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Residual Concentration of Lead, Iron and Calcium in Chicken Meat Paste: Indicative Parameters for Identifying Deboning Procedure and Health Risk Assessment

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KEYWORDS

Calcium;
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ABSTRACT: Deboning method has a major effect on quality and safety of poultry carcasses or animal bones.The aim of this study was to determine residual concentration of lead, iron and calcium in chicken meat paste as indicative parameters for identifying deboning procedure. The test samples were eight whole chicken carcasses collected randomly in a three-month period from main retail markets in Tehran. All of the collected samples divided into the two groups: deboning of one group well done by hand and the other was performed mechanically by machine. Sample preparation and analyses were performed by wet digestion and atomic absorption analysis, respectively. According to the results the mean concentration of lead, iron and calcium in manually deboned samples were 2.94 (±0.56), 4.21 (±0.31) and 65.77 (±2.11) mg Kg^{-1} and in mechanically deboned samples were 3.79 (±0.52), 23.05 (±3.47) and 275.83 (±16.31) mg Kg^{-1}, respectively. In all cases, concentration of investigated elements in mechanically deboned treatments were significantly higher than manual ones ($P<0.05$) and obviously indicated the difference in deboning procedures. More ever the results of calculating estimated daily intake for lead due to consumption of chicken meat paste showed that in both of deboning methods, the concentration of lead was more than that recommended by provisional tolerable daily intake.

INTRODUCTION

Due to the desirable flavor of poultry products and as a result of the increased demand for replacing red meat with white meat due to their salutary effect and lower costs, recently the use of mechanically deboned poultry meat (MDPM) has increased in industrialized countries [1]. The similar demands for chicken parts with lower commercial value such as back and neck in less developed countries were reported [2,3]. Mechanically

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recovered meat (MRM), mechanically separated meat (MSM) and mechanically deboned meat (MDM) are common synonyms used to name the product, obtained by applying pressure to the poultry carcasses or animal bones for faster producing final meat paste with higher production yield than chicken pasted that was produced from hand-deboned meat (HDM) [4]. These machines separate the muscular tissue from the bones by broking and pulverizing forcing them to pass through a strainer. The final paste is a mix of soft tissues having specific nutritional value and relatively different chemical properties. It contains also marrow calcium and high levels of hem pigments that have high amounts of iron [2]. Bone separation for the first time was performed in Japan in early 1940s for fish and subsequently it was done for poultry in the U.S.A. [5]. The MDM contains many more lipids than HDM which was produced from the same animal. This excess lipid comes from bone marrow and bone tissue [4]. Due to this difference in chemical composition, some problems such as rapid oxidation that lead to off-odors and off-flavors may happen in MDM [1]. Also over the time of deboning, parts of the meat may be exposed to other pollutions with machines and production site that are important factors in occurring food-borne diseases [6]. In general, the wish of consumer for improving the quantity and quality of products must be fulfilled for success in the poultry industry [7]. However, many functional properties such as water retention, ability to emulsify and jelly consistency belong to MDPM [8]. In several studies some properties such as fat content (18-26%), protein contents (14-15%), moisture content (60-65%), collagen contents (about 3%), total saturated fatty acids (26.2%), total unsaturated fatty acids (73.8%), calcium (0.06 – 0.28%), ash content (about 1.2%) and pH ranges (6.8-7.4) were pointed out for mechanically separated meats [5]. This demonstrates that meat paste is a main source of trace elements and the best source for iron with the highest bioavailability. Iron and calcium due to their important role in biological systems are essential elements. However essential elements can also impair toxic effects at high concentrations [9-11]. Due to growing effect of food anthropogenic pollutants such as heavy metals from man-made sources, many researchers have discussed about it [12-18]. Knowing broiler chickens are assailable to lead inebriation so that only 1.0 ppm of lead in the chicken diet can cause significant growth prevention. The intake of lead can aggregate it in the soft tissues, eggs and even chicken bones [19] and it must be considered that mechanically deboned meat can result in entry of bone marrow and consequently iron and calcium into the final chicken paste product [20]. The risk of excessive amounts of lead in foodstuffs such as reducing mental performance in children and hypertension in adults was reported before [14]. In a previous work published by members of this group, the accumulation of lead and cadmium in Kiwifruit due to contamination of soil and irrigation water was evaluated [21]. Recently, we also reported the contamination of different honey brands which were produced in an industrial zone [22]. The elemental contamination of Macaroni and Pasta during production technology [23] and effect of production process on contamination of sunflower oil with some heavy metals [24] which were also reported before revealed that production technique has a major role in accumulation of concerning elements in the final product. Due to the safety importance of marine food stuffs that provide the required protein for body, some concerning elements were evaluated in *Penaeus Semisulcatus* after its fresh fishing [25] and after samples distributed in retail markets and collected in a post-market surveillance program [26]. The purpose of this study was evaluating the residual concentration of lead as a concerning element as well as iron and calcium as nutrient elements in mechanically and hand-deboned chicken meat paste and identifying the indicative role of them in differentiating between deboning procedures. The related health risks associated with their consumption were also fully evaluated.

