



## DETERMINATION OF TRACE ELEMENTS IN SOME BRANDS OF GREEN TEA USING WET DIGESTION, INFUSION AND DRY ASHING METHODS

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### ABSTRACT

The concentrations of trace elements (Cu, Zn, Fe, Mn and Ni) in some commercially available brands of green tea sold within Katsina metropolis coded (SGT, HGT, LGT, AGT and GGT) were determined using wet acid digestion, water infusion, and Dry Ashing methods by Atomic Absorption Spectroscopy (AAS). Among the trace elements analyzed Nickel and Manganese recorded the highest value ranged 1.0 – 3.5 mg kg<sup>-1</sup> and 1.0 – 5.5 mg kg<sup>-1</sup> in dry ashing and wet digestion respectively. Fortunately, almost all the elements analyzed were within the standard permissible limit of WHO. The content of trace elements was found highest in the Dry Ashing method and lowest values recorded in tea infusion. The contributory factors responsible for the differences found in tea samples could be from a varying concentration of metals in the soil, conditions of cultivation and preparations, and these vary between both countries of origin and for the type of tea. Further work is recommended on these brands to study other important parameters.

**Keywords:** Green Tea, infusion, wet digestion, dry ashing, trace elements, *Camellia sinensis*.

### INTRODUCTION

Tea, derived from the leaves of the *Camellia sinensis* plant, is the most commonly consumed beverage in the world and, depending on the degree of oxidation, can be classified into three main types: green (unfermented), oolong (partly fermented) and black (fermented) tea (Khokhar and Magnusdottir, 2002). Green tea's chemical composition varies with genetic strain, climatic condition, soil properties, plucking season, and leaf location, processing and storage. Tea contains mineral elements such as manganese, iron, magnesium, sodium, potassium, copper, titanium, zinc, aluminum, strontium, bromine, iodine, phosphorus and fluorine (Sabhapondit *et al.*, 2012).

Green tea and black tea are the two most popular types worldwide. The drying of the steaming leaves produces green tea and it is obtained after the fermentation stage (Zarabruk *et al.*, 2010). Tea has a medicinal benefit that is known. It helps to avoid and heal many illnesses due to its antibacterial and antioxidant properties. Many elements that are present in food at major, minor and trace levels are said to be vital to man's well-being. Nevertheless, their intake in excessive quantities can cause serious health problems (Kumar *et al.*, 2005).

The ingestion of such herbal teas was associated with a decrease in serum cholesterol, prevention of low-density lipoprotein oxidation, and a decrease in the risk of

degenerative diseases such as cardiovascular disease, diabetes and cancer. The ingestion of herbal teas may also have a positive and negative influence on human health (Narin *et al.*, 2004). There is very little protein, vitamins & carbohydrates in the tea infusion, but maybe a source of essential dietary metals and binding polyphenols (Fiedman, 2007). Green tea's polyphenol composition is similar to that of fresh tea leaves, but steaming or roasting activates enzymes involving the oxidation and hydrolysis of chemical components of the leaves at the initial stage of green tea production (Karak and Bhagat 2010).

However, numerous studies have shown that the presence of trace elements in tea can have both beneficial and detrimental effects on human health (Tanaka and Kouno, 2003). Tea is now considered a nutritious drink with bioactive molecules and high antioxidant levels (Fiedman, 2007). Previous research has shown that the presence of trace elements in tea can have a positive and negative influence on human health (Rober & Gustav, 2014). Trace elements such as Fe, Cu, Zn, Ni and Mn are considered essential metals but can cause toxic effects on humans if their concentrations are higher than their allowable limits (Fung *et al.*, 2002).

### MATERIALS AND METHODS

**Apparatus:** Atomic Absorption Spectrophotometer was used for the determination of the trace metals. The operating parameters were set as recommended by the manufacturer.

