or Ag-110m to Cs-137 showed specific regional features.
Dose rates in the air have been measured in two different ways: [1] measurements at a level of one meter above undisturbed flat fields, and [2] measurements on roads using mobile survey systems. Dose rates in the air have decreased more than would be expected from physical decay. Moreover, decreases on roads were found to be much larger than those on undisturbed flat fields.

S-16-2
Behavior of radiocesium in ecosystems and its impact on products

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After the releases of radionuclides from Fukushima Daiichi Nuclear Power Plant, extensive efforts have been put into the monitoring of different environments and their products. Radioactivity monitoring of food products are especially important, in order to estimate internal radiation doses of public and to assure the food safety. Radiocesium concentrations in most of foods decreased sharply with time, and fell under the standard limit for General Foods (100 Bq/kg). On the other hand, some specific food products still keep relatively high concentration of radiocesium. They are mushrooms, edible wild plants, freshwater fishes, ocean-floor fishes, etc. This report summarizes radioecological features of the environments producing such specific products, focusing on the behavior of radiocesium. Impact on forest tree, which is one of the important non-food products, is also discussed.

S-16-3
Effects of Prussian blue on radioactive cesium degradation by mushrooms

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Mushrooms are known for their capacity to accumulate radioactive cesium in their fruiting bodies. The present study examined how adding Prussian blue to the mushroom cultivation media affected radioactive cesium degradation. It is known that Prussian blue can efficiently absorb cesium.
We measured radioactivity in the fruiting bodies of *Lentinula edodes, Grifola frondosa, Pholiota nameko, and Auricularia polytricha* cultivated on media containing Cs-134 and Cs-137. Radioactive cesium in fruiting bodies of the four mushrooms dropped below the detectable threshold when Prussian blue was added to the cultivation media (0.1%).
After a bed-log of *L. edodes* was sunk into water containing Prussian blue (0.05% w/w), radioactive cesium levels in the fruiting bodies dropped to less than half of the control (without Prussian blue).
In conclusion Prussian blue is effective for use against absorption of radioactive cesium by mushrooms.

S-16-4
Evaluation of committed effective dose due to food ingestion for Japanese adults before the Fukushima nuclear plant accident and the ongoing dose evaluation for children in the post-Fukushima era

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In 2008, the committed effective dose due to food ingestion was estimated at 0.80 mSv for Japanese adults. This value was based on the radioactivity of U-238, Th-238, Ra-226, Pb-210, Po-210, Sr-90, Cs-137 and Pu-239+240 in food products in Japan from 1989 to 2005. These data are essential as background values for dosage evaluation following the Fukushima disaster.
In this post-Fukushima nuclear disaster era, an evaluation of the committed effective dose for children living in the post-Fukushima era is of great importance.We started the research in 2013 to evaluate the dose among Japanese children due to ingestion of school-provided lunches. The analysis program for dietary samples has just begun, so some time is required for sufficient results to be produced. Here we show some of the newly obtained results from the period after the Fukushima nuclear disaster in addition to the committed effective dose as evaluated in 2008.

S-16-5
Radioactivity Ingestion Dose Estimation Following the Fukushima Nuclear Disaster in Japan

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The present study used a duplicated food sample method to measure radioactive cesium concentrations in 82 two-day food samples, including 37 from Fukushima, collected in late 2012 to early 2013. Estimated committed effective doses ranged from 0.1 μSv to 7.5 μSv for one year of food consumption. While the attitude surrounding food selection significantly affected ingestion dose ($p=0.026$), no significant difference was detected between subjects from Fukushima versus those from other areas.
We also used the food monitoring dataset provided by the Ministry of Health, Labour and Welfare (N=445,516) and the Japanese National Food Consumption Survey dataset (N=8,815) to estimate ingestion radiation doses up to March 2013. Assuming full implementation of food restrictions, the median effective doses in children aged 1-6 years and adults were estimated to be roughly 0.15 mSv and 0.21 mSv, respectively.

MS-1-1
Metal profiles in segmented hair samples obtained in arsenic-polluted areas

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The hair metal analysis is of great importance in estimating exposure levels to pollutant metals. Since the hair grows 1 to 1.5 cm per month, the metal analysis in each segment of hair from the root to peripheral end may provide information on exposure history to metals. However, qualities of the hair including protein contents may be different among each segment. In this study, we determined the concentrations of metals and S in segmented human hair samples obtained from the subjects in arsenic-polluted areas in Cambodia. The results of ICP-MS analyses show that S contents are consistent along the hair strands. The segmental concentrations of Mn and Fe showed increasing tendencies while those of Se showed a decreasing tendency toward the peripheral end. On the other hand, the segmental concentrations of hair As showed inconsistent tendencies suggestive of individual differences in previous exposure to As. The metal analysis in segmented hair may provide useful information on exposure history of pollutant metals.

MS-1-2
Associations of circulating molecules involved in atherosclerosis with arsenic exposure

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Exposure to arsenic and subsequent development of atherosclerosis has been suggested to be responsible for increased mortality. However, biochemical events for arsenic exposue-related atherosclerosis remain unknown. This study was aimed at investigating the associations of circulating biomarkers for atherosclerosis with arsenic exposure in the individuals exposed to arsenic in Bangladesh. A total of 324 study subjects from arsenic-endemic and non-endemic areas in Bangladesh were recruited. Total cholesterol, LDL and HDL levels were lower in arsenic-endemic subjects than in non-endemic subjects. Oxidized LDL, CRP, ICAM-1 and VCAM-1 levels were significantly higher in arsenic-endemic subjects than in non-endemic subjects. These circulating molecules also showed significant correlations with arsenic exposure metrics. Further, HDL, oxidized LDL and CRP showed dose-response relationships with arsenic exposure. All these associations may be the major features of arsenic-related atherosclerosis.

MS-1-3
Comparative Nutritional Evaluations in Global Arsenic Endemic Areas

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Nutritional involvement in the development of human arsenic toxicity is of interest. Recently several evaluations of arsenic endemic populations for nutritional status have been published. These studies used the methodology of food frequency, 24 hour dietary recall, and BMI assessments. In most cases deficiencies in one or more nutrients and low socioeconomic status seemed prevalent.

The present study is to develop a comparative evaluation of nutritional findings in order to further tease out etiologic factors to arsenic disease.

We determined in earlier studies that vitamin A intakes were substantially below the RDA (1/3 the RDA in retinol equivalents) for a Latin American population studied. Values were also lower (2/3 the RDA in retinol equivalents) in a U.S. population experiencing arsenic exposure. This nutrient and others, zinc, selenium, and protein for example, will be compared across global boundaries of India, Bangladesh, and other countries including Taiwan. These reports will be compared and the contributing factor of nutrition to arsenic human pathogenesis evaluated.

MS-2-1
Drinking Water Intake Evaluations-a Review for Developed and Developing Countries

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Human intake of drinking water can serve as a significant source of toxic chemicals. Arsenic exposures via drinking water in the countries of India and Bangladesh serve as examples. Risk assessment needs accurate determinants of water intake when understanding human health and toxicological evaluations. We present here a review of water intake estimates.

Data from our population surveys found an enhanced water intake (plain water and prepared beverages) greater than 2 L/day for selected residents in the southwest and northwest areas of the United States. Approximately 2.9 L/day of water was consumed by participants, 3.1 L/day by males and 2.7 L/day by females. Researchers have reported individual intakes up to 9.2 L/day for males and 8.7 L/day for females in developing countries.

Correlations will be examined between water intake, corresponding dosages, and severity of arsenic-related health outcomes around the globe. Seasonal variations, age relationships, and those of diet and socioeconomic determinants will be considered. In the study of human drinking water intake and health, elevated symptoms in males vs. females have resulted from toxic chemical exposure.

MS-2-2
Public health impact of low-environmental exposure to cadmium: the emerging risks of metabolic confounding

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The current risk assessment of cadmium (Cd) largely relies on the assumption that urinary Cd (U-Cd) is a reliable biomarker of the long-term exposure to the metal. Recently, we found that at low exposure levels variations of U-Cd over lifetime are mainly driven by the recent Cd intake and the physiological variations in the renal function. We here further investigated the significance of U-Cd in a population of 260 schoolchildren (mean age, 5.7 years). We measured urinary Cd, β2-microglobulin, retinol-binding protein and α1-microglobulin. The mean U-Cd (0.24 µg/g creatinine) was similar to that of middle-aged adults in the same area. The covariate-adjusted concentrations of the three urinary proteins correlated with U-Cd through associations as strong as those reported previously for elderly and adults in Belgium or Sweden. This means that these associations are unlikely to be the reflection of long-term Cd toxicity. Other possible sources of metabolic confounding found in these children were diuresis and urinary zinc excretion that emerged as much stronger determinants of U-Cd than did age or gender.

MS-2-3
Changes in Metal Ion Metabolism as Indicated by Erythrocytes in Patients with Alzheimer's Disease

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Much research on Alzheimer's disease has attempted to understand the disease's pathophysiological processes by focusing on β-amyloid, Tau, protein folding, heat shock proteins, and metallostasis. Some models based on protein metabolism may be too distant from the intended targets in the body and the brain. Thus far, limited consensus has been achieved. The authors have utilized elemental profiles of erythrocytes from patients with Alzheimer's disease as biomarkers. Erythrocyte elemental profiles may be examined during the approximately 120 days that they circulate in blood vessels to indicate changes not only in their own metabolism, but also in that of surrounding cells. The resulting elemental profiles are indicative of the metabolism of proteins, carbohydrates, lipids, and metal ions. Significant changes in concentrations of lead, cadmium, silver, and uranium were observed in comparison with controls. Inductively coupled plasma mass spectrometry (ICP-MS) was used to analyze elemental profiles. Aspects of accumulated metal ions in erythrocytes will be discussed in the paper.

MS-2-4
Metabolomic approach for tributyltin-induced toxicity.

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Organotin compounds such as tributyltin (TBT) are known to cause various cytotoxicity, including developmental toxicity and neurotoxicity. However, the mechanism by which nanomolar levels of TBT induce cytotoxicity has not been fully understood.
In the present study, we studied the effect of 100nM TBT on cell growth using human NT2/D1 embryonic carcinoma cells. Exposure to 100 nM TBT induced growth arrest without causing cell death. To examine the mechanism by which TBT induce growth arrest, we performed comprehensive metabolomic analysis. We found that exposure to TBT decreased ATP in the cells. In addition, exposure to TBT reduced the levels of TCA cycle metabolites, such as ketoglutarate, succinate and fumarate, in the cells.
In conclusion, these results suggest that exposure to nanomolar levels of TBT causes growth arrest by targeting energy supply in human embryonic carcinoma cells. Thus, metabolimic approach may provide new insights into the mechanisms of metal-induced cytotoxicity.

