Ind J Clin Biochem
DOI 10.1007/s12291-017-0693-y

ORIGINAL RESEARCH ARTICLE

Investigation of Trace Elements in the Hair and Nail of Patients with Stomach Cancer

Ghasem Janbabai[1] · Abbas Alipour[1] · Sina Ehteshami[1] · Seyedeh-Samaneh Borhani[1] · Touraj Farazmandfar[1,2,3]

Received: 28 June 2017 / Accepted: 31 August 2017

Abstract Trace metals are beneficial nutrient materials that act as essential cofactors in physiological processes. Recent evidence suggests that increase or decrease in certain trace metals may be related with risk and development of chronic diseases such as cancer. This study analyzed some trace elements level in hair and nail of patients with stomach cancer, and compared with their level in healthy controls. Trace elements (Cu, Fe, K, Li, Mg, Mn, Na, P, Se, Sr and Zn) are estimated in hair and nail of the 73 cancer patients and 83 controls by atomic absorption spectrophotometric method. The levels of Cu, K, Li, P and Se in hair and nail samples, were significantly higher in cases than controls. Levels of Mg and Sr were significantly lower in cases than controls. Fe level in hair samples was significantly higher in cases than controls. The mean concentrations of Fe, Se and P significantly increased with increasing cancer stage in the hair of patients. The average concentration of k also significantly increased with increasing cancer stage in the nail of patients. The results of our study show that there is an association between the increase in Cu, K, Li, P, Se and Fe, and stomach cancer development. Our results reveal that the increase in the trace elements could be a potential diagnostic marker to predict cancer progression and its etiology.

✉ Touraj Farazmandfar
tourajf@yahoo.com

[1] Molecular and Cell Biology Research Center, Mazandaran University of Medical Sciences, Sari, Iran

[2] Cancer Research Center, Golestan University of Medical Sciences, Gorgan, Iran

[3] Medical Cellular and Molecular Research Center, Golestan University of Medical Sciences, Shastkola Road, Falsafi Complex, Gorgan 4934174611, Iran

Keywords Stomach cancer · Trace element · Hair · Nail · Association

Introduction

The importance of trace metals in human health issues has been increasingly recognized in recent decades. Trace metals are beneficial nutrient materials that act as essential cofactors in physiological processes by affecting enzymatic reactions or by influencing the permeability of cell membrane [1]. Therefore, trace elements might affect directly or indirectly on the carcinogenic development [2–4]. Moreover, some metals may be toxic to human health. Recent evidence suggests that increase or decrease of certain trace metals may be related with risk and development of chronic diseases such as cancer [1]. On this subject, many investigations have been carried out in which was focused on metal-induced carcinogenicity and oxidative stress as the most important mechanisms [5, 6]. Finding in recent studies have revealed that there was a significant difference in the concentration of some trace metals in cancerous tissues in comparison with normal tissues [7–9]. Stomach cancer is one of the common types of cancer worldwide, and mainly related to environmental factors rather than genetics [10, 11]. Thus, the role of environmental contaminants become important in the screening and prognosis of stomach cancer [12].

Human nails are largely comprised of keratin-rich proteins, which accommodate trace metals in proportion to their intake by various mechanisms such as synthesis of proteins with sulfhydryl groups. Therefore, the nails are a useful marker for trace metals investigation and are increasingly used in clinical studies. Many hypotheses involve trace metals, either as a required nutrient or a toxic

Published online: 11 September 2017

material. Nail pieces can be often collected and stored for years prior to being retrieved and used in epidemiological studies to analyze the concentration of the trace elements in future clinical approaches [1, 7, 13, 14].

Hairs are the metabolic end products which have a specific potential to reflect the body metal load. The concentration of a certain metal in hair demonstrates a balanced mineral content of body in a long period of time. This content can be modified by intake exposure of trace metals in high quantities [15, 16]. Thus, the quantification of trace metals in hairs can be used for estimation of long-term exposure to trace metals [17].

This study analyzed some trace element levels in hair and nail of patients with stomach cancer and healthy controls, thereby investigating whether these trace elements had any possible values in stomach cancer prognosis.

