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POTENTIAL TOXIC EFFECTS TRIGGERED BY RADIATION EXPOSURE AMONG MEDICAL RADIOGRAPHERS THROUGH AN IMBALANCE IN TRACE ELEMENTS AND REDOX STATUS

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ABSTRACT

Prolonged exposure to radiation may be associated with high incidence of health hazards. The purpose of this study was to evaluate the potential toxic effects of radiation on the level of trace elements and structure of hair among radiographers. The participants were divided into three equal groups (20 per group) (control group, male medical radiographers with X-ray exposure group, and radiographers exposed to radiation from multiple sources. The levels of urinary 8-hydroxy-2'-deoxyguanosine (8-OHdG), conjugated dienes (CD), thiobarbituric acid reactive substances (TBARS), superoxide dismutase (SOD), and reduced glutathione (GSH) were determined. Electron microscopy was employed for examination of hair samples. The radiographers exposed to X-ray and multiple radiation sources exhibited considerably significant elevation in the levels of studied trace elements in the blood, imbalance in their systemic redox status which was reflected by a significant increase in the levels of nitrite, CD, and TBARS and a significant decrease in SOD activity and GSH. Electron microscopy examination of hair samples revealed that the lamellae underwent cuticle disorganisation deterioration and the whole cell was vacuolated. Prolonged exposure to radiation induces accumulation of toxic concentration of trace elements in blood and alteration in hair structure.

KEY WORDS : Male, Radiographers, Trace element, DNA, Radiation.

INTRODUCTION

Over the past thirty years, there have been fast-paced advances in medical imaging technologies like X-ray, computed tomography (CT) and magnetic resonance imaging (MRI), the use of which has become common practice in hospitals. The key benefit of such technologies is that they afford structural detail of the human body, which improves disease diagnosis, examination of internal body tissue, monitoring and therapeutic interventions. Consequently, patient care quality and safety depend greatly on this technologies (Songurand Top, 2016).

All medical imaging modalities are accompanied by the risk of radiographers being exposed to radiation, but this risk has a great deal of variation, according to the traits of individual radiographers. Radiation exposure can make it more likely for radiographers to develop cancer or increase the levels of trace elements in the blood, ultimately resulting in DNA damage (Mattsson, 2016; lukoff *et al*., 2017).

There are various mechanisms of DNA change; one mechanism involves reactive species, causing DNA strand breakage and DNA protein cross-linking. Recent research suggested that DNA change may be indirectly caused by radiation, through aggregation of trace elements and metals (Jena *et al*., 2012; Ravanat and Douki, 2016; Sage and

Shikazono, 2017).

Trace elements occur in small levels all through the body, contributing to body functions. According to the needs of the body, trace elements are divided into essential and non-essential elements (Tchounwou *et al.*, 2012). Taking up 0.02% of body weight, essential elements are involved in body functions such as metabolism, hemopoiesis, reproduction and immunity. Occurrence of trace elements in amounts that are too low or too high can result in metabolic disorders and cellular growth disruption, including development of mutations and cancer (Al-Fartusie *et al.*, 2017).

A correlation between trace elements and cancer development was suggested by Eken *et al.*, (2016) based on the observation that prostatic cancer patients had higher levels of trace elements than healthy individuals (Ekan *et al.*, 2016). Furthermore, the authors argued that oxidative stress may result from high levels of trace elements like copper (Cu), zinc (Zn), cobalt (Co), manganese (Mn) and molybdenum (Mo), while the incidence and progress of malignant tumours might be associated with the essential trace elements selenium (Se), iron (Fe) and manganese (Mn).

According to research conducted by the National Institute of Health (NIH), toxicity and carcinogenicity develop with the major involvement of production of reactive oxygen species (ROS) and oxidative stress. However, owing to the singular properties of every element and physical-chemical qualities, the manner in which that involvement unfolds is still uncertain. Several trace elements, such as cadmium (Cd), chromium (Cr), lead (Pb) and mercury (Hg – the metal with the highest toxicity), are of particular importance for public health due to their extreme toxicity (Tchounwou *et al.*, 2012).

The reason why trace elements are toxic is that they can interfere with protein activity and alter the expression pattern of several genes, thus disrupting cellular events and heightening the risk of disease occurrence (Al Bakheet *et al*., 2013).

There are several ways in which trace elements can interact with DNA. Nucleic acids contain large amounts of firmly bound metal ions. The binding of some metals may alter DNA polymerase activity, increasing the incorporation of genetic errors. Trace elements hinder the activity of reparative proteins, thus changing the outcome of the repair process of double-strand breaks (DSBs), whilst also affecting how mutated DNA is distributed, thus potentially enhancing the risk of cancer development DNA damage can occur as a result of exposure to trace elements via ROS production (Al Bakheet *et al*., 2013; Morales *et al.*, 2016).

Li *et al.*, (2018) reported that exposure to Pb altered the motility and morphology of sperm and reduced its quality, whilst also inducing modifications in DNA structure and integrity, leading to infertility (Li *et al.*, (2018). Moreover, Fazli *et al.*, 2014 indicated that Pb contributed to neurobiological defects as well, like schizophrenia and Alzheimer's disease incidence (Fazli *et al.*, 2014).

Meanwhile, Hg exposure was found to cause renal damage by changing the expression of the HO-1 gene and heightening hydrogen peroxide levels (Al Bakheet *et al*., 2013). In combination with other metals (e.g. Cd and Pb), Hg may become even more toxic. Investigating Cd exposure, Boonprasert *et al.*, (2018) reported that, owing to the impact on the cytochrome P450 enzyme (CYP) with involvement in blood regulation, exposure to this element in low levels over the long term could lead to heightened risk of hypertension (Boonprasert *et al.*, 2018). Moreover, Nordberg *et al.*, (2016); found that Cd toxicity contributed to the development of several conditions, including osteoporosis, osteomalacia, anaemia, diabetes, cardiovascular disease and cancer (Nordberg *et al.*, 2016).

