THE CONTAMINATION AND EXPOSURE OF MERCURY IN HONEY FROM SINGIDA, CENTRAL TANZANIA

Ali D. Maggid [1, 2], Martin E. Kimanya [2*] and Patrick A. Ndakidemi [2].

[1] Tanzania Forest Service (TFS) Agency, P.O. Box 40832, Dar Es Salaam, Tanzania
[2] The Nelson Mandela African Institution of Science and Technology, School of Life Sciences and Bio-engineering, P.O. Box 447, Arusha, Tanzania.
Corresponding author Email: martin.kimanya@nm-aist.ac.tz.

ABSTRACT

Honey from regions where artisanal mining, using mercury is practiced, might be contaminated with mercury at levels of public health concern. This study was designed to determine the prevalence and levels of mercury contamination in honey from Singida, Tanzania and explore its extent of exposure in humans. Sampling was done in three villages where ninety (90) honey samples were collected from beekeeper households. Honey samples were tested for the presence of mercury, by using Inductively Coupled Plasma – Optical Emission Spectroscopy (ICP-OES). Mercury presence was detected in 43% of honey samples in London, 28% in Mwamagembe and 13% in Nduamughanga. The overall observed levels of contamination ranged from 0.38 µg/kg to 31.69 µg/kg; with a mean of 10.28 µg/kg. The levels of contamination were well below the permitted levels set by different countries. Based on prevalence of contaminated samples, the risk of mercury contamination and exposure was significantly ($p=0.03$) higher in London compared to the other villagers. The overall exposures of mercury ranged from 0.00015 – 15.975 µg/person/week and none of the exposure levels exceeded the Provisional Tolerable Weekly Intake (PTWI) of 1.6 µg Hg/kg Bwt/week set by the Joint FAO/WHO Expert Committee for Food Additives. Thus honey from the study villages can be considered fit for human consumption. However, more studies are needed to examine these areas to pinpoint sites of pollution and make honey from study villages a mercury free quality product.

Keywords: honey; artisanal mining; mercury; prevalence; contamination; exposure; Singida-Tanzania; ICP-OES

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INTRODUCTION

There is considerable evidence that human foods are frequently subjected to some form of contamination such as mercury as a result of environment pollution and anthropogenic activities (Berka, 2009). Heavy metals such as mercury are important potential harmful environmental pollutants (Khanna, 2011; Orina, 2012) and the widespread of contamination with heavy metals in last decades has raised public and scientific interest due to their dangerous effects on human health (Ibrahim, 2011). Mercury, one of the heavy metals, has toxic properties and severely affects the environment and humans, especially developing fetuses and infants. It damages the developing brain, and causes a lifelong negative effect in exposed population (Bose-O'Reilly *et al*., 2008). When exposed to high levels of mercury vapor, children exhibit a syndrome known as acrodynia (painful limbs) or pink disease (Davidson *et al*., 2004). Garcá-Fernández *et al*. (1996) reported that the health problems caused by mercury toxicity include headache, metabolic abnormalities, respiratory disorders, nausea and vomiting.

Just like other foods, originating from polluted environment, honey may be contaminated by mercury in honey (Ajibola *et al*., 2012). Honey is the natural sweet substance produced by honey bees from the nectar of blossoms or from secretions of or on living parts of plants, which they collect, transforms and combines with specific substances of their own, store and leave in the honey comb to ripen or mature (Anyanwu, 2012). As a natural product, honey is famous for its richness in nutrients and being a valuable remedy as it is used to treat diseases such as gastrointestinal disorders, cancer or wound healing in Egypt and Greece (Inoue *et al*., 2005).

Contaminants can reach the raw materials of bee products (nectar, honeydew, pollen, plant exudates) by air, water, plants and soil during cultivation and then be transported into the bee hive by the bees (Bogdanov, 2006; Husain *et al*., 1995; Ozores *et al*., 1997; Ellen *et al*., 1990). Buldini et al. (2001) explained that honeybees' accretions are related to air, water, and soil; they depart from flower to flower, touch branches and leaves, and drink water from pools and their hairy bodies collect contaminants particles. The presence of mercury in honey may also be caused by external sources such as industrial smelter pollution, emissions from factories, non-ferrous metallurgy, incorrect procedures during the honey harvesting, processing and preservation phases (Bratu and Georgescu, 2005; Perna *et al*., 2012; Mbiri *et al*., 2011; Pisani *et al*., 2008; Rashed *et al*., 2009). Use of agrochemicals in growing of flowers is another factor causing contamination of nectar with metals (White Jr, 1975). Rapid increase in gold mining especially small scale gold mining has led to an increase in use of mercury and hence environmental pollution and might result in increase of these metals in honey. In gold mining, mercury is used to separate gold from ore by forming an amalgam with gold. UNEP (2002) reported that, emission in the developing world have been increasing, mainly due to intensive use of mercury in artisanal gold mining and the absence of restrictive legislation.