MS-3-1
Prevention of intestinal alterations caused by *Trypanosoma cruzi* infection by association of Selenium and Plant-based Product

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Chagas disease is caused by the protozoan *Trypanosoma cruzi*. The symptomatic stages present distinct clinical forms, such as cardiac and digestive. We showed the pharmacological effect of selenium (Se) against this disease progression. Given that the use of phenolic compounds and fibers naturally present in plants are restricted, we evaluated the association between Se and plant-based product (PBP) in a murine model of Chagas colopathy. Infected (I) male mice received diets formulated with PBP and Se in drinking water (ISePBP) starting two weeks before infection. Other infected groups were used as controls (ISe,IPBP, I), as well as, uninfected ones (NSe, NPBP, NSePBP, N). Preliminary results show that the combination of Se and PBP was able to reduce parasitemia, intensity of inflammation in intestinal tissues, improved intestinal motility and consequently the frequency of defecation. Other studies are in progress to confirm these results, and PBP is under patent application. These results indicate that this nutritional approach is a new therapy perspective for intestinal disorders caused by *T. cruzi*.

MS-3-2
Selenium Status in the Elderly: Relationship with Cognitive Decline

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Studies show that reduced activity of antioxidant systems is related to cognitive decline. We measured selenium (Se) status in elderly patients with Alzheimer's disease (AD; n=27) and mild cognitive impairment (MCI; n=17) compared with a control group (CG; n=28). Se concentrations were determined from plasma and erythrocytes using hydride generation atomic absorption spectroscopy. Erythrocyte Se concentration in the AD group was lower than in the CG (43.73±23.02 µg/L and 79.15±46.37 µg/L; p=0.001), but not significantly different from the MCI group (63.97±18.26 µg/L; p=0.156). The AD group exhibited the lowest plasma Se level (34.49±19.94 µg/L) compared to patients with MCI (61.36±16.08 µg/L; p=0.000) and the CG (50.99±21.06 µg/L; p=0.010). Erythrocyte Se decreased relative to cognition function. Because erythrocytes reflect longer-term nutritional status, these data emphasize the important relationship between Se exposure and cognitive function. These findings suggest that Se deficiency may contribute to cognitive decline in the elderly.

MS-3-3
Zinc regulation of phosphorylation signalling in health and disease

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A major hypothesis of how we age and how degenerative diseases develop was formulated 30 years ago by the late Nobel Laureate Sir Macfarlane Burnet. It considers a compromised homeostasis of metal ions, especially zinc, as a cause for suboptimal cellular functions and adverse chemical reactivities. The identification of at least three dozen cellular proteins involved in zinc transport, sensing, and subcellular compartmentalization has provided support for Burnet's hypothesis. Total cellular zinc concentrations of a few hundred micromolar are buffered to yield remarkably low picomolar zinc(II) ion concentrations. Yet induced zinc(II) ion transients serve in cellular control, eg. by modulating protein tyrosine phosphatase activities in phosphorylation signalling. Perturbations of such zinc signals are implicated in many diseases. We surmise that states of "zinc deficiency" relate primarily to inadequate control of cellular zinc(II) ions rather than to the role of zinc as a permanent cofactor of proteins. 1. W Maret (2013) Met Ions Life Sci 12:479; 2. W Maret (2013) Adv Nutr 4:82; 3. W Maret (2013) BioMetals 26:197.

MS-3-4
The role of ZIP14 in the transport of manganese in SH-SY5Y cells

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Exposure to manganese is known to cause symptoms related to Parkinsonism. However, the precise mechanism of manganese transport in nervous systems remains unclear. Recently, we reported that not only DMT1, the transporter for divalent metals, but also ZIP8 and ZIP14, the zinc transporters, may play roles in the transport of Mn^{2+}. To explore the roles of these metal transporters in the accumulation of manganese in neuronal cells, we introduced siRNAs of each metal transporter to SH-SY5Y cells, a model cell line for dopaminergic cells, and examined the uptake rates of Mn^{2+}. The transfection of siRNA of DMT1 significantly reduced the uptake of Mn^{2+}, as is similar to the previous observations. The transfection of siRNA of ZIP14, but not that of ZIP8, significantly reduced the uptake of Mn^{2+}. Pretreatment of SH-SY5Y cells with IL-6 markedly increased the expression of ZIP8 and ZIP14, but not that of DMT1, and resulted in higher accumulation of manganese. These results suggest that ZIP14 may play a significant role in the transport of manganese in neuronal cells especially in the presence of inflammatory cytokines.

MS-3-5
Metal-induced conformational changes of prion protein fragment peptides and its neurotoxicity

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Prion diseases are progressive neurodegenerative diseases with the accumulation of amyloidogenic prion protein (PrP) in the patient brain. The conformational conversion of normal cellular proteijn (PrP^{C}) to an abnormal scrapie-type (PrP^{SC}) is suggested to be based on its pathogenesis. Here, we investigated the effects of trace elements on conformational changes of PrP fragment peptide, PrP106-126. PrP106-126 exhibited ß-sheet structures and formed amyloid-like fibers after the aging process. Aged PrP106-126 caused marked death of primary cultured rat hippocampal neurons. The co-existence of Zn^{2+} or Cu^{2+} during the aging inhibited ß-sheet formation by PrP106-126 and attenuated its neurotoxicity. We also found that Carnosine (ß-alanyl histidine) significantly inhibited ß-sheet formation and neurotoxicity of PrP106-126. Our results suggest the implication of trace elements in the pathogenesis of prion diseases.

MS-3-6
Trace elements status in a rural population of Bolivia, in relation to the intake and the presence of parasitic diseases

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Micronutrient deficiencies lead to an increased risk for development and chronicity of infectious and parasitic diseases. The status of zinc, iron and copper were evaluated in adults (patients with leishmaniasis and healthy controls) and in children (96% diagnosed with intestinal parasites).
Results regarding to the presence of leishmaniasis showed no differences in iron and copper serum levels however, serum zinc was significantly lower in patients than in controls. Although their median zinc intake met the recommendations, serum zinc in both groups was below the reference values. In children, high prevalence of intestinal parasites was found, and their zinc intake did not meet the recommendations thus, serum zinc levels were below the suggested lower zinc cut off, also iron was below the reference but not copper. Dietary intake showed molar ratios phytate:zinc at levels to compromise the zinc status and negative correlated to serum zinc. Conclusions: patients with leishmaniasis and children with intestinal parasites had low serum zinc levels due to presence of the disease per se, phytate in the diet and in case of children and inadequate zinc intake.

MS-4-1
Successful Treatment of Fulminant Wilson Disease without Liver Transplantation: A Case Report and Review of Literature

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Fulminant Wilson disease (FWD) is a life-threatening condition. The scoring system, new Wilson predictive index (NWPI), has been used well in the clinical setting. According to NWPI, a score ≥11 indicates a poor chance of survival and liver transplantation (LTx) is recommended. We present a 10-year-old FWD female who fully recovered without LTx in spite of high NWPI score of 16 and has remained well for four-year follow-up. Plasma exchange and continuous hemodiafiltration were considered to play an important role for improvement of her prognosis. Furthermore, we reviewed the clinical findings, therapeutic modalities, and outcomes of 4 previously reported patients with FWD, who didn't need LTx despite their high NWPI score of 11 or more.

MS-4-2
Utility of Zinc Acetate Treatment in 15 Patients with Childhood Onset Wilson Disease: A Single Center Experience

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Treatment of Wilson disease is based on the use of copper chelators to promote copper excretion from the body, or use of zinc to reduce copper absorption in the ileum. Here we report the utility of zinc acetate therapy for Wilson disease in children. Fifteen patients including 6 males and 9 females, were treated with zinc acetate. Before treatment, nothing was given to 6 patients, 5 were treated with penicillamine, and 4 were with trientine. Median levels of liver function measured at zero months, 6 months, and 1 year were as follows: aspartate aminotransferase 48, 35, and 33 IU/l; alanine aminotransferase 71, 25, and 46 IU/l; γ-glutamyltranspeptidase 49, 46, and 55 IU/l. The median value of spot urinary copper was 1112 μg/l (0.15 μg/mg/creatinine) with the use of a chelator, and 141 μg/l (0.10 μg/mg/creatinine) without the use of a chelator. Three patients experienced nausea, and one experienced oral cavity discomfort. This study shows that zinc acetate therapy is an effective treatment for childhood onset of Wilson disease.

MS-4-3
Long-term Outcomes of Wilson Disease Neurological Phenotypes: A Nationwide Survey

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Wilson disease (WD) is one of the very few chronic neurological diseases for which specific and effective treatment is available. The authors examined factors affecting the prognosis of WD patients, based on the results of a Wilson disease nationwide survey. For 114 cases, age at onset, age at diagnosis, onset phenotype, presence of neurological symptoms, and outcomes were investigated. Results were analyzed using the Cox proportional hazards model. The significant difference in relative risk was found to be dependent on phenotype and the existence of neurological symptoms. Besides, the significant difference was also found between the hepatic group and the neurological group in the terms of diagnosis. In this examination, long-term outcomes of patients who experienced neurological symptoms were relatively poor.

MS-4-4

Zinc concentrations and zinc transporter expression in blood linked to age and Systemic Inflammatory Response Syndrome (SIRS)

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Zinc levels in plasma alter with ageing and disease. We compared zinc concentrations in plasma and red blood cells from people of different age groups and in patients with SIRS. Zinc concentrations were compared to mRNA expression in blood of all human zinc transporters and on data on zinc intake and a battery of clinical variables. A group of young athletes (average 26 y), a group of middle-aged individuals (average 51 y), and a group of critically ill patients (average 56 y) were recruited. Zinc contents in plasma and blood cells were lower in the 51 y controls than in the 26 y group. Plasma but not erythrocyte zinc was further reduced in SIRS patients. Transcripts for zinc transporters in blood cells were generally highest in the 51 y control group dropping precipitously in patients with SIRS. Only ZIP1 mRNA increased in SIRS patients compared to the 51 y old controls. Zinc transporters and zinc levels in blood correlated with a number of clinical variables.

P-1
Adding Zinc to Vitamin A on IGF-1, Bone Age in Stunted Children

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A randomized double blind placebo controlled trial of a single dose of 200,000 I.U of vitamin A with daily Zinc supplementation was done in children of Mojo village, Surabaya City. Children aged from 48 to 60 months were randomized to receive a single dose of 200,000 I.U. of vitamin A and zinc sulfate six day a week (n=12), and placebo(n=12) for period of six months. Children were evaluated weekly for nutritient intake, and IGF-1, bone age, index Height for Age at six months.At the end of the study there were a significant increase in IGF-1 ($p<0.04$) and Z-Score Height for Age ($p<0.001$), bone age ($p<0.01$).These results suggest that combined vitamin A and Zinc supplementation increase linear growth among children, and thus may play a key role control and strategies for stunted children under five years old.

KEY WORDS :
Zinc and vitamin A ,IGF-1, Bone Age and Height for age ,stunted

P-2
Cranial CT and MRI findings in nutritional tremor syndrome and malnourished children with zinc deficiency

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In this study, cranial magnetic resonance imaging (MRI) and computerized tomography (CT) findings were investigated in children of nutritional tremor syndrome (NTS) with moderate and severe protein energy malnutrition (PEM) who had zinc deficiency to determine cerebral abnormalities. Thirty children aged 6-36 months were enrolled in NTS and malnutrition group. Serum zinc in NTS children was low 93.23±15.95 µg/dL compared with control children 101.27±6.16 µg/dL ($p<0.05$). Serum zinc in grade III-IV malnutrition children was also low 88.80±18.99 µg/dL compared with control children 99.97 ±8.94 µg/dL ($p<0.01$). Brain scan in 11 NTS and moderate to severe malnourished (grade III-IV) children (37%) had abnormal CT and MRI showing brain atrophy. Cerebral atrophy give rise to important compromise of the child's higher brain functions.Therefore, it is suggested that children with moderate/severe malnutrition and NTS should be evaluated for cerebral atrophy because if detected early it is possible that changes are reversible. Zinc containing foods and medicinal zinc supplementation should be considered for better outcome.

