

Effects of Dietary Selenium Against Lead Toxicity Are Related to the Ion Profile in Chicken Muscle

Xi Jin¹ • Chun Peng Liu² • Xiao Hua Teng¹ • Jing Fu^{1,3}

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Abstract Complex antagonistic interactions between Selenium (Se) and heavy metals have been reported in previous studies. However, little is known regarding the effects of Se on lead (Pb)-induced toxicity and the ion profile in the muscles of chickens. In this present study, we fed chickens either Se or Pb or both Se and Pb supplement and later analyzed the concentrations of 26 ions in chicken muscle tissues. We determined that a Se- and Pb-containing diets significantly affected microelements in chicken muscle. Treatment with Se increased the content of Se but resulted in a reduced concentration of Cu, As, Cd, Sn, Hg, and Ba. Treatment with Pb increased concentrations of Ni while reducing those of B, V, Cr, Fe, Co, Cu, Zn, and Mo. Moreover, Se also reduced the concentration of Pb, Zn, Co, Fe, V, and Cr, which in contrast were induced by Pb. Additionally, we also found that synergistic and antagonistic interactions existed between Se and Pb

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 \boxtimes Xiao Hua Teng 364928357@qq.com

 \boxtimes Jing Fu jf699@cornell.edu

> Xi Jin Yaoyaojiu119@126.com Chun Peng Liu

liuchunpeng@neau.edu.cn

- ¹ College of Animal Science and Technology, Northeast Agricultural University, Harbin 150030, People's Republic of China
- ² Department of Veterinary Medicine, Northeast Agricultural University, Harbin 150030, People's Republic of China
- ³ Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14850, USA

supplementation. Our findings suggested that Se can exert a negative effect on Pb in chicken muscle tissues and may be related to changes in ion profiles.

Keywords Selenium . Lead . Chicken muscle . Ion profiles

Introduction

Lead (Pb) is a metal element that can be toxic to living organisms. Along with economic and technological advances, Pb pollution has become a growing concern. Agricultural products are largely exposed to lead, which can result in high Pb residues in livestock and vegetables. Excessive Pb levels in humans and animals could lead to problems in the reproductive system in males along with different types of cancer and intellectual damage affected children [[1](#page-6-0)–[3](#page-6-0)]. A previous study documented the suppressive effects of Pb on various enzymes, which can lead to the destruction of the antioxidant system and cause oxidative damage in muscles [\[4](#page-6-0)]. Digestion of Pb by animals can cause Pb accumulation in meat and milk [\[5](#page-6-0)] and eventually hurt consumers. However, the exact mechanism whereby Pb mediates toxicity and results in the accumulation of Pb in vivo remains unclear and warrants further study.

Selenium (Se) is an essential micronutrient for living organisms and is closely related to both human and animal health. Se plays an important role in anti-cancer activities through the induction of apoptosis [[6](#page-7-0)–[8\]](#page-7-0). Se deficiency reduces the expression of selenoproteins in chicken muscles [\[9](#page-7-0), [10](#page-7-0)] and can damage the liver by inducing oxidative stress [\[11,](#page-7-0) [12](#page-7-0)]. Recent studies have shown that Se shows a strong affinity for metals and, thus, can reduce the toxicity of heavy metals, such as Cd and Hg [[13](#page-7-0)–[15](#page-7-0)]. Se can alleviate the destruction of the cell membrane integrity induced by Cd in plants [[16](#page-7-0)] and reduce methyl mercury poisoning in marine mammals [[17\]](#page-7-0). Complex antagonistic interactions exist between Se and both other heavy metals and multi-ions under various physical and pathological conditions [\[18](#page-7-0)]. Tong indicated that the protective effects of Se against heavy metalinduced toxicity may be related to the ion profile in the chicken liver [\[19\]](#page-7-0).

