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Full Length Research Paper

Chromium and copper in toenails of some Kano inhabitants

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Toenail chromium and copper concentrations in toenails of some inhabitants resident in Kano for at least six months were assessed by atomic absorption spectrometry. Average toenail chromium and copper concentrations were 1.33 ± 0.68 and 27.62 ± 13.29 µg/g respectively. Both chromium and copper concentrations in nails decreased with age indicating that these metals may be playing some physiological functions during the formative years. Their concentrations were inversely related to age with approximate average of 10% decline with each decade of age.

Key words: Chromium, copper, toenails, Kano, Nigeria.

INTRODUCTION

The classification of trace elements includes a group of non-essential or toxic trace elements because their biological significance is confined to their toxic properties only. These include lead, cadmium, mercury and aluminium, acquired through environmental contamination, modern agriculture, industrial practices and contaminated food/water supplies (Barnes and Bradley, 1994; Bradley and Bennett, 1995).

Chromium is an essential trace element required for normal carbohydrate, lipid, and protein metabolism (Mertz, 1993; Katz and Salem, 1994; WHO, 1996; Anderson, 1997; Stoecker, 1999; NAS, 2001; Cefalu and Hu, 2004; Vincent, 2004). Chromium deficiency results in impaired glucose tolerance, hyperglycemia, and glycosuria that cannot be controlled with insulin (Anderson, 1998; Jeejeebhoy, 1999). Although overt chromium deficiency is rare and its low intake may be a cause of subclinical insulin resistance and an adverse lipid profile in the Western populations (Katz and Salem, 1994; Anderson, 1997; Anderson, 1998). Use of chromium supplements is increasingly popular in some countries (Simon, 1994; Lukaski, 1999) although their effects on glucose tolerance, body composition and lipid parameters are still unclear (Vincent, 2004; Cefalu and Hu 2004, Vincent, 2003; Nissen and Sharp, 2003; Pittler et al., 2003).

Chromium, in the naturally occurring dinicotinic acid gluthationes complex is vital for carbohydrate metabolism as it potentiates the action of insulin (Underwood, 1977; Mertz, 1982) and normalizes blood sugar levels in subjects with tendencies toward blood-sugar fluctuations associated with diabetes and low blood sugar (Anderson, 1980).

When chromium is assessed in the context of age, sex, race, cholesterol, triglycerides, systolic blood pressure and diastolic blood pressure its low concentrations proved to be the best predictor of coronary artery disease (Newman et al., 1978). The role of chromium in the function of nucleic acids metabolism and synthesis is indicated by the high concentrations of this trace element in nuclear proteins relative to other transition metals (Underwood, 1977; Borel and Anderson, 1984). It is concentrated largely in the nuclear fraction of the cells, the remainder is divided between the mitochondria and the microsomes (Underwood, 1977).

Chromium deficiency depresses nucleic acid synthesis, lower sperm count and decreased fertility. Chromium is essential for maintaining the structural stability of proteins

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and nucleic acids. Premature infants and those with evidence of intrauterine growth retardation have lower hair chromium status compared to infants born full-term (Hambidge, 1971). Similarly multiparous women have lower body chromium levels compared to nulliparae (Hambidge, 1974). It is an essential trace element during fetal growth and development (Barnes and Bradley, 1994; Bradley and Bennett, 1995).

Copper deficiency leads to anaemia defective wool keratinization, abnormal bone formation, arterial and cardiac aneurysm (Hart et al., 1928; Shields et al., 1962). Other features of severe copper deficiency include neurological problems such as ataxia, seizures and episodic apnea due to lack of myelination leading to reduced nerve cell formation during embryonic development (Underwood, 1977; Iwata et al., 1979; Zimmerman et al., 1976). Copper deficiency, as a syndrome in infants is characterized by poor growth, white brittle hair with peculiar twisting, arterial defects, focal cerebral degeneration and mental retardation (Danks et al., 1971). Copper deficiency in infants may lead to pathological bone fractures (Cordano et al., 1964) and these defects are related to reduced activity of a copper-dependent enzyme, lysyl oxidase, vital for the cross-linking of collagen (Mason, 1979; Gallop et al., 1972). Cardiovascular disorders are evident in all species subjected to severe copper deficiency whether genetic or nutritional in origin (Underwood, 1977). These are caused by impairment of cross-link formation of soluble elastin and collagen due to depression of the lysyl oxidase activity (O`Dell, 1976; Gallop et al., 1972).