P-3
Infants and elders are liable to zinc deficiency.

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We have reported that many of infantile patients with autistic disorders are suffered from zinc deficiency 1). In this study, we show that many of zinc deficiency are detected in infants and elders. Hair trace elements including zinc were measured with ICP-MS for 28,424 subjects (9,612 males and 18,812 females). In 1,754 out of the subjects (6.2 %), their zinc concentrations were lower than –2 S.D. level of the reference range. In the child group of less than 10-year-old, the rate of zinc deficiency was estimated 30.8 %, and 43.7 % in the infantile group aged 0-3 year-old. The prevalence rate of zinc deficiency decreased to minimal (0.8 %) at high-teens, and age-dependently increased to maximal (16.4 %) at the 7-8th decade of life. The lowest concentration of 9.7 ppm was observed in a 51-year-old woman, corresponding to about 1/13 of the mean reference level. In contrast, toxic metals such as lead, cadmium and aluminum were detected at high levels in children. A significant inverse relationship was observed between zinc and lead concentrations.
These findings suggest that infants and elders are liable to zinc deficiency.

1) Yasuda H et al: Sci Rep 1: srep129 (2011); 3: srep1199 (2013).

P-4
Evaluation of zinc complexes' dynamics by using the Gamma-Ray Emission Imaging (GREI)

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Zinc (Zn) complex that has a hypoglycemic effect is candidate metal-based medicine for the treatment of type 2 diabetes. However, no methodology is established for noninvasively analyzing the dynamics of zinc complex in whole body. Here, a novel nuclear medicine modality of GREI was applied to the noninvasive imaging analyses of the dynamics of metal drugs. In this study, di(1-oxy-2-pyridinethiolato)Zn complex ($Zn(opt)_2$), di($_L$-histidino)Zn complex ($Zn(His)_2$)), and $ZnCl_2$ were used. The dynamics study of Zn complexes radiolabeled with ^{65}Zn were carried out by GREI for 8 hours. From this GREI experiment, we succeeded in visualizing the different distribution of these two zinc complexes and $ZnCl_2$.

P-5
Rice fortified with zinc through parboiled process

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Rice is the staple food for more than half of the world's population and, hence, the main source of a vital micronutrient, zinc (Zn). This study evaluated the Zn fortification of rice through a parboiling process intended to increase the concentration of this nutrient in the grain. Samples of paddy rice were treated with the following zinc solutions: 0; 100; 200; 300 and 400 (mg L^{-1}). The results showed that the parboiling process with solutions containing from 100 to 400 mg L^{-1} of Zn increased from 1.2 to 1.4 times the Zn content in polished grains (white rice) and from 1.8 and 3.5 times the Zn content of unpolished rice (brown rice). The Zn concentration in the parboiled rice was linearly correlated with the concentration of Zn (mg L^{-1}) in both unpolished (r = 0.98) and in polished rice (r = 0.95). Our results indicate that the rice parboiling process is a quick and effective way to increase the concentrations of Zn in rice grains. However, further studies are required to evaluate rice quality and Zn bioavailability in human diets.

P-6
A novel splicing variant of zinc transporter ZIP2 controls mucus hypersecretory phenotype in CF and CF-like airway epithelial cells

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Zinc ion (Zn^{2+}) is an essential dietary metal ion that has pleiotropic effects in airway epithelial cells. Although patients with cystic fibrosis (CF), a common human hereditary pulmonary disease characterized by mucus hypersecretory phenotype, have a defect in regulation of intracellular Zn^{2+} level, the cellular and molecular mechanisms of Zn deficiency are largely unknown. Here, we identified in CF and CF-like airway epithelial cells that expression of the novel splicing isoform of a zinc transporter ZIP2 (ΔC-ZIP2) is increased, which results in a production of C-terminus-deleted immature ZIP2 protein. Consistently, ZIP2 protein expression and intracellular Zn^{2+} level were decreased in the CF-related cells. Finally, reduced intracellular zinc concentration with a zinc chelator TPEN up-regulated MUC5AC gene expression in normal airway epithelial cells, suggesting an important role of intracellular Zn^{2+} level in the regulation of mucus hypersecretory phenotype in CF airway.

P-7
Effects of carnosine on Zinc-induced neurotoxicity and its analysis in rat's brain

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Excess, zinc (Zn) is neurotoxic and plays a crucial role in the pathogenesis of vascular-type dementia (VD). We previously found that carnosine (β-alanyl histidine) inhibited Zn^{2+}- induced and prion induced neuronal death. Here we investigate the molecular mechanism of carnosine for prevention of zinc-induced neuronal death. The DNA microarray revealed that co-existence of carnosine with Zn inhibited the expressions of several genes which affected after Zn exposure.
Furthermore, we investigated the amount of carnosine in parts of rat's brain by using HPLC. Carnosine abundantly expressed in the olfactory bulbs and increased according to developmental change. Considering that olfactory bulb neurons are less sensitive to Zn^{2+}-induced neurotoxicity, it is possible that carnosine plays protective roles in zinc-induced neurodegeneration after ischemia in the olfactory bulb and proposed carnosine as a possible target for drug of VD.

P-8
Zn^{2+} signal in dentate granule cells is required for LTP maintenance and memory recall

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The real-time relationship between the extinction of LTP maintenance and the extinction of recognition memory was examined focusing on the loss of Zn^{2+} signal with zinc chelators in the dentate gyrus. Dentate gyrus LTP maintenance was erased by the transient loss of Zn^{2+} signal in the dentate gyrus. The irreversible erasure was rescued not only by amelioration of the loss of Zn^{2+} signal, but also by pretreatment with Jasplakinolide, a stabilizer of F-actin. It is likely that synaptic Zn^{2+} participates in the formation of F-actin and is required for LTP maintenance in the dentate gyrus. Simultaneously, acquired space recognition memory was affected by the loss of Zn^{2+} signal, but not by pretreatment with Jasplakinolide prior to the loss of Zn^{2+} signal. The present study indicates that the extinction of acquired memory coincides with the extinction of LTP maintenance under the transient loss of Zn^{2+} signal in the dentate gyrus. It is likely that dentate gyrus LTP maintenance is real-timely linked to the retention of space recognition memory.

P-9
Amyloid β-mediated enhancement of the hypothalamo-pituitary-adrenal axis activity and behavioral abnormality

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In patients with Alzheimer's disease, behavioral and psychological symptoms of dementia (BPSD) such as aggression, anxiety, and hallucinations are known to frequently occur in addition to the core symptoms, i.e., cognitive dysfunction and memory deficit. BPSD is a serious problem for caregivers. However, the mechanisms of pathogenesis of BPSD, as well as the core symptoms, are unclear. Amyloid β (Aβ) is involved with the pathophysiology of Alzheimer's diseases in cooperation with zinc. When human $A\beta_{1-42}$ protein was injected into the lateral ventricle of mice, the hypothalamo-pituitary-adrenal (HPA) axis activity was enhanced; serum corticosterone level was significantly higher than the control. Simultaneously isolation-induced aggressive behavior and anxiety-like behavior were increased as well as cognitive dysfunction. These results suggest that Aβ-mediated enhancement of the HPA axis activity is involved in behavioral abnormality. It is likely that Aβ affects the HPA axis activity.

P-10
Effect of iron and zinc deficiencies on blood pressure in rats

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Hypotension frequently associates with iron deficiency anemia (IDA), and some scientists reported that Zn deficiency induces hypertension, but the mechanisms of abnormal blood pressure are unknown. To investigate the physiological role of Fe and Zn in blood pressure control, we carried out a 2x2 factorial experiment with Fe and/or Zn (Exp. 1), and a pair-fed experiment of Zn deficiency (Exp. 2) using rats. Exp. 1: Blood pressures were significantly decreased in Fe/Zn-deficient rats compared to Control. Plasma aldosterone concentration was significantly decreased in Fe deficient rats and Fe/Zn-deficient rats compared to Control. Exp. 2: Blood pressures of Zn-deficient rats were significantly higher than the pair-fed control. Angiotensin II of Zn-deficient rats was marginally lower than the pair-fed control. These results suggest that secondary hypotension in IDA is actually evoked by simultaneous deficiency of Fe and Zn deficiencies, and that "zinc deficiency hypertension" is not caused by augmentation of renin-angiotensin system.

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P-11
Impairment of LTP maintenance and memory recall by excess of synaptic Zn^{2+} signal in the hippocampal dentate gyrus

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Synaptic Zn^{2+} signal in the hippocampus is involved in recognition memory and the excessive release in the hippocampal CA1 is involved in recognition memory deficit. However, it is unknown whether excess of synaptic Zn^{2+} signal erases LTP maintenance and acquired recognition memory. In the present study, long-term potentiation (LTP) maintenance and acquired space memory were assessed focusing on excessive Zn^{2+} signal in the dentate gyrus. Dentate gyrus LTP maintenance was erased by the transient increase in Zn^{2+} signal in the dentate gyrus, which was selectively induced with injection of exogenous Zn^{2+} or KCl into the dentate gyrus. Simultaneously acquired space memory was erased. The erasure was not restored even when the Zn^{2+} signal was returned to the basal level. In contrast, the erasure was rescued by co-injection of zinc chelators into the dentate gyrus. The present study indicates that excess of synaptic Zn^{2+} signal in the dentate gyrus erases not only LTP maintenance but also acquired space memory.

P-12
Fatigue-related Changes in Serum Trace Elements during a Continuous Work Loading Process

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The purpose of this study was to examine the effects of physiological fatigue during continuous fatigue and the recovery process, and to identify changes in trace elements such as zinc (Zn) with the fatigue index. Subjects (n=12) received a continuous desk work load for 18 hours, and took rest time 4 hours after work. Blood samples were collected every six hours, and again after resting. Concentrations of serum trace elements during the period of continuous fatigue and the recovery process were determined using inductively coupled plasma mass spectrometry (ICP-MS). Three fatigue indexes (flicker value, subjective symptoms, and Visual Analog Scale (VAS)) were also examined. The flicker value decreased, while both subjective symptoms and VAS increased during the fatigue period and decreased after resting. Serum Zn levels also increased during the fatigue load process, but remained high level in the recovery process. The behaviors of other trace elements in serum were also discussed in the paper.

P-13
A novel mechanism for zinc deficiency in a breast-fed infant: low milk zinc concentrations caused by compound heterozygous mutations in *ZnT2*

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Zinc concentrations in breast milk are considerably higher than those of the maternal serum to meet infant's requirements for normal growth and development. Thus, effective mechanisms ensuring secretion of large amounts of zinc into the milk operate in mammary gland during lactation. The zinc transporter ZnT2 was found to play an essential role in the secretion of zinc into milk. Here we identified a Japanese mother with low milk zinc concentrations (>90% reduction) whose infant developed severe zinc deficiency. Sequencing of the genomic DNA isolated from the mother's blood detected two novel missense mutations, W152R and S296L, on different alleles in the *ZnT2* gene. Biochemical characterization indicated that the W152R mutant abolished to transport zinc and to form dimer complex and that the S296L mutant retained both abilities but was extremely destabilized. Taken together, these results show compound heterozygous mutations in the *ZnT2* gene causing zinc deficiency in a breast-fed infant.