Exposure to radiation also has an impact on human body tissues and cells, particularly haemotological cells (Pizent *et al*., 2012). Taqi *et al.*, (2018) provided evidence that haematological profile was impacted to a significant extent by long-term X-ray exposure (Taqi *et al.*, 2018). The general assumption is that the effects of ionising radiation on human blood cells contribute to a haematopoietic syndrome that has been revealed to occur in both humans and animals following total body irradiation (Billings *et al*., 2014).

Human exposure to radiation has an impact on hair, and in particular the scalp, with long-term exposure even leading to alterations in scalp morphology. Therefore, hair is currently employed as a biological indicator to determine dose distribution in the body in the case of an accident (Oreby *et al*., 2012). Radiation affects hair growth due to the extreme radio sensitivity of hair follicles. Thus, hair follicle analysis is an accurate means of establishing the radiation dose that radiographers can be safely exposed to (Man *et al.*, 1998). Radiation damages hair bulb cells, leading to a reduction in

hair diameter, which becomes noticeable in 1-2 weeks, when the hair protrudes through the skin (Schlak, 2007).

The aim of this study was to evaluate of potential toxic effects of radiation on the level of trace elements and structure of hair among Saudi medical radiographers.

MATERIALS AND METHODS

Study population

The research participants consisted on 60 male individuals in the age range 26-40 years old who worked at governmental and private hospitals in Buraydah, Saudi Arabia. No participant had exposure to trace elements or had used chronic medication for six months prior to the research. The participants were equally divided into three groups (20 participants per each group) ; the first group represented the control group and comprised healthy participants without radiation exposure, the second group included medical radiographers with X-ray exposure, and the third group comprised radiographers exposed to radiation from multiple sources (MRI, CT scan, nuclear medicine, X-ray).

Participants were included in the study if they were of male sex of 26-40 years of age, worked as medical radiographers and had been exposed to radiation for one year or more. On the other hand, participants were excluded from the study if they were of female sex, had exposure to metals, or smoked cigarettes. Whereas, the inclusion criteria of control group include male sex of 26-40 years of age, no exposure to any type of radiation, did not exposed to metals, non-cigarettes smoker and were age-& life style(diet, smoking, residences, hygiene)-matched to the radiation exposed groups.

Ethical consideration and consent form

The rules of the General Directorate of Al-qassim Health Affairs Planning and Training Administration were followed to formulate the consent form and the approval of the Qassim Region Research Ethics Committee (QREC) was obtained prior to commencing research. The consent form informed the participants regarding the research objectives, significance and risks. The participants were included in the study only after they signed the consent form. Furthermore, the participants were required to complete a data collection tool regarding their personal details (e.g. age, gender, length of employment in the radiology department, and types of radiation sources they were exposed to) and health status (e.g. liver, kidney, heart or diabetic disease, and prior metal exposure).

Sample collection

Following overnight fasting, around 10 mL of blood was extracted from the antecubital vein of each participant before being divided into two equal aliquots introduced in a vacutainer tube with no anticoagulant to obtain the serum and in a plasma vial with heparin to obtain the plasma.

Determination of conjugated dienes (CD) level

Levels of conjugated dienes in serum were assayed according to Recknagel and Glende method. The absorbance was measured at 234 nm (Recknagel and Glende, 1984).

Determination of thiobarbituric acid reacting substance (TBARS) level

Levels of TBARS in serum were determined according to Satoh method. The absorbance was determined at a wavelength of 530 nm (Satoh, 1978).

Enzymatic antioxidant assay – SOD

Estimation of plasma SOD was performed using the method reported by Kakkar *et al.,* (1984). The absorbance was recorded at 560 nm (Kakkar *et al.,* 1984).

Nonenzymatic antioxidant assay – reduced GSH

The concentration of GSH was determined using a fluoro-metric method described by Hissin and Hilf. The, fluorescence detection was performed with excitation and emission at 350 and 418 nm, respectively (Hissin and Hilf, 1976).

Determination of blood level of trace elements

The approach proposed by Bazzi *et al.*(2008) was adopted for the measurement of the trace elements via Inductive Coupled Plasma-Mass Spectrometry (ICP-MS; Agilent 4500, Agilent Technologies, CA)(Bazzi *et al.,* 2008). The blood samples were mixed with 2 mL nitric oxide and 0.01% TritonX-100 for three hours at 25 °C, followed by dilution of the mixtures with Millii-Q water to obtain a volume of 10 mL. Ionisation of the generated mixture was achieved via ICP-MS, while the mass charge was taken into account in ion extraction. Expression of the Cu and Pb concentrations in the blood took the

form of mg/dl and mcg/L, respectively (Bazzi *et al*., 2008).

Determination of urinary 8-OHdG concentration

The key result of the research was the urinary 8-OHdG concentration. The DNA Damage EIA kit (Enzo Life Sciences, Switzerland) was employed in keeping with the manufacturer's instructions to assay that concentration. This involved loading every sample in duplicate and interpolating the sample mean absorbances with the absorbances derived from a standard curve. Expression of data took the form of ng/mL.

Determination of haematological profile

The fully automatic blood cell counter Model PCE 210 N (ERMA INC, Japan) was used to measure the haematological parameters (i.e. white blood cells, lymphocytes, haematocrit, haemoglobin, and red blood cells) after the blood samples were collected.

Hair sampling preparation

Scalp hair nape was obtained from every participant. To make sure that they were not contaminated with cosmetics, the samples were washed and stored at room temperature. The hair was cut into pieces of about 0.3 cm and was mixed thoroughly (George *et al*., 2010).