Interactions among ion profiles in the tissues of some animals have been documented in recent studies, such as antagonistic interactions between Se and Zn in rats [[20\]](#page-7-0), and reduced retention of methyl mercury induced by dietary Se in freshwater fish [\[21\]](#page-7-0). However, the intramuscular ion profiles have not yet been investigated when Se and/or Pb was introduced as a dietary supplement. In this study, we established a model of Se and Pb interactions in chicken muscle tissues and measured the effects of Se and Pb supplementation on ion profiles, and we discuss the antagonistic function of Se against Pb in chicken muscle tissues. This present study reveals antagonistic effects of Se on Pb toxicity in chicken tissues.

Materials and Methods

Birds and Diets

All procedures used in this study were approved by the Institutional Animal Care and Use Committee of Northeast Agricultural University. A total of 360 male broiler chickens (1 day old; Weiwei Co. Ltd., Harbin, China) were randomly divided into four groups (90 chickens per group). Each group was separated into six pens with 15 chickens per pen. Throughout the entire experimental period, chickens were allowed ad libitum consumption of food and water. In this present study, we fed chickens a supplemental diet of Pb, Se, and $Pb + Se$ to control Pb-induced toxicity and the antagonistic functions of Se in chicken muscle tissues. The Se group was fed a Se-adequate (sodium selenite) diet that contained 1 mg/kg Se and 0.5 mg/kg Pb. The Pb group was fed a Pbsupplemented (Pb acetate) diet containing 0.2 mg/kg Se and 350 mg/kg Pb. The Se + Pb group was fed a Se and Pb compound diet containing 1 mg/kg Se and 350 mg/kg Pb. Finally, the control group was fed a basic diet containing 0.2 mg/kg Se and 0.5 mg/kg Pb. Chickens were euthanized when they were 90 days old. The muscle tissues were quickly removed and rinsed with ice-cold sterile deionized water, frozen immediately in liquid nitrogen, and stored at −80 °C until required for subsequent experiments.

Detection of Mineral Elements

We assessed the ion profiles [including those of calcium (Ca), vanadium (V), lithium (Li), boron (B), natrium (Na), magnesium (Mg), aluminum (AI), silicium (Si), kalium (K), chromium (Cr), manganese (Mn), ferrum (Fe), cobalt (Co), nickel (Ni), copper (Cu), zinc (Zn), arsenic (As), selenium (Se), molybdenum (Mo), cadmium (Cd), stannum (Sn), stibium (Sb), barium (Ba), hydrargyrum (Hg), thallium (Tl), and plumbum (Pb)] in chicken muscle tissues.

Mineral elements in chicken muscle tissues were examined by inductively coupled plasma mass spectrometry (ICP-MS; Thermo iCAPQ, USA). The parameters of the equipment used were as follows: frequency (MHz) 27.12, reflect power (KW) 1.55, sampling depth (mm) 5.0, torch-H (mm) 0.01, torch-V (mm) −0.39, carrier gas (L/min) 1.05, nebulizer pump (rpm) 40, S/C temperature (°C) 2.7, oxide ions (156/140) <2.0 %, doubly charged $(70/140)$ <3.0 %, and nebulizer-type concentric.

Mineral element concentrations were determined in the acid digest of samples according to the method of Uluozlu et al. [\[22](#page-7-0)]. Then, 1 g of each sample was digested with 2 mL H_2O_2 (30 %) and 5 mL HNO₃ (65 %) and then diluted to a final volume of 10 mL with deionized water. All sample solutions were clear. Samples were digested in a microwave system applied at 3 min for 1800 W at 100 \degree C, 10 min for 1800 W at 150 °C, and 45 min for 1800 W at 180 °C. A blank digest was conducted in the same manner. All digested samples were filled with ultrapure water to the specified final volume before analysis by ICP-MS.

Statistical Analysis

Data were analyzed using SPSS statistical software for Windows (version 13; SPSS Inc., Chicago, IL, USA). All data were checked for normal distribution and equal variance. Differences between the differently treated groups were assessed by one-way ANOVA. All data were plotted as means ± standard deviation. Differences were considered to be significant at $P < 0.05$. Additionally, the most important parameters were defined by principal component analysis (PCA) that could be used as key factors for individual variations using the program Statistic 6.0.