P-14
Functions of the C-terminus in Zinc Transporter-1 (ZnT1) as assessed by Mutant Zebrafish, slc30a1^{sa17}

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Zinc homeostasis is maintained by the action of zinc transporters. ZnT1 (SLC30a1) is present in the plasma membrane and exports zinc [from the cell to the extracellular matrix]. The zebrafish TILLING mutant slc30a1^{sa17} lacks the 40 last C-terminal amino acids of Znt1 due to a point mutation. These fish display a subtle phenotype that sheds light on the functions of ZnT1, some of which are not readily interpreted with impaired zinc transport. The zinc content of homozygous slc30a1^{sa17} fish was not different from that of the wild-type. However, these fish showed changed expression of several zinc transporters, altered distribution of free zinc(II), delayed embryonic development, yolk sack resorption, and reduced ERK1/2 phosphorylation. Remarkably, they also displayed a disturbed circadian rhythm associated with changes in expression of clock genes. Ongoing experiments are now linking molecularly the observed phenotype and functions of the truncated Znt1 protein.

P-15
Significant contribution of carbonic anhydrase, zinc enzyme, to normal taste discrimination.

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Zinc deficiency induces growth retardation, reduced reproductive function, dermatitis, taste abnormality, immune dysfunction, and so on. In the present paper we report the effect of dietary zinc deficiency on the taste discrimination in SD rats. At an early stage of zinc deficiency after 3 to 7 days of the feeding, abnormal increase of taste preference for NaCl solution and abnormal decrease of food intake itself were observed. After 7 to 14 days feeding of zinc-deficient diet, the rats showed the decreased chorda tympani and lingual trigeminal nerves sensitivity to carbonated water, in addition to the decreased carbonic anhydrase (CA) activity in the submandibular gland. At a severe stage, i.e., after 3 weeks or much longer-term of the zinc deficiency, decreased CA activity was observed, and considered that this is indispensable factor to maintain the taste discrimination ability of bitter substance (quinine-HCl) that is an aversive taste solution for healthy control rats. We found also that there is a positive correlation of saliva secretion with salivary gland CA activity, and finally it was suggested that salivary CA activity could be used as a taste abnormality index.

P-16
New aminobenzopyranoxanthene-based colorimetric sensor for Cu^{2+} with dual-color signal detection system

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Copper is required for body where it causes diseases in abundance, so rapid determination of copper is needed. New Aminobenzo-pyranoxanthene dyes (ABPX) have three chemical species of a colorless and two heterochromatic forms. We hypothesized this unique color system can be applied to sensor showing not only turning color on but also shifting of color dependent on Cu^{2+} concentration. The hydrazide group has high affinity to Cu^{2+} and thus, ABPX01-hydrazide (ABPX01-hy) was designed, synthesized and applied to absorptiometric Cu^{2+} chemosensor.
The absorption spectra were measured upon the addition of Cu^{2+} to ABPX01-hy. With a small amount of Cu^{2+}, absorption bands of the mononuclear complex species were observed. Further addition of Cu^{2+} caused dinuclear species to be emerged. Solution color firstly clear was turned on to pinkish-red and gradually shifted to purple, thus ABPX01-hy is suitable for naked eye Cu^{2+} sensing. There was a linear relationship in Cu^{2+} concentration of 0.064–93.5 μg mL^{-1} and absorbance.[1]

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P-17
Thermodynamic stability of 4N-Cu^{2+} complex is governed by basicity of N-terminal amine in ATCUN peptide

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The Xaa-Yaa-His tripeptide, named ATCUN motif, forms stable 4N-Cu^{2+} complex via 4N {NH_2, $2N^-_{amide}$, $N_{imdazole}$} binding mode. The complexes are of particular interest for applications in metallodrugs due to their interesting functions, such as DNA cleavage and inactivation of enzymes. However, it is still unclear what factors affect the stability of such complex.
Hence, we synthesized new derivatives of ATCUN peptides: NNHG5-NH_2 **1**, FFHG5-NH_2 **2**, RRHG5-NH_2 **3**, KKHG5-NH_2 **4**, VVHG5-NH_2 **5**, and DDHG5-NH_2 **6**. The stability of their 4N-Cu^{2+} complexes was quantitatively evaluated by pH titration.
In this study, we observed that low basicity of the N-terminal amine of the peptide led to the enhanced stability of the 4N-Cu^{2+} complex. These findings may accelerate the design of new ATCUN-based metallodrugs.

P-18
Utility evaluation of ^{64}Cu-labeled anti-CTLA-4 antibody as the probe for imaging CTLA-4 in the tumor

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Ipilimumab, anti-human Cytotoxic T-Lymphocyte Antigen-4 (CTLA-4) monoclonal antibody, is extremely useful for cancer treatment. Therefore, to aim the development of the probe that can potentiate CTLA-4 blockade therapy, we have synthesized the probe visualizing the expressions of CTLA-4 in the tumor.
DOTA-anti-CTLA-4 antibody was prepared and its binding affinity for CTLA-4 was evaluated by ELISA. The binding affinity of DOTA-anti-CTLA-4 antibody for CTLA-4 was 86.3 % of that of original anti-CTLA-antibody, whereas that of DOTA-isotype IgG was only 0.35 %. This result indicated DOTA-anti-CTLA-4 maintained the affinity to CTLA-4. PET imaging showed that ^{64}Cu-DOTA-anti-CTLA-4 antibody more accumulated in the tumor expressing CTLA-4 than ^{64}Cu-DOTA-IgG and noninvasive visualization of CTLA-4 was succeeded. These results suggested ^{64}Cu-DOTA-anti-CTLA-4 antibody leads to valuable diagnosis for evaluation of CTLA-4 expression in the tumor.

P-19
Is chaperone of superoxide dismutase (CCS) a copper marker?

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Abundance of mRNA transcripts of Cu-Zn-SOD chaperon (CCS) increase during copper deficiency, both in cell lines and rodent models. To assess this chaperone as potential copper marker useful in nutritional studies, it is necessary to test it in humans, in real life situations, when variable degrees of inflammation that modify other copper markers, are present. 566 healthy adults were screened and 211women and men without firm clinical diagnoses were selected and divided using their C Reactive protein (CRP) values, as clinical indicator of inflammation. Group1: 61 cases with CRP <3mg/dL; Group 2: 150 cases with CRP > 3mg/dL. Blood mononuclear cells were isolated and mRNA transcripts of CCS, SOD1, MT IIA and TNFα were determined. Results showed that abundance of transcripts of all proteins measured were not different between groups G1 and G2. These results further support that CCS may be a copper marker useful for human studies.

Key words: copper, CCS, humans

P-20
Studies on Cu complexes as therapeutic drugs for Menkes disease

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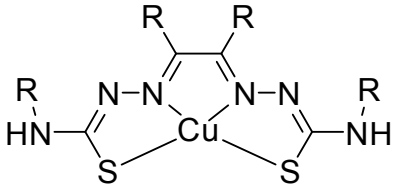
Menkes disease (MD) is a genetic neurodegenerative disorder characterized by copper deficiency. The current standard treatment, which involves parenteral copper-histidine administration, is less effective for neurologic symptoms. Toward a development of new therapeutic drugs for MD, various copper coordination complexes (Figure) were designed and synthesized concerning several factors such as charge, lipophilicity, and redox. The synthesis and preliminary evaluation of these copper complexes will be presented.



Figure

P-21
Underweight status increases risk for iron deficiency among women of reproductive age

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Many individuals in Indonesia are underweight and iron deficient, but the association between an underweight status and certain parameters for iron deficiency remains unclear. We investigated the association between body mass index (BMI) and iron deficiency in young women. A cross-sectional study was conducted in 9 subdistricts, from which 80 young women of apparent good health were randomly selected as study subjects. Body weight and height were measured, and BMI values were calculated and compared with age-specific BMI reference values for adolescent subjects. Underweight was defined as a BMI within the 5th percentile. Iron deficiency was noted for those with anemia (Hb<12g/dL) and iron depletion (serum ferritin<12 µg/L). We found that underweight subjects were roughly twice as likely to be anemic (OR:2.15; 95%CI: 0.74-6.17) and three times more likely to show iron depletion (OR: 3.167; 95% CI: 0.7-14.335) relative to non-underweight individuals. We conclude that iron deficiency is more prevalent among underweight young women, with underweight individuals showing a 2-3 fold increase in risk for this deficiency over their non-underweight counterparts.

P-22
Stable Neutron-Activatable Isotopes to Study the Efficacy of Fe Supplementation in Individuals with Iron Disorders

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Research on the role of nutrition with regard to trace element behavior in humans and animals is hampered by the limitations of radiotracer use. The use of neutron activatable isotopically enriched stable tracers received relatively little follow-up. Such enriched tracers have much to offer, and the approach is applied for a project on the efficacy of Fe supplements to people with iron metabolism disorders such as hemochromatosis and anemia. Use is made isotopically enriched 58Fe to label the supplement. The study requires analysis of the daily intake and of blood, urine and feces. The Fe in the dietary intake is measured using large sample neutron activation analysis. Neutron activation of blood and its components urine and feces is followed by the measurement of 59Fe as an indicator of supplement and 54Mn (from 54Fe) as an indicator for the naturally present Fe in the body-pool. The analytical strategy of this study will be outlined for discussion, including the challenges and problems to be expected.

P-23
Iron deficiency without anaemia is associated with anger and fatigue in young Japanese women

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Iron deficiency without anaemia (IDNA), the most prevalent nutritional deficiency worldwide, affects young women of reproductive age. This study aimed to elucidate the relationship between IDNA and mental and somatic symptoms, including anger and fatigue, using the Japanese version of the Cornell Medical Index Health Questionnaire (CMI-J). Data regarding demographic characteristics, anthropometry, haematological and biochemical indices of iron status, frequency of non-specific symptoms and neurotic traits were collected from 72 female college students, aged 18-22 years. In the IDNA group, sections A-R (overall complaints) and I (fatigability) of CMI-J showed significantly higher scores than those in the normal and IDA groups, while sections B (respiratory system) and Q (anger) scored significantly higher than in the normal group, even though IDA subjects showed no significant differences in those sections compared to normal subjects. These findings suggest that IDNA may be a risk factor for anger and fatigue in this population.

P-24
Effects of iron chelator from plant food upon iron absorption in human

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The plant foods such as beans and broccoli contain a large amount of nicotianamine (NA), which mainly transports iron in plants. For elucidating the effects of NA upon iron absorption in human, we carried out *in vitro* assays for examining the uptake of NA and ^{59}Fe–labeled NA complex using caco-2 monolayer cells. Quantitative analysis of NA was carried out with LCMS-8030 (Shimadzu) after derivatization with FMOC. For *in vivo* assays, mice were oral administrated with ^{59}Fe or ^{59}Fe labeled-NA complex. After one, two or five hours (each n=5), blood and tissues were taken from mice, and their iron concentrations were measured ICP-AES (ICPE-9000, Shimadzu). The results revealed that Fe-NA complex transports iron to the basal side (blood vessel side) via caco-2 cells and *in vivo* assays using mice showed that Fe-NA administration induces approximately two times increases of iron contents in lung, stomach and skeleton muscle as compared with Fe only administration. The data further suggest that NA is implicated in iron trafficking in the human intestine.

