



## ASSESSMENT OF ANTIOXIDANT VITAMIN STATUS IN NIGERIANS OCCUPATIONALLY AND ENVIRONMENTALLY EXPOSED TO ELECTRONIC WASTE IN SOUTHWESTERN NIGERIA

Igharo, O. G. and Idomeh, F. A

Department of Medical Laboratory Sciences, School of Basic Medical Sciences, College of Medical Sciences, University of Benin, Benin City, Nigeria

Corresponding Author: [osaretin.igharo@uniben.edu](mailto:osaretin.igharo@uniben.edu); Phone number: +2348038664896

### ABSTRACT

**Background:** The menace of unregulated dumping of electronic waste (e-waste) has been a major public health concern in both developing and developed countries. In Nigeria where e-waste reprocessing is substantial, the art has remained unregulated. The possible contribution of optimum antioxidant vitamins in mitigating the effects of occupational metal exposure in Nigerian e-waste workers has received little attention, hence this study.

**Aim:** The present study was aimed at investigating antioxidant vitamin Status in e-waste workers and environmentally exposed participants in Lagos, Benin and Ibadan cities in Southwestern Nigeria

**Methods:** Six hundred and thirty-two consenting participants from three major cities in Nigeria were recruited for this study. They included e-waste workers (EW) (381), environmental e-waste exposed (EEEP) individuals (120) and age- matched apparently healthy and unexposed individuals (131). Vitamins A, C and E were determined in serum using spectrophotometry. Data were analyzed using ANOVA at  $\alpha_{0.05}$ .

**Results:** Vitamins A and C in EEEP ( $2.20 \pm 0.04 \mu\text{mol/L}$ ;  $116 \pm 2.03 \mu\text{mol/L}$ ) were significantly higher than EW ( $2.08 \pm 0.04 \mu\text{mol/L}$ ;  $88.75 \pm 3.33 \mu\text{mol/L}$ ) and control ( $1.97 \pm 0.09 \mu\text{mol/L}$ ;  $92.19 \pm 1.68 \mu\text{mol/L}$ ), respectively while vitamin E in EW ( $8.91 \pm 0.19 \mu\text{mol/L}$ ) and EEEP ( $8.05 \pm 0.18 \mu\text{mol/L}$ ) were significantly lower than control ( $12.36 \pm 0.34 \mu\text{mol/L}$ ). Compared by location, levels of antioxidant vitamins in the three participants groups varied significantly from location to location.

**Conclusion:** Nigerians exposed to e-waste through occupational and environmental means may experience decreased levels of vitamin E. Antioxidant supplementation may be recommended with the aim of mitigating the risk of antioxidant vitamins depression in jobs with suspected mixed metal exposure like crude e-waste recycling.

**Keywords:** Antioxidant vitamins, e-waste, Metals, Occupational exposure.

### INTRODUCTION

Electronic waste (e-waste) generation and unregulated disposal have become a critical global environmental health issue. This has been partly attributed to its massive production volume and insufficient management policy in many countries (Ogunseitan *et al.*, 2009). E-waste products contain a variety of plastics and chemicals, which when not properly handled can be harmful to humans and pose environmental hazard (Leung *et al.*, 2006). There are over 1000 different chemical substances including toxic elements like lead, arsenic, cadmium, mercury, selenium and hexavalent

chromium and flame retardants in a given electronic waste, depending on its size and function (Leung *et al.*, 2006)

Owing to the growing influx of e-waste to Nigeria and an estimated 75% of the 400,000 computer monitors or 175,000 TV sets entering Lagos, Nigeria each month alone, and the accompanied inadequate recycling, parts salvaging, poor distribution and disposal of discarded or obsolete electronic devices creates new set of environmental and public health challenges in Nigeria (Nnorom and Osibanjo, 2007; Terada, 2012)

Exposure to metals in the course of crude e-waste reprocessing has been documented (Igharo *et al.*, 2014) and high body burden of toxic metals has been associated with high oxidative stress and low antioxidant defenses (Igharo *et al.*, 2016). There are not much investigation to look into the possible association of optimum antioxidant vitamin levels in mitigating the effects of occupational metal exposure in Nigerian e-waste worker.

