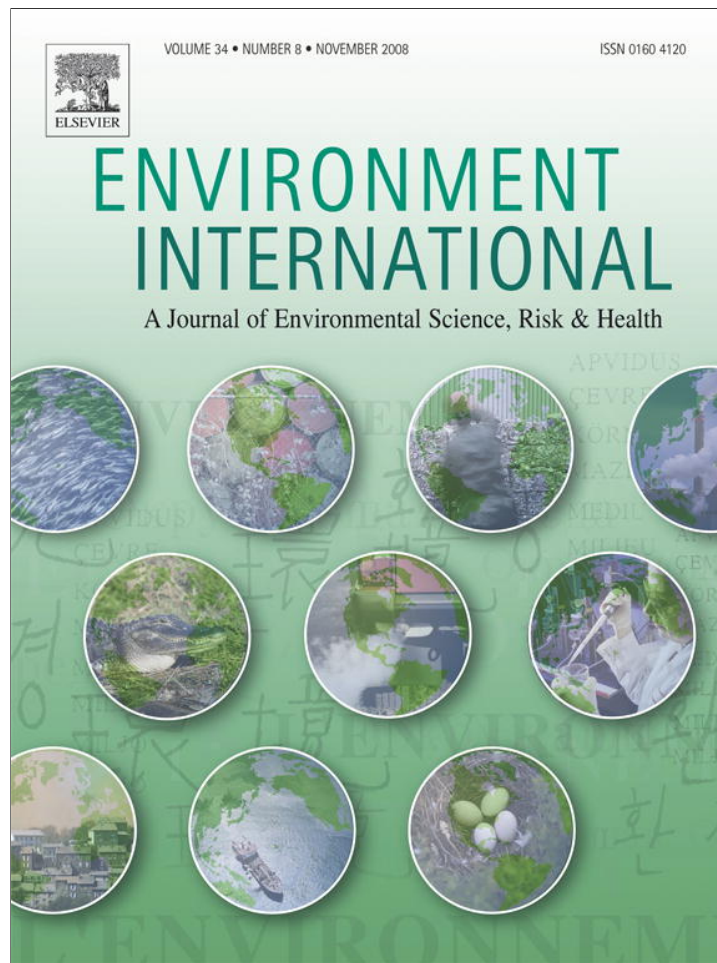


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Essential and non-essential trace metals in scalp hair of men chronically exposed to volcanogenic metals in the Azores, Portugal

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ABSTRACT

Volcanoes produce and release large amounts of toxicants, and the concentration of metals in hair can be used as a biomarker of exposure to trace metals. In order to investigate whether humans exposed to volcanic emissions demonstrate higher concentrations of essential and non-essential trace metals, scalp hair of men (3–89 years) living in two areas of the Azores, one exposed to volcanic emissions and the other not, was sampled, and their content in Cd, Cu, Pb, Rb, Se, and Zn was quantified by using High Resolution Inductively Coupled Plasma Mass Spectrometry. In average, men exposed to volcanic emissions presented higher concentrations of Cd (96.9 ppb), Cu (16.2 ppm), Pb (3417.6 ppb), Rb (216.3 ppb), and Zn (242.8 ppm), but not Se (469.6 ppb). Also, strong and significant correlations were found between Cd–Rb and Pb–Rb in the exposed men. In conclusion, humans living chronically exposed to volcanic emissions show high concentrations of essential and non-essential trace metals in scalp hair, and is suggested that this type of exposure may be as harmful as living close to industrial facilities.

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1. Introduction

Metals are ubiquitous elements, being present in the environment and in all living organisms. Some metals are essential to life as they are involved in multiple functions, such as enzymes structure and activity, transport proteins, hormonal function and specific receptor sites, being crucial for the well functioning of the biological systems, if maintained at required levels (Apostoli, 2002). For example, Zn that is the most abundant intracellular component is involved in genetic stability and gene expression as well as in DNA repair and programmed cell death (Dreosti, 2001). Deficiency in Zn has been associated with growth retardation and increasing cancer risk, but in excess it can be neurotoxic (Cai et al., 2005; Dreosti, 2001). Cu is a component of various enzymes, is involved in collagen synthesis and in the normal development of connective tissues, nerves and immune system. In excess, Cu can be extremely toxic due to its pro-oxidant activity, causing DNA damage, and it has been associated with neurodegenerative changes such as Alzheimer's disease (Cai et al., 2005; Llanos and Mercer, 2002). Se is present in various enzymes,

which are responsible for cell protection against oxidative damage, and is considered indispensable for normal growth and development of mammals (Ventura et al., 2005). Other metals are not considered essential and their presence, even at very low doses, may result in impairment of biological functions (Apostoli, 2002). For instance, Cd is carcinogenic and Pb neurotoxic, and even in small amounts both may interact with calcium and replace it in the skeleton or Zn in the heme enzymes and metallothioneins (Goyer, 1997; Shukla et al., 1998; Waalkes, 2003).