The symptoms of copper poisoning frequently associated with suicidal intent include: nausea, vomiting, diarrhoea, hypotension, jaundice, hematuria, anuria, coma and death (Chuttani et al., 1965). Prominent sources of copper are from drinking water supplies in areas where the water is soft and acidic, as this corrodes layers of copper from the pipes (Barnes and Bradley, 1994; Bradley and Bennett, 1995). Cigarette smoking is another source of excessive copper accumulation (Davidoff et al., 1965). Similarly oral contraceptives are notorious in raising the body's copper burden (Barnes and Bradley, 1994; Bradley and Bennett, 1995; Crew et al., 1980; Carruthers et al., 1966). A high body copper burden can be responsible for such disorders as hypotension, heart disease, premenstrual tension, postpartum depression, paranoid and hallucinatory schizophrenias, childhood hyperactivity and autism (Pfeiffer, 1979).

Toenails are preferred markers for assessment of long term exposure and as measures of absorption. They are preferred biological medium because of ease of collection, storage convenience, their usefulness in estimating intake of minerals in nutritional studies, ease of handling, reproducibility of later analysis results and the potential for less external contamination compared with hair or fingernails (Garland et al., 1993; Hunter, 1990; Karagas et al., 1996; Takagi et al., 1988). Each clipping represents several weeks of growth and because nails from various toes vary in the time between formation and clipping these are likely to reflect integrated exposure (Hunter, 1990). This study was aimed to investigate chromium and copper in toenails of inhabitants residents in Kano and at identifying the sources of exposure that make important contributions to the concentrations of these elements.

MATERIALS AND METHODS

Sample collection

The toenails collected were washed using the standard procedure (IAEA, 1997). Volunteers were asked to wash their hands and feet thoroughly with distilled water and medicated soap devoid of metal contamination, followed by drying with clean towel or tissue paper to remove any external contamination. Using clean stainless steel scissors, toenail samples of the volunteers (age 1 – 60 years) were collected from the toes. For subsequent analysis, each of the nail samples was sealed in plastic cover till it was washed, dried, digested and converted into a water-clear solution.

Procurement of requisite details of subjects

The personal and medical history, along with relevant details of the subjects was obtained through a questionnaire based on the recommendation of the World Health Organisation (WHO). The information required to be filled in the pro-forma included sex, age, place of residence, occupation and any possible prior metal exposure.

Sample treatment

The procedure involved 6 steps of washing each sample in a clean beaker in an ultrasonic bath, with 25 cm^3 portions of, successively, water, acetone, 1% detergent solution, water, acetone, water, decanting the wash liquid after 10 min wash (Gammelgaard et al., 1991; Kucera et al., 1996). After washing the samples were air dried and kept in a plastic container.

Analyses

The concentrations of the metals were assayed by flame atomic absorption on a Buck Model 210 VGP spectrophotometer with air acetylene flame. A series of standards were prepared in deionised water for instrumental calibration by diluting commercial standards containing 1000 mg/dm^3 of the metals. All reagents used were of analytical grade and were free from any metal contamination. A number of blanks were prepared for minimization of contaminated errors. The main instrumental parameters like band width, lamp current and wavelength for estimation of metals by atomic absorption spectrophotometer were set-up separately for each metal (Mehra and Juneja, 2005).

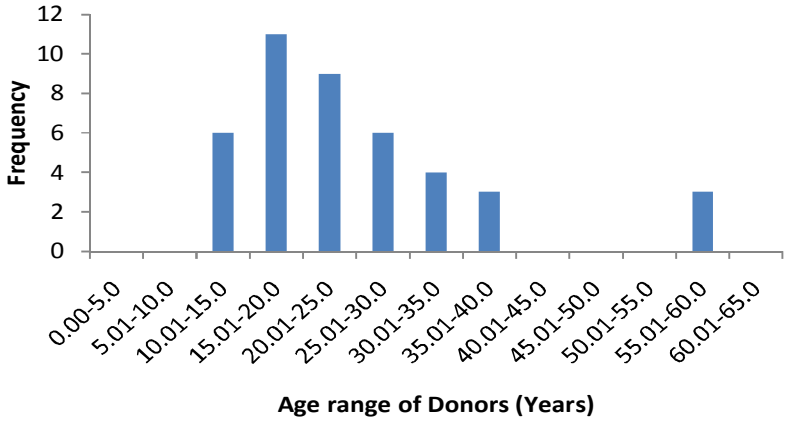

Figure 1. Frequency distribution pattern for age (years) of donors.