Oxidative stress is a phenomenon caused by an imbalance between production and accumulation of oxygen reactive species (ROS) in cells and tissues and the ability of a biological system to detoxify these reactive products (Pizzino *et al.*, 2017). Reactive oxygen species are involved in several physiological roles example of which is cell signaling, and they are normally generated as by-products of oxygen metabolism (Pizzino, *et al.*, 2017). Additionally, environmental stressors such as ultra-violet radiation, ionizing radiations, pollutants, and heavy metals contribute to greatly increase ROS production, therefore causing the imbalance that leads to cell and tissue damage (oxidative stress) (Pizzino *et al.*, 2017). Several antioxidants have been exploited in recent years for their actual or supposed beneficial effect against oxidative stress, such as vitamin E, flavonoids, and polyphenols (Cook-Mills, 2013; Rajendran *et al.*, 2014; Pizzino *et al.*, 2017).

The central and local stress limiting systems, including the antioxidant defense system involved in defending the organism at the cellular and systemic levels from excess activation response to stress influence, leading to damaging effects (Kodentsova, *et al.*, 2013). The development of stress, regardless of its nature [cold, increased physical activity, aging, the development of many pathologies (cardiovascular, neurodegenerative diseases, diseases of the gastrointestinal tract, ischemia, the effects of burns), immobilization, hypobaric hypoxia, hyperoxia, radiation effects etc.] leads to a

deterioration of the vitamin status (vitamins E, A, C). Damaging effect on the antioxidant defense system is more pronounced compared to the stress response in animals with an isolated deficiency of vitamins C, A, E, B1 or B6 and the combined vitamins deficiency in the diet. Addition missing vitamin or vitamins restores the performance of antioxidant system. Thus, the role of vitamins in adaptation to stressors is evident. Antioxidant vitamins are valuable in enhancement of the body's defences against damage arising from exposures to substances such as e-waste-borne metals (Cook-Mills, 2013; Rajendran *et al.*, 2014; Pizzino *et al.*, 2017). This study assessed the impact of e-waste on the antioxidant vitamin status of exposed and unexposed individuals.

## MATERIALS AND METHODS

### Study Design

This was a cross-sectional study with purposive approach in the recruitment of participants into the study groups. The groups comprised of e-waste occupationally exposed participants (e-waste workers), participants trading or working around the e-waste repair/ recycling sites/locations considered for the study (environmentally exposed), and non-occupationally exposed, apparently healthy participants (unexposed or control participants).

### Study Areas

The study was carried out in three urban cities (Lagos, Ibadan and Benin). These have been identified and reported as high impact locations for e-waste activities in South-West Nigeria (Osibanjo and Nnorom 2007).

### Study Participants

A total of six hundred and thirty two participants were enrolled into the study: viz, three hundred and eighty-one e-waste workers; one hundred and twenty environmental e-waste exposed participants and one hundred and thirty-one age-matched unexposed individuals, serving as controls.

**E-waste workers**

Male waste electric and electronic equipment (WEEE) workers who were involved daily with four main tasks, purchasing/reception, dismantling, repair and resale, formed the group of e-waste workers enrolled into the study.

Of this group, one hundred and seventy-eight e-waste workers from Alaba International Market and Ikeja Computer village, Lagos state; one hundred and twenty e-waste workers from Ogunpa Market and environs, Ibadan, Oyo state; and eighty-three e-waste workers from Lawani Close/Upper Mission Road/ New Benin Market Area, Benin City, Edo state met the inclusion criteria. Consenting participants with a minimum of five years of occupational exposure to toxic substances in WEEE were enrolled into the study.

**Environmentally Exposed Participants**

The environmentally exposed group comprised of traders and non-e-waste workers involved in work and business activities around the e-waste high impact areas in the aforementioned three study locations selected for this study. Fifty-two participants from Lagos; thirty-eight from Ibadan; and thirty participants from Benin.

**Unexposed Participants**

Non-occupationally and minimally environmentally exposed, and apparently healthy age- and sex -matched participants formed the unexposed or control group. These included forty participants from Lagos State University community, Lagos Badagry Expressway, Ojo, Lagos state; fifty participants from the University College Hospital/University of Ibadan communities, Ibadan, Oyo State; and forty-one participants recruited from the Ugbowo Campus Community of the University of Benin, Benin City, Edo State.