Rb is a rarely studied but abundant alkali metal that competes with K for enzymatic sites (Meltzer, 1991) and that has been found in high levels in malignant and benign thyroid nodules (Zaichick et al., 1995). Though safe levels are not known, toxic responses linked to high concentrations of Rb have been found in mammals with high-Rb low-K diets (Kosla et al., 2002).

The effects of environmental exposure to trace metals on human health have been discussed in the last decades, especially with respect to toxic metals originating in anthropogenic activities, but little has been said concerning natural pollution and especially volcanic activity. Volcanic emissions during eruptions and post-eruption periods may represent a significant risk to human health because of acute and/or chronic exposure to toxic gases and metals (Amaral et al., 2006a; Amaral and Rodrigues, 2007). Among others, Cd, Cu, Pb, Rb, and Zn are some of the metals emitted by volcanoes during and after the eruption (Delmelle and Stix, 2000; Durand et al., 2004).

Abbreviations: Cd, cadmium; Cu, copper; Pb, lead; ppb, parts per billion; ppm, parts per million; Rb, rubidium; Se, selenium; S.E., standard error; Zn, zinc.

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As metals are present in the air, soil, food and water, they enter the organism through skin, respiratory tract and digestive system, being the excess excreted through the sweat, hair, urine and faeces (Apostoli, 2002). Interest in the hair as a marker of basic levels of metals in the human body has recently increased due to some advantages over urine or blood, particularly because it is a metabolic end product that has a recognized ability to reflect the body metal burden. Moreover, the elemental composition of the hair reflects long-term exposure, while blood or urine represents a current, or acute, situation (Chojnacka et al., 2005). The concentration of a certain metal in hair reveals a balanced mineral content of the individual body pool over a long period of time, which can only be significantly modified by exposure or intake of high quantities of trace metals. Thus, the quantification of trace elements in hair has been used as a biomarker enabling the assessment of both long-term environmental exposure to trace metals and metabolic status concerning chronic nutritional deficiencies (Hopps, 1977; Khalique et al., 2005; Rivai, 2001).

The objective of this study was to compare the concentrations of Cd, Cu, Pb, Rb, Se, and Zn in scalp hair of men from two rural areas in the Azores distinguishable by the occurrence of volcanic activity. In one area (Furnas) volcanic activity is present as hydrothermal waters, degassing soils and fumaroles, while in the other area (St. Maria) there is no volcanic activity since 3 million years ago.

2. Materials and methods

2.1. Study population

The Azores is an archipelago of nine volcanic island located in the North Atlantic Ocean. Presenting fumarolic fields and hydrothermal vents (Booth et al., 1978; Cruz, 2003), and the always present “rotten egg” odor, the rural village of Furnas, in S. Miguel Island, is built upon actively degassing ground inside a volcanic caldera (Baxter et al., 1999), where the last eruption occurred at 1630 AD (Cole et al., 1995). This village has about 1541 inhabitants (Serviço Regional de Estatística dos Açores, 2003), where the age groups 0–14, 15–24, 25–44, 45–64, and over 64 represent 22%, 16%, 27%, 19%, and 16% of the local population, respectively. Santa Maria, which is one of the smallest islands (92 km²), has about 5578 inhabitants (Serviço Regional de Estatística dos Açores, 2003), and is also rural but, in contrast, has had no volcanic activity since 3 million years ago (Feraud et al., 1984). In Santa Maria, the age groups 0–14, 15–24, 25–44, 45–64, and over 64 represent 24%, 16%, 30%, 18%, and 12% of the local population, respectively. Both populations are similar in access to medical care, socioeconomic status, economy, and home construction, but dissimilar in terms of smoking, where Furnas and Santa Maria differ since they present 25% and 44% prevalence, respectively (Peixoto, 2005).

2.2. Hair sampling and analysis

Samples of scalp hair of 158 men aged 3–89 were obtained from Furnas ($n=68$) and Santa Maria ($n=90$), but only from those who did not have dyed or treated hair. Previous to sample collection, all participants or legal representatives signed an informed consent authorizing it. Also, tobacco smoking habit of each donor was recorded. Those who smoked one or more cigarettes per day were considered smokers.