The indiscriminate use of mercury in a gold mining process has resulted in the contamination of many aquatic and terrestrial environments (Harada, 1996; Harada *et al*., 1999). Limbong *et al*. (2003) pointed out that all related processes are undertaken with a low level of technical knowledge and skills, no regulation and disregard to safety of human and environmental health. Chibunda *et al*. (2010) observed that during dry season the mercury contaminated rivers in most of artisanal gold mining areas in Tanzania are the only watering points for animals. As a result of the environmental pollution, the nectar from which the honey is made contains metals absorbed by the roots from the polluted soil and may also contain metals carried by the bees from polluted water sources (Mbiri *et al*., 2011).

The risk of heavy metals to human health has been known for long time but exposure to it continues, taking an example from most of gold mines in the country (Tanzania) where mercury is still used. Presently, less is known with regard to risks of human exposure to mercury emanating from mining and other industries which are close or near beekeeping (honey) production areas such as those found in Singida region in Tanzania.

The present study was aimed at assessing the prevalence and levels of mercury contamination in honey from Singida, Tanzania and explores its extent of exposure in humans.

MATERIALS AND METHODS

Study sites

The study was conducted in Singida region, central Tanzania. Singida was purposely selected because it is among the high honey producing regions in the country (Mwakatobe and Mlingwa, 2005) and there are small scale gold mining operations in the region which can be a source of environmental contamination with mercury. Londoni, Mwamagembe and Nduamughanga villages were selected for the study. Londoni was selected based on the fact that it produces both honey and gold. Mwamagembe and Nduamughanga also produce honey but were selected as control villages because they are located away from mining operation; more than 300 km from small scale gold mining places.

Collection of honey handling and consumption data

Information on honey handling practices and consumption pattern was collected from 90 beekeepers (households) that were randomly selected, 30 from each of the three villages, using questionnaire. Interview was conducted to household heads or any other adult member of the beekeeper house faced provided he or she was considered being aware of beekeeping activities. The questionnaire was administered by a researcher to enable any clarifications to respondents once required and this approach facilitated mutual understanding between the two (Gwao, 2013). Questions focused on general honey production process (harvesting, extraction and storage), and honey consumption patterns. Interviewee body weight was measured using standard procedures (mechanical personal scale, Nicai Japan). Other information collected includes; distances from apiary (beehives site) to the nearby water source and mining site and forest. The handling practices information was necessary to explore the sources of mercury contamination in honey.

Information on honey consumption pattern; quantity and on daily and weekly frequency was also collected. A repeat 24-h recall technique was used to collect information from each beekeeper. During the 24h dietary recall the interviewee was also asked to recall the number of days he/she consumes honey in the previous week. The information was necessary to determine the weekly honey intake.

Estimation of weekly honey intake

Information on weekly honey intake for an individual was obtained by multiplying the daily consumption times number of days in week the individual consumed honey. Since consumption by respondents was expressed in local measurement scale and units such as a cup of tea or spoonful; honey amount contained on those scales was measured using analytical balance (Ohaus pioneer) into grams to get actual quantity of honey consumed.

Sample collection, preparation and analysis

A sample of honey was taken from each of the family interviewed. All samples were kept in air tightly closed sample bottles, labeled and stored in a cool dry place awaiting transport to laboratory for analysis.

At the laboratory, the first and second day collected honey sample, from each beekeeper, was mixed together, labeled and stored ready for mercury testing. Honey samples were digested based on a method used by Jomo Kenyatta University of Agriculture and Technology for bees honey (Mbiri *et al*., 2011). The digestion aimed at determining levels of mercury in the samples. Approximately 5g of honey was accurately weighed to 4 decimals using analytical balance, transferred to a 250 ml beaker and digested using 1:1 nitric/perchloric acid. A digested sample was then filtered to 50mls volumetric flask using de-ionized water. Mercury analysis was done by using Inductively Coupled Plasma – Optical Emission Spectroscopy (ICP-OES), with automatic introduction of 3 replicates for each sample. Limit of detection (LOD) was defined as the concentration corresponding to three times the standard deviation of ten blanks (Tuzen, *et al*., 2007). This was determined to be 0.01µg/kg. Mercury concentrations in honey samples were reported in µg/kg.