Histopathological examination

The histological structure was examined with an electron transmission microscope (JEOL electron microscope at 80 kV at EM unit). Hair sample fixation was achieved with 2.5% glutaraldehyde in 0.1 M phosphate buffer, while after fixation, phosphate buffer (1% osmium tetroxide) was employed for two hours. The samples were subsequently washed and introduced in ethanol. Afterwards, the samples were cut again into ultrathin, 50-60 nm pieces. Uranyl acetate and lead citrate were used for staining the samples twice before electron microscopy examination (Bozzola *et al.*, 1999).

Statistical analysis

Expression of results took the form of mean ± SD of three replicates. Parametric data were analyzed by ANOVA followedby Tukey-Kramer post-hoc test were conducted to compare the research groups. A p value of 0.05 or lower indicated significance and every statistical analysis was undertaken in SPSS (Version 21). Correlations between different parameters were evaluated by Pearson correlation (r) for parametric data and Ranked Spearman correlation (r) for non-parametric data. Multiple linear regression analyses were applied to study the association between demographic characteristics, oxidative stress, and DNA fragmentation.

RESULTS

In the present study, serum CD levels and TBARS, which are markers for oxidative stress, were found to be significantly increased in the radiographers exposed to X-ray and multiple sources compared with the control group. However, the levels of plasma SOD, a marker of enzy-matic antioxidant activity, were found to be significantly decreased in the radiographers exposed to X-ray and multiple sources compared with the control. In addition, the levels of serum GSH, a marker of non-enzymatic antioxidant activity, were found to be significantly decreased in the radiographers exposed to X-ray and multiple sources compared with the control group. Unlike control, the groups with exposure to radiation from X-ray and from multiple sources exhibited higher levels of Ca, Cl, Na, Fe, Cu, Pb and Cd in the blood. Also, the groups with X-ray and multiple-source exposure had higher levels of white and red blood cells, haematocrit, and lymphocytes, but lower levels of hemoglobin. By contrast to control, the groups with exposure to radiation from X-ray and multiple sources exhibited considerably higher urinary 8-OHdG, which is an indicator of oxidative DNA damage (Table 1).

In the group with X-ray exposure, a significant correlation existed between urinary 8-OHdG concentration and Pb levels, while in the group with multiple-source exposure; 8-OHdG was closely correlated with levels of Fe, Pb and Cd as presented in Table 2.

Participants' age was positively correlated with the levels of white blood cells, haematocrit, the serum levels of Ca, Cu and Cd. Whereas length of exposure was significantly negatively correlated with Cl level in serum as shown in Table 3.

The levels of Ca, Na, Cl, Fe, Pb and Cd were significantly positively correlated with the haematological parameters of white blood cells, haematocrit and lymphocytes. The red blood cell count was significantly positively correlated solely with Ca level in serum. By contrast, haemoglobin level in serum was significantly negatively correlated with all trace elements (Table 4).

Table 1. Blood biomarkers of oxidative stress, levels of trace elements, urinary 8-OHdG concentration and hematological parameters of participants

Items	Control	X-ray	Mixed
Blood biomarkers of oxidative stress			
TBARs (nmol/mL)	7.5±0.03	9.8±1.4*	8.7±1.8*
SOD (U/mL)	4.2±0.9	2.7±0.2*	2.9±0.3*
GSH (µmol/mL)	2.3±0.3	1.9±0.2*	1.7±0.1*
CD (nmol/mL)	1.8±0.1	2.7±0.3*	2.6±0.1*
Blood trace elements			
Calcium (mmol/L)	7.1±0.2	9.4±0.3*	9.6±0.4*
Chloride (mmol/L)	84.3±3.5	104.9±1.4*	104.7±3*
Sodium (mmol/L)	112.3±2.7	143.9±3.3*	140.8±3.4*
Iron (ug/dL)	64.7±2	70.7±7.2*	93.3±11.4*
Copper (mcg/dL)	58.8±1.1	85.7±8.8*	86.9±3.8*
Lead (mcg/dL)	11.2±0.2	21.1±6.7*	22.5±1.4*
Cadmium (µg/dl)	0.4±0.04	0.7±0.06*	0.7±0.06*
Oxidative DNA damage			
8-OHdG (ng/mL)	59.6±1.26	78.9±*1.66	79.9±*1.20
Haematological parameters			
WBCs (10^3/µL)	5.2±0.3	6.1±0.5*	7.8±0.6*
RBCs (10^6/µL)	4.4±0.2	4.8±0.5*	4.9±0.6*
Hematocrit test (%)	35.4±2.5	43.2±3.1*	42.2±2.3*
Hemoglobin (g/dl)	15.7±0.8	11.5±0.4*	11.4±0.4*
Lymphocyte (10^3/µL)	1.4±0.3	2.7±0.6*	2.3±0.3*

Expression of data takes the form of mean ± SD; each group comprised twenty participants; *significant by comparison with control at p≤0.05

Thiobarbituric acid reactive substances (TBARS); Superoxide dismutase (*SOD);* Glutathione (GSH) and *conjugated diene (CD);*white blood cells (WBCs); red blood cells (RBCs);

Table 2. Pearson correlation coefficient for 8-OHdG and levels of trace elements in the groups with X-ray and multiple-source exposure

Items	x-ray	Mixed
Fe	0.298	0.900**
Cu	0.624	0.437
Pb	0.851**	0.887**
Cd	0.327	0.915**

**Correlation has significance at p<0.01

*Correlation has significance at p<0.05

Data in Table 5 and 6 showed the multiple linear regression analysis between TBARs, SOD, GSH, CD, Calcium, Chloride, Sodium, Iron, Copper, lead, Cadmium and 8-OHdGas dependent variables and the other demographic characteristics, oxidative stress, and DNA fragmentation as independent variables. All biochemical parameters were significant with each other whereas there was no significant change between oxidative stresses, 8-OHdG with level of chloride and sodium respectively. Also, there was no significant change between demographic characteristics with level of sodium.