P-25
Reproductive toxicity of iron oxide nanoparticles in CD-1 mice

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Interest exists in nanoparticles (NP) because of their small size, unique properties, and customizability. Iron oxide NPs are currently being investigated for many uses including drug delivery and magnetic imaging. Yet, the risk to pregnant women is high as the small NPs have the potential to cross the placenta. Studies in our lab show that iron oxide nanoparticles can cross the placenta, and low doses (10 mg/kg body mass) of NPs given intraperitoneal (IP) for 8 consecutive days result in significant decrease in maternal weight and increase in fetal death. This study was performed to evaluate the reproductive toxicity of iron oxide NPs in mice. Female CD-1 mice were given an IP dose of 100 mg/kg on gestation 8, 9, or 10. Mice were allowed to litter, and reproductive organs were examined on postnatal week 6-10. Edematous uteri and decreased testicular seminiferous tubule epithelium were observed in the offspring of NP-treated dams.

P-26
The Effects of Zinc (Zn) and Iron (Fe) for Increase IL -10 and IL-12 on Elemetary Students after Inffected Malaria Deases Who Lived in Endemics Area

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Previous research on Malaria only focussed on children who suffering the desease. However, we have not study for children after infected of Malaria deases who lives in endemics area. The aimed of this research to analyze Zn and Fe for increase IL – 10 and IL – 12 for elementary students as subject who inffected after Malaria. The result of this research found that Zn and Fe concentrations can increase IL – 12 protective. However, IL – 10 unnecessary as Sitokin Anti inplammatory for elementary students after inffected Malaria who lives in endemics area. More Study for checking others Sitokin by using Zn and Fe suplement for prevent Malaria deases coming up.

Keywords: Malaria, Zn, Fe, IL – 10 , IL - 12

P-27
Blood zinc, copper & calcium in pre- & term infants & mother's blood & milk and effect on growth on feeding mother's milk and preterm formula

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Study was conducted to assess zinc, copper and calcium status of preterm neonates to ascertain growth output when fed on mothers' milk and on preterm formula. Group I-28 neonates-15 PreAGA and 13 PreSGA and fed on their mother's milk. Group II-20 neonates-10 PreAGA and 10 PreSGA and fed on preterm formula. Neonate's weight, length, and cranial circumference were serially recorded with neonate's blood, mother's blood, breast milk for estimation of zinc, copper, & calcium at birth, 30, 60, and 90 day in group I and II. In group I infants weight gain on 30, 60 and 90 day was 690±64, 1530±109, and 2200±172 g; length gain on 30, 60, 90 day was 3.5±0.28, 7.6±0.48, and 11.1±0.53 cm; cranial circumference increase on 30, 60 and 90 day was 1.9±0.13, 3.9±0.14, and 5.8±0.16 cm, respectively. In group II infants weight gain on 30, 60 and 90 day was 800±77, 1735±99, and 2621±53 g; length gain and cranial circumference increase on 30, 60, and 90 day was similar with both groups. Serum zinc, copper, and calcium levels were low in preterm neonates.

P-28
Comparison of element levels of blood, teeth, hair, nails and urine of subjects from Mysore, India

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Elements causing harm are to be monitored. In the present study, whole blood, permanent teeth, urine, scalp hair and fingernails of subjects from Mysore and adjoining villages were analyzed for Cd, Co, Cu, Cr, Fe, Mn, Ni, Pb and Zn levels with ICP for evaluating status, sources and health impact. The results revealed positive and negative element interaction, rural and urban exposure gradient, occupational exposure of workshop and cement factory workers. Lifestyle factors (food, water, alcoholism and smoking) also influenced element values. The higher Pb and lower Zn in urban diabetics and higher Pb in type-1 diabetics were ascribed to mode of intake. Elemental correlation showed that nails and blood were comparable respectively to hair and urine, but teeth were unique. Different exposure nature elucidated, relied on sample features. Cd and Ni were within the baseline levels in all the samples, except urinary nickel. The lower Pb levels of urine, hair and nails were considered as reference values. Their use for human exposure assessment is found in the following order of choice: nail, blood, hair, urine and teeth. Hence, nails are an ideal sample.

P-29
Epidemiological study: Interaction between iron deficiency and lead poisoning on Moroccan children's health

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Iron deficiency and lead poisoning are common among infants and children in many parts of the world and often these two problems are associated to one another. Both conditions are known to cause anemia and if they are combined, they tend to cause a more severe form of anemia. Even though the nature of their relationship is not completely elucidated, characterization of a common iron-lead transporter and epidemiological studies among children strongly suggest that iron deficiency may increase susceptibility to lead poisoning. Recent human studies suggest that high iron intake and sufficient iron stores may reduce the risk of lead poisoning. Future clinical trials are necessary to assess the effect of iron supplementation in the public health prevention of lead poisoning.

Indeed, the supplementation iron's study done in Marrakech, Morocco on an anemic infant population (3 to 13 years) living in a lead polluted area confirms this fact.

P-30
Use of human primary tooth enamel as a biomarker for lead, iron, copper and zinc exposure during prenatal and neonatal development

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Lead (Pb) may affect neurodevelopment of children by direct toxicity. There is also evidence that zinc (Zn), copper (Cu) and iron (Fe) status affect early development. In this work, we examine pre- and neonatal exposures to trace metals using the enamel of 151 primary teeth gathered from children in Quebec, Canada, as a biomarker for Pb, Zn, Cu and Fe exposure.
In primary tooth enamel, divalent cations may substitute for isovalent calcium (Ca) sites in hydroxyapatite [$Ca_{10}(PO_4)_6(OH)_2$], the main component of which enamel is formed. The highly conservative nature of the hydroxyapatite crystal may allow for its use as a biomarker of cumulative exposure to Pb, Fe, Zn, and Cu. Once teeth are shed naturally, they can be analyzed in an energy dispersive x-ray fluorescence spectrometer. Trace element quantities in deciduous tooth enamel will be compared to measured neurological outcomes including full WASI IQ test scores and Santa Ana motor neuron functionality test scores.

P-31
Trace elements and inflammatory and oxidative markers in hemodialysis patients

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Oxidative stress, inflammation and trace element imbalance occur in most hemodialysis (HD) patients.
In order to test trace element status and the possible influence of inflammatory state and oxidative stress in HD patients on biomarkers in plasma and erythrocytes, we assessed related parameters in 100 HD patients. They included: Fe, Cu, Zn, and Se levels in plasma and erythrocytes; total and non-transferrin Fe, ferritin, C-reactive protein, albumin/globulin, ceruloplasmin, protein carbonylation and superoxide dismutase.
Plasma Fe and Cu markers were clearly affected by inflammatory state and oxidative stress, while erythrocyte concentrations remained unaffected. HD patients showed Zn and Se deficiency and apparently normal Fe and Cu plasma levels.
In HD patients, Fe and Cu plasma level was not a reliable marker of body status. Using erythrocytes as a marker of trace element status should be evaluated, especially in relation to the role of Fe (indirectly of Cu, as well) for efficient anemia treatment in HD patients.

P-32
Assessment of Human Body Iodine Status using Hair, Blood, and Urine: Scaling Matters

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Currently, human iodine (I) status is assessed by analyzing urinary I with the *a priori* assumption that the geometrical organization of data is linear. This study involved the analysis of hair I (H_I) levels in 870 Croatians, whole blood I (B_I) levels in 311 Croatians, and urinary I (U_I) levels in 98 Cameroon men and women from a WHO 2007 study. Data were analyzed via the median derivative method (the power function). All resulting bio-indicators exhibited a characteristic sigmoid curve with a linear section, having medians of 0.05, 0.12, and 0.50 $\mu g \cdot g^{-1}$ for B_I, U_I, and H_I, respectively. This linear region can be graded into low adequate (LA), adequate (A), and high adequate (HA) ranges, depending on the degree of bio-indicator saturation with iodine. We estimated optimal I saturations (70%) to be: B_I (0.068), U_I (0.156) and H_I, (0.857) $\mu g \cdot g^{-1}$. The current linear scaling used in geometrical organization of bio-indicator iodine data is false, and should be replaced with the power function.

P-33
Median Concentrations of Phosphorus in Human Hair Do Not Differ between Croatia, Macedonia, Japan, and the United States

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Median concentrations of phosphorus (P) in human hair in subjects from two different prospective studies in Croatia (C_J, n = 311; C_V, n = 761), one from Macedonia (n = 91), one from Japan (n = 33) and one from a pooled group in the United States (n = 271) were compared. Hair was washed using different procedures and P concentrations were analyzed via different analytical instruments, including ICP-ES and ICP-MS at different time intervals over 25 years. The median hair P level for all tested groups ranged from 147–153 $\mu g \cdot g^{-1}$, with no difference between men and women. Evidently, the analytical variability of hair P concentration is negligible and unrelated to possible external environmental contamination, type of shampoo used, pre-analytical washing procedures, hair biological matrix destruction method, place of residence, or race. Hair P concentration reflects the constant rate of hair protein synthesis from the replication of DNA phosphate backbone structures.

P-34
The effects of low dose sodium selenite supplementation to normal human esophageal cell line (CHEK-1 cells) against radiation treatment

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Administration of radioprotective agents has been suggested for prophylaxis against side effects of radiation therapy. Selenium is a suggested preventive approach for radical detoxification. The aim of this study is to investigate the effects of sodium selenite supplementation on CHEK-1 cells treated with X-ray radiation. GPx activity assay results showed that the activity increased dose-dependent manner and reached plateu at 50nM sodium selenite supplementation for 72h. Citotoxicity assay results showed that the IC50 of sodium selenite to CHEK-1 cells was 3.6μM. Cell survival assay results showed that the cells supplemented with 50nM sodium selenite for 72h and treated with 2Gy X-ray survived better than the cells treated with 2Gy X-ray radiation alone ($p<0.05$). These results suggest that sodium selenite is a promising agent as a radioprotector of normal cells.

P-35
Selenoenzymes iodothyronine deiodinases: novel radiometric methods for determination of their activities

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General properties of selenoenzymes iodothyronine deiodinases (IDs) of types 1, 2 and 3 and their functions are described. In particular, we show several applications of our newly developed radiometric enzyme assays for determination of IDs activities in specific samples of biological material. These assays are based on the use of ^{125}I-labeled iodothyronines of high specific radioactivity as substrates; TLC separation of radioactive products from the unconsumed substrates; film-less autoradiography of radiochromatograms using storage phosphor screens; and quantification of the separated compounds with laser scanner.
With the use of these assays we followed, for example, the alterations in IDs activities caused by short-time incubation of cultured astroglial cells in a chemically defined medium with various concentrations of several effectors, such as retinoic acid, adrenergic and purinergic agonists, etc. Further, we determined changes in IDs activities in various rat tissues as a response to the effects of an antidepressant fluoxetine.