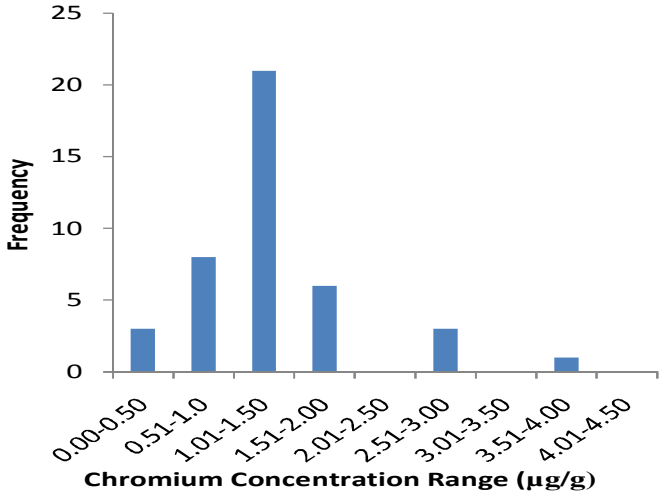

Figure 2. Frequency distribution pattern for chromium in toenails.

0.5 g of each sample was digested in 10 cm^3 concentrated HNO_3 and the resulting solution was evaporated to dryness and redissolved in 0.1 M nitric acid. Trace metal concentrations were determined by flame atomic absorption on a Buck Model 210 VGP spectrophotometer. The result of the absorbance of each sample was the average of ten sequential readings. Background light absorption and scattering were compensated for by deuterium hollow cathode lamp. Distilled water was digested as blank using the same procedure previously described (Ayodele and Abubakar, 1998; Ayodele and Abubakar, 2001; Ayodele and Bayero, 2009).

Statistical analyses

All statistical computations either were on the PC 486 66 MHz microcomputer using the integrated statistical package for windows from Umstat Ltd. (London) or dedicated micro instructions for the Excel spread sheets from Microsoft. The approach enabled the advantages of the various computational and graphical facilities of both types of software's to be used with the ability to read different file formats. The analyses of variance (ANOVA) were carried out according to described procedures (O'Mahony, 1986).

RESULTS AND DISCUSSION

The frequency distribution pattern for the age of toenail donors is as shown in Figure 1. The distribution is multimodal with a mean age of 25.01 ± 11.46 years. The frequency distribution pattern for chromium in toenail is as shown in Figure 2. The distribution is multimodal and is skewed towards low frequency of high concentration with a mean and standard deviation of 1.3 3± 0.68 µg/g while the frequency distribution pattern for copper in toenails

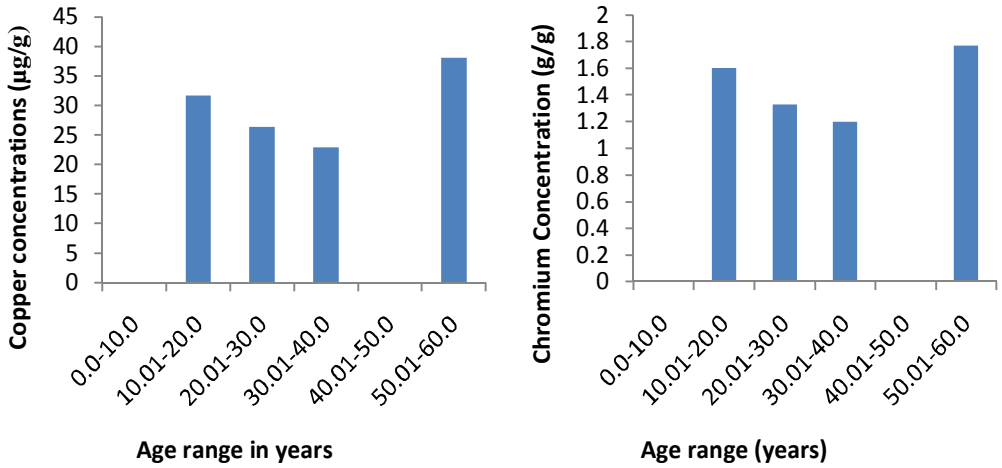

Figure 4. Chromium and copper concentrations (µg/g) in toenails with respect to age.

Table 1. Parametric correlation coefficients for chromium and copper in toenails.