Materials and Methods

Case group in this study consisted of 73 stomach cancer patients with ages between 46 and 74 years and a median value of 61 years. All the patients subjected in this study had surgery with histologically confirmed TNM (Tumor, Node, Metastases staging system) stage from the Bagheban Clinic, Sari, Iran, on the volunteer basis. In agreement with the Helsinki Declaration, all participants were aware of the study details, and signed the relevant written informed consent. This study was approved by the Ethics Committee of Mazandaran University of Medical Sciences. Clinicopathological features of patients were obtained from medical records. Healthy group (n = 83) were also collected from the same region with matched age groups.

About 2 g of hair and nail was cut by using plastic scissors. The samples were stored in polythene bags and were labelled. The samples were then washed twice with 4% (w/v) detergent solution and twice rinsed with distilled water to remove exogenous contamination. The samples were dried overnight in an oven at 50 °C and cooled to 25 °C in a desiccator containing desiccant silica gel. A weighed portion about 1 g of the samples was treated with 8 ml of concentrated nitric acid (65%) and heated 10 min at 80 °C and was cooled to 25 °C. Then, 5.0 ml of perchloric acid was added to samples with subsequent heating until white dense fumes evolved. Finally, the samples were cooled to 25 °C and diluted to 10 ml with distilled water. The blank was prepared in the same way but without nail and hair sample. An inductively coupled plasma mass spectrometer (ICP-mass) ELAN 6000 (Perkin-Elmer, Norwalk, USA) was used for the determination of trace elements and metals including Copper (Cu), Iron (Fe), Potassium (K), Lithium (Li), Magnesium (Mg), Manganese (Mn), Sodium (Na), Phosphorus (P), Selenium (Se), Strontium (Sr) and Zinc (Zn).

Descriptive statistic was applied for demographic data. Concentration of Trace elements and metals were expressed as a mean and median value (25 and 75 percentile) in parts per million (ppm). The statistical significance of median values between case and control groups was analyzed by independent sample *t* test and Mann–Whitney test. A p value of less than 0.05 was considered statistically significant.

Results

In this study, 156 subjects, aged 45 to 82 years, consisted of 73 patients with stomach cancer and 83 healthy controls. Basic clinical and demographic characteristics of the participants in Table 1 showed that the difference between the two groups of case and control in terms of age and gender was not significant ($p > 0.05$). As shown in Table 1, 40 (57.1%) of patients were in stage IV disease. Moreover, the most frequent tumor location in the stomach located in the Body and Antrum (any location 18, 32.1%).

Basic statistical distribution of parameters in selected trace metals (Cu, Fe, K, Li, Mg, Mn, Se, Sr, Zn, Na, P) in hair and nail samples of patients and controls have been shown in Table 2. At the present study, level of trace elements and metals was determined in samples (hair and nail), and was compared in two groups (case and control) (Table 3). As shown in Table 3, the level of Cu, K, Li, P and Se in hair and nail samples, was significantly higher in cases than controls. In contrast, level of Mg and Sr was significantly lower in cases in comparison with controls.

Table 1 Characteristics of the subjects

Variable	Case	Control	*p* value
Age, mean ± SD	60.84 ± 13.9	60.22 ± 13.36	0.790
Sex, F/M	39/34	44/39	0.560
TNM stage, N (%) (n = 70)			
I	4 (5.7)	–	–
II	19 (27.1)		
III	7 (10)		
IV	40 (57.1)		
Location of tumor, N (%) (n = 56)			
Cardia	16 (28.6)	–	–
Fundus	4 (7.1)		
Body	18 (32.1)		
Antrum	18 (32.1)		

TNM tumor, node, metastases staging system

Table 2 Distribution of trace elements in hair and nail of the cancer patients and healthy donors