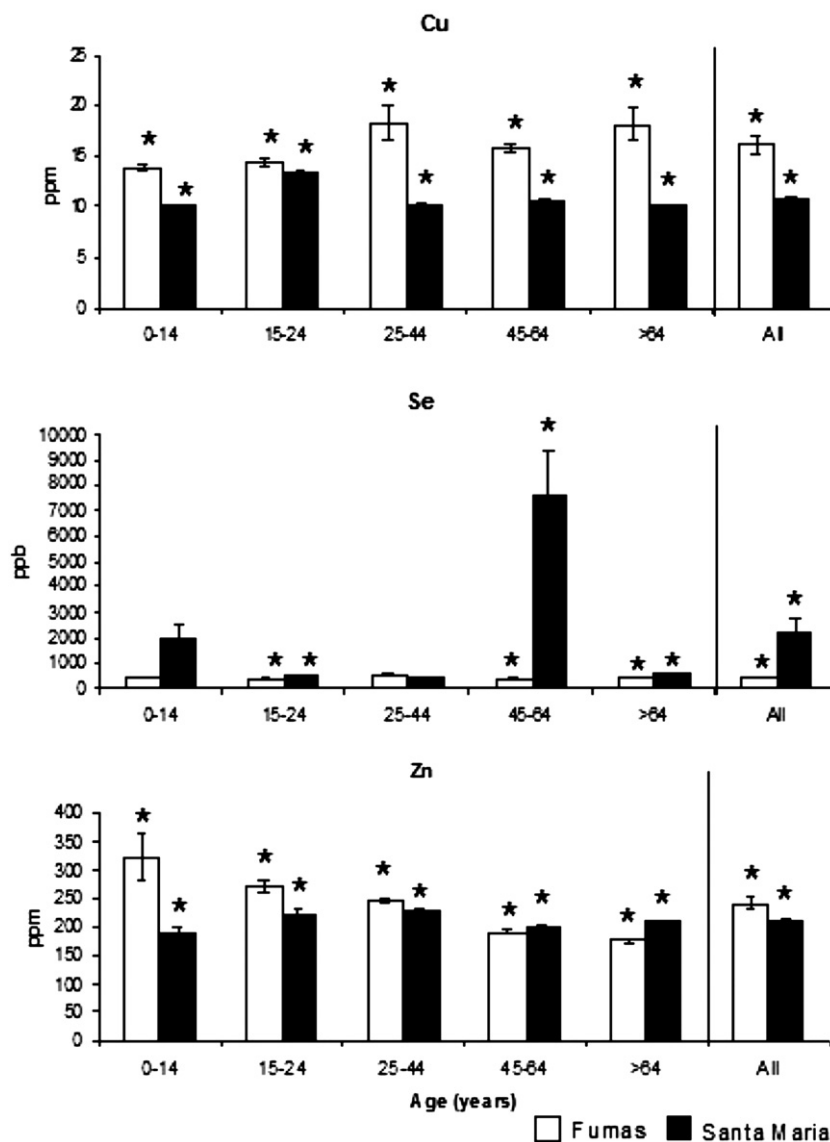


Fig. 1. Mean concentrations (\pm S.E.) of copper (Cu), selenium (Se) and zinc (Zn) in hair samples of men from Furnas and Santa Maria. Asterisks indicate significant differences ($p < 0.05$) between sites for each age group.

