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### Analysis of essential heavy metals in ready-to-eat chicken meat products of Chennai city

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#### Abstract

A study was carried out to analyse the essential heavy metals viz. cobalt, copper, iron, manganese, nickel and zinc in commercially available Ready-To-Eat chicken meat products (chicken 65, chilly chicken, grilled chicken and tandoori chicken) collected from street food outlets at different regions of Chennai city. Totally 288 samples were collected and analysed using inductively coupled plasma-optical emission spectrometry (ICP-OES). Results revealed that, among the three regions southern region had higher concentration of cobalt  $(4.19\pm6.35 \text{ ppm})$ , copper  $(2.20\pm0.31 \text{ ppm})$ , manganese  $(2.34\pm0.37 \text{ ppm})$  and nickel  $(8.09\pm2.42 \text{ ppm})$  whereas higher concentrations of iron  $(12.86\pm2.02 \text{ ppm})$  and zinc  $(10.42\pm1.56 \text{ ppm})$  were noticed in central region. Among the products grilled chicken samples had higher concentrations of zinc  $(10.42\pm1.56 \text{ ppm})$  and nickel  $(8.09\pm2.42 \text{ ppm})$ . Based on the results it was concluded that, heavy metals like copper, iron and zinc in ready-to-eat chicken meat products were within the limits of standard whereas cobalt, manganese, and nickel were slightly higher than the permissible limits set by different regulatory agencies for meat. The reason for these higher values may be due to the ingredients, cooking methods and cooking utensils used for the preparation of the chicken meat products and also the extraneous contaminants.

Keywords: Street food outlets, chicken meat products, essential heavy metals, ICP-OES

#### 1. Introduction

The food chain is contaminated with heavy metals which results in food safety issues and human health risk. These heavy metals are transferred into the environment through anthropogenic activities such as mining, industrial processing, waste water irrigation, agricultural activities, transport and fuel combustion, iron and steel production, coal and oil combustion, waste incineration, non-ferrous manufacturing and cement kilns (Dietz *et al.* 1998) <sup>[1]</sup>. The constituents like air, water, soil, and food are vital to the human lives which directly influence the quality of human life and risk of contamination with various pollutants in this industrialized area. The practices like production, manufacture, processing, preparation, treatment, packaging, transport, or holding of such food or as a result of environmental contamination the contaminants are present in such food (Codex Alimentarius, 1995) <sup>[2]</sup>. Therefore, all food products are at risk of contamination from several resources, and there are no exemptions for chicken meat and products.

The development of a primary production and processing standard for poultry meat uses an approach that investigate the sources of potential risks, which may be introduced at different points through the primary production and processing chain. Chicken meat and chicken meat products supply chain is divided into four distinct steps viz. primary production, processing, retail, and consumer. At each of these steps, chicken meat and products probably directly or indirectly exposed to contamination (Reyes-Herrera, 2012)<sup>[3]</sup>. Direct exposure of contamination means a compound is present in raw meat, whereas in indirect exposure, contaminants cross into meat during processing, storage, packaging, or preparation like procedures. Indirect contaminants also include substances that become toxic and harmful to people due to food-processing practices. Indirect pollution is most frequently the result of unawareness, lack of education of food handlers or inappropriate handling applications (Botsoglou and Fletouris, 2001)<sup>[4]</sup>.

Many heavy metals are essential for plants, animals and humans when present in the low concentrations and they become toxic only when a concentration limit is exceeded in which case the term 'heavy metals' rather than 'micronutrients' is used (Rengel, 1999)<sup>[5]</sup>. Essential

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heavy metals are naturally present in variety of foods and also in the environmental components like soil, air, water etc. which are entering into the food chain through some anthropogenic activities. The major essential heavy metals viz. Copper, Cobalt, Iron, Manganese, Nickel and Zinc which are beneficial to the human body and also essential to maintain the metabolism of human body.