	Cu	Fe	K	Li	Mg	Mn	Se	Sr	Zn	Na	P
Hair											
Cancer patients											
Min	6.1	3.1	1.1	1.2	7.8	0.1	1.4	0.7	32.1	3108.8	3827.2
Max	111	917	4753.3	243.4	1552	31.2	171.5	482	530.8	18,364.5	17,834
Mean	52.1	417.8	637	89.1	386.4	3.6	77.2	47.8	237.5	8825.5	9951.4
SD	33.3	247.7	1184.7	75.2	386.6	5.9	47.6	84.1	113.1	3858.3	4375.6
Healthy controls											
Min	1.4	69.9	1.3	1.5	172.4	0.1	1.1	7.3	78.1	496.2	191.8
Max	34.1	1272.1	867.2	77.8	1846.8	577	78.7	122.6	1052.3	20,321.5	16,382.8
Mean	13.3	275.2	154.3	6	787.5	21.5	4.8	47.6	256.8	7605.5	6048.4
SD	8.6	252.8	229.4	17.4	401.5	97	15.8	26.6	198.7	4037.7	3549.5
Nail											
Cancer patients											
Min	1.1	39	1.2	1.1	37.2	0.1	1.2	0.6	74.3	1501.1	3506.1
Max	108.1	930	2044.7	337.9	1664	89	188.2	113.8	460.2	17,165.4	18,496
Mean	61.3	391.1	342	109	410.3	6.3	82.6	25.6	160.8	8943.6	11,175.6
SD	31.9	258.4	444.2	74.4	335.6	15.6	51.3	27.5	78.4	3414.2	4365.8
Healthy controls											
Min	1.3	102	1.1	1.3	373.4	0.1	1.3	11.1	70.1	548.6	1199.6
Max	33.9	656	2950	65.6	1887.3	19.3	63.8	89.3	316.7	13,327.9	13,520.4
Mean	6.7	297.9	202	4.1	852.5	4.8	2.5	38.6	158.1	7442.8	6589.7
SD	7.2	135.5	579.4	11.7	367.9	5.5	9.8	20.5	55.3	3236.5	3106.2

All numbers are reported in ppm

However the Fe level in hair samples was significantly higher in cases than controls, but no significant difference was seen in nail Fe level between cases and controls. Moreover, No significant difference was also observed in levels of Mn, Na and Zn between case and control groups.

The average of trace elements concentration based on cancer stage in the hair and nail are displayed in Figs. 1 and Fig. 2, respectively. The mean concentrations of Fe, Se and P significantly increased with increasing cancer stage in the hair of patients ($p = 0.021$, $p = 0.033$ and $p = 0.037$, respectively) (Fig. 1). The average concentration of k also significantly increased with increasing cancer stage in the nail of patients ($p < 0.001$) (Fig. 2).

Discussion

It is known that metals and trace elements have a key role in a large number of biological mechanisms. They have some regulatory functions such as affecting immune reactions and free radical production. The extremely low quantities of some of the these trace elements imply that their molecular effects are specific and unique [1–4]. Some studies on experimental animals, revealed that a higher Zn concentrations was associated with more tumor growth [18]. A significant correlation have been also observed between blood Zn concentration and mortality in several different cancers [19]. Furthermore, a few studies reported that there is a significant association between higher Zn concentration and cancer progression [7, 9]. Schrauzer [18] reported that breast cancer development was directly correlated with the dietary intake of zinc and inversely correlated with Se. In addition, they showed that Iron is a key factor for the numerous physiological functions in human because it is an essential part of many enzymes and proteins. The excessive accumulation of iron in the body may be correlated with an increased risk of human cancers [20]. It causes tissue damage by increasing the generation of free-radical ions from hydrogen peroxide. These ions attack cellular membranes, inactivate enzymes, cause DNA strand breaks, initiate lipid peroxidation, and depolymerize polysaccharides [21, 22]. Iron also causes inflammation and increase the growth of cancer cells [23]. Copper is an essential element, and its high concentration can induce cell proliferation and cancer, specially through the production of highly reactive oxygen species that unfavorably modify proteins, nucleic acids, and lipids [24]. The high level of Cu concentration in serum of cancer patients probably promote tumor growth through oxidative DNA damage and angiogenesis [25, 26]. Selenium is an essential

Table 3 Concentrations of trace elements in the hair and nail of study participants and comparison of those between the cancer patients and healthy donors