Correlations		Chromium	Copper
Chromium	Pearson correlation	1	.864**
	Sig. (1-tailed)	.	.000
	N	42	42
Copper	Pearson correlation	.864**	1
	Sig. (1-tailed)	.000	.
	N	42	42

**. Correlation is significant at the 0.01 level (1-tailed).

toenails (Figure 3) is multimodal and is skewed towards low frequency of high concentration with a mean and standard deviation of 27.62 ± 13.29 µg/g. Pearson parametric correlation showed a significant correlation between the chromium and copper contents in toenail ($p < 0.05$) (Table 1). The analysis of variance (ANOVA) revealed that the mean chromium concentration in toenail is significantly different from that of copper $p > 0.05$ (Table 2). The levels obtained in this study are in agreement with mean copper in toenails reported by other authors worldwide (Table 3).Chromium and copper concentrations in toenails with respect to age is as shown in Figure 4. Both Cr and Cu concentrations in toenails decreased with age indicating that these metals may be playing some physiological functions during the formative years. There was inverse relation in their concentrations with age with approximate average of 10% decline in their concentrations with each decade of age (Guallar et al., 2005) (Figure 4).Toenails can record the level and changes of elements in the body over a long period of time (Saiki et al., 1998; Khuder et al., 2008). Changes in the elemental composition of toenails therefore depend on alterations of external and internal media of the human body and is considered that toenails of healthy individuals contain each element and are potential indicator of both external and internal long-term exposure to pollutants. The idea of toenail analysis is inviting, since it is painlessly removed, normally discarded, easily stored and transported to the laboratory for analysis. Analysis is simple and painless trace metal concentrations are not subjected to rapid fluctuations due to diet or other variables and therefore reflect a long – term nutritional status. Samples are stable at room temperature, analytical methods are easy because metal concentrations in toenails are relatively high (Borel and Anderson, 1984; Ayodele and Bayero, 2008; Ayodele and Bayero, 2009).

The main route of exposure to chromium and copper in the general population is dietary intake. Most chromium in diet is trivalent chromium and any hexavalent chromium in food or water is reduced to trivalent chromium in the acidic environment of the stomach (Guallar et al., 2003).

Foods with high chromium concentrations include whole

Table 2. Non-parametric correlations coefficients for chromium and copper in hair and nails.

Correlation			Chromium	Copper
Kendall's tau_b	Chromium	Correlation coefficient	1.000	.847**
		Sig. (1-tailed)	.	.000
		N	42	42
	Copper	Correlation coefficient	.847**	1.000
		Sig. (1-tailed)	.000	.
		N	42	42
Spearman's rho	Chromium	Correlation coefficient	1.000	.915**
		Sig. (1-tailed)	.	.000
		N	42	42
	Copper	Correlation coefficient	.915**	1.000
		Sig. (1-tailed)	.000	.
		N	42	42

**Correlation is significant at the 0.01 level (1-tailed).

Table 3. Results of chromium concentrations in toenails from different countries.

Country	Mean	Unit	References
Granad-a Spain	0.7		
Malaga-Spain	1.87		
Europe (mean)	1.10		Guallar et al. (2005)
Europe (control	1.30		
Moscow Russia	1.85	µg/g	
USA	1.6 - 4.0		Saiki et al. (1998)
USA	2.43 ± 3.16		Garland et al. (1993)
USA	2.39 ± 2.91; 1.72 ± 2.10		Garland et al. (1993; 1996)
Nigeria	1.326 ± 0.678		This study

Table 4. Results of copper concentrations in toenails from different countries.

Country	Mean	Unit	References
Brazil	29 - 175; 34 - 292; 40 - 101	µg/g	Menezes et al. (2002)
USA	5.21 ± 3.34; 4.33 ± 2.29		Garland et al. (1993)
USA	5.61 ± 3.88; 5.68 ± 4.47		Garland et al. (1996)
Nigeria	27.624 ± 13.2		This study

grain products, green beans, broccoli and bran cereals (Anderson et al., 1992). The use of stainless steel equipment in food processing may add measurable amounts of chromium (Offenbacher et al., 1997) especially in acidic foods, salted foods and foods that are in close contact with stainless steel when processed. The chromium content of grain products, fruits and vegetables varies because of soil properties (Offenbacher et al., 1997; Pfeifer et al., 2000).

Conclusion

This study has shown that toenails may be useful biomarkers for exposure to contamination and has also confirmed that factors such as age and sex are important and should be considered when investigating the exposure of human populations to environmental trace metal concentrations. Toenail trace metals may therefore be a better surrogate because of the correlations with

environmental concentrations compared with hair. It is not possible to conclude from the results of this study that residents with high toenail chromium and copper concentrations are absorbing them to a greater extent because of the likelihood of external contamination. However, residents are certainly exposed to these metals from these sources as measured by their toenail concentrations. Further work is recommended to characterize chronic exposure in residents known to be exposed to high concentrations.

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