As shown in (Figure 1A), when ultrathin sections of hair follicles derived from the participants in the control group were examined, it was observed that the innermost hair layer cells made up the keratinised medulla, which was enveloped by two keratinised layers, namely, the cortex and the cuticle. The cuticle lamellae exhibited focal disorganisation in the participants with radiation exposure from X-ray, while the cytoplasm of certain IRS and ORS cells displayed vacuolar spaces (Figure 1B). In some cases, sizable cytoplasmic vacuoles were exhibited by ORS cells, mainly in the proximity of nuclei, which was the reason for their indentation (Figure 1C). In the case of participants exposed to radiation from multiple sources, there was significant multifocal alteration, with irregular indentation, compression, or chromatin content of numerous nuclei. Furthermore, cells displayed multiple cytoplasmic vacuoles and extensive cytoplasmic inhomogeneous masses (Figure 1D).

DISCUSSION

Increased generation of free radicals and decreased

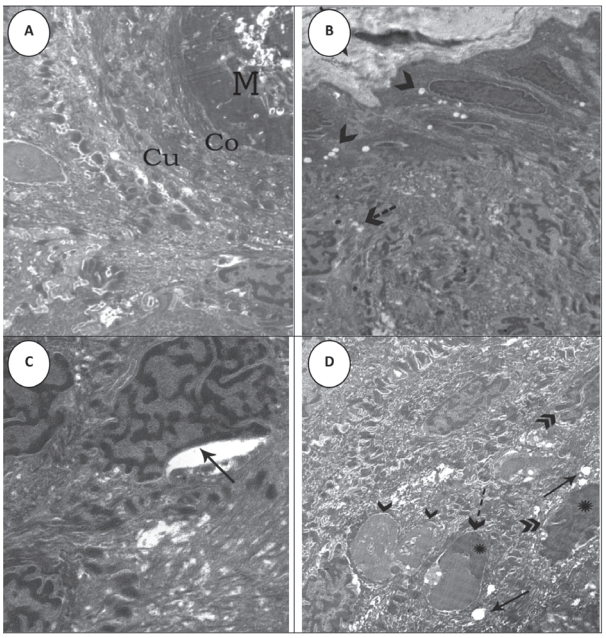

Fig. 1A. An electron micrograph of a human scalp hair root from a control males showing the innermost layer of the hair constituting the medulla (M) which is surrounded by two layers, the cortex (Co) and the cuticle (Cu); **Fig. 1B:** An electron micrograph of a human scalp hair root from a male radiographer (X rays) showing disorganization of the cuticle lamellae (→) as well as vacuolar spaces in the cytoplasm of some cells of IRS (>) and ORS (- ->); **Fig. 1C:** An electron micrograph of a human scalp hair root from a male radiographer (mixed) showing cells of ORS with large cytoplasmic vacuolation causing indentation of the nucleus (→); **Fig. 1D.** Multiple cytoplasmic vacuoles in cells of ORS (!), large cytoplasmic heterogenous masses (>) as well as irregularly indented (>>) and compressed nuclei (- ->) and condensed chromatin (*) Multiple cytoplasmic vacuoles in cells of ORS (→, large cytoplasmic heterogenous masses (>) as well as irregularly indented (>>) and compressed nuclei (- ->) and condensed chromatin (*) (mixed).

antioxidant activity has been proposed to play a major role in the development of age-related disease (Alissa and Ferns, 2011). In the present study, serum CD levels and TBARS, which are markers for oxidative stress, were found to be significantly increased in the radiographers exposed to X-ray and multiple sources compared with the control group. However, the levels of plasma SOD, a marker of enzy-matic antioxidant activity, were found to be significantly decreased in the radiographers exposed to X-ray and multiple sources compared

Table 3. Correlation between trace element levels, haematological profile, age and duration of exposure

Items	Ca (mg/dl)	Cl (mmol/L)	Na (mmol/L)	Fe (ug/dl)	Cu (mcg/dl)	Pb (mcg/L)	Cd (µg/dl)	WBC (10^3/µL)	RBC (10^6/µL)	HCT (%)	Hb (g/dl)	Lymphocyte (10^3/µL)
Age in years	0.516**	0.193	0.271	0.285	0.499*	0.223	0.455*	0.410*	0.167	0.363*	-0.314	0.279
Duration of exposure in radiation field	0.087	-0.482**	-0.307	0.054	-0.299	-0.157	-0.054	0.083	-0.134	-0.031	0.163	-0.341

Spearman correlation (r) was used to determine the correlation coefficient
Ca-calcium, Cl-chloride, Na-sodium, Fe-iron, Cu-copper, Pd-lead, Cd-cadmium.
WBC–white blood cells, RBC–red blood cells, Hct–Haematocrit, Hb–Haemoglobin
*p<0.05; **p<0.01

with the control. In addition, the levels of serum GSH, a marker of nonen-zymatic antioxidant activity, were found to be significantly decreased in the radiographers exposed to X-ray and multiple sources compared with the control group. It can be proposed that a successful alteration in an adaptive phenomenon against oxidative stress caused an elevation in the levels of plasma SOD in the aged persons. This could have resulted from either genetic or environmental factors, or both (Menke *et al*., 2016).

Medical radiographers are regularly exposed to radiation and therefore have a high risk of toxicity. Evaluation of potential toxic effects of radiation among Saudi medical radiographers through measurement of some trace elements levels, haematological parameter, hair histopathological examination. A range of biological processes are dependent on trace elements. However, if their levels are too high or too low, these elements can trigger metabolic disorders and cellular growth disruption, including development of mutations and cancer (Al-Fartusie *et al.*, 2017; Sage and Shikazono, 2017). The levels of metals in the body can be increased by various risk factors, including smoking and liver, kidney, heart and diabetic disease (Alissa *et al.*, 2011; Menek *et al*., 2016; El-samad *et al.*, 2017).

According to Barbosa *et al.*, (2005) the rise of trace element levels in the blood was a biomarker for protracted exposure and indicated the burden on the body (Barbosa *et al.*, 2005). Meanwhile, Keil *et al.* 2015 argued that exposure to toxic elements (e.g. Pb) closely adhering to intracellular proteins could be

Table 4. Correlation between levels of trace element levels and haematological profile.