Element	Sample type	Participant median (25–75%) (ppm)		*p* value
		Case	Control	
Cu	Hair	43 (24–83.5)	12 (7–19)	<0.001
	Nail	49 (39–93)	5 (0.5–9)	<0.001
Fe	Hair	394 (208.5–571)	204 (120–290)	0.003
	Nail	365 (187.5–549)	259 (200.5–358.5)	0.34
K	Hair	98 (11.25–283.25)	1 (1–327)	0.009
	Nail	191 (68–438.5)	1 (1–55)	<0.001
Li	Hair	82(11.5–140.5)	1 (1–1)	<0.001
	Nail	100(58.75–136.25)	1 (1–1)	<0.001
Mg	Hair	218 (135–579)	731.5 (492.8–979.5)	<0.001
	Nail	320 (220.5–583)	793.5 (615.8–101.3)	<0.001
Mn	Hair	1.9 (0.6–3.8)	0.5 (0.05–9)	0.340
	Nail	2.7 (0.8–4.2)	4 (0.05–10)	0.541
Na	Hair	8171 (5856–11,677)	6699 (5506–8796)	0.241
	Nail	8735 (5540–11,565)	7183 (4978–10,907)	0.060
P	Hair	9813 (6401–1366)	5778 (4338–7807)	<0.001
	Nail	10,849 (7535–15,779)	6631 (4703–8725)	<0.001
Se	Hair	71 (50.5–113)	1 (1–1)	<0.001
	Nail	72 (55–133)	1 (1–1)	<0.001
Sr	Hair	20 (12.5–51)	40 (29–67)	0.023
	Nail	16 (7.5–35)	35 (22.5–45)	<0.001
Zn	Hair	224 (164–285)	205 (138–280.5)	0.57
	Nail	134 (115.5–177.5)	150 (117.5–200)	0.62