Mercury intakes assessment

For the purpose of estimating Hg intake/exposure, all honey samples below LOD were assigned half the value of the limit of detection which is 0.005 µg/kg (Govaerts *et al*., 2005). Dabeka *et al*. (2003) cautioned that, the LOD and the way results less than the LOD are reported have strong influence over dietary intake estimates since many foods contain mercury concentrations below the LOD of the analytical methods used and if these concentrations are reported as zero value will underestimate dietary intakes or, overestimating it if reported as the LOD. The dietary intakes or exposure of mercury on a body weight basis were obtained by multiplying the mercury concentration in the sample from a household by the weekly honey intake by an adult individual in the household and divided by the individual body weight (kg bw) as proposed by Chien *et al*. (2007);

Yi = Ci. Di. Xi,

Whereby:

Yi = the weekly intake by an adult individual "i" of mercury (µg/kg bw/week)

Ci = the mercury (µg/kg) in the honey sample from the family of an adult "i".

Di = the number of days an individual consumed honey in a week.

Xi, = the weekly consumption of honey (kg/kg bw) by an adult individual "i"; as estimated by the 24h dietary recall.

Statistical analysis of the data

Statistical analysis was done using Microsoft Excels spread sheet and R. Statistical differences for mean were tested using non – parametric Kruskal-Wallis test. A probability level of $p \leq 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

Levels of mercury in honey samples

Occurrence of mercury in the honey samples is presented in Table 1. A significant ($p<0.05$) difference was observed among percentage of positive samples from the three studied villages. The result shows that the prevalence of Hg contaminated honey is higher in London compared to

Nduamughanga and Mwamagembe. This finding confirms our worries that honey from areas where artisanal gold mining involves use of mercury is contaminated with the heavy metal. Conversely mean contamination in Nduamughanga was higher than in London. It should be recalled that, in addition to contamination of the environment by this element, the concentration of mercury in food depends on actions during honey processing and conservation steps (Toporcák *et al*., 1992; Akbari *et al*., 2012). Pisani *et al*. (2008) narrated that the acidic property of honey could lead to release of heavy metals from metallic tools and containers. Observations from study villages showcased that, some beekeepers used a 200 litres uncovered metallic drum for storing honey which is poor storage technique that might lead to mercury contamination in honey. The two commonly used methods for extracting honey from honey comb in studied villages are honey pressing machine and gravitation force. However, more studies are needed to scan out the possible source of mercury contamination in each of the study villages.

Table 1: Occurrence of mercury (µg/kg)

	Villages			***P-value***
Variable	**Nduamughanga**	**Mwamagembe**	**Londoni**	
Percentage of positive samples	13.3	27	43.3	0.034*
Mean of positive samples	16.187	7.633	7.023	0.789**
Range of positive samples	4.31 – 31.69	0.72 – 21.88	0.38 – 30.01	

*, Comparison by Chi-square test

**, Comparison by t-test

Mercury content in honey from study villages was lower than those reported in similar studies of mercury contamination in honey (Akbari *et al*., 2012; Carrero *et al*., 2013). For example, Akbari *et al*. (2012) found a mean value of 3030 µg/kg of honey collected from Iran. Likewise, Carrero *et al*. (2013) found the level of mercury in honey collected from Argentina, Australia, Brazil and Venezuela to vary from a minimum value of 12.96 µg/kg to maximum of 145.21 µg/kg.

Other studies revealed lower mercury content of 1.65 µg/kg in honey samples from southeastern China (Ru *et al*., 2013) and 0.67 – 2.93 µg/kg in samples from Czechoslovakia (Čelechovská and Vorlova, 2001). Bilandžić *et al*. (2012) found 0.13 µg/kg in black locust honey to a maximum of 6.11 µg/kg in chestnut honey. Bilandžić, *et al*. (2012) and Toporcák, *et al*. (1992) found levels of 1 – 3 µg/kg mercury content in honey samples from uncontaminated area. The concentration of metals in honey varies from one sample/location to another depending on plant source (botanical origin of the nectar) visited by bees, season, environmental conditions and production methods (harvesting, extraction and storage techniques) (Mbiri *et al*., 2011; Bertoncelj *et al*., 2007; Guler *et al*., 2007; Osman *et al*., 2007; Rashed and Soltan, 2004; Sanz *et al*., 2004).

There limited regulatory limits for honey (Piro and Mutinelli, 2003).The results of the present study were compared with levels of mercury in food set by different countries. The levels of mercury from study areas comply with the Canadian and Czech guideline on level of mercury in food of 500 µg/kg (Dabeka *et al*., 2003). Assuming honey is the only source of mercury in these communities, and then there is no safety concern for contamination of mercury in them.

Dietary intake of honey and mercury

Dietary intake of honey

All participants in this study reported to consume honey either as food stuff and or medicine with an average consumption of 315.58 g/person/ week (approximately to 16.41 kg/person/year). The minimum and maximum intake was recorded to be 30 g/person/week and 1575 g/person/ week, respectively (Table 2).