Presently, the society is sparing little attention and time towards food and health due to the technology advancement and busy chores at home as well as at work place. There is a rapid development and attraction towards the street foods, fast foods and foods from restaurants due to their spicy and tasty preparation in a short time. Most of the urban people living a mechanical life and they don't find time to prepare food regularly due to their work nature. Mainly for time saving and to reduce the work load they prefer to eat outside. The outside foods are mainly consumed from street foods, roadside food outlets, fast food restaurants etc. which have more chances to be contaminated with extraneous contaminants like heavy metals. In Chennai, commonly consumed ready- to-eat foods are chicken meat products which are available at any time in street shops as well as most of the people are like to have these products. Hence, this study has been designed to screen the essential heavy metals levels in the ready-to-eat chicken meat products in street food outlets of Chennai city.

#### 2. Materials and Methods

The study was conducted to analyse the essential and nonessential heavy metals in Ready-To-Eat chicken meat products samples viz. chicken 65 (72), chilly chicken (72), grilled chicken (72) and tandoori chicken (72). A total of 288 Ready-To-Eat chicken meat products were collected from street food outlets of northern, central and southern regions of Chennai as mentioned by Greater Chennai Corporation. The samples each weighing approximately 200g were carefully collected and homogenized thoroughly using blender. The homogenized samples were then packed in polyethylene ziplock bags and stored at the temperature of -18°C in the laboratory until further use as per the procedure of Hoha *et al.* (2014) <sup>[6]</sup>.

#### 2.1 Preparation of glassware and plastic ware

Glassware used for the study were initially soaked in detergent solution and thoroughly cleaned using tap water. Subsequently, they were rinsed and soaked in 5 per cent nitric acid solution overnight. Then, the glassware soaked in the nitric acid were rinsed with single glass distilled water and soaked again overnight. The next day it was rinsed with Millipore water and allowed to remain soaked overnight. Finally, they were drained and dried in hot air oven at 70°C for 8-10 hours.

#### 2.2 Wet digestion method

The homogenized frozen samples were thawed overnight in a refrigerator at  $4\pm1^{\circ}$ C before starting the digestion process. The samples were digested by wet digestion method for the analysis of various heavy metals by following the procedure of Jankeaw *et al.* (2015)<sup>[7]</sup>. One gram of sample was taken in a 25ml glass screw cap test tube. Five ml of 69% concentrated nitric acid (Sigma aldrich) was added to the sample and kept in water bath at 80°C for 15 minutes subsequently the test tubes were cooled to room temperature and dried in hot air oven at 135°C for 3 minutes. The samples were cooled to room temperature and 1 ml of 30% hydrogen peroxide

solution was added to the contents and filtered through Whatman filter paper No.1 and followed by No.42 filter paper. The extracted solution was quantitatively transferred into a 50 ml volumetric flask and the volume was made upto the mark with millipore water and immediately used for heavy metal analysis.

#### 3. Analysis of heavy metals

Samples were analysed for the estimation of heavy metals using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES) (The Agilent Technologies 720 series).

Instrument set up					
Parameters	Ignite	Run			
Plasma (L/min)	1.5	15			
Auxillary (L/min)	1.5	1.5			
Mass Flow Control (L/min)	0.000	0.900			
Power (kw)	2.00	1.20			
Pump (RPM)	50	7			
Time (s)	5	3			
Nebulizer flow (L/min)	0	0.75			
Instrument stabilization delay (s)	15				
Replicates	3				
Replicate Read time (s)	1				
Sample uptake delay (s)	30				
Pump rate (RPM)	15				
Rinse time (s)	10				

#### 4. Estimation of heavy metals

Calibration was done with multi-element calibration standard for the heavy metals viz. cobalt, copper, iron, manganese, nickel and zinc in aqueous medium linear range of 0 ppb to 1 ppm.

Table 2: Wavelength used for quantification

S. No.	Elements	Wavelength (nm)
1	Cobalt (Co)	228.615
2	Copper (Cu)	324.754
3	Iron (Fe)	259.940
4	Manganese (Mn)	257.610
5	Nickel (Ni)	221.648
6	Zinc (Zn)	206.200

#### 5. Quantitative determination of heavy metals

Multi-element including cobalt, copper, iron, manganese, nickel, and zinc were recorded directly from the analysis scale of ICP and were calculated by the following equation according to ASTM, (2002)<sup>[8]</sup>.