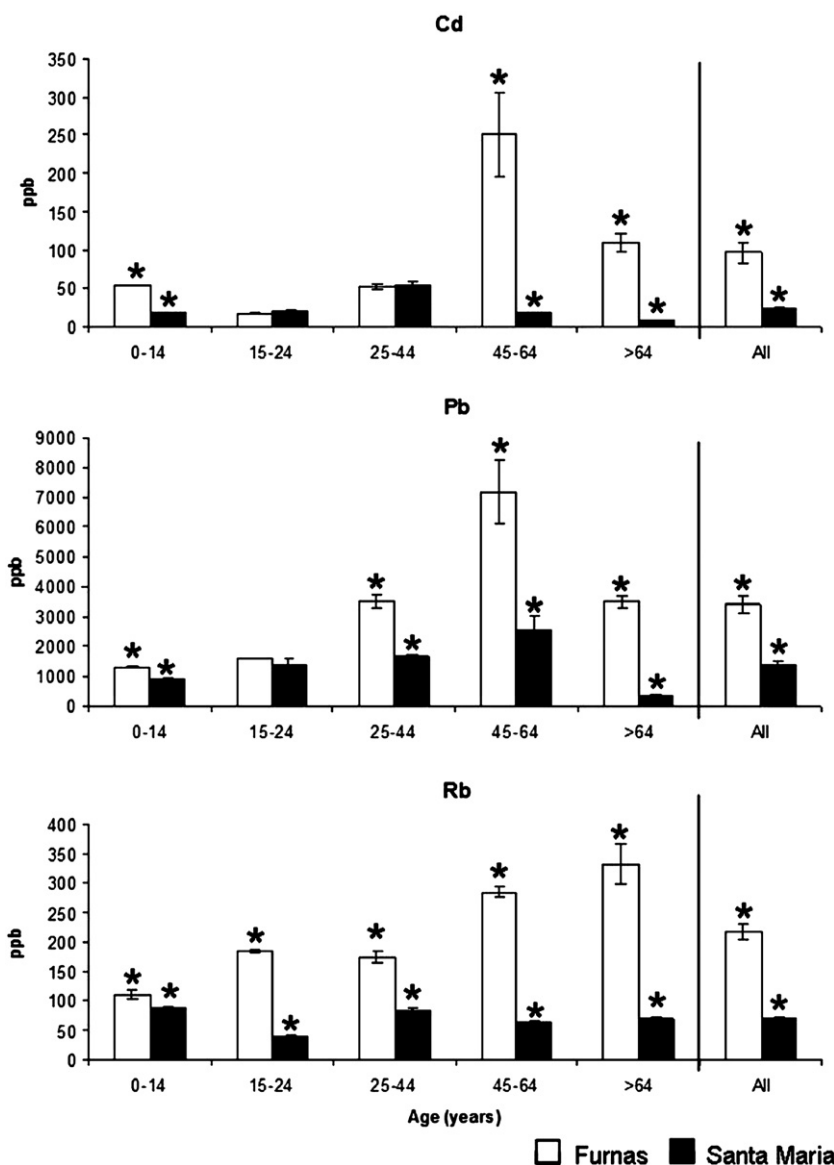


Fig. 2. Mean concentrations (\pm S.E.) of cadmium (Cd), lead (Pb), and rubidium (Rb) in scalp hair of men from Furnas and St. Maria. Asterisks over the bars indicate significant differences at $p \leq 0.05$.

In order to eliminate external contamination, each hair sample was washed in a sequence of acetone, bi-distilled water, and acetone (Ryabukkin, 1978). After washing, each sample was air-dried at room temperature in a dust-free area. Then they were digested in aqua regia at 95 °C, for 2 h, and with microwave inside a closed Teflon vessel, for 45 min and 1 h plus. Prior to analysis samples were always kept away from metallic materials and dust to avoid contamination. For analysis of Cd, Cu, Pb, Rb, Se, and Zn, the resultant sample solutions were diluted and analyzed on a Finnegan Mat Element 2 High Resolution ICP/MS. For quality control, internal standards and reference materials were run together with the samples. At least, six different reference materials covering all the elements in the study were used. Duplicate samples were also used in order to determine precision of the analysis. For each element, a minimum of three standards were used to cover the analytical working range of the instrument. Ultrapure water was used to prepare calibration standards and blanks, and three replicate determinations were performed for each sample.

2.3. Statistical analysis

Differences in metal concentrations in hair between groups were examined using the Mann–Whitney *U* test and considered significant when $p \leq 0.05$. Also, the Spearman's correlations between all analyzed metals found in hair were assessed. To perform this statistical analysis the SPSS 14.0 (SPSS Inc.) was used.

3. Results

In general, hair of men from Furnas (with volcanic activity) presented significantly high concentrations of all trace metals, with the exception of Se (Figs. 1 and 2), when compared to that of men from Santa Maria (without such activity).

While the concentrations of Cu and Zn were stable along the different age groups in Santa Maria, in Furnas they showed different behaviors with Cu increasing and Zn decreasing with increasing age, respectively (Fig. 1). The concentrations of Cd, and especially Pb and Rb in hair of men from Furnas showed a pattern of increase along with age (Fig. 2). On the other hand, Se concentrations were higher in hair of men from Santa Maria, when compared to that of men from Furnas (Fig. 1).

Concerning the six trace metals, in neither sites we found significant differences between smokers and non-smokers, except for the group of 45–64 years. Among smokers, we found that men from Furnas presented significantly (Mann–Whitney *U*, $p < 0.05$) high concentrations of all trace metals when compared to those from Santa Maria. Among non-smokers, we did not find a pattern transversal to all age groups, but we found that children (0–14 years) from Furnas presented higher (Mann–Whitney *U*, $p < 0.05$) concentrations of all trace metals, except Se, than those from Santa Maria.