**Chemicals:** All the chemicals used during the experiments were of analytical grade.

**Sample Collection:** Five (5) different brands of green tea that are commercially consumed in Nigeria were obtained from local retail in Katsina metropolis, Katsina State Nigeria. The brands were coded as SGT, HGT, LGT, AGT, and GGT, respectively. The tea samples were coded to conceal the originality.

**Digestion:**

Accurately weighed 0.5g of each sample was transferred into a 100 cm<sup>3</sup> beaker. 10.0 cm<sup>3</sup> concentrated nitric acid was added, the beaker was covered with a watch glass and the mixture was boiled gently on a hot plate until digestion was complete in about 1 hour. A 10.0 cm<sup>3</sup> portion of perchloric acid was then added and gentle heating was continued for another 1 hour. A

Small amount of deionized water was added to prevent dryness due to evaporation, after which the digest was allowed to cool, filtered into a 100 cm<sup>3</sup> volumetric flask and transferred into a sample bottle.

**Infusion:**

Water from the well water was used for the infusion, a 250 cm<sup>3</sup> beaker was weighed and zero, a tea bag was also weighed and poured into the beaker with the addition of hot water (100cm<sup>3</sup>). The sample was kept for 10 minutes and filtered into a 100 cm<sup>3</sup> volumetric flask.

**Dry Ashing**

Accurately weighed 0.5 g of each sample was transferred into a silica crucible and kept in a muffle furnace for ashing at 450°C for three hours and then 5.0 cm<sup>3</sup> of 6.0 mol/dm<sup>3</sup> HCl was added to the crucible. Furthermore, the crucible containing the mixture was kept on a hot plate and digested to obtain a clean solution. The final residue was dissolved in 0.1 mol/dm<sup>3</sup> HNO<sub>3</sub> solution and made up to mark in 50 cm<sup>3</sup> volumetric flask.