P-36
Role of glutathione in the efflux of selenium from cells

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Selenite is a source of selenium for the synthesis of selenoproteins in mammalian cells. Although the involvement of glutathione (GSH) and/or thioredoxin reductase in selenite metabolism has been suggested, intracellular selenite metabolism remains unknown. In this study, we investigated the role of GSH in selenium metabolism in Hepa 1-6 cells. The depletion of GSH did not affect the amount of selenoprotein, suggesting that GSH is not essential for the reduction of selenite in selenoprotein biosynthesis. Our data suggest that GSH is involved in the efflux of low-molecular-weight selenium compounds from cells. Moreover, the addition of selenite inhibited the efflux of a fluorescent bimane-GS conjugate that may be mediated by ATP-dependent multidrug-resistant proteins. These results suggest that GSH plays a role in the excretion of selenium from cells by forming a GSH-conjugate, which may contribute to the distribution, detoxification, and homeostasis of selenium in the body.

P-37
Selenium Removal from Wastewater Using Ti^{3+} as a Reducing Agent

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Selenium is considered an essential element for animals, but is also known to be toxic. Se is used in semiconductors and regulated not to exceed 0.01 mg/L in industrial wastewater. Se exists in -2, 0, +4, and +6 oxidation states, of which Se^{4+} and Se^{6+} are toxic. There are currently no methods available to reduce Se^{4+} and Se^{6+}. We studied the dissolution states of Ti^{3+} and Ti^{4+}. $TiCl_3$ could be utilized to reduce Se compared with other reagents, such as $SnCl_2$ and ascorbic acid.
Reducing agents, $TiCl_3$, $SnCl_2$, and ascorbic acid were used in these studies. Dissolution states of Se^{6+}, Sn^{2+}, Ti^{3+}, and Ti^{4+} were identified by ESI-MS. Se^{6+} was not reduced by ascorbic acid. With $SnCl_2$, Se^{6+} changed to Se(metal), but the reaction rate was slow. Sn gradually precipitated during the process. With $TiCl_3$, the reaction $Se^{6+} + 6Ti^{3+} \rightarrow Se\downarrow(red) + 6Ti^{4+}$ was rapidly observed and TiO_2 was produced, as reflected by the red coloring of the solution. Using $TiCl_3$ to reduce Se^{6+} resulted in Se and Ti precipitates that were easily removed from the solution. Detailed procedures are described.

P-38
Agronomic biofortification of upland rice with selenium to improve human health

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Evaluating selenium (Se) intake in specific human populations is seldom precise due to differences in geography, agronomic practices, food availability, and preferences. Se contents of plants vary according to the element's soil availability. Low soil Se results in Se deficiency within the food chain (i.e., in plants grown in these soils and livestock and people fed with the plant products). This study evaluated genotypic variations in Brazilian upland rice regarding Se accumulation in edible parts and tested an agronomic biofortification strategy to enhance Se content in rice grains. Significant varietal differences in Se content of rice grains was noted, ranging from 15 to 122 µg kg^{-1} DW (dry weight) without Se application. Application of 0.5 mg dm^3 of Se (sodium selenite) significantly increased the Se concentration in rice grains (up to 9237 µg kg^{-1} DW), demonstrating that agronomic biofortification is important in improving human nutrition.

P-39
The development of a next generation PET for multiple molecular imaging

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Positron Emission Tomography (PET) is a molecular imaging technique used in nuclear medicine and life science, producing functional images of radioactive probes. Iodine-124 is one of the positron-emitting isotopes used in PET imaging. The principle of the PET system is based on detection of positron-electron annihilation gamma rays, which have constant energies of 511 keV. Because of this principle, conventional PET cannot identify different nuclear species and multiple radiotracers are not used simultaneously. However, multiple molecular imaging - simultaneous imaging analysis of multiple biological processes provides detailed information about pathologies and numerous life phenomenon. Therefore, we have been developing a next generation PET system that enables multiple molecular imaging. This system utilizes a de-excitation gamma ray having an intrinsic energy in nuclide, emitted with positron. We examined the new system's performance using ^{124}I and other isotopes.

P-40
Status of Iodine Deficiency Amongst Pregnant Mothers in Himachal Pradesh, India

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Iodine is an essential micronutrient. Iodine Deficiency (ID) in Pregnant Mothers (PMs) leads to compromised mental and physical development of fetus. Himachal Pradesh (HP) is a known endemic region for ID. Present study was conducted to assess current status of Iodine Nutrition amongst PMs in HP. Three districts (Kangra, Kullu and Solan) one from each region of HP was selected. In each district, 30 clusters were selected by utilizing population proportionate to size cluster sampling. Total of 1711 PM were included. Clinical examination of thyroid of each PM was conducted. Urine and Salt samples were collected from a sub sample of PMs. Total Goiter Rate (TGR) was 42.2% (Kangra), 42.0% (Kullu) and 19.9% (Solan). Median Urinary Iodine Excretion (UIE) levels were 200µg/l (Kangra), 150µg/l (Kullu) and 49.9µg/l (Solan). Percentage of PMs consuming salt with iodine content of 15ppm and more was 68.3% (Kangra), 60.3% (Kullu), and 48.5% (Solan). Study revealed that according to TGR all districts had ID as a public health problem. However, as per UIE district Solan had severe ID which was substantiated by intake of inadequate iodized salt by PMs.

P-41
Altered expression of Mn-SOD and CuZn-SOD in diabetic condition

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The superoxide dismutase (SOD) family serves as a defense system against oxidative stress and plays a critical role in eliminating superoxide radicals. In mammals, there are three isoforms of SOD, including CuZn-SOD, Mn-SOD, and EC-SOD. The reported activity of SOD in the diabetic state varies based on the gender, rodent species, the duration of diabetes. Diabetes is characterized by disturbance of glucose metabolism due to absolute or relative insulin deficiency. Hyperglycemia reveals oxidative stress is strongly involved. In the current study, we analyzed hepatic Mn-SOD and CuZn-SOD expression and oxidative status in type 1 and type 2 diabetic conditions.
In Goto-Kakizaki rats (type 2 diabetic model), hepatic Mn-SOD and CuZn-SOD expression were not altered compared with controls whereas thiobarbituric acid reactive substances (TBARS) level in liver was increased. In streptozotocin induced type 1 model rats, hepatic Mn-SOD expression and TBARS level were increased compared with controls. We speculate that the change of hepatic Mn-SOD is related with factors other than lipid peroxidation.

P-42
Analysis of gene expression profiles in the heart of magnesium-deficient model mice

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Recently, relationship between heart disease and homeostasis of magnesium (Mg) concentration is considered significant. In previous studies, it was observed that cardiac tissue structures were disrupted in Mg-deficient mice (MgD). In this study, to clarify interactions between Mg and biomolecules related pathogenesis, we studied changes of gene expression in the heart of MgD.
We analyzed comprehensive gene expression changes in heart tissue among normal mice, MgD and MgD+Mg using DNA microarray analysis. MgD affected the expression of 5,496 genes, with 3,003 genes up-regulated and 2,493 genes down-regulated as compared to control. In addition, comparison of MgD with MgD+Mg indicated 2,669 gene up-regulated and 2,827 genes down-regulated. A lot of genes altering expression are related to signaling transduction, substrate metabolism and/or morphogenesis when we classified the altered genes with gene ontology. It is suggested that these expression changes are related impaired myocardial energy gain and function.

P-43
Serum magnesium in preterm and term neonates

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We have analyzed magnesium concentration in cord blood after birth of 35 term and 35 preterm neonates. Mothers of these neonates were selected for blood magnesium. Infants born before 37 weeks from the first day of LMP were termed preterm. Infants born between 24-35 weeks of gestation were selected for cord blood. Mother's venous blood was also taken. Serum magnesium level in term neonates was 1.85±0.28 mEq/L compared with preterm neonates 1.68±0.19 mEq/L ($p<0.05$). Serum magnesium level in term delivered mothers was 1.99±0.31 and in preterm delivered mothers was 1.76±0.19 mEq/L ($p<0.001$). Results show relationship in serum magnesium at birth in different gestational age. Serum magnesium level in preterm neonates was significantly low compared to term neonates. Serum magnesium in preterm delivered mothers was also low compared with mothers who delivered at term. Since magnesium is an important element for fetal tissue, therefore during pregnancy it should be measured when its deficiency is expected and therapeutic measures should be taken.

P-44
In vivo quantification of strontium in bone using handheld X-ray fluorescence spectrometers

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Questions still remain as to strontium's potential essentiality with regards to bone health and the use of strontium-based drugs for the treatment of osteoporosis presents a problem in regards to the determination of bone mineral density by DEXA. The latter problem can be resolved if a reliable estimate of subject bone strontium levels is known. This study evaluated three handheld X-ray fluorescence (HHXRF) spectrometers for the purpose of quantifying bone strontium focusing on the development of comparative analytical figures of merit, as a function of measurement time. The systems consisted of Ag, W and Rh anode X-ray tubes and SDD and SiPiN detection systems. Measurements were made on a series of plaster of Paris as well as novel hydroxyapatite phantoms to assess the analytical figures of merit as a function of measurement time. It was found that HHXRF units can potentially be used to quantify strontium in bone with as little as a one second scan time.

P-45
Vitamin A Fortification of Edible Oils and Vanaspati Ghee in Pakistan

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Malnutrition, including micronutrient malnutrition, is a major contributor of child mortality in Pakistan. Surveys have shown the prevalence of vitamin A deficiency among preschoolers to be over 30%, highlighting it as a subclinical and biochemical issue in children in Pakistan. Vitamin A fortification in cooking oils provides approximately one-third of the recommended daily intake (RDI). There is an approximately 16 kg consumption of edible oils per capita in Pakistan. Low variation makes it an ideal food vehicle for producing efficient results. Pakistan has legislated vitamin A fortification of ghee and oils. Laws in Pakistan specify vitamin A fortification at 16000 IU/lb fat, which provides about 33% of the RDI to the average adult. It is an affordable option at US $2.00 per ton of production or PRS 2.00 per person per year. However, outcomes have not been satisfactory. Important initiatives include increasing awareness among producers and consumers, creating public-private sector partnerships, forming a fortification task force, and emphasizing training, monitoring, analysis, reporting, and sampling.

P-46
Titanium measurement in clinical samples

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Due to its resistance to attack from body fluids and high strength, titanium has numerous medical applications. Every year over 1 million kilograms of titanium in the form of hip joints, bone screws, knee joints, bone plates, dental implants, surgical devices and pace makers are implanted into humans. Blood titanium measurements can be used as a means of assessing the rate and severity of prosthesis wear in much the same way as blood chromium and cobalt concentrations have been used. In this presentation I will outline some of the chemical properties of titanium that make it a popular choice for biomedical applications and highlight some of the challenges associated with its measurement in clinical samples.

P-47
The quantification of low levels of noble metals in biological matrices by total reflection X-ray fluorescence spectrometry

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The use of noble metals such as Pt and Au for cancer therapy is rather well established. Current work is now focusing on increasing the uptake of said elements into cells using ultrasound-assisted methods and monitoring the general elemental changes that such approaches induce in cell cultures and tissues. One of the major challenges with such studies is quantifying elemental concentrations in cell cultures given their very low concentrations and very small sample volumes. This work describes a total reflection X-ray fluorescence spectrometry (TXRF)-based method for the *in situ* quantification of the noble metals in biological matrices. The method is based on the use of a molybdenum target X-ray tube and the use of high purity quartz reflectors as sample carriers. Small sample volumes (*i.e.* 1 µL) can be measured directly, with minimal sample pre-treatment, resulting in detection limits (as Au and Pt) on the order of 10^{16} atoms/mm^2.