MATERIALS AND METHODS

Chemicals and reagents

All standards and samples were prepared in double-distilled deionized water; using analytical grade reagents. Deionized water with 18 MΩ.Cm, prepared by Milli-Q Water System (Millipore, Le montsur-Lausanne, Switzerland). Standard stock solutions of lead, iron and calcium ions at the concentration of 1000 mg mL^{-1} (Merck, Darmstadt, Germany) were used to prepare working solutions after appropriate serial dilutions. The glassware used for standards and sample preparations were soaked in detergent solution, acid-washed in concentrated hydrochloric acid and rinsed with deionized water before subsequent analysis.Nitric acid and perchloric acid were purchased from Merck, Darmstadt, Germany. Wherever needed, the pH was adjusted by a digitalpH meter (Metrohm, model 744) equipped with a combinedglass–calomel electrode. The accuracy of the analytical method was evaluated by the use of certified reference material (FAPAS, Canned Fish, Metallic Contaminants, Test material 07225).

Collection and handling of samples

To prepare the required HDM and MDM samples, a total of four duplicate whole chicken carcasses were collected randomly from several main retail markets, in Tehran, Iran. Sampling was conducted in a three months period. The selected samples were from the most abundant and highly consumed chicken carcasses which were distributed in this area. All of the samples were packed individually in plastic bags, immediately placed on ice, transported to the laboratory where they were stored at -18 °C until processing. All of the collected samples divided into the two groups: deboning of one group performed on skinned chicken carcasses using sharp knife (HDM samples) and deboning of other skinned chicken carcasses well done by machine (MDM samples). Four samples obtained from each treatment were mixed together to prepare the fifth homogenized sample. The later samples was fully homogenized and labeled as representative sample from each group. Then all the five samples from each treatment were divided into the three parts and analyzed individually as triplicate runs.

Sample preparation and analysis

Exactly 100.0 g of each sample was dried in an oven at 100 °C. The dried samples were crushed with a pestle porcelain and mortar and kept in acid washed nylon bags in desiccators. Exactly, 2.0 g of each dried sample was added to 10 mL of the digestion mixture (65% HNO_3 and 70% $HClO_4$, 3:2 v/v). Digesting of samples was done by convection current heating for 3 h in a water bath at 70ºC and then were cooled and carried out into clean flask and 20 mL de-ionized water was added to it. This destroys the organic matter in the sample and releases metal ions into the solution. The solutions were transferred into acid-leached polyethylene bottles and then centrifuged and the supernatant was analyzed. In the treated sample, residual concentrations of iron and calcium with nutritive value and lead as major concerning element were individually determined by atomic absorption spectrometry (Shimadzu, model AA-6200, Japan) using an air-acetylene with flame temperature of 2800°C, acetylene pressure of 0.9–1.0 bar, air pressure of 4.5–5.0 bar, reading time of 1–10 sec, flow time of 3-4 sec and using the wavelength of 217.0, 324.8 and 228.8 nm for detection of lead, iron and calcium, respectively. All standards and samples were analyzed three times. In calibration curve, at least five standard solutions for each metal were considered. Blank sample was also prepared by taking 10 mL of the reagents mixture through the same procedure [21].