Element in  $mg/kg = R \times D/W$ 

R= Reading of elemental concentration (mg/kg) from the digital scale of ICP system

D = Dilution of prepared sample

W = Weight of the sample

The results obtained in this study (ppb ( $\mu g/kg$ )) were converted into ppm (mg/kg) for the calculation.

**Statistical analysis:** The raw data were analysed by using one way ANOVA for statistical analysis with statistical package SPSS 14.0 version (SPCC Inc., Chicago, USA 2005).

#### 6. Results and Discussion

The results of essential heavy metals in the Ready-To-Eat

chicken meat products of street food outlets at different regions of Chennai city, Tamil Nadu were as follows:

**Essential heavy metals:** The results of essential heavy metals viz. cobalt, copper, iron, manganese, nickel and zinc are presented in Table No.3.

Table 3: Mean ± SE values of essential heavy metals (ppm) in chicken meat products collected from the street food outlets of di	ifferent
Regions of Chennai	

Elements (ppm)	Products	Northern Region	Central Region	Southern Region	F Value
Cobalt (Co)	Chicken 65	0.97±0.94 <sup>a</sup>	1.30±1.06 <sup>a</sup>	4.18±6.52 <sup>b</sup>	5.04**
	Chilly chicken	0.94±0.74 <sup>a</sup>	1.14±1.21 <sup>a</sup>	4.19±6.35 <sup>b</sup>	5.65**
	Grilled chicken	1.00±0.89 <sup>a</sup>	1.21±1.19 a	2.22±2.41 <sup>b</sup>	3.85**
	Tandoori chicken	1.27±0.76 <sup>a</sup>	1.11±1.13 a	3.97±4.82 <sup>b</sup>	7.37**
Copper (Cu)	Chicken 65	1.40±0.09 <sup>a</sup>	1.61±0.10 <sup>ab</sup>	2.11±0.29 <sup>b</sup>	3.93*
	Chilly chicken	1.41±0.08 <sup>a</sup>	1.61±0.08 <sup>ab</sup>	2.15±0.34 a	3.33*
	Grilled chicken	1.43±0.09 <sup>a</sup>	1.44±0.08 <sup>a</sup>	2.10±0.29 <sup>b</sup>	4.42*
	Tandoori chicken	1.40±0.07 <sup>a</sup>	1.55±0.09 a	2.20±0.31 <sup>b</sup>	4.91**
	Chicken 65	8.88±1.41	7.45±0.99	6.29±1.62	0.91 <sup>NS</sup>
	Chilly chicken	10.71±1.47 <sup>a b</sup>	12.86±2.02 <sup>b</sup>	6.67±2.03 <sup>a</sup>	2.86*
IIOII (Fe)	Grilled chicken	10.19±1.34	8.10±1.17	6.93±1.94	1.19 <sup>NS</sup>
	Tandoori chicken	8.24±1.15	8.77±1.52	6.88±2.15	0.34 <sup>NS</sup>
Manganese (Mn)	Chicken 65	2.02±0.19	1.52±0.14	2.34±0.37	2.65 <sup>NS</sup>
	Chilly chicken	1.71±0.16	1.44±0.15	2.08±0.29	2.37 <sup>NS</sup>
	Grilled chicken	1.44±0.13	1.25±0.17	1.86±0.29	2.26 <sup>NS</sup>
	Tandoori chicken	1.57±0.10	1.31±0.17	2.01±0.29	3.13 <sup>NS</sup>
Nickel (Ni)	Chicken 65	0.83±0.47	1.83±0.97	2.85±1.38	0.99 <sup>NS</sup>
	Chilly chicken	0.79±0.33	2.85±1.29	5.73±2.07	3.06 <sup>NS</sup>
	Grilled chicken	0.99±0.49 a	2.51±1.14 a	8.09±2.42 <sup>b</sup>	5.68**
	Tandoori chicken	0.79±0.44 <sup>a</sup>	2.28±0.96 a	6.94±2.31 <sup>b</sup>	$4.79^{*}$
Zinc (Zn)	Chicken 65	6.65±0.66 <sup>a</sup>	9.42±0.95 <sup>b</sup>	6.16±1.09 <sup>a</sup>	3.65*
	Chilly chicken	3.42±0.52 <sup>a</sup>	6.62±1.23 <sup>b</sup>	4.15±0.73 <sup>a</sup>	3.65*
	Grilled chicken	6.41±0.77	10.42±1.56	8.89±1.47	2.36 <sup>NS</sup>
	Tandoori chicken	7.01±1.02	9.38±1.09 <sup>b</sup>	5.66±1.28	2.76 <sup>NS</sup>