	WBC (10^3/μL)	RBC (10^6/μL)	HCT (%)	Hb (g/dl)	Lymphocyte (10^3/μL)
Ca (mg/dl)	0.702 **	0.410*	0.731**	-0.759**	0.592**
Cl (mmol/L)	0.621**	0.228	0.679**	-0.678**	0.708**
Na (mmol/L)	0.492**	0.341	0.725**	-0.626**	0.757**
Fe (ug/dl)	0.685**	0.320	0.396*	-0.564**	0.422*
Cu (mcg/dl)	0.596**	0.221	0.564**	-0.638**	0.794**
Pb (mcg/L)	0.676**	0.292	0.543**	-0.574**	0.486**
Cd (μg/dl)	0.499**	0.125	0.622**	-0.602**	0.676**

Spearman correlation (r) was used to determine the correlation coefficient
WBC-white blood cells; RBC-red blood cells; Hct-haematocrit; Hb-haemoglobin
*p<0.05; **p<0.01

Table 5. Multiple linear regression analysis between different investigated parameters in mixed group

Independent /Dependent	Age (Years)	Duration of service in radiation field (Years)	TBARs (nmol/mL)	SOD (U/mL)	GSH (μmol/mL)	CD (nmol/mL)	8-OHdG (ng/mL)
TBARs (nmol/mL)	0.565[b]	0.761[a]	-	0.649[a]	0.587[b]	0.627[a]	0.615[a]
SOD (U/mL)	0.686[a]	0.882[a]	0.682[a]	-	0.631[a]	0.548[b]	0.886[a]
GSH (μmol/mL)	0.805[a]	0.725[a]	0.705[a]	0.632[a]	-	0.622[a]	0.825[a]
CD (nmol/mL)	0.749[a]	0.619[a]	0.601[a]	0.645[a]	0.644a	-	0.703[a]
Calcium (mmol/L)	0.643[a]	0.633[a]	0.533[b]	0.630[a]	0.507[b]	0.603[a]	0.621[a]
Chloride (mmol/L)	0.711[a]	0.691[a]	0.322	0.302	0.320	0.245	0.310
Sodium (mmol/L)	0.211	0.321	0.126	0.235	0.339	0.325	0.317
Iron (ug/dL)	0.764[a]	0.684[a]	0.530[b]	0.567[b]	0.557[b]	0.630[a]	0.804[a]
Copper (mcg/dL)	0.698[a]	0.708[a]	0.508[b]	0.634[a]	0.631[a]	0.668[a]	0.548[b]
Lead (mcg/dL)	0.712[a]	0.682[a]	0.521[b]	0.722[a]	0.641[a]	0.641[a]	0.672[a]
Cadmium (μg/dL)	0.699[a]	0.639[a]	0.751[a]	0.547[b]	0.743[a]	0.797[a]	0.829[a]
8-OHdG (ng/mL)	0.754[a]	0.654[a]	0.758[a]	0.711[a]	0.798[a]	0.984[a]	-

Results are expressed as standardized coefficients (b).
[a]Correlation is significant at the 0.01 level (P 0.01).
[b]Correlation is significant at the 0.05 level (P 0.05).

most effectively identified through blood analysis (Keil *et al*., 2015). The finding of this study that individuals exposed to radiation from X-ray and multiple sources had levels of Ca, Cl, Na, Fe, Cu, Pb and Cd that were respectively 27.85%, 21.77%, 24.66%, 8.86%, 37.23%, twofold, and 54.54% higher than in individuals without exposure is consistent with the results reported by Oreby *et al.,* (2012), who found that the levels of trace elements in the body were increased by radiation exposure (Oreby *et al*., 2012). Furthermore, in the case of the two exposure groups in the present study, there was a statistical increase in the levels of Ca, Na, Cu, Pb and Cd in proportion with participants' age. Similarly, a positive correlation was found between service length and increase in the levels of Ca, Cl, Na, Cu, Pb and Cd.

The increase in Cd and Pb levels due to radiation exposure has been demonstrated to lead to ROS production, causing DNA damage in human spermatozoa (Adams *et al*., 2014; Agarwal *et al*., 2014). Furthermore, exposure to Cd in a work context has a negative impact on lung function or can cause emphysema (Moitra *et al.,* 2013; Oh *et al.,* 2014).There is also research evidence that the male reproductive system is adversely impacted by high blood concentration of Pb, with decrease in sperm quality, sperm morphological alterations and DNA fragmentation (Wu *et al*., 2012; Li *et al*., 2018). Moreover, it has been suggested that high levels of Cu and Fe in blood could enhance the risk of Alzheimer's disease (Wang *et al*., 2015; Padovani *et al.,* 2017).

Urinary 8-OHdG is considered a reliable biomarker for determining the risk of cancer and degenerative disease development, for determining the impact of endogenous and exogenous oxidative damage on DNA, and as a factor of carcinogenesis onset and promotion (Valavanidis *et al*., 2009). Furthermore, Flora *et al*., 2008 suggested that urinary 8-OHdG could help to determine the DNA damage caused by carcinogenic agents like tobacco smoke, asbestos fibres, trace elements, and polycyclic aromatic hydrocarbons (Flora *et al.,* 2008). In the present study, urinary 8-OHdG concentration was measured to determine the DNA damage associated with oxidative stress engendered by trace elements, owing to the capability of those pollutants to produce ROS (Valavanidis *et al*., 2009). In keeping with the results obtained by Bakheet *et al.,* 2013, it was determined that exposure not only to Cd, but also to Fe and Pb, was closely correlated with urinary 8-OHdG concentration (Al Bakheet *et al.,* 2013). This finding was consistent with the results reported by Flora *et al.,* 2008 regarding the fact that the lack of balance between pro-oxidant and antioxidant homeostasis, known as oxidative stress, was the cause of the toxic effect of Cd (Flora *et al.,* 2008). Meanwhile, Pizzino *et al.* 2014 revealed that the gene expression profile was adversely affected by trace elements (Pizzino *et al.,* 2014).