RESULTS

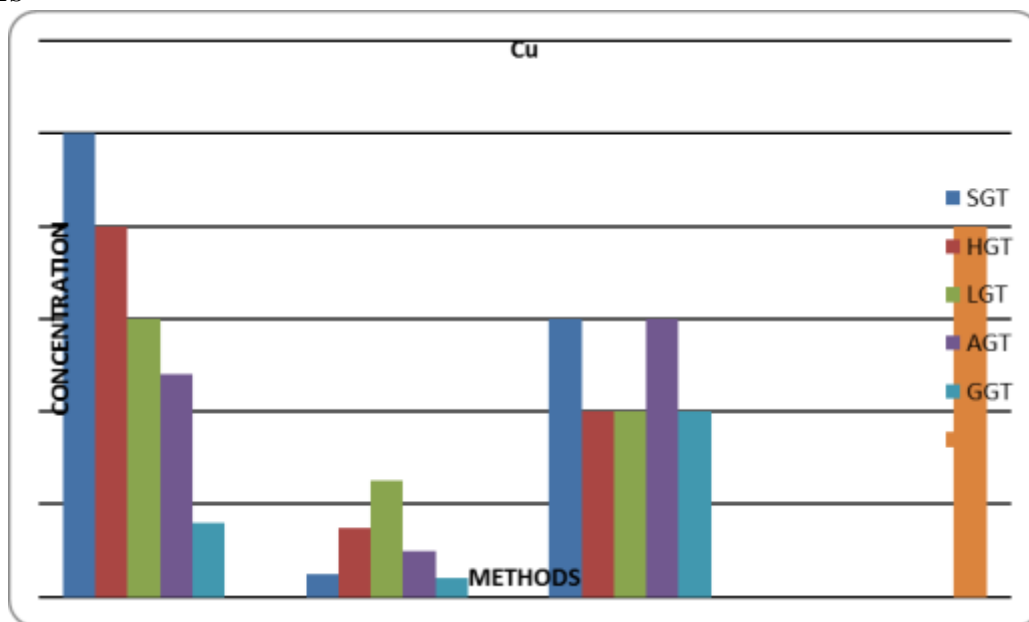


Figure 1. Copper Concentration of Green Tea Samples

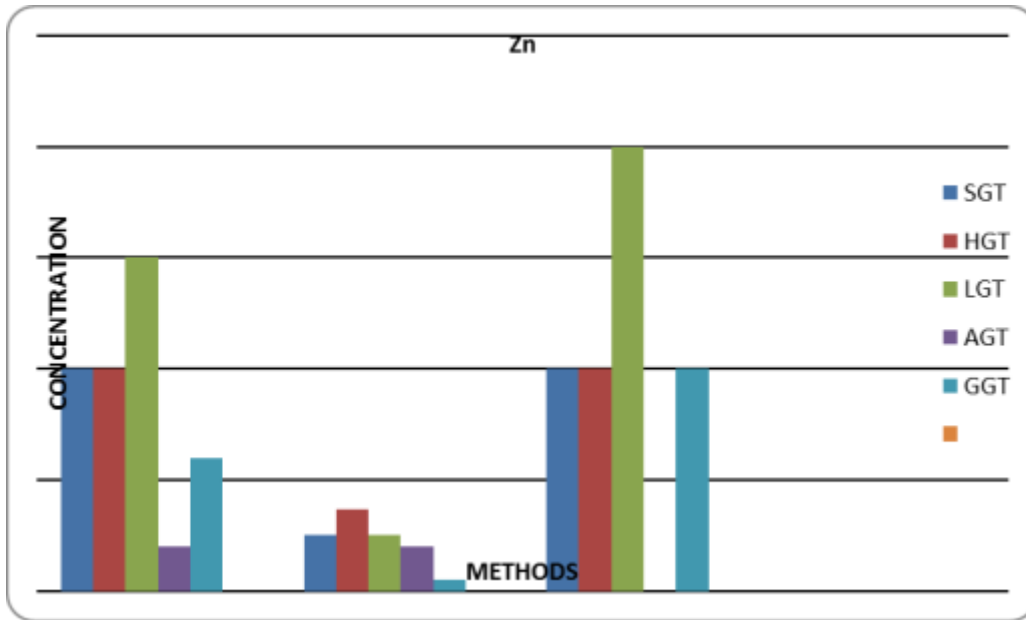


Figure 2. Zinc Concentration of Green Tea Samples

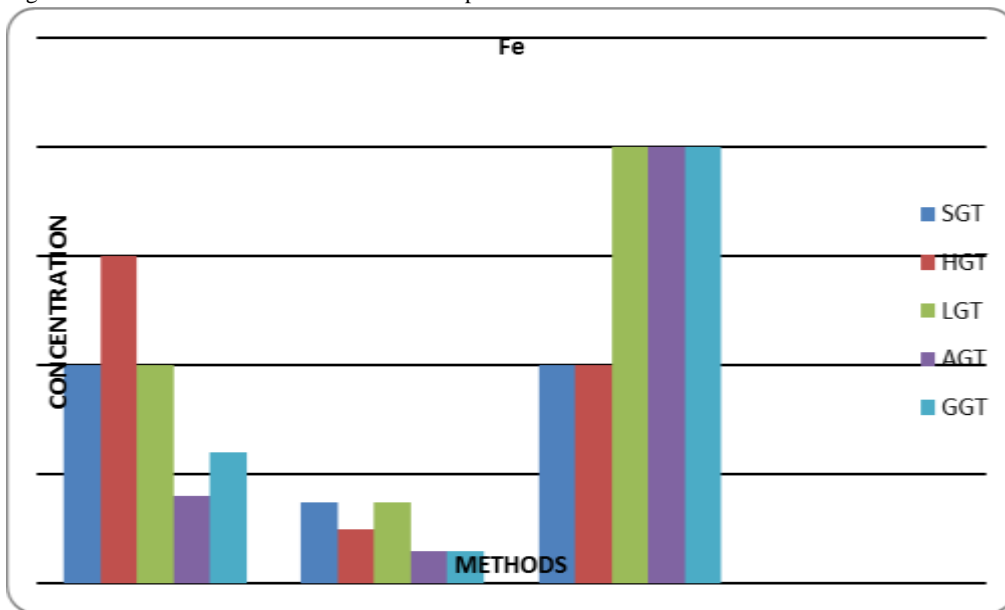


Figure 3. Iron Concentration of Green Tea Samples

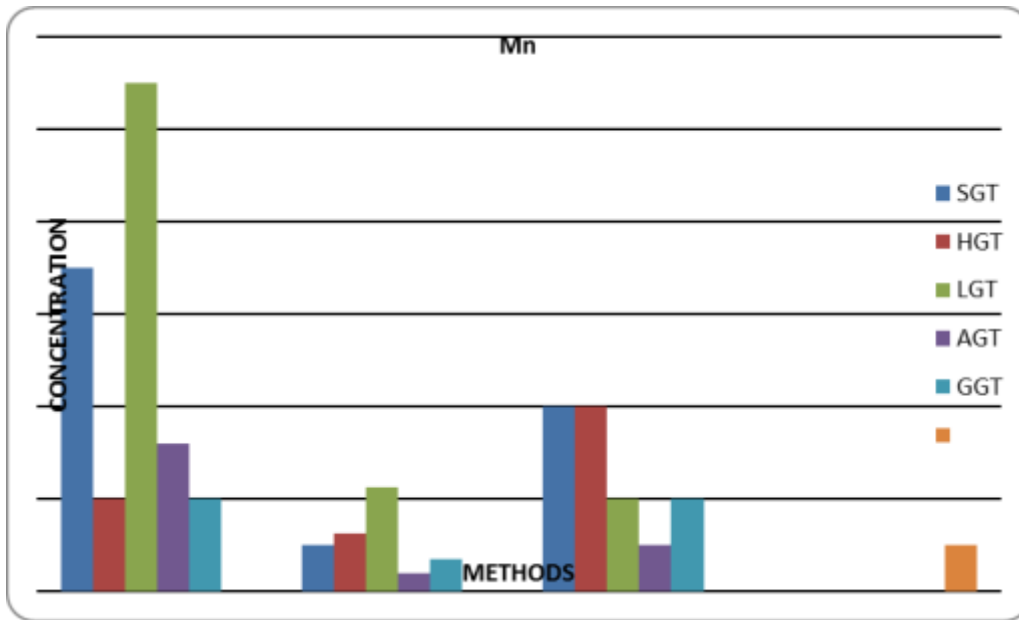


Figure 4. Manganese Concentration of Green Tea Samples

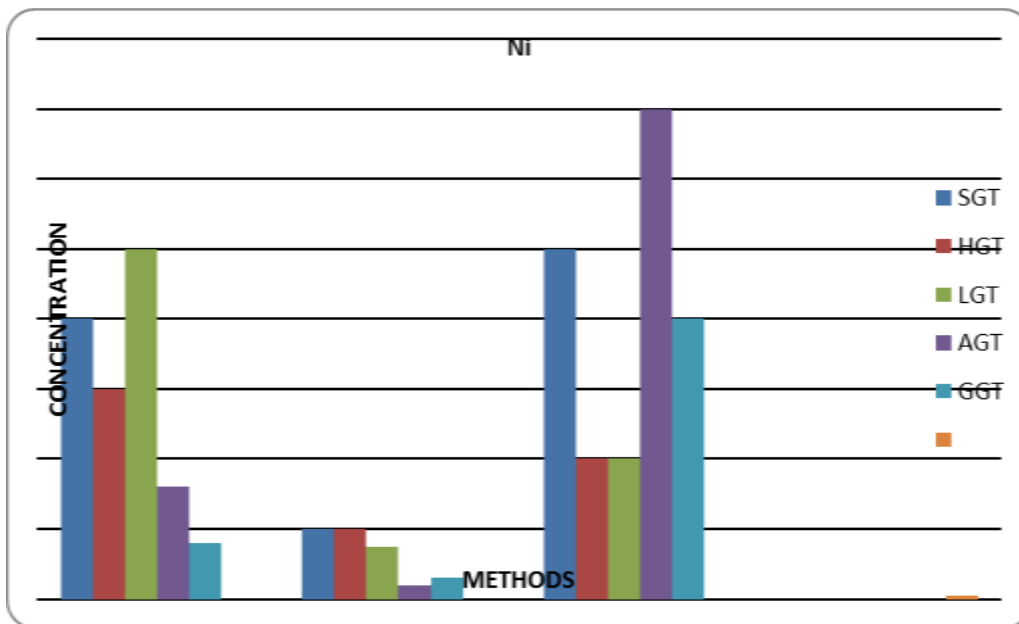


Figure 5. Nickel Concentration of Green Tea Samples

Table 1. WHO Drinking water standard Comparative Table (1993)

Metals	WHO (1993)
Cu	2 mg/L
Fe	No Guideline
Mn	0.5 mg/L
Ni	0.02 mg/L
Zn	3 mg/L

**Table 2. Tolerable daily intake of metals (mg/day) USEPA (1992)**

Trace Elements	<sup>a</sup> TDI oral (mg/kg-d)	<sup>b</sup> ATDI (mg/day)	<sup>c</sup> CTDI (mg/day)
Cu	0.0400	2.8000	0.8000
Fe	0.7000	49.0000	14.0000
Mn	0.1400	9.8000	2.8000
Ni	0.0200	1.4000	0.4000
Zn	0.3000	21.0000	6.0000

<sup>a</sup>TDI: Tolerable Daily Intake, <sup>b</sup>ATDI: Tolerable Daily Intake for adults, <sup>c</sup>CTDI: Tolerable Daily Intake for children.