**Inclusion Criteria**

(a) Workers who were occupationally exposed to e-waste for a period of five years and above at the time of sample collection were enrolled into the study. The five-year duration of exposure used in this

study was based on E-waste Risk Assessment Report (Adaramodu *et al.*, 2012); suggesting that a five-year duration is sufficient for the health risks and effects of e-waste crude reprocessing to become apparent in exposed human populations.

(b) Environmentally exposed participants comprised individuals involved in work and business activities around the e-waste high impact areas for a minimum of five years, and as such were environmentally exposed to e-waste-borne toxicants (including metals), due unregulated and environmentally unfriendly WEEE disposal practices previously identified (Grau *et al.*, 2003).

(b) Control subjects were healthy male individuals with minimal or no occupational exposure and with no hobby involving e-waste exposure or other toxic substances.

**Exclusion Criteria**

E-waste workers who were not exposed to e-waste for a period up to five years at the time of sample collection were excluded from the study. Participants with demographic or medical history of any form of cancer, frequent/ habitual tobacco smoking and alcohol consumption were excluded from the study. Demographic and medical history of incidence of cancer, frequent tobacco smoking and alcohol consumption also served as a basis of exclusion for recruiting the apparently healthy control participants.

**Ethical Clearance/Informed Consent**

The protocol for this study was approved by the Health Research Ethics Committee of University of Ibadan/University College Hospital, Ibadan, Nigeria, with a reference number UI/UCH EC: NHREC/05/01/2008a and the Ethical Clearance Committee, Edo State Ministry of Health, with reference number HA.577/Vol.11/164.

Participants for this study were adults who were adequately educated on the benefits of the study, sufficiently briefed of the research protocol, and informed consent was obtained from them prior to sample collection.

The informed consent form used for this study was explicitly explained to each participant in English and Pidgin English languages.

### Sample Collection and Preservation

Prior to blood collection, participants were urged to abstain from using herbal medications, drugs and vitamin/mineral supplements for 12 - 24hours. About ten (10) millilitres (mL) of venous blood was collected from each participant using standard phlebotomy technique. Blood sample obtained was dispensed into ethylene diamine tetra acetic acid (EDTA) anticoagulant specimen bottles (5 mL). Another 5 millilitres was dispensed into anticoagulant-free specimen bottles to obtain serum. Blood samples were allowed to clot, centrifuged at 3000 revolution per minute for 3 minutes and serum collected and stored in another anticoagulant-free bottle. Samples were immediately analyzed and when delay anticipated, serum samples were kept frozen (0 to -4°C) until analysis.

### Determination of antioxidant vitamins using spectrophotometry

Vitamin A was determined by the method as described by Rutkowski *et al.*, (2006). Vitamin C was determined by the method of (Rutkowski and Krzysztof, 2007) using phosphotungstate reagent. Vitamin E was determined according to the method of Rutkowski *et al.*, (2005).

### Statistical Analysis

Using SPSS version 21 IBM, data were analyzed using ANOVA at  $\alpha_{0.05}$ .

## RESULTS

The results in this study were presented in Tables 1 and 2 respectively. Comparison of levels of antioxidant vitamins in all study participants is presented in Table 1, which showed that levels of

vitamin A in environmentally exposed participants was higher and significantly different compared with e-waste workers and the unexposed groups, ( $p = 0.045$ ); and post hoc test showed that the difference observed in vitamin A level between e-waste workers and unexposed group was not significant. Vitamin C level was lower but not significantly different in e-waste workers compared with unexposed groups, whereas, its level in environmentally exposed participants was significantly higher compared with other two groups ( $p = 0.000$ ). Vitamin E level varied significantly amongst the three participants groups, viz, unexposed > e-waste workers > environmentally exposed, ( $p < 0.001$ ).

Compared by location, levels of antioxidant vitamins in the three participants groups are summarized in Table 2. Vitamin A level in Ibadan e-waste workers was significantly higher compared with Lagos and Benin e-waste workers ( $p = 0.017$ ). Vitamin C levels in the e-waste workers across the three study locations did not vary significantly. Vitamin E levels in Benin and Ibadan e-waste workers did not vary significantly, but were significantly higher compared with Lagos e-waste workers.