Unlike the control, the groups with exposure to radiation from X-ray and multiple sources had significantly higher counts of white blood cells, haematocrit and lymphocytes, but significantly lower levels of haemoglobin. These results corroborated the findings of earlier research, which indicated a direct correlation between increase in the levels of trace elements in the blood and reduction in haemoglobin levels (Al Bakheet *et al.,*

Table 6. Multiple linear regression analysis between different investigated parameters in x-ray group

Independent /Dependent	Age (Years)	Duration of service in radiation field (Years)	TBARs (nmol/mL)	SOD (U/mL)	GSH (µmol/mL)	CD (nmol/mL)	8-OHdG (ng/mL)
TBARs (nmol/mL)	0.685[b]	0.761[a]	-	0.611[a]	0.617[a]	0.614[a]	0.606[a]
SOD (U/mL)	0.706[a]	0.882[a]	0.682[a]	-	0.641[a]	0.599[b]	0.801[a]
GSH (µmol/mL)	0.711[a]	0.725[a]	0.705[a]	0.630[a]	-	0.608[a]	0.832[a]
CD (nmol/mL)	0.688[a]	0.549[b]	0.541[b]	0.660[a]	0.631a	-	0.711[a]
Calcium (mmol/L)	0.643[a]	0.734[a]	0.343	0.638[a]	0.509[b]	0.611[a]	0.619[a]
Chloride (mmol/L)	0.741[a]	0.541[b]	0.262	0.252	0.229	0.211	0.219
Sodium (mmol/L)	0.321	0.252	0.246	0.139	0.319	0.229	0.237
Iron (ug/dL)	0.698[a]	0.544[b]	0.610[a]	0.537[b]	0.525[b]	0.622[a]	0.794[a]
Copper (mcg/dL)	0.711[a]	0.609[a]	0.322	0.655[a]	0.611[a]	0.640[a]	0.522[b]
Lead (mcg/dL)	0.742[a]	0.722[a]	0.321	0.642[a]	0.621[a]	0.605[a]	0.542[b]
Cadmium (µg/dL)	0.649[a]	0.599[a]	0.691[a]	0.612[b]	0.723[a]	0.703[a]	0.739[a]

2013; Korashy *et al.*, 2017; Taqi *et al.*, 2018). Haematological parameter alterations serve as early biomarkers for radiation-related toxicity. In this study, exposure groups had 31.9% higher white blood cell levels than control, with a number of different implications for immunity (Velickova, 2017). Furthermore, in the case of participants with X-ray exposure, the levels of white blood cells and lymphocytes increased in direct proportion with age and service length (Nureddin *et al*., 2016). Also reported this, observing that X-ray technicians with 10-20 years' service duration exhibited higher levels of white blood cells than control.

The increase in participants' age was found to be positively correlated with the levels of Ca, Na, Cu, Pb and Cd, while the longer participants were in service, the higher the levels of Ca, Cl, Na, Cu, Pb and Cd. This was consistent with Lin *et al.*, 2018, who observed that accumulation of Cd was directly proportional with age advancement (Lin *et al.*, 2018). Furthermore, the increase in participants' age was also positively correlated with the counts of white blood cells and haemoglobin, while the longer participants were in service, the higher the counts of white blood cells, haematocrit, haemoglobin and lymphocytes.

This study found that the levels of the trace elements Ca, Na, Cl, Fe, Pb and Cd were positively correlated with the haematological parameters white blood cells, haematocrit and lymphocytes. Earlier research also pointed out the existence of a correlation between heightened Pb and Cd levels and increased counts of white blood cells, haematocrit and lymphocytes (Al Bakheet *et al.*, 2013; Korashy *et al.*, 2017). Furthermore, the count of red blood cells and Ca level in serum were significantly positively correlated, which is consistent with Hertz *et al.* 2017 who reported the same for individuals with anaemia (Hertz *et al.*, 2017). On the other hand, both groups with radiation exposure exhibited a significant negative correlation between haemoglobin levels in serum and all trace elements. Likewise, earlier studies indicated that, as the levels of Cd and Pb increased, the level of haemoglobin decreased (Al Bakheet *et al*., 2013; Korashy *et al*., 2017).

By causing certain metals to bind to the protein structure (Oreby *et al.*, 2012), radiation exposure may lead to disruption in communication between cells, enzymatic functions, the integrity of chromosomes and lipid membranes, which in turn have implications for haematopoiesis, tissue respiration and cell division.

An additional finding of this study was that radiation exposure engendered different modifications in scalp hair roots, such as disorganisation of cuticle lamellae, cytoplasmic vacuolation of cells of both the inner root sheath (IRS) and outer root sheath (ORS), and nuclear indentation and fragmentation in certain ORS cells. All these are indicative of degeneration in cuticle layer, IRS and ORS. Oreby *et al.*, 2012 reported similar findings, distinguishing structural damage to radiographers' hair follicles through EM scanning of identified dispersed areas of cellular exfoliation and erosion of the cuticle, suggesting that low-dosage radiation was damaging (Oreby *et al*., 2012). Likewise, Freites-Martinez *et al.*, 2018 observed that numerous cellular alterations in hair follicles were exhibited by individuals subjected to palliative radiotherapy or exposed to acute radiation dosage (Freites-Martinez *et al*., 2018).