## DISCUSSION

### Copper (Cu)

Cu is one of the native metals present in tea, essential to the enzyme polyphenol oxidase, a micronutrient for plants, but at high concentrations it is also extremely phytotoxic (Brun *et al.*, 2001). Overconsumption of food and beverages with Cu is also harmful to human health (Kawada *et al.*, 2002). The concentration of Cu ranges from 0.4 - 2.5 mg kg<sup>-1</sup> in wet digestion, 0.1- 0.625 mg kg<sup>-1</sup> in tea infusion and 1.0 – 1.5 mg kg<sup>-1</sup> in Dry Ashing respectively. The result indicated that the amount of Cu was recorded highest in the wet digestion method and the lowest level was shown in the infusion method, for the five samples coded as SGT, HGT, LGT, AGT and GGT respectively. The results found in both the three methods were within the standard permissible limit of 2mg/L (Table 1) except sample SGT under the Digestion method having the highest value of 2.5 mg kg<sup>-1</sup>. Moreover the result obtained was above the tolerable daily intake of 0.0400 mg/kg-d (Table 2). These variations could be attributed to different types, grades and producing areas of the tea.

### Zinc (Zn)

Zn is an important mineral essential to biological and public health and is also considered to be involved in the majority of human metabolic pathways. Zinc deficiency can lead to loss of appetite, retardation of development, changes in the skin and immune abnormalities (Ramakrishna *et al.*, 1986). In the three methods, the range of zinc concentration was 0.2-1.5 mg kg<sup>-1</sup>, 0.05-0.375 mg kg<sup>-1</sup> and 1.0-2.0 mg kg<sup>-1</sup> for wet digestion, infusion, and Dry Ashing, respectively. The highest value was observed under Dry ashing method and the lowest was recorded in the infusion method both values are within the permissible limit of 3mg/L by WHO (Table 1). However, most of the result observed under the three methods was below the tolerable daily intake of 0.3000 mg/kg-d (Table 2).