P-48
The element composition of cow's hair of Moscow region

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The element composition of hair of Black Motley cows from the Moscow region has been studied by ICP – AES and ICP – MS methods. According to our investigations the levels (min-max) of essential and toxic chemical elements in the terminal hair of tail of cows are as follows (mg/kg): Al 22-289; As 0,0502- 0,1515; Be 0,005-0,0248; Ca 818-2326; Cd 0,0108-0,0329; Co 0,0605- 0,3103; Cr 0,1661-0,9405; Cu 6,84-12,08; Fe 36-578; Hg 0,0012-0,0203; K 519-8223; Li 0,0734-0,2284; Mg 385-1818; Mn 7,25-44,55; Na 26,5-677; Ni 0,2847-1,1007; P 165- 960; Pb 0,0418-0,2429; Se 0,2030-0,4235; Si 1,35-141,6; Sn 0,009-0,1024; Ti 1,0248-8,1935; V 0,0713-0,6806; Zn 48,5-120,6. It is shown, that the element composition of hair changes depends on type of feeding and biogeochemical features of territory of facilities.
As an express-method of eco-biogeochemical estimation of territory of agrolandskape it is expedient to use the chemical element analysis of hair cover of cattle, feeding of which is carried out by sterns, grown only on the studied territory. Data on chemical composition of this test-object allows exposing ecological connections of living organisms with the geochemical environment of their dwelling.

P-49
Assessment of environmental exposure to methylmercury in Polish women on the basis of hair mercury determination

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The purpose of this study was to assess the total hair mercury (THg-H) concentration in 306 women living in three regions of Poland with different fish consumption: Lodz - market fish, Gizycko - freshwater fish, Wladyslawowo - Baltic See fish. The geometric mean of THg-H concentrations were 0.115 µg/g in Lodz, 0.235 µg/g in Gizycko, 0.185 µg/g in Wladysławowo and demonstrated a statistically significant correlation ($p<0.0001$) with the frequency of fish meals consumed: 3.0 ($\beta=0.0124$); 10.5 ($\beta=0.0145$,) and 6.6 times/month ($\beta=0.0204$), respectively. Comparison of the slope coefficient β did not showed statistically significant differences ($p>0.06$), indicating that the concentration of THg-H did not depend on the type of fish consumed. Though results of THg-H concentrations were rather low they exceeded the US EPA reference dose of 1 µg/g in 4% of women in Gizycko and 1% in Wladyslawowo. There are no results that exceeds the 1 µg/g in Lodz. The results of our study revealed that fish consumption in Poland is not a threat to the health of women, also in the event of their pregnancy.

P-50
Cr (VI): Modified Extraction from Soil by Ascorbic Acid

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Use of ascorbic acid to reduce Cr (VI) was evaluated as a method to control soil contamination. This study discusses the application of ascorbic acid to reduce and remove Cr (VI) from soil.
A total of 1 ml 10 ppm Cr (VI) was added to 1 g soil sample aliquots and incubated for one day. Ascorbic acid and water were added separately to different sample aliquots, which were shaken for 1-14 days. The solution containing chromium and the sediment containing Cr (VI) of the soil were extracted with 5 mM sodium carbonate and 10 mM sodium hydrogen carbonate and measured by ICP-MS and an absorptiometric method with diphenylcarbazide.
Cr (III), which was reduced from Cr (VI) by ascorbic acid, complexes with negatively charged ascorbate. A total of 20-30% Cr was recovered from 1 g soil samples following addition of 1 ml 10 ppm Cr (VI), 1 mM ascorbic acid, and water. The extraction rate was 20- to 30-fold higher than the extraction process using water. After shaking for three days using this procedure, Cr (VI) soil concentrations were below the environmental standard. Cr (VI) remained in the sediment, but was not eluted by water and the ascorbic acid solution after shaking for 14 days.

P-51
Fixing Cesium to Soil with Silicic Acid

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The earthquake in east Japan led to the Fukushima Daiichi Nuclear Power Plant disaster, causing ^{137}Cs and ^{90}Sr fallout on the ground surface. After two and a half years, significant amounts of Cs have been incorporated into clay soil structure. In addition, Cs in ground water will be inevitable in the future. In this study, we examined silicic acid as a fixative for Cs in soil.
Soil was meshed to 1 mm and dried at 110°C for one week. One ml of a 10 ppm Sr or Cs solution was absorbed to 1 g of soil for one day. The soil was washed with 20 mL of solution, which consisted of pure water and silicic acid without any metal ions. Chemical species were identified by three mass spectrometers: FAB, ESI, and ICP.
Cs recovery from the soil using pure water was between 3% and 5%. Recovery using a water silica solution was between 0.5% and 1.5%. Cs recovery decreased against increasing extraction time in pure water and the water silica solution. Addition of silicic acid fixes Cs in the soil. That is, Cs is trapped in clay soil structure, and silicic acid prevents Cs release.

P-52
Erythrocytes as biomarkers of changed metal ion homeostasis in patients with Parkinson's disease

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In neurological disorders like Parkinson's disease the etiology is not well known. Erythrocytes may be useful as biomarkers by their travel between different organs gathering information during their life span of about 120 days. Blood test by isolating erythrocytes and screening elements by ICP-MS may be used for a preliminary test of the disease. Twelve patients with Parkinson's disease showed significant elevated concentrations of silver and lead in the erythrocytes. The accumulated lead and silver ions in the erythrocytes may interfere with metal ion homeostasis. Silver ions are known to react with thiol, selenol groupes disturbing the antioxidant systems. The observations indicate that erythrocytes may be used as a biomarker of elemental changes of homeostasis at the cellular level. The accumulated lead and silver in the erythrocytes will be discussed in more details in the paper.

P-53
First case of isolated sulfite oxidase deficiency in Japan

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Background: Isolated sulfite oxidase (SO) deficiency is a very rare and devastating inborn error of sulfur metabolism, characterized by refractory seizure, rapid progressive encephalopathy and dislocated ocular lenses.
Case report: We report a case of infant boy with isolated SO deficiency who presented with generalized seizures at one month old.
Brain CT scan showed bilateral multiple subcortical cystic lesions. MRI revealed severe cystic leukomalacia, cortical atrophy with ulegyric pattern on ten days after onset.
Herpes virus detection in CSF using PCR is negative. Other studies for CNS infection are all negative.
Serum uric acid level is 3.3mg/dl. Plasma levels of cystine and total homocysteine are very low. Urinary excretion of sulfite is over 40mg/ml detected by sulfite test. Urinary excretion of sulfocystein is very high (174µmol/mmol Cre.).
The mutations are c.1040T>C and c.796G>A in SUOX, the gene that encodes the molybdohemoprotein SO.
Conclusion: When we found multifocal leukoencephalopathy and uric acid in plasma is not low, it necessary to consider isolated SO deficiency.

P-54
Trans-Platinum(II) Complex with Flavone Containing Ligand: Spectroscopic Identification and Cytotoxicity *in vitro*

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Cisplatin is one of the most widely administrated anticancer drugs, used particularly for ovarian and testicular tumors. In order to diminish side-effects caused by cisplatin the search for new platinum drugs is ongoing. Recently platinum complexes with *trans* geometry have been found to possess significant antitumor properties [1].
The aim of the study is the synthesis of *trans*-$PtCl_2(3\text{-af})_2$ with 3-aminoflavone ligand and evaluation cytotoxic activity against ovarian cancer cell lines.
The characterization of this complex has been undertaken using reversed HPLC-MS and ICP-MS. The complex has comparable toxic effect with that of cisplatin against ovarian resistance cells A2780cis and 3-4 times less active towards A2780 cells.
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P-55
Ethylmercury indirectly produces Ca^{2+} influx by the stimulation of P2X receptor

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Ethylmercury increased intracellular Ca^{2+} level of C8-B4 cell, but thimerosal didn't it. Ca^{2+} channel blocker did not change the increase of intracellular Ca^{2+} level induced by ethylmercury, and Ca^{2+} influx induced by ethylmercury was not observed after the pretreatment of EGTA. In the whole cell patch clamp method, ethylmercury did not induce Ca^{2+} current. Ethylmercury may not induce Ca^{2+} influx by the action on the extracellular membrane, although Ca^{2+} in extracellular fluid is penetrated into the cell. Then, we observed the participation of the P2X receptor which affects Ca^{2+} influx. Suramin, non-selective P2 antagonist, inhibited Ca^{2+} influx induced by ethylmercury, and both P2X4 and P2X7 mRNAs were expressed in C8-B4 cells by the addition of ethylmercury. From these results, it is thought that thimerosal is divided into ethylmercury and thiosalicylate after the penetration into the cell, and ethylmercury may increase cytosolic ATP content. ATP activates P2X receptor after ATP is released into the extracellular fluid, and Ca^{2+} influx may be induced.

P-56
Geographic Determinants of Trace Element Status

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Trace elements play a critical role in optimal health. Their status is affected by genetics, physiology, biochemistry, disease, and diet intake /bioavailability. On a global basis, impact of geographic determinants must be considered, as soil composition, water, vegetation, and culture vary. In China, selenium (Se) deficiency resulting from inadequate soil levels coupled with a mutated virus has produced Keshan disease. This congestive cardiomiopathy also has been observed in Se-deficient areas of New Zealand and Finland. In mountainous locations (Himalayas, European Alps, Andes) and lowland (Central Africa, Eastern Europe) regions far from the ocean (a replenishment source), iodine (I) deficiency is prevalent as it is washed away by glaciation or flooding. A deficiency also can occur in areas where plants containing goiterogens (cassava) are consumed, such as Congo-Brazzaville in Africa. In terms of culture, widespread vegetarianism due to religious beliefs in India is associated with a high rates of anemia and stunting due to inadequate intakes of bioavailable iron and zinc, respectively. This paper will address the impact of geographic determinants on trace element status.

P-57
Analysis of total arsenic and its species in Australian rice

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There has been an increased concern on elevated levels of total arsenic (As) and its inorganic species in rice in many countries, including Australia. The aims of this study were to analyse and compare the arsenic levels in the Australian rice with the imported rice, and to check for compliance for As content in cereal grain. The samples (n=36) of Australian and imported rice were obtained from various market outlets in Brisbane, Australia. The total As level was analysed by ICP-MS after microwave digestion, and the As speciation analysis was performed by hyphenated system of HPLC-ICPMS. A wide variation of total As levels (range: <0.05 – 0.42 mg/kg) in Australian and imported rice was found. The mean level of total As (0.24 ± 0.09 mg/kg, n=10) in the Australian rice was relatively higher that the imported rice from other countries (0.09 ± 0.04 mg/kg, n=26). The mean level (0.25 ± 0.08 mg/kg, n=7) of dimethylarsinic acid (DMA) was considerably higher than the inorganic As (III) (0.07 ± 0.03 mg/kg, n=7) in the Australian rice.

P-58
The speciation of cadmium binding proteins for the cadmium transport and the protection to the cadmium toxicity in the male and female reproductive organs.