Dicelio *et al.* (2006) suggested that fragmentation of the hair protein fractions (keratin) was the reason for the hair structural damage caused by radiation (Dicelio *et al*., 2006). Additional cell layers are subsequently affected as they are permeated by the deteriorated protein fractions with low molecular weight migrating from the cuticle cells (Ruetsch *et al*., 2000; Ruetsch *et al.*, 2003). The findings of the present study provide strong evidence that radiographers are at a high risk of certain diseases due to long-term radiation exposure, which has a different impact on haematological parameters, levels of trace elements, and histopathological modifications in scalp hair. However, further study is required to explore in greater depth how long-term exposure to radiation is correlated with the levels of trace elements in the human body. The findings could inform health protection and prophylactic measures that medical radiographers should adopt, such as protective gear and shielding, use of the beam-limiting device, and suitable beam filtration. Furthermore, fundamental rules of distance, time and shielding should be complied with to diminish radiation exposure in a work context (Sherer *et al.*, 2017). Moreover, for purposes of disease prevention or detection, radiographers with protracted radiation exposure should have their blood levels of trace elements and haematological parameters checked on a regular basis.

CONCLUSION

Radiographers are highly likely to experience toxic effects because exposure to radiation causes trace elements to accumulate in dangerously high levels in the blood and hair. This study found that there was a direct correlation between radiographers' age and exposure length and DNA damage, haematological alterations and modifications in the ultrathin structure of hair follicles.

REFERENCES

Adams, J.A., Galloway, T.S. and Mondal, D. 2014. Effect of mobile telephones on sperm quality: a systematic review and meta-analysis. *Environment International*. 70 : 106-112.

Agarwal, A., Virk, G., Ong, C. and du Plessis, S.S. 2014. Effect of oxidative stress on male reproduction. *The World Journal of Men's Health*. 32 : 1-17.

Al Bakheet, S.A., Attafi, I. and Maayah, Z. 2013. Effect of long-term human exposure to environmental heavy metals on the expression of detoxification and DNA repair genes. *Environmental Pollution*. 181 : 226-232.

Al-Fartusie, F.S., Hafudh, A. and Mustafa, 2017. Levels of some trace elements in sera of patients with lung cancer and in smokers. *Indian Journal of Advances in Chemical Science*. 5 : 344-352.

Alissa, E. and Ferns, G. 2011. Heavy metal poisoning and cardiovascular disease. *Journal of Toxicology*. 2011: 1-21.

Barbosa Jr, F., Tanus-Santos, J. E. and Gerlach, R.F. 2005. Critical review of biomarkers used for monitoring human exposure to lead: advantages, limitations, and future needs. *Environmental Health Perspectives*. 113 (12) : 1669-1974.

Bazzi, A., Nriagu, J. and Linder, A. 2008. Determination of toxic and essential elements in children's blood with inductively coupled plasma-mass spectrometry. *Journal of Environmental Monitoring*. 10 : 1226-1232.

Billings, P., Romero-Weaver, A. and Kennedy, A. 2014. Effect of gender on the radiation sensitivity of murine blood cells. Gravitational and space research: publication of the *American Society for Gravitational and Space Research*. 2 : 25-31.

Boonprasert, K., Vesey, D. and Gobe, G. 2018. Is renal tubular cadmium toxicity clinically relevant?. *Clinical Kidney Journal*. 11 : 681-687.

Bozzola, J. and Russell, L. 1999. *Electron Microscopy: Principles and Techniques for Biologists*. 2nd ed. Sudbury, MA, USA: Jones & Bartlett Learning.

Dicelio, L. E. and Joekes, I. 2006. About photo-damage of human hair. *Photochemical & Photobiological Sciences*. 5 : 165-169.

Eken, A., Kaya, E., Nlu Endirlik, B. 2016. Evaluation of trace element levels in patients with prostate cancer, benign prostatic hyperplasia and chronic prostatitis. *Gulhane Medical Journal*. 58: 27.

El-Samad, M.A. and Hanafi, H.A. 2017. Analysis of toxic heavy metals in cigarettes by Instrumental Neutron Activation Analysis. *Journal of Taibah University for Science*. 11 : 822-829.

Fazli, D., Malekirad, A. and Mirzaee, M. 2014. Study on the link between lead exposure and hematological, psychological, and memorial parameters in automobile repair workers. *Health*. 6 : 712-719.

Flora, S.J., Mittal, M. and Mehta, A. 2008. Heavy metal induced oxidative stress and its possible reversal by chelation therapy. *Indian Journal of Medical Research*. 128 : 501-523.

Freites-Martinez, A., Shapiro, J. and van den Hurk, C. 2018. CME Part 2: Hair disorders in cancer survivors Persistent chemotherapy-induced alopecia, persistent radiotherapy-induced alopecia, and hair growth disorders related to endocrine therapy or cancer surgery. *Journal of the American Academy of Dermatology*. 120 : 401-410.

George, G., Singh, S., Myers, G. 2010. The chemical forms of mercury in human hair: a study using X-ray absorption spectroscopy. *JBIC Journal of Biological Inorganic Chemistry*. 15: 709-715.

Hertz, L., Huisjes, R., Llaudet-Planas, E. 2017. Is increased intracellular calcium in red blood cells a common component in the molecular mechanism causing anemia?. *Frontiers in Physiology*. 8 : 673.

Hissin, P. and Hilf, R. 1976. A fluorometric method for determination of oxidized and reduced glutathione in tissues. *Anal Biochem*. 74 : 214-226.

Jena, N.R. 2012. DNA damage by reactive species: mechanisms, mutation and repair. *Journal of Biosciences*. 37 : 503-517.

Kakkar, P., Das, B. and Viswanathan, P. 1984. A modified spectrophotometric assay of superoxide dismutase. *Indian J Biochem Biophys*. 21 : 130-132.

Keil, D. E., Berger-Ritchie, J. and McMillin, G.A. 2015. Testing for toxic elements: a focus on arsenic, cadmium, lead, and mercury. *Laboratory Medicine*. 42 : 735-742.

Korashy, H.M., Attafi, I.M., Famulski, K.S. 2017. Gene expression profiling to identify the toxicities and potentially relevant human disease outcomes associated with environmental heavy metal exposure. *Environmental Pollution*. 221 : 64-74.