Means bearing different superscripts in a row differs significantly (P < 0.01)

\*\* Highly significant \* Significant NS Non significant

**Cobalt:** Chilly chicken samples collected from southern region had the highest cobalt content  $(4.19\pm6.35 \text{ ppm})$  whereas the lowest cobalt content  $(0.94\pm0.74 \text{ ppm})$  was observed in chilly chicken collected from northern region. There was highly significant differences (*P*<0.01) observed

between the regions and all products (Fig 1). These results were in accordance with the study made by Chowdhury *et al.* (2011) <sup>[9]</sup> who also found higher cobalt contents of  $1.05\pm0.001$  mg/kg and  $2.03\pm0.001$  mg/kg in chicken samosa and chicken sandwich respectively.



Fig 1: Cobalt concentration (ppm) of Ready-To-Eat chicken meat products (Street food outlets) in different regions of Chennai city

Cobalt is considered to be an essential element necessary for the formation of vitamin  $B_{12}$ . The general population are exposed to cobalt primarily via food. On the other hand, trace amounts of cobalt are necessary for the normal activity of human body (Stanojkovic-Sebic *et al.* 2015) <sup>[10]</sup>. Cobalt is the least accumulated metal and the highest accumulation occurred in liver while muscle and integument were least influenced. According to the WHO, the maximum permissible limit of cobalt is 1.5 mg/kg (Valadez-Vega *et al.* 2011) <sup>[11]</sup>. Cobalt is not known to bio-magnify the food chain therefore the meat generally does not have high amounts of cobalt. This may be due to the environmental pollutants like vehicle exhaust, industrial activities, cobalt containing ores etc. Wherein the cobalt gets attached to small particles and settles down in the soil and water (ATSDR, 2004)<sup>[12]</sup>.

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**Copper:** Tandoori chicken collected from southern region had the highest content of copper  $(2.20\pm0.31)$  whereas the lowest content  $(1.40\pm0.07)$  was observed in Tandoori chicken collected from northern region. There was a significant difference (P<0.05) between the regions and the products viz. chicken 65, chilly chicken and grilled chicken. Whereas, a highly significant (P<0.01) difference was observed between the regions in Tandoori chicken (Fig 2).



Fig 2: Copper concentration (ppm) of Ready-To-Eat chicken meat products (Street food outlets) in different regions of Chennai city

Iwegbue *et al.* (2008) <sup>[13]</sup> in their study they found copper content ranging from 0.01-5.15 mg.kg-lin chicken meat samples collected from different locations of southern Nigeria. Chowdhury *et al.* (2011) <sup>[9]</sup> also found higher copper content in Chicken patties (16.90±0.002) and in Chicken roll (97.28±0.006). The copper levels found in the present study were much lower than the values prescribed by WHO, 1996 (10 ppm) <sup>[14]</sup>.

**Iron:** Chilly chicken collected from central region had the highest content of Iron  $(12.86\pm2.02)$  compared to other regions whereas the lowest content of Iron  $(6.29\pm1.62)$  was observed in chicken 65 collected from southern region. There was significant difference (*P*<0.05) between the regions of chilly chicken and no significant difference (*P*>0.05) was

observed between the regions in chicken 65, grilled chicken and tandoori chicken (Fig 3).

The values obtained in this study were lower than the results of Iwegbue *et al.* (2008) <sup>[13]</sup> who found the iron content ranging from 22.07–97.72 mg.kg-1 in chicken meat samples collected from different locations of southern Nigeria. Demirezen and Uruc (2006) <sup>[15]</sup> also found a very low iron content (156.4±14.7  $\mu$ g/g) in sausage.

**Manganese:** Chicken 65 collected from southern region had the highest content of Manganese  $(2.34\pm0.37)$  whereas the lowest content of Manganese  $(1.25\pm0.17)$  was observed in Grilled chicken collected from Central region. There was no significant difference (P>0.05) observed between regions and products (Fig 4).