Table 2: Summary of weekly honey intake (g honey/person/week)

	Villages			
Variable	**Nduamughanga**	**Mwamagembe**	**Londoni**	***P-value***
Number of samples	30	30	30	
Mean	318.5	272.75	355.5	0.7427
Range	30 – 900	30 – 675	60 - 1575	

The weekly honey consumption in the study village is 45 times higher than that of Iran (weekly per capita honey consumption of 7 g/person) **(**Akbari *et al*., 2012**).** Generally, honey consumption in studied villages was higher as compared with findings from other researchers. China and Argentina had weekly per capita consumption of 1.92 to 3.84g (Bogdanov *et al*., 2008). The weekly consumption of honey in Italy, France, Great Britain, Denmark and Portugal varies between 2.1 – 7.67g, Germany, Austria, Switzerland, Portugal, Hungary and Greece 19.18 – 34.52g, USA, Canada and Australia 11.51 to 15.34g [available on http://www.apiservices.com/]. There is no information on per capita honey intake in Tanzania. A simple estimation of honey intake in Tanzania based on the country population of 45 million people (The United Republic of Tanzania (URT), 2013) and country's annual honey consumption of 14,795,000 kg (Belgium Technical Cooperation **(**BTC) Tanzania, 2014), honey consumption per person per week is 6.31g. The estimated value of honey intake from study village (Table 2) is still much higher as compared with estimate at country level. The difference in honey intake could be associated with culture, product availability, consumption pattern, taste and preference.

Dietary intake of mercury

Table 3 shows the results of the weekly mercury intake (µg Hg/person/week). When the mercury intake data was compared, it was observed that the differences among the villages was significant ($P<0.05$). The difference in mercury intake among the three villages is consistent with the differences in Hg contamination and honey intakes. Whereas Hg contamination was significantly higher in Nduamughanga, honey intake was considerably higher in London.

Table 3: Weekly dietary intake of mercury (µg/person/week)

Variable	Villages Nduamughanga	Mwamagembe	Londoni	*P-value*
Number of samples	30	30	30	
Mean	0.9497	0.3973	0.9093	0.03186
Range	0.00015 – 15.975	0.00015 – 4.071	0.0006 – 10.804	

The minimum and maximum weekly mercury intakes are 0.00015 and 15.98 µg/person/week respectively (Table 3). The observed average results of weekly mercury intake from study villages were low when compared with results of other studies**.** Lee *et al*. (2006) in assessing dietary exposure of the Korean population to some selected heavy metals, observed mercury dietary intake of 11.27µg Hg/person/week. In the 1994 Total Diet Study in the UK, Ysart *et al*. (1999) observed that the UK population was exposed at 28 µg Hg/person/week. The highest observed weekly mercury intake from studied villages in this study (15.98µg Hg) was higher when compared with results estimated in Korea and UK. Honey consumption pattern and levels of mercury in honey samples are two major reasons that caused differences in mercury intakes data from area of study.

Furthermore, results showed that total mercury intake in the studied villages did not exceed Provisional Tolerable Weekly Intake (PTWI) of 1.6 µg/kg Bw/week – mercury as recommended by the Joint FAO and WHO Committee on Food Additives and Contaminants (JEFCA) in (2003).

Codex Alimentarius Commission (2001) emphasized that honey shall be free from heavy metals in amounts which may represent a hazard to human health. However, the population in study area used varieties of types of food in their daily diet and honey being one of them. Maize, rice and sorghum are some of staple food commonly used by most Tanzanians. A study conducted by Qian *et al*. (2010) in China found level of mercury in rice to be 0.0058µg/g. For the purpose of avoiding adverse health risk in future, it is advised that immediate action should be taken to assess the contribution of other foods in their total daily intake of mercury and identify and control possible source of mercury contamination in honey from study villages, possibly using total dietary studies. The World Health Organization (1999) pointed out that, total diet studies are one of the most reliable techniques for estimating dietary intakes of chemicals by large population groups and recommends each country to conduct total diet studies and that mercury be included in the obligatory list of toxicants to be measured.

CONCLUSION

The study generated useful information on honey consumption patterns and mercury contamination in honey in Singida region of Tanzania. It shows that despite the unusual high intake of honey in the region, there is no imminent risk of mercury exposure from honey consumption in Singida. Results further showed that, based on the significant higher prevalence of mercury contaminated samples, people in Londoni are at a higher risk of consuming mercury contaminated honey compared to those in Nduamughanga and Mwamagembe villages. The higher prevalence of mercury in honey from Londoni compared to the other villages of Nduamughanga and Mwamagembe suggests that the environment in this village is contaminated by the artisanal gold mining operations taking place in the village. Additionally, the study found a significantly higher mean contamination of mercury in Nduamughanga compared to the other villages, despite its location away from gold mining sites. This finding suggests that there are sources of contamination, other than mining operations to be considered. Importantly, the study shows that, more studies are needed to examine these areas to pinpoint sites of pollution, identify the extent of contamination and exposure of mercury for other foods and recommend measures to minimize mercury exposure in Singida and other areas nationally and internationally.

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