Li, C., Zhao, K. and Zhang, H. 2018. Lead exposure reduces sperm quality and DNA integrity in mice. *Environmental Toxicology*. 33 : 594-602.

Lin, X., Peng, L. and Xu, X. 2018. Connecting gastrointestinal cancer risk to cadmium and lead exposure in the Chaoshan population of Southeast China. *Environmental Science and Pollution Research*. 1-9.

Lukoff, J. and Olmos, J. 2017. Minimizing medical radiation exposure by incorporating a new radiation "vital sign" into the electronic medical record: quality of care and patient safety. *The Permanente Journal.* 21 : 17-007.

Man, A., Zheng, Y. and Mak, P. 1998. Structural and trace element changes in scalp hair of radiographers. *Biological Trace Element Research.* 63 : 11-18.

Mattsson, S. 2016. Need for individual cancer risk estimates in x-ray and nuclear medicine imaging. *Radiation Protection Dosimetry.* 169 : 11-16.

Menke, A., Guallar, E. and Cowie, C. 2016. Metals in Urine and Diabetes in United States Adults. *Diabetes.* 65 : 164-171.

Moitra, S., Blanc, P. and Sahu, S. 2013. Adverse respiratory effects associated with cadmium exposure in small-scale jewellery workshops in India. *Thorax.* 68 : 565-570.

Morales, M., Derbes, R., Ade, C. 2016. Heavy metal exposure influences double strand break DNA repair outcomes. *PLOS ONE.* 11 : e0151367.

Nordberg, G., Bernard, A. and Diamond, G. 2018. Risk assessment of effects of cadmium on human health (IUPAC Technical Report). *Pure and Applied Chemistry.* 90 : 755-808.

Nureddin, A. S. and Alatta, N. O. 2016. Effects of Long-term exposure low X-ray on The blood Consists of Radiology Department staff of health centers in Libya. *International Journal of Information Research and Review.* 37 : 1-60.

Oh, C., Oh, I. and Lee J. 2014. Blood cadmium levels are associated with a decline in lung function in males. *Environ Res.* 132 : 119-125.

Oreby, M., El-Maddah, E. and Mousa, A. 2012. Evalution of radiation-induced alterationin macro and micro trace elements and ultrastructural changes in hair of Egyptian radiographers. *The Egyptian Journal of Forensic Sciences and Applied Toxicology.* 12 : 43-61.

Padovani, A., Rozzini, L. and Lanfranchi, F. 2017. Free copper levels and cerebrovascular damage in alzheimer's disease. *Alzheimer's & Dementia.* 13 : 361.

Pizent, A., Tariba, B. and •ivkovie, T. 2012. Reproductive toxicity of metals in men. *Archives of Industrial Hygiene and Toxicology.* 63 : 35-46.

Pizzino, G., Bitto, A. and Interdonato, M. 2014. Oxidative stress and DNA repair and detoxification gene expression in adolescents exposed to heavy metals living in the Milazzo-Valle del Mela area (Sicily, Italy). *Redox Biol.* 2 : 686-693.

Ravanat, J. and Douki, T. 2016. UV and ionizing radiations induced DNA damage, differences and similarities. *Radiation Physics and Chemistry.* 128: 92-102.

Recknagel, R. and Glende, E. 1984. Spectrophotometric detection of lipid conjugated dienes. *Meth Enzym.* 105 : 331-337.

Ruetsch, S., Kamath, Y. and Weigmann, H. 2000. Photodegradation of human hair: An SEM study. Journal of the Society of Cosmetic Chemists 51.

Ruetsch, S., Kamath, Y. and Yang, B. 2003. Chemical and photo-oxidative hair damage studied by dye diffusion and electrophoresis. *Journal of the Society of Cosmetic Chemists.* 54 : 379-94.

Sage, E. and Shikazono, N. 2017. Radiation-induced clustered DNA lesions: Repair and mutagenesis. *Free Radical Biology and Medicine.* 107 : 125-135.

Satoh, K. 1978. Serum lipid peroxide in cerebrovascular disorders determined by a new colorimetric method. *Clin Chim Acta.* 90 : 37-43.

Schlake, T. 2007. Determination of hair structure and shape. *Seminars in Cell & Developmental Biology.* 18: 267-273.

Sherer, M. A. S., Visconti, P. J. and Ritenour, E. R. 2017. Management of imaging personnel radiation dose during diagnostic X-ray procedure. *Radiation Protection in Medical Radiography-E-Book.* Elsevier Health Sciences 273-299.

Sonður, C. and Top, M. 2016. Regional clustering of medical imaging technologies. *Computers in Human Behavior.* 61 : 333-343.

Taqi, A., Faraj, K. and Zaynal, S. 2018. Effects of occupational exposure of X-Ray on hematological parameters of diagnostic technicians. *Radiation Physics and Chemistry.* 147 : 45-52.

Tchounwou, P., Yedjou, C. and Patlolla, A. 2012. Heavy Metals Toxicity and the Environment. *EXS.* 101 : 133-164.

Valavanidis, A., Vlachogianni, T. and Fiotakis, C. 2009. 8-hydroxy-22 -deoxyguanosine (8-OHdG): A critical biomarker of oxidative stress and sarcinogenesis. *Journal of Environmental Science and Health.* Part C 27:120-139.

Velickova, Nevenka 2017. Environmental impact of heavy metals on the blood cells in professionally exposed workers. *Journal of Environmental Protection and Ecology.* 18 : 363-374.

Wang, Z. X., Tan, L. and Wang, H. F. 2015. Serum iron, zinc, and copper levels in patients with Alzheimer's disease: a replication study and meta-analyses. *Journal of Alzheimer's Disease.* 47 : 565-581.

Wu, H.M., Lin-Tan, D.T. and Wang, M.L. 2012. Lead level in seminal plasma may affect semen quality for men without occupational exposure to lead. *Reproductive Biology and Endocrinology.* 10 : 91.þ