ppm parts per million

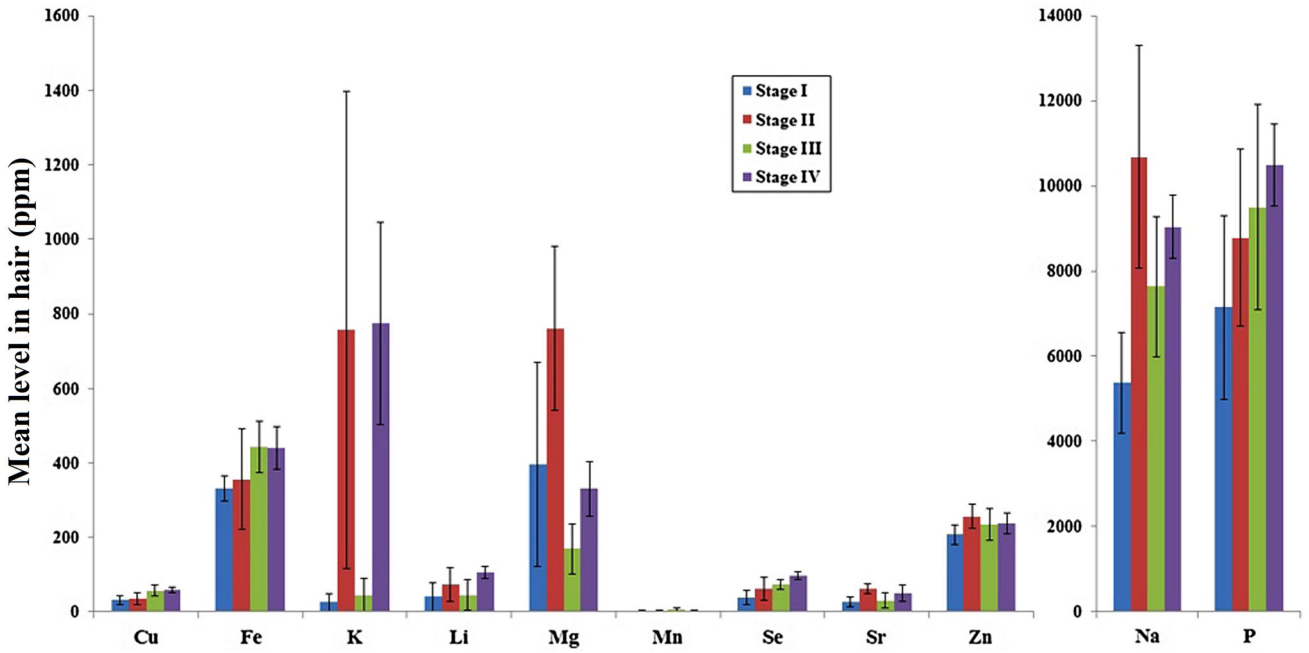

Fig. 1 Stage-based comparison of trace elements level in hair of the cancer patients and healthy donors. The average concentrations of Fe, Se and P significantly increased with increasing cancer stage in the hair sample of patients

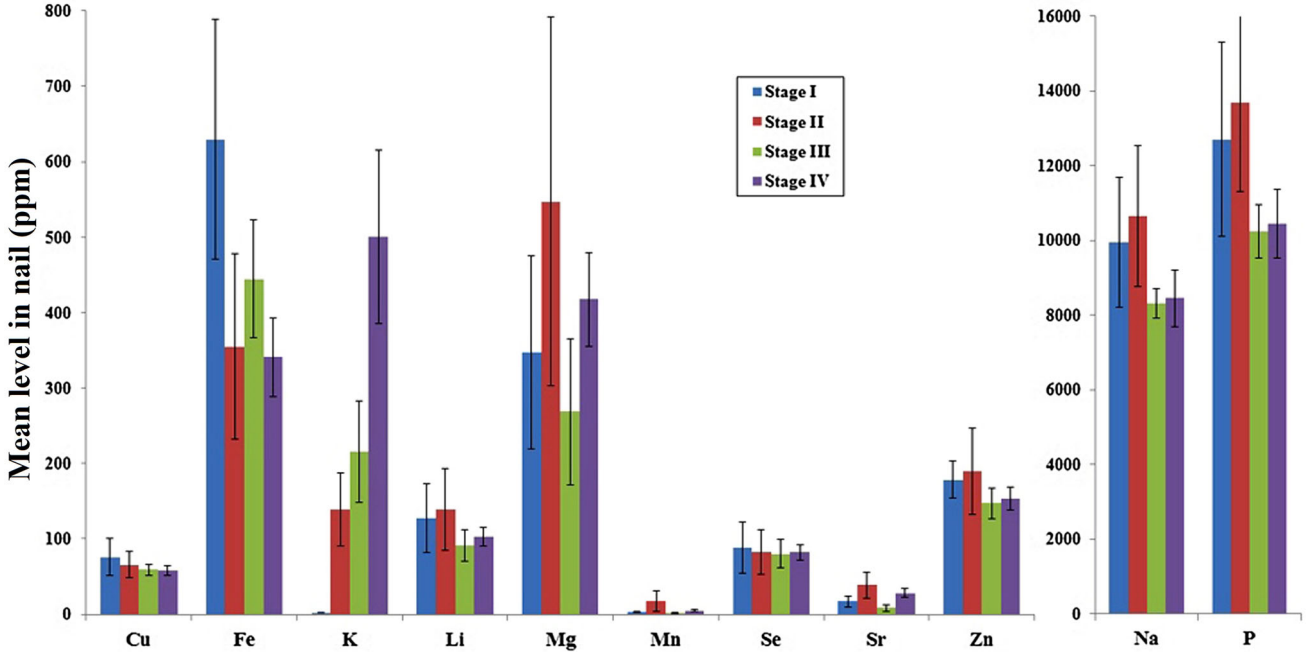

Fig. 2 Stage-based comparison of trace elements level in nail of the cancer patients and healthy donors. The average concentration of k also significantly increased with increasing cancer stage in the nail of patients