Risk Assessment

To estimate the potential risk of lead due to consumption of chicken meat paste and study its risk due to releasing from bone marrow and come into the meat paste during mechanically deboning, the value of Provisional Tolerable Daily Intake (PTDI) was considered for risk assessment studies. It was used by joint FAO/WHO expert committee on food additives (JECFA) that divided by 7 to evaluate PTDI and estimate the contaminants that may accumulate in the body. The PTDI value was calculated by the (1.1) equation in which Cons. is the average consumption of chicken in Iran (that its average was recorded as 32 g per day), C is the heavy metal concentration in chicken paste and B_W is the mean body weight of an Iranian adult person (that was considered as 60 kg). The final results were calculated from 1.1 equation and compared with that reported by WHO/FAO (3.6 µg Kg^{-1} B_w) [27-28].

$$PTDI = C \times Cons. / B_W \quad (1.1)$$

Statistical analysis

All of the calculated data were expressed as mean± standard deviation and analyzed using one-way analysis of variance (ANOVA) followed by Duncan's test as a post-hoc test to determine whether Pb, Fe and Ca concentrations varied significantly between MDM and HDM, with values less than 0.05 ($p<0.05$) considered statistically significant. This was done using statistical package for the social sciences (SPSS) program Ver. 22.0 software (SPSS Inc., Chicago, IL, USA) and the charts was created by Microsoft Excel 2013 software.

RESULTS AND DISCUSSION

The minimum, maximum and average concentration of lead, iron and calcium in HDM and MDM chicken paste and their standard deviations for analyzing three replicate treated samples were depicted in Table 1. Statistical grouping of the concentrations for each element in either HDM or MDM groups by ANOVA test revealed that mean concentration of all investigated elements was higher in MDM chicken paste than HDM ones, significantly ($p<0.05$). This statistically difference was depicted by assigning (a) and (b) letters to the mean concentration values for each element (Table 1).

The obtained results for each duplicate prepared samples that categorized in HDM and MDM groups were depicted in Figure 1 for lead, iron and calcium, individually. The results for the fully homogenized sample was also included in this Figure as the fifth sample. The standard deviations for these results were also depicted as the error bars for each triplicate runs and any significant difference between HDM and MDM groups was depicted in table 1.

Determination of iron in various foodstuffs is a common test in food quality control laboratories and can be used for differentiating chicken meat paste produced in various techniques. Iron in low concentration is an essential element for human health and its insufficiency causes anemia disease. For overdose cases, the upper tolerable intake level of iron in male/female (14-70 years) and children (from birth onwards up to 8 years) was considered as 45 and 40 mg day^{-1}, respectively [29]. Excessive mechanical force and extraction of hem and red marrows from the bone, causes higher content of iron in mechanical deboned meat [29-30] and this can be a potential risk for iron overdoses especially if it accompanied with other foodstuffs with high levels of iron. The mean and standard deviaion of iron content in HDM with 4.21±0.31 mg Kg^{-1} value showed significant difference with its related value in MDM that was increased up to 23.05±3.47 mg Kg^{-1} ($p<0.05$).

Calcium is another valuable element that is suitable for differentiating between MDM and HDM with two different production techniques. Bone ash in poultry contains about 37 percent of calcium [31] varies by age of animal, presence or absence of tissues such as marrow or cartilage that are associated with bone, and state of bone hydrationand so its determination in chicken paste can be useful criteria for entry the bone into the final product. During mechanical deboning, some bone marrow come in to the chicken paste and so calcium content in MDM treated samples showed greater concentration than HDM ones (Table 1). The mean calcium content with its standard deviation in HDM with 65.77±2.11 mg Kg^{-1} value showed a good difference with its corresponding value in MDM that was increased significantly up to 275.83±16.31 mg Kg^{-1} ($p<0.05$). Several studies have investigated this issue and the reported results deal with lower calcium content in HDM samples than MDM treated meats that are in agreement with our reported results for the processed chicken carcasses in this study [32-33]. The higher intake of calcium due to its treatment by MDM is not a considerable concern and some bone particles that comes into the chicken paste due to this special treatment can dissolved in high concentrated acid in stomach and there is no concern in this regard. The appropriate intake of calcium suggested by FAO/WHO is 400-500 mg day^{-1}, therefore the excessive residual of calcium due to applying mechanically separation technique can result in a nutritional advantage except for a few calcium hyper-absorbing cases [5]. Furthermore, the amount of calcium represents generally the bone content in MSMs. This is in compatible with some other report by Ang & Hamm (1982) for calcium contents for different cuts (chicken neck, with and without skin, and back) as (17-34 mg $100g^{-1}$ sample) and (53-91 mg/100g sample) for MDM and MSMs, respectively [34]. In another study the content of calcium in MSMs was higher than manually deboned hen (162.5 versus 16.75 mg/100g) [35].The mean concentration of calcium which was reported in this study (65.78 mg Kg^{-1} for HDM and 275.83 mg Kg^{-1} for MDM) in chicken paste has a good compatibility with other reported studies and can be used as a suitable indicator for identifying the type of chicken meat paste processing.