Fig 3: Iron concentration (ppm) of Ready-To-Eat chicken meat products (Street food outlets) in different regions of Chennai city

Iwegbue *et al.* (2008) <sup>[13]</sup> found the highest Manganese content in fresh turkey meat  $(1.37\pm0.21 \text{ mg.kg-1})$ . These values were higher than the permissible limits (0.5 ppm) of FAO/WHO (2000) <sup>[16]</sup> for fresh meat. The reason for increased level of manganese was may be due to the

environmental contaminants or extraneous contaminants with manganese which also occurs naturally in most foods and used in variety of products as ingredient in crackers, dry cell batteries, fertilizers, paints, medical imaging agent and cosmetics (ATSDR, 2012)<sup>[17]</sup>.



Fig 4: Manganese concentration (ppm) of Ready-To-Eat chicken meat products (Street food outlets) in different regions of Chennai city

**Nickel:** Grilled chicken collected from southern region had the highest content of nickel  $(8.09\pm2.42)$  whereas the lowest content of nickel  $(0.79\pm0.33)$  was found in chilly chicken collected from northern region. There was a highly significant difference (*P*<0.01) observed between all the regions in grilled chicken and a significant difference (P < 0.05) was observed in chilly chicken and tandoori chicken between the regions. There was no significant difference (P > 0.05) observed between regions in chicken 65 (Fig 5).



Fig 5: Nickel concentration (ppm) of Ready-To-Eat chicken meat products (Street food outlets) in different regions of Chennai city

Iwegbue *et al.* (2008) <sup>[13]</sup> who also found the highest nickel content ranging from  $(1.20\pm0.16$  to  $9.02\pm0.12$  ppm) in chicken meat samples collected from southern Nigeria. These contents were much higher than the permissible limit of (0.5 ppm) WHO/FAO (2000) <sup>[16]</sup>. In India, there is no standards limit for nickel content in fresh meat as well as meat products (FSSAI, 2011) <sup>[18]</sup>. The reasons for the nickel contamination in food chain may be due to the environmental contaminants or extraneous pollutants. Nickel can be released in industrial waste water which ends up in soil or sediment where it strongly attaches to particles containing iron or manganese.

Under acidic conditions, nickel is more mobile in soil and might seep into groundwater (ATSDR, 2005)<sup>[19]</sup>.

**Zinc:** Grilled chicken collected from central region had the highest content of zinc (10.42±1.56) whereas the lowest zinc content (3.42±0.52) was found in chilly chicken collected from northern region. There was a significant difference (P<0.05) noticed between regions in chicken 65 and chilly chicken. No significant difference observed between regions in Grilled and Tandoori chicken (Fig 6).



Fig 6: Zinc concentration (ppm) of Ready-To-Eat chicken meat products (Street food outlets) in different regions of Chennai city

Similar study made by Djinovic-Stojanovic *et al.* (2017) <sup>[20]</sup> who also found zinc values in chicken meat products viz. pates ( $6.96\pm1.72$  ppm), canned meats ( $7.33\pm2.78$  ppm) and cooked sausages ( $11.\pm2.58$  ppm) and these values were in the range of USFDA reported data (20.10-20.50mg/kg) for different chicken products. The values of this study were within the permissible limits ANFZA, 2001 <sup>[21]</sup> (150 ppm).

#### 7. Conclusion

Based on the results it can be concluded that the presence of higher levels of some heavy metals in Ready-To-Eat chicken meat products collected from street food outlets of Chennai city mostly due to the extraneous contaminants only not from the chicken meat. Because the chickens are slaughtered around 37 days old that is not possible to accumulate the heavy metals in the meat especially the cumulative poisoning like arsenic, cadmium, mercury and lead. The levels exceeded in this study were not of health risk, as the products were not consumed daily as a whole diet to accumulate in the body. Hence, it can be concluded that the Ready-To-Eat chicken meat products sold in the street food outlets of Chennai can be consumed as a part of the diet in regular interval and also the contamination of heavy metals in meat products were mainly through some extraneous contaminants.

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