Comparison of vitamin levels in environmentally exposed group in three study sites showed that vitamin A levels were statistically similar in Benin and Lagos environmental groups, but were higher and significantly different from Ibadan environmental participants ( $p < 0.001$ ). Vitamin C and E did not significantly vary by location in the environmentally exposed group ( $p > 0.05$ ).

In the unexposed control, vitamin A in Ibadan participants was higher and significantly different from Benin and Lagos participants ( $p < 0.001$ ). Vitamins C and E in the unexposed control did not significantly vary by location ( $p > 0.05$ ).

**Table 1 Antioxidant vitamins levels in e-waste exposed and unexposed participants**

Antioxidant Vitamins	All Participants			F value	P value
	E-waste workers (n=381)	Environmentally exposed (n=120)	Unexposed (n=131)		
Vitamin A (µmol/L)	2.08 <sup>B</sup> ±0.04	2.20 <sup>A</sup> ±0.04	1.97 <sup>B</sup> ±0.09	3.12	0.045
Vitamin C (µmol/L)	88.75 <sup>B</sup> ±3.33	116.31 <sup>A</sup> ±203	92.19 <sup>B</sup> ±1.68	41.147	0.000
Vitamin E (µmol/L)	8.91 <sup>C</sup> ±0.19	8.05 <sup>B</sup> ±0.18	12.36 <sup>A</sup> ±0.34	77.78	0.000

Keys: Abbreviations and units as previously defined  
P<0.001= highly significant, P>0.05= not significant

**Table 2 Antioxidant vitamin levels in e-waste workers, environmentally exposed and unexposed (control) in all study locations**

Antioxidant vitamins	E-waste Workers			F value	P value
	Benin-MiIA (n=83)	Ibadan-MoIA (n=120)	Lagos-HiIA (n=178)		
Vitamin A (µmol/L)	2.01 <sup>B</sup> ±0.04	2.27 <sup>A</sup> ±0.09	2.05 <sup>B</sup> ±0.06	4.20	0.017
Vitamin C (µmol/L)	88.15 ±3.69	91.13 ±4.85	87.85 ±8.94	0.08	0.927
Vitamin E (µmol/L)	9.69 <sup>A</sup> ±0.27	9.15 <sup>A</sup> ±0.24	7.96 <sup>B</sup> ±0.37	8.63	0.000
Environmentally Exposed					
Antioxidant vitamins	(n=30)	(n=38)	(n=52)		
Vitamin A (µmol/L)	2.32 <sup>A</sup> ±0.08	1.85 <sup>B</sup> ±0.07	2.36 <sup>A</sup> ±0.08	11.49	0.000
Vitamin C (µmol/L)	120.04 ±1.13	115.79 ±5.73	112.87 ±3.60	1.16	0.316
Vitamin E (µmol/L)	7.92 ±0.19	8.21 ±0.51	8.08 ±0.30	0.20	0.821
Unexposed (Controls)					
Antioxidant vitamins	(n=42)	(n=50)	(n=40)		
Vitamin A (µmol/L)	1.34 <sup>B</sup> ±0.07	3.04 <sup>A</sup> ±0.18	1.68 <sup>B</sup> ±0.06	66.95	0.000
Vitamin C (µmol/L)	93.27 ±1.67	94.12 ±3.77	89.47 ±2.75	0.75	0.473
Vitamin E (µmol/L)	12.78 ±0.24	12.73 ±0.92	11.69 ±0.60	1.14	0.321

Keys: Abbreviations and units as previously defined  
P<0.001= highly significant, P>0.05= not significant  
MiIA = Mild e-waste impact area  
MoIA = Moderate e-waste impact area  
HiIA = High e-waste impact area

## DISCUSSION

Vitamins A, C and E are known to have antioxidant properties; and antioxidant vitamins from supplements and natural sources have been enlisted to be useful in cancer prevention (Palace, 1999; IOM, 2000; Bjelakovic *et al.*, 2008).

In the present study, Comparison of levels of antioxidant vitamins shows that levels of vitamin A in environmentally exposed participants was higher and significantly different compared with e-waste workers and the unexposed groups, and the post hoc test showed that the difference observed in vitamin A level between e-waste workers and unexposed group was not significant. Vitamin C level was lower but not significantly different in e-waste workers compared with unexposed groups, whereas, its level in environmentally exposed participants was significantly higher compared with other two groups. Vitamin E level varied significantly amongst the three participants groups, viz, unexposed > e-waste workers > environmentally exposed. This suggests a considerable low levels of both vitamins E and C in the e-waste groups, and by implication, depressed antioxidant status from the contributions of these antioxidant vitamins.