Table 1
Spearman's correlations coefficients between all six trace metals

	Cd	Cu	Pb	Rb	Se	Zn
Cd	–					
Cu	0.357**	–				
Pb	0.698**	0.571**	–			
Rb	0.648**	0.558**	0.570**	–		
Se	–0.188*	–0.295**	–0.049	–0.323**	–	
Zn	–0.069	0.286**	–0.040	0.016	–0.081	–

**Correlation is significant at $p \leq 0.01$. *Correlation is significant at $p \leq 0.05$.

In average, we found strong and significant correlations between Rb and Cd, Cu, and Pb, as well as between Pb and Cd, and Cu (Table 1).

4. Discussion

In general, concentrations of Cd, Cu, Pb, Rb, and Zn were higher in the scalp hair of men from Furnas. According to age, the highest levels of these metals were found in the oldest groups, which may indicate a possible bioaccumulation effect through aging. Cd and Pb levels in men of both areas were lower than those from developing countries (Khalique et al., 2005; Rivai, 2001; Sharma and Kumar, 2005) and the surrounding area of an abandoned mine in Portugal mainland (Pereira et al., 2004), but in contrast the concentrations in men from Furnas were higher than those of developed countries, such as Germany, Italy or Canada (Goullé et al., 2005; Seifert et al., 2000; Violante et al., 2000). These differences between countries may be due to different types of industries and also to the late banning of leaded gasoline in the developing countries. Men from Furnas present Cd and Pb concentrations in-between developing and developed countries most likely because of the volcanic input, which sets this area to a moderately polluted area, and less likely because of smoking habits.

The concentrations of Rb in men from Furnas were not only higher than the levels of men from St. Maria, but also extremely higher than levels found in Canadian men (Goullé et al., 2005). Although Rb is not considered extremely toxic it can exert toxic effects when in high concentrations, mainly because it competes with K for enzymes, such as the Na⁺/K⁺-ATPase (Meltzer, 1991; Mitchell et al., 1921). Accumulation of Rb has been observed in several organs of rats, and reported effects of this accumulation include spasms and tissue disorganization (Mitchell et al., 1921; Salaün and Truchet, 1996). Strong and significant correlations found in donors from Furnas between Rb–Cd and Rb–Pb show that Rb hair concentrations may be used as biomarker of exposure to volcanic emissions, composed of more harmful gases and aerosols, as previously suggested by Durand et al. (2004).

In the case of Cu, the concentrations found were almost always below the reference levels, being particularly low in scalp hair of men from Santa Maria (Fig. 1).

Concentrations of Zn were always above the maximum reference level, being higher in men from Furnas, particularly in scalp hair of children where it reaches a maximum of 321 ppm (Fig. 1).

Hair of men from Furnas showed lower concentrations of Se than that of men from Santa Maria, but nevertheless men from the latter presented concentrations below reference levels, with the exception of age groups 0–14 and 45–64 (Bleise et al., 2000; Goullé et al., 2005). Values observed in these two age groups may be due to high concentrations of Se in very few individuals, probably as a consequence of dietary supplements or any chemical medication used as cancer chemopreventive agent, not assessed in this study, which may explain the high standard errors. The low Se concentrations detected in men from Furnas were in accordance to data referred by Pavão et al. (2003) that, comparing a group of people from this site with a group of people from a fishing community, concluded blood Se levels were lower in the individuals living in Furnas.

According to the present results, it seems that the concentrations found in the hair of men from Furnas and from Santa Maria were more influenced by the exposure to volcanic emissions than to tobacco smoking. Although it is known that smokers usually tend to present higher levels of trace metals, such as Cd, than non-smokers, the lack of significant differences between them, in most age groups, in hair metal concentrations strengthens the former hypothesis.

Previous studies in Furnas showed that: (i) soil invertebrates and mice present high levels of trace metals, such as Cd and Zn (Amaral et al., 2006b, 2007); and (ii) humans present high incidence of chronic bronchitis and some cancer types, which may be related to volcanic emissions (Amaral et al., 2006a; Amaral and Rodrigues, 2007). Since the cancer chemopreventive role of Se has been well documented

(Brinkman et al., 2006; Kellen et al., 2006), it is reasonable to speculate as to a possible association between the low concentrations of this element, the high concentrations of Cd, which is carcinogen, and Pb, which is a probable carcinogen, and some of the cancers found previously. Although further studies should be performed to better understand the possible associations between chronic exposure to volcanogenic pollutants and health outputs, our present and previous results suggest that living chronically exposed to volcanic emissions may be as harmful as living close to industrial facilities.

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