### Iron (Fe)

Iron (Fe) is the most needed micronutrient in plant. The bioavailability of this element is influenced by polyphenol found in tea that can markedly inhibit the absorption of iron (Salahinejad & Aflaki 2010). The concentration of Iron were, 1.0 mg kg<sup>-1</sup>, 1.5 mg kg<sup>-1</sup>, 1.0 mg kg<sup>-1</sup>, 0.4 mg kg<sup>-1</sup>, 0.6 mg kg<sup>-1</sup> in wet digestion while in tea infusion were 0.37 mg kg<sup>-1</sup>, 0.25 mg kg<sup>-1</sup>, 0.37 mg kg<sup>-1</sup>, 0.15 mg kg<sup>-1</sup>, 0.15 mg kg<sup>-1</sup> and in dry

ashing the results were 1.0 mg kg<sup>-1</sup>, 1.0 mg kg<sup>-1</sup>, 2.0 mg kg<sup>-1</sup>, 2.0 mg kg<sup>-1</sup>, 2.0 mg kg<sup>-1</sup>. However, figure 3. Has shown the increases in concentration in dry ashing and lowest values was obtained in the infusion method. Both the three methods are within the TDI, ATDI and CTDI of 0.7000 mg/kg-d, 49.0000 mg/kg-d 14.0000 mg/kg-d respectively.

### Manganese (Mn)

Due to its effect on acceptability as an important element for humans and animals, manganese (Mn) has widespread significance. However because of its biochemical value, Mn was the most studied element in tea from different countries and according to the literature, Mn was the only element in tea with a large dietary amount (Powell *et al.*, 1998). The range of concentration of zinc in the three methods was; 1.0 – 5.5 mg kg<sup>-1</sup>, 0.20 – 1.125 mg kg<sup>-1</sup> and 0.5 – 2.0 mg kg<sup>-1</sup> for wet Digestion, infusion and Dry Ashing respectively. Most of the results (figure 4) were above the permissible limit of 0.5 mg/L by WHO (Table 1), except in sample AGT in dry ashing which recorded 0.5 mg kg<sup>-1</sup> and 0.50 mg kg<sup>-1</sup>, 0.20 mg kg<sup>-1</sup>, 0.35mg kg<sup>-1</sup>, for SGT, AGT and GGT respectively in the infusion method. Also SGT & LGT in wet digestion exceeded the 2.8000 mg/kg-d for tolerable daily intake for children. Manganese therefore is known to cause neurological effects following exposure to inhalation, particularly in occupational settings. Epidemiological studies have been performed to report adverse neurological symptoms following repeated exposure to very high drinking water levels (Powell *et al.*, 1998).

### Nickel (Ni)

Nickel (Ni) is almost impossible to visualize soil without a trace level of heavy metals like nickel (Ni) (Adriano, 1986) which is one of the most important heavy metals in terms of its potential toxicity to plants and animals. Activities such as mining and agriculture have polluted extensive areas throughout the world by Ni. It is well known that an element like Ni is essential for plant growth at low concentrations. Nevertheless, beyond certain threshold concentrations, this element becomes toxic for most plant species (McLaughlin, 2002). The results obtained in this study were in the range of 0.4 – 2.5 mg kg<sup>-1</sup>, 0.10 – 1.5 mg kg<sup>-1</sup> and 1.0 – 3.5 mg kg<sup>-1</sup> for wet Digestion, infusion and Dry Ashing respectively. The result has shown that the concentration of Nickel in both

methods exceeded the permissible limit of 0.02 mg kg<sup>-1</sup> (WHO, 1993). Additionally, the result was above the tolerable daily intake of 0.0200mg/kg-d. Therefore, the element may affect health.

## CONCLUSION

The results of concentrations of the studied trace elements (Cu, Zn, Fe, Mn and Ni) in the tea samples coded (SGT, HGT, LGT, AGT and GGT) show that the concentrations are significantly vary among the tea brands. However, the contributory factors responsible for the differences found in tea samples could be from varying concentrations of metals in the soil, conditions of cultivation and preparations, and these vary between both countries of origin and for the type of tea. The level of concentration was compared with WHO (1993) permissible limit and most of the results were found below the standard with few exceeded the limit and this may probably be a source of health implication on humans. Hence further work should be done on these brands to study the effect of temperature, pH, and other important physical and chemical parameters.

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