dietary element for human, and has been shown that certain forms of Se might be an efficient anticancer nutrient [27]. In Pasha et al. [7] study, the levels of some trace elements were investigated in the blood and hair of patients with gastrointestinal cancer. They showed that in hair samples, levels of Fe and Cu were significantly higher in the patients group than controls group. In blood sample, however Cu levels was higher in the patient group in comparison with the control group, but iron level was vice versa. Pasha et al. [7], also reported that in hair samples, Zn level was higher in patients group than control group and in blood sample was inversely. In this study, no significant difference was observed in Zn level between case and control. Magalhaes et al. [28], examined the levels of trace elements in cancer tissue and showed that levels of P and K were higher in all types of cancerous tissues than normal tissues. In addition, they revealed that level of Se was lower in colon cancer tissue than normal tissue, in contrast to breast cancer tissue. Pirincci et al. [29], showed that levels of Zn, Fe and Mn were lower in kidney cancer tissue than normal tissue. Gurusamy et al. [30], investigated liver tumor tissue and showed that the levels of Zn and Fe were lower in liver cancer tissue than healthy tissue. In our study, results showed that these elements were significantly higher in the nails and hair of the patients group than the controls group. In a prospective study, Steevens et al. [31], evaluated the level of trace elements in nail and investigated risk of various cancers. Their study showed that individuals with higher Se levels in nail were prone to esophageal squamous cell carcinoma, although risk of gastric cardia adenocarcinoma was low in them. Our findings showed that levels of this element were significantly higher in patients group than controls group. This result is in contrast to the results of the Steevens et al. [31], study which may be due to factors affecting stomach cancer in this region. Copper level was found to increase in hair and decrease in nail stage wise showing inverse relationship between the two samples which could be interesting in further management and prognosis of the stomach cancer. This could also be helpful in diagnosis of early and advanced stages of stomach cancer patients. Considering the increased concentrations of Fe, Se, P and decreased concentrations of K in accordance with the increase in the stage of cancer, and also the important role of some of these elements in oxidative stress, it is suggested that these elements may be helpful biomarkers for diagnosis and prognosis in cancers. Although its expansion requires more comprehensive researches.

Conclusion

The results of our study showed that there was an association between the increase in Cu, K, Li, P, Se and Fe, and stomach cancer development. Our results reveal that the increase in the trace elements could be a potential diagnostic marker to predict cancer progression and its etiology.

Acknowledgements The authors sincerely thank the Research and Technology Department in Mazandaran University of Medical Sciences.

Funding This study was funded by the Research and Technology Department in Mazandaran University of Medical Sciences.

Compliance with Ethical Standards

Conflict of interest All authors declare that they have no conflict of interest.

Ethical Approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed Consent Informed consent was obtained from all individual participants included in the study.