Results

Effects of Se, Pb, and $Se + Pb$ on Levels of Macroelements in Chicken Muscle Tissues

In this present study, we measured the concentrations of Na, Mg, K, and Ca after treatment with Se and Pb. We found that the macroelement with the highest concentration was Ca, followed by Mg, K, and Na (Table [1\)](#page-2-0). Among these elements (Fig. [1](#page-3-0)), compared with the control group, the Se-treated, Pbtreated and Se + Pb-treated groups did not show any changes in the Na, K, or Mg concentrations ($p > 0.05$). Additionally, Ca concentrations were slightly affected in the Pb-treated

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Table 1 Ion content in chicken muscle

 $\frac{b}{\gamma}$ of control was the relative ion content to control

group ($p < 0.05$). Generally, our data indicated that macroelements were not influenced by treatment with Se or Pb.

Effects of Se, Pb, and Se + Pb Treatment on Essential Microelement Concentrations in Chicken Muscle Tissue

In this present study, we measured a number of the essential microelements, including B, Si, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, Se, and Mo. We found that the five ions present at the highest concentrations were Si, Fe, Zn, Cu, and Cr (Table [1\)](#page-2-0). Among these top five ions, Fe, Zn, Cu, and Cr were significantly increased or decreased after the treatments ($p < 0.05$); however, the content of Si was not markedly changed compared with the control group. In the Se-treated group (Fig. 2), compared with the control group, the concentration of Se increased, while that of Ba and Cu decreased ($p < 0.05$). In the Se + Pb group, compound treatment of Se and Pb resulted in reduced concentrations of V, Cr, Fe, Co, Cu, and Mo ($p < 0.05$). In the Pb-treated group, the concentration of B, V, Cr, Fe, Co, Cu, Zn, and Mo decreased, while that of Ni increased ($p < 0.05$). Nevertheless, other essential microelemental ions were not affected by treatment with either Se, Pb, or $Se + Pb$ in chicken muscle tissues ($p > 0.05$).

In this present study, we also detected ten other toxic microelements, including Li, Al, As, Cd, Sn, Hg, Tl, Sb Ba, and

Fig. 2 The ion profiles of essential microelements in chicken muscle. Bars without a shared common letter are significantly different ($p < 0.05$). Data are means \pm SDs, $n = 6$

ings (Table [1\)](#page-2-0) revealed that Al and Pb were present at a higher level compared with any of the other elements. Se treatment alone significantly reduced the content of As, Cd, Sn, Hg, and Ba ($p < 0.05$; Fig. [3](#page-4-0)). Moreover, compared with the control group, Pb was decreased by 125 ppb (approximately 20 % of the control group concentration) and the content of Pb in the $Se + Pb$ group decreased by 2497 ppb compared with the Pb group (approximately 50 % of the Pb group). Compared with the concentrations in the control group, the content of Al, Tl, and Pb increased in the Pb-treated group, whereas that of As and Cd decreased ($p < 0.05$). In the Se + Pb group, As, Cd, Sn, and Ba levels decreased; however, levels of Tl and Pb increased compared with those in the control group ($p < 0.05$).

Pb, in addition to the essential microelement ions. Our find-

Principal Component Analysis

Using PCA, all parameters could be distinguished on ordination plots that corresponded to the first and second principal components (58.696 and 30.90 %, respectively; Fig. [4\)](#page-4-0). A correlation between the different ions was confirmed and quantified using Spearman's test (Table [2\)](#page-5-0), indicating that there were both positive and negative correlations between different ions.

Our data indicated that Al and Zn had a high positive correlation with Pb, whereas Mg, Si, Ni, Co, Cu, Cr, Mn, and Fe

Essential microelement

Fig. 3 The ion profiles of toxic microelements in chicken muscle. Bars without a shared common letter are significantly different $(p < 0.05)$. Data are means \pm SDs, $n = 6$

showed a high negative correlation with Pb. Moreover, Se exhibited a high positive correlation with Mg and a high negative correlation with Na. Taken together, our findings showed that the association among ions in chicken muscle tissues was convoluted and warrants further investigation.