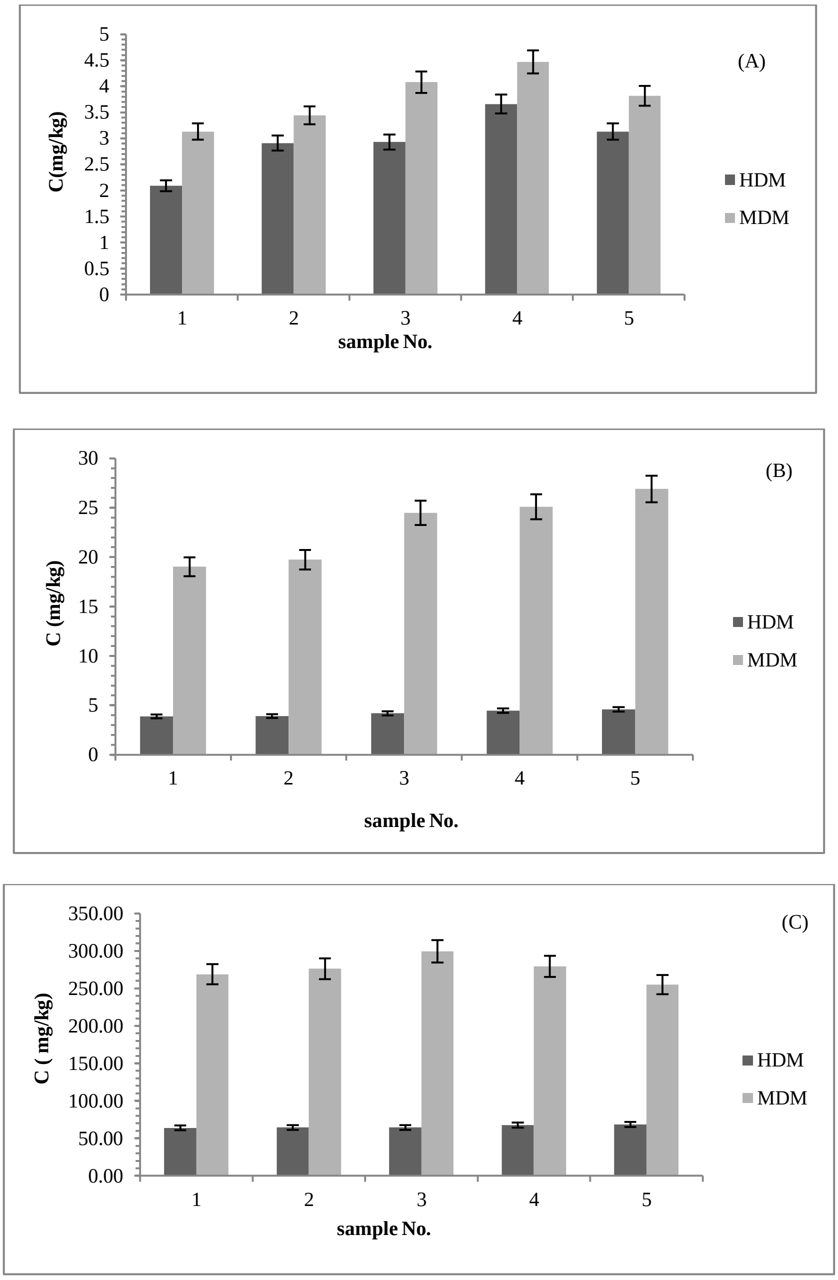

Figure 1. The concentration of investigated elements in poultry carcasses treated by HDM and MDM, A) Lead; B) Iron and C) Calcium