References

1. He K. Trace elements in nails as biomarkers in clinical research. Eur J Clin Invest. 2011;41:98–102.
2. Wolff MS, Weston A. Breast cancer risk and environmental exposures. Environ Health Perspect. 1997;105(Suppl 4):891–6.
3. Singh V, Garg AN. Trace element correlations in the blood of Indian women with breast cancer. Biol Trace Elem Res. 1998;64:237–45.
4. Trush MA, Kensler TW. An overview of the relationship between oxidative stress and chemical carcinogenesis. Free Radic Biol Med. 1991;10:201–9.
5. Navarro Silvera SA, Rohan TE. Trace elements and cancer risk: a review of the epidemiologic evidence. Cancer Causes Control. 2007;18:7–27.
6. Karadag F, Cildag O, Altinisik M, Kozaci LD, Kiter G, Altun C. Trace elements as a component of oxidative stress in COPD. Respirology. 2004;9:33–7.
7. Pasha Q, Malik SA, Shaheen N, Shah MH. Investigation of trace metals in the blood plasma and scalp hair of gastrointestinal cancer patients in comparison with controls. Clin Chim Acta. 2010;411:531–9.
8. Yaman M. Comprehensive comparison of trace metal concentrations in cancerous and non-cancerous human tissues. Curr Med Chem. 2006;13:2513–25.
9. Kuo HW, Chen SF, Wu CC, Chen DR, Lee JH. Serum and tissue trace elements in patients with breast cancer in Taiwan. Biol Trace Elem Res. 2002;89:1–11.
10. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2015. CA Cancer J Clin. 2015;65:5–29.
11. Freeman HJ. Risk of gastrointestinal malignancies and mechanisms of cancer development with obesity and its treatment. Best Pract Res Clin Gastroenterol. 2004;18:1167–75.
12. Janbabai G, Oladi Z, Farazmandfar T, Taghvaei T, Naghshvar F. The prognostic impact of EGFR, ErbB2 and MET gene amplification in human gastric carcinomas as measured by quantitative Real-Time PCR. Oncol: J. Cancer Res. Clin; 2015.
13. Xun P, Liu K, Morris JS, Daviglus ML, He K. Longitudinal association between toenail selenium levels and measures of subclinical atherosclerosis: the CARDIA trace element study. Atherosclerosis. 2010;210:662–7.
14. Xun P, Liu K, Morris JS, Daviglus ML, Stevens J, Jacobs DR, et al. Associations of toenail selenium levels with inflammatory biomarkers of fibrinogen, high-sensitivity c-reactive protein, and interleukin-6: the CARDIA trace element study. Am J Epidemiol. 2010;171:793–800.
15. Wang T, Fu J, Wang Y, Liao C, Tao Y, Jiang G. Use of scalp hair as indicator of human exposure to heavy metals in an electronic waste recycling area. Environ Pollut. 2009;157:2445–51.
16. Pereira R, Ribeiro R, Gonçalves F. Scalp hair analysis as a tool in assessing human exposure to heavy metals (S. Domingos mine, Portugal). Sci Total Environ. 2004;327:81–92.
17. Borella P, Bargellini A, Caselgrandi E, Piccinini L. Observations on the use of plasma, hair and tissue to evaluate trace element status in cancer. J Trace Elem Med Biol. 1997;11:162–5.
18. Schrauzer GN. Interactive effects of selenium and chromium on mammary tumor development and growth in MMTV-infected female mice and their relevance to human cancer. Biol Trace Elem Res. 2006;109:281–92.
19. Hayes RB. The carcinogenicity of metals in humans. Cancer Causes Control. 1997;8:371–85.
20. Reddy SB, Charles MJ, Raju GJN, Reddy BS, Reddy TS, Lakshmi PVBR, et al. Trace elemental analysis of cancer-afflicted intestine by PIXE technique. Biol Trace Elem Res. 2004;102:265–82.
21. Andrews NC. Disorders of iron metabolism. N Engl J Med. 1999;341:1986–95.
22. McCord JM. Effects of positive iron status at a cellular level. Nutr Rev. 1996;54:85–8.
23. Lieu PT, Heiskala M, Peterson PA, Yang Y. The roles of iron in health and disease. Mol Aspects Med. 2001;22:1–87.
24. Armendariz AD, Gonzalez M, Loguinov AV, Vulpe CD. Gene expression profiling in chronic copper overload reveals upregulation of Prnp and App. Physiol Genomics. 2004;20:45–54.
25. Brem S. Angiogenesis and cancer control: from concept to therapeutic trial. Cancer Control. 1999;6:436–58.
26. Hu GF. Copper stimulates proliferation of human endothelial cells under culture. J Cell Biochem. 1998;69:326–35.
27. Ji JH, Shin DG, Kwon Y, Cho DH, Lee KB, Park SS, et al. Clinical correlation between gastric cancer type and serum selenium and zinc levels. J Gastric Cancer. 2012;12:217–22.
28. Magalhães T, Carvalho ML, Von Bohlen A, Becker M. Study on trace elements behaviour in cancerous and healthy tissues of colon, breast and stomach: total reflection X-ray fluorescence applications. Spectrochim Acta Part B At Spectrosc. 2010;65:493–8.
29. Pirincci N, Gecit I, Gunes M, Kaba M, Tanik S, Yuksel MB, et al. Levels of serum trace elements in renal cell carcinoma cases. Asian Pac J Cancer Prev. 2013;14:499–502.
30. Gurusamy KS, Farquharson MJ, Craig C, Davidson BR. An evaluation study of trace element content in colorectal liver metastases and surrounding normal livers by X-ray fluorescence. Biometals. 2008;21:373–8.
31. Steevens J, van den Brandt PA, Goldbohm RA, Schouten LJ. Selenium status and the risk of esophageal and gastric cancer subtypes: the Netherlands cohort study. Gastroenterology. 2010;138:1704–13.