Discussion

Previous studies have shown that Pb exerts its toxic effects by accumulating in the tissues of living organisms and entering the gastrointestinal tract of humans and animals. When Pb gains entry into organisms, it is to be toxic to the kidney and is mainly deposited in calcareous tissues, such as the bones [\[23\]](#page-7-0). Animals, such as fish, can absorb Pb through internal organs, after which Pb will eventually penetrate into and accumulate in muscle tissues [[24](#page-7-0)]. In this current study, we measured the content of Pb in chicken muscle tissues after exposure to Pb. The intramuscular content of Pb in the Pb group increased significantly compared with that of the control

Fig. 4 Ordination diagram of the principal component analysis (PCA) of parameters measured in chicken muscle

group. This finding is consistent with that of previous studies, indicating that Pb indeed accumulated in chicken muscle tissues.

A previous study revealed that a complex antagonistic interaction existed between Se and other heavy metals. Se can reduce the content of certain heavy metals, such as Cd and Hg [\[21](#page-7-0), [25\]](#page-7-0). The study of Li also showed that the antagonistic effect of selenite on lead-induced neurotoxicity could be observed in *Caenorhabditis elegans* [\[26\]](#page-7-0). In this present study, we determined that there was a significant difference in the content of Pb when Se was supplemented in the diet. This finding is consistent with a previous study that indicated that Pb accumulation could be alleviated by Se treatment. Therefore, we hypothesized that Se may have a negative effect on Pb and could alleviate Pb-induced toxicity in chicken muscle tissues.

Many previous studies have shown that ions play an important role in some biological processes, such as signal transduction [\[27](#page-7-0)] and structural integrity [\[28](#page-7-0)]. Ions profiles are also related to various pathological processes. Se exerts antagonistic effect on Cd-induced oxidative stress in humans [\[29\]](#page-7-0) and reduces the concentrations of Hg in freshwater fish [\[21](#page-7-0)]. Additionally, Se can potentially mitigate As-induced toxicity as a consequence of its antioxidant and antagonistic properties [\[30](#page-7-0)]. A previous study indicated that the addition of Se often showed antagonistic effects on certain essential elements, such as P, Ca, Mg, K, P, Fe, Cu, and Zn under some conditions [[19,](#page-7-0) [31\]](#page-7-0). In this present study, aside from the content of Se and Pb, we also examined the profiles of 24 other ions in chicken muscle tissues after exposure to Se, Pb, and both Se and Pb. Our study showed that Se and Pb generally had no effect on macroelements in chicken muscle tissues, whereas they significantly influenced microelement concentrations. Our findings are consistent with those of previous studies of the antagonistic effects of Se on heavy metals and of the ion profiles that are involved in the antagonistic function of Se. Thus, we deduced that the toxicity of Pb may be related to the altered profiles of essential microelements, including reduced concentrations of B, V, Cr, Fe, Co, Cu, Zn, and Mo, and increased

Table 2

levels of Ni. Moreover, the antagonistic function of Se against Pb may be related to the increased levels of V, Cr, Fe, Co, and Zn and the decreased amounts of Ni. Additionally, the toxic metals Al and Tl may also play an important role in Pbdependent toxicity, while the reduced amounts of Hg, Sn, and Ba may be related to the antagonistic function of Se. However, many other synergetic interactions are present between different ions in living organisms. In this present study, Pearson 's correlation coefficient showed both negative and positive correlations among different ions. We cannot exclude that interactions exist among many other ions that we did not detect, which may play an important role in the regulation of biological processes. Thus, following treatment with Se and Pb, the changes in the ion profiles were sufficiently complex that it was difficult to determine which ions played a primary or secondary role.

In summary, we demonstrated that Pb accumulates in chicken muscle and Se can alleviate Pb-dependent toxicity. Moreover, changes in the ion profile are indeed associated with the antagonistic function of Se that protects against Pbinduced toxicity. Additionally, complex interactions that we observed indicated that both synergistic and antagonistic effects persisted among these ions. The antagonistic function of Se that protects against Pb-induced toxicity may be related to changes in the profiles of these ions in chicken muscle tissues.

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Compliance with ethical standards All procedures used in this study were approved by the Institutional Animal Care and Use Committee of Northeast Agricultural University.

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