Control of specific toxic elements can be another suitable subject for estimating the processing that was made on chicken meat. The results of this research showed that the mean concentrations of lead in manual and mechanical deboned chicken paste samples were 2.94 and 3.79 mg Kg^{-1}, respectively. The obtained values showed a significant difference in both treatment techniques ($p<0.05$) and were higher than codex criteria as well as UK and Australia standards. The body intake of heavy metals in toleration is dependent on the frequency of their consumption, the amount of food consumed and the rate of detoxification of contaminant in the body. Actually, polluting chicken meat paste with toxic elements depends on the production techniques as well as how to feed the poultry. The average concentration of lead in the chicken meat samples were reported up to 0.088 mg Kg^{-1} in our previous report [36]. Generally, lead concentration in chicken meat paste depends on the food production methods and environmental conditions [37]. The obtained results showed that the mechanical deboning of poultry meat increased the lead content in final product. In some reports claimed that garlic feeding can be applied to protect consumers by decreasing lead concentrations in chicken meat because of chelating compounds of garlic and elimination of Pb [38-39]. However, due to its potential for bioaccumulation in chicken tissue and harmful effects on nervous system, according to EU Directive 466.2001, the permitted level for concentration of lead in meat that obtained from different animal sources was considered as 100 µg Kg^{-1} [40]. The Food and Agriculture Organization (FAO)/World Health Organization (WHO) (1993) has established a provisional tolerable weekly intake (PTWI) equivalent to 1500 µg lead/week for a person weighing 60 kg (25 µg/kg/day). This recommendation was maintained at the meeting of the Joint WHO/FAO Expert Committee of Food Additives in 1999 (WHO 2000) [40]. Considering the daily intake of chicken meat per day to be 32 g, the mean daily intake of lead from chicken meat was calculated and showed in Table 2. As it is clear, the obtained results were more than the PTWI values recommended by JECFA. Although the deboned chicken meat not used directly by all age groups but it may be formulated in sausages and other meat products. Since the children are more susceptible to the harmful effects of lead intake than adults and considering that they as well as adults tend to consume these products, it is clear that more strict criteria needs to be developed for control of high levels of lead in chicken paste due to applying inappropriate processing technique. So, primarily poultry feed and after that, the air pollution around maintenance area, their processing site (deboning and meat product factories) and the status of production equipments must be carefully checked. This can also directs the health regulatory laboratories for recognizing the safter chicken meat paste which was produced by hand deboing method and improves healthy consumption of it considerably.

Table 1. The mean concentration (mg Kg^{-1}) of investigated elements in mechanically and manually deboned meat paste.

Element	Concentration in Deboned Chicken Paste					
	HDM			MDM		
	Min	Max	(mean ± S.D.)	Min	Max	(mean ± S.D.)
Iron	3.88	4.59	4.21 (±0.31)a*	19.03	25.90	23.05 (±3.47)b
Calcium	63.90	68.40	65.77 (±2.11)a	254.95	299.6	275.83(±16.31)b
Lead	2.09	3.66	2.94 (±0.56)a	3.13	4.47	3.79 (±0.52)b

* Letters a and b show statistically significant difference ($p<0.05$)

Table 2. Provisional Tolerable Daily Intake (PTDI) for lead due to consumption of chicken paste.

Deboning Method	Mean Value (mg Kg^{-1})	Consumed chicken meat (g day^{-1})	Mean body weight (Bw) (Kg)	Daily intake due to consumption of chicken meat (µg.Kg^{-1} Bw.day^{-1})	PTDI (µg/Kg Bw/day) (JECFA)
MDM	3.79	32	60	2020	3.6
HDM	2.94	32	60	1560	3.6

CONCLUSIONS

Evaluating the presence of essential minerals and some heavy metals in chicken meat paste can be useful and informative for determining type of processing and its potential health and safety. In this study the deboning method and its effect on residual concentration of lead, Iron and calcium in the chicken meat paste were investigated for the first time. The results revealed that in all cases, concentration of investigated elements in mechanically deboned treatments was significantly higher than manual samples and this process can be a potential source for identifying the deboning procedure and the potential contamination of meat paste by lead. Calculation of estimated daily intake for lead specially for consumtion of chicken meat paste showed that in both processing methods, the concentration of lead was more than that recommended by provisional tolerable daily intake and any health promotion in this field would be helpful by better control of feed quality and using detoxification techniques.

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Conflict of interest

The authors declare that they have no conflict of interest.

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