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An induction of metallothionein (MT) and MT like cadmium (Cd) binding protein (MT-like Cd-BP) in male and female reproduction organs were investigated for the role of Cd toxicity protection and of the Cd transport. Male and female Wistar strain rats were given Cd ip injection or oral administration. MT proteins were determined separately by the ELISA method for the iso- MT (I&II), and by the Cd-Hem method for total MT (T-MT). A hemorrhagic inflammation disorder by Cd toxicity was protected by Cd pretreatment. MT-like Cd-BP concentration increased significantly. These results suggested that the contribution of MT-like Cd-BP alleviate in the Cd toxicity. The remarkable manifestation of the MT-II gene in placenta suggested that MT protein in placenta was caused by the increase of MT-II gene expression, as suggesting that MT in placenta prevents the Cd transport into fetus. It was suggested that the different Cd protein species were carrying the different role in a male and female reproductive organs.

P-59
Prostatic damage by the cadmium toxicity and the protective role of metallothionein.

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The acute and chronic toxicity of cadmium(Cd) in the prostate was examined in the relation of metalloprotein metallothionein(MT)and various gene expression. Wistar strain male rats(6 weeks of age)were given cadmium chloride($CdCl_2$)of 20mgCd/kg orally for 6 day a week for 5-10 weeks. On the other hand, animals of the ip-group were injected intraperitoneally Cd of 2mgCd/kg, and in the ip-ip group, Cd of small dose(0.2mgCd/kg)was injected at before 24hr of the 2.0mgCd/kg Cd injection. The concentration of Cd and metallothionein and the gene expression of inflammation index were measured using AAS, Cd-Hem, and RT-PCR-method. A reduced tendency of prostate damage by the pretreatment of small dose of Cd was shown, and the participation of the Cd binding protein MT was thought from the above-mentioned results. Also, in the protection of the chronic toxicity, MTI gene expression increased dose-dependently of Cd accumulation.

P-60
Placental cadmium concentration as an indicator of maternal tobacco smoking

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Tobacco smoke is a mixture of toxic chemicals with reproductive endocrine-disrupting potential including cadmium.
We monitored tobacco smoke exposure in healthy subjects (mean age 28) after vaginal delivery at term in a clinical hospital. Based on self-ascertainment and maternal hair nicotine concentrations (measured in min. 12 cm long samples by GC-MS) 3 groups were identified: 32 smokers, 35 passive smokers and 19 nonsmokers. Cadmium concentrations were analyzed (by ICPMS) in maternal blood, umbilical cord blood and the placenta.
We found increases in hair nicotine of ca. 7 times and maternal blood cadmium of ca. 4 times in smokers *vs.* passive smokers and nonsmokers. Cord blood cadmium concentrations were equally low (< 0.1 ppb) in all of the groups. Placental cadmium was significantly higher in smokers than in passive smokers and nonsmokers.
In conclusion, apart from hair nicotine, placental cadmium could serve as an alternative marker of past tobacco smoking in postpartum smokers.

P-61
Assessment of dietary cadmium exposure in kindergarten children from Zadar County, Croatia

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Children are prone to the adverse effects of toxic metals, including cadmium, due to specific physiological needs during growth and development.
We evaluated the level of cadmium in daily meals offered to children (*N*=1000, age 4 to 7) who attended public nurseries in the coastal area, where they received 80 % of their daily dietary needs. The duplicate portion technique for sampling total food and beverage during 15 consecutive workdays over three seasons (during 2008) was used. Cadmium was analysed in representative samples by electrothermal atomic absorption spectrometry.
The estimated levels of ingested cadmium in the meals (expressed as average daily cadmium concentrations) in winter, spring and autumn were: 2.40±1.02, 1.53±1.36, and 0.57±0.38 µg/kg b. wt.
Based upon a comparison with the dietary exposure of an average child consumer to cadmium, which is between 2.56 and 3.46 µg/kg b. wt. a week (according to EFSA, 2011), we concluded that cadmium levels in the meals can be considered safe.

P-62
Trace element content of foods and estimated intake by poor and non poor households in Dhaka

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Eighty food samples and beverages commonly consumed by poor and non-poor households in Dhaka were analyzed for selected trace elements. The intake of Fe, Mn, Cu, Zn, Mo and Ni by adults in poor and non-poor households was estimated using the Household Income Expenditure Survey 2010. Cereals were the primary contributors of dietary intake of Fe (55.45%), Mn (76.34%), Cu (71.28%), Zn (60.0%), Mo (53.85%) and Ni (76.95%). Vegetables were in second position to supply dietary intake of Fe (17.55%), Mn (7.94%), Cu (9.14%), Zn (9.16%) and Ni (13.62%). Pulses provided with 9.93% Fe, 9.81% Mn, 9.77% Cu, 9.88% Zn and 1.76% Ni of the dietary intake of trace elements. Diets of males from poor household are estimated to be more deficient in trace elements (Fe, Cu, Zn, Co and Se) than non-poor household males. The intake of Mn and Mo was higher than the Recommended Dietary Intake for both poor and non-poor households.

P-63
Increment of all types of tumors in Kuwait after exposure to massive pollutions of combusting oil fields in 1991

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All kinds of tumors spread in Kuwait community after the massive exposure to pollutants (trace elements;(TEs), mono and poly aromatic hydrocarbons;(MAH; PAH)), expelled in the environment , as a result of incomplete combustion of oil fields by Iraq in 1991..At present the brain tumors occurrence is phenomenal , compared to the time before 1991.L. Hayat (1996) reported the deposition of Ni & V at higher levels in malignant brain tumors than benign. The malignant brain tumors manifested an increase in the ratio of trans/cis F.A relative to benign.. L. Hayat 1999 reported that Ni & V can replace the Fe of the hemoglobin , which might lead to overcoming the blood brain barrier .The research on Ku food basket, reported the presence of dangerous and carcinogenic TEs , MAH and PAH .in local plants, fruits, marine creatures , animals, avian and their by products . A study of rate of local food consumption resulted in determining L50 of pollutants From the above studies it was concluded that humans in Ku are exposed to levels of pollutants way above normal accepted limits., which explains the continuously ascending numbers of tumor cases per year starting 1991.till now.

P-64
Zinc biofortification of cassava through agronomic management in Brazil

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Zinc (Zn) deficiency is a well-known nutritional problem in agricultural soils and human populations, especially in developing countries. Biofortification is a strategy that aims to increase the content of selected micronutrients, including Zn, in staple foods. Cassava is a staple crop in many developing countries, feeding some 600 million people a day worldwide. The nutritional quality of the cassava root is not sufficient to meet all dietary needs. Thus, more studies on biofortification of this crop can lead to an improvement of its nutritional quality. We evaluated Zn contents in cassava accessions cultivated under different soil management in different areas from the central Brazilian savanna. The Zn content in cassava tubers ranged from 4 to 24 mg kg^{-1}, with higher values found in cassava accessions cultivated in an area receiving application of animal manure, which showed high soil Zn content (76 mg kg^{-1}). These results suggests that biofortification through agronomic management can lead to improved zinc contents in cassava.

P-65
Interference of Exogenous Bromide and Perchlorate Ions with Iodine and Thyroid Hormones Metabolism

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Presumed goitrogenic and thyrotoxic effects of excessive bromide and perchlorate ions were followed in adult male rats, as well as in lactating rat dams (and their breast-fed pups), maintained on diets with different iodine content, ensuring either sufficient iodine supply or mild to severe iodine deficiency.
In rats administered with various bromide and perchlorate doses we measured a consistent increase in relative weight of their thyroid glands with increasing time and concentration of applied bromide, and a sharp reduction of the 24-h uptake of [^{131}I]-iodide by their thyroids. In these animals, we also determined a steady decline in serum total thyroxine levels. Moreover, excessive bromide in the mothers substantially depressed the extent of [^{131}I]-iodine transfer from the dams through mother's milk to the suckling. As a consequence, a marked decline in the body weight increments of afflicted pups was observed. We have also confirmed our earlier observation that bromide toxicity is dependent on the state of iodine supply into the organism.

P-66
Granulated Barely β-Glucans Intake May Promote Excretion of Toxic Substances from Human Bodies

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The present study was performed to elucidate whether β-glucans intake promotes the excretion of toxic substances from human bodies and dynamics of plasma lipid levels.
Twenty-six of healthy volunteer women were subjected under the enough informed consent. Then, 3.6g/day of barely β-glucans granules (ADEKA Company, Tokyo, Japan) was administered for six months, and plasma various lipids levels were measured before and after intake of β-glucans. The trace elements (6 harmful and 20 essential minerals) in hair were also analyzed by the ICP-MS method.
Plasma total and HDL-cholesterol levels tended to decrease by the intake of β-glucans (227.8±51.7 to 212.8±47.0mg/dl, 73.6±19.6 to 69.1±15.6mg/dl, respectively). The trace elements in hair, arsenic (41.7±43.2 to 30.4±26.6ppb) and aluminum (6824.7±4154.5 to 2702.3±351.8ppb) also tended to decrease, however, there were no distinct changes in the other trace elements.
Conclusion: It was suggested that the intake of barely β-glucans granules improved the environment of human bodies through the decreasing of plasma lipids and the harmful trace elements.

P-67

Effect of Oral Exposure to Tungsten on Molybdenum Status and Serum Uric Acid Concentration in Rats

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It has been reported that animals exposed to tungsten shows low tissue molybdenum and decreased activities of xanthine oxidase and sulfite oxidase due to the competition of tungsten with molybdenum in vivo. In the present study, we examined a molybdenum status and serum uric acid concentration in rats exposed to tungsten orally.

Twenty-four male weaning Wistar rats were divided into 4 groups according to diets (AIN93G diet (control diet) or the control diet except for ammonium molybdate (low Mo diet)) and drinking water (deionized water or deionized water containing tungsten at a level of 200 ppm as sodium tungstate). Mo content in the control and low Mo diet were 0.2 and 0.05 ppm, respectively. Orally exposure to tungsten did not effect on body and tissue weights, as well as serum biochemistry (liver and kidney functions, lipid metabolism and iron status). Tissue molybdenum contents and hepatic xanthine oxidase activity were varied with molybdenum intake. The tungsten exposure dramatically decreased tissue molybdenum and hepatic xanthine oxidase activity; tungsten exposure brought low molybdenum status. Serum uric acid was also decreased by the tungsten exposure irrespective of molybdenum intake. These results indicate a possibility that sodium tungstate is used as a therapeutic medication for hyperuricemia.

P-68

Trace element status of population and demography in Russia: possible linkage

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During last decades there was a dramatic worsening of demographic situation in Russia. In frame of a national project evaluating influence of trace element status on population health we investigated the connection between hair elemental profile and demographic indices in European part of Russia. Totally 37578 adult inhabitants of 36 regions were observed; hair samples were collected and analyzed by ICP-MS for 20 chemical elements. The results were compared to regional demographic data. We found that life expectance in regions correlated negatively with lowered hair Co, excess Fe, Mn, Cr, Hg, and positively - with normal Co, Hg, lowered hair Fe, Mn and excess Se. Correlations of mortality rate were generally found to be inverted against life expectance. Birth rate correlated negatively with Fe, Cu, Cr excess. Most correlations were more pronounced in men vs women. Remarkable, that birth rate negatively correlated with normal Zn in men, and positively – with supernormal level of this element. The obtained data can suggest necessity of additional Se, Co supplementation in adults for enlargement of life expectance and Zn supplementation of men for birth rate increasing.