Oxidative processes have been implicated in both initiation and promotion of cancer. Likewise, animal studies have revealed effects of the antioxidant nutrients on both processes. Vitamin E exhibits antioxidant properties by acting as a lipid-soluble free radical scavenger in cell membranes. Thus, vitamin E may be involved in both initiation and promotion stages. Among the other potentially anti-carcinogenic effects of vitamin E are its ability to inhibit formation of the carcinogenic chemical nitrosamine from nitrites in some foods, and its ability to promote immune system function (van Poppel and van den Berg, 1997; IOM, 2000).

Vitamin C (ascorbic acid) also acts as an antioxidant, and through its ability to scavenge free radicals, it may have

protective effects on biopolymers such as DNA. Like vitamin E, vitamin C may be protective for both initiation and promotion of carcinogenesis. Also, like vitamin E, it is thought to prevent formation of nitrosamine (by converting nitrite to nitrous oxide) and to influence immune system function (van Poppel and van den Berg, 1997; IOM, 2000). Vitamin C has also been reported to affect liver enzymes responsible for detoxification and transformation of carcinogens, Coenzyme Q10 (also termed ubiquinone and ubidecarenone) is an endogenously synthesized chemical (called a quinone) that is also obtained through food intake (van Poppel and van den Berg, 1997; IOM, 2000). In addition to its role as an electron carrier in the mitochondrial electron transport chain, it can also function as a soluble antioxidant (Overvad *et al.*, 1999). Also, its role as an antioxidant (possibly in conjunction with vitamin E) and as a free radical scavenger, and other roles have been proposed for Q10 (Overvad *et al.*, 1999). These include acting as a nonspecific stimulant for the immune system, (Hattersley,1996) and playing a role in membrane stabilization, prostaglandin metabolism, inhibition of intracellular phospholipases, and stabilization of calcium-dependent slow channels. Decreased levels of Q10 have been noted with aging and in such disorders as congestive heart failure, cardiomyopathy, cancer, hypertension, Parkinson's disease, spontaneous abortion, male infertility, chronic hemodialysis, and periodontal disease (Overvad *et al.*, 1999). Vitamins C and E and coenzyme Q10 have been implicated in a variety of potential anti-carcinogenic processes.

Epidemiological studies have suggested an inverse correlation between cancer development and dietary consumption of vitamin A. Pharmacological concentrations of vitamin A decreased the incidence of chemically induced experimental tumours (van Poppel and van den Berg, 1997; IOM, 2000).

Natural and synthetic retinoids have been demonstrated to inhibit the growth and the development of different types of tumours, including skin, breast, oral cavity, lung, hepatic, gastrointestinal, prostatic, and bladder cancers (Niles, 2000; Altucci and Gronemeyer 2001; Arrieta *et al.*, 2010; Bryan *et al.*, 2011; Siddikuzzaman *et al.*, 2011). Moreover, the addition of retinoic acid or synthetic retinoids to human cancer cell lines or human tumour xenografts in nude mice result in growth arrest, apoptosis, or differentiation (Niles, 2000). It is noteworthy that natural retinoids act as chemotherapeutic agents for the treatment of acute promyelocytic leukemia (APL). Moreover, vitamin A reduced the induction of carcinoma of the stomach by polycyclic hydrocarbons (Shibata *et al.*, 2014) and vitamin A-deficient rats were more susceptible to induction of colon tumours by aflatoxin B than normal animals (Rogers *et al.*, 1973). Genomic functions of the retinoids are mediated via their nuclear DNA-binding receptors, RARs, and RXRs, which regulate gene transcription through recruitment of corepressors and coactivators. Natural and synthetic retinoids have been used as potential chemotherapeutic or chemopreventive agents because of their

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## CONCLUSION

Nigerians exposed to e-waste through occupational and environmental means may experience decreased levels of vitamin E. Antioxidant supplementation may be recommended with the aim of mitigating the risk of antioxidant vitamins depression in jobs with suspected mixed metal exposure like crude e-waste recycling.

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