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Author's Affiliation:
Kassala University - Sudan

***Corresponding Author:**
Ahmed A. Osman
Email:
sudanup.ao@gmail.com

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Organophosphorus Pesticide Residues Contaminating Vegetables and Fruits in Sudan are a Major Public Health Hazard

Gasmelseed Y. Ahmed, Ahmed A. Osman*, Ahlam Mukhtar, Wala M. Awad

Abstract

Background: Organophosphorus pesticides (OP) are a diverse group of compounds. Extensive application of these chemicals in Sudan increases the risk of food and water contamination. We aimed to identify the occupational hazard on sellers and consumers resulting from exposure to organophosphorus through measurement of blood (AChE) enzyme activity.

Methods: This is a community-based study conducted at the Khartoum vegetable market, where 219 sellers and consumers were consented and recruited consequently. Data on demographics and pesticides exposure was collected and blood for acetylcholinesterase enzyme (AChE) activity was withdrawn and tested in a Lovibond machine.

Results: 139 (63.5%) of the participants were males with a mean age of 41.6 ± 12.9 years. Educational levels: 83 (37.9%) elementary school, 52 (23.8%) middle and high school, 54 (24.6%) college and above, and 30 (13.7%) were illiterate. More than half 129 (58.9%) were consumers and the remaining 90 (41.1%) were sellers. Univariate analysis revealed sellers having a significantly more proportion of impaired acetylcholinesterase (AChE) activity compared to consumers, [33 (36.6%) versus 9 (7%), p-value 0.001]. Moreover, the mean (AChE) activity for whole respondents was lower than the physiologic mean of unexposed people (82.3% versus 87.5%). Independent t-test revealed significantly impaired (AChE) activity among sellers (74.9% versus 87.5%), (P-value = 0.001).

Conclusion: People exposed to agricultural chemical pesticide residues in vegetables and fruits have impaired (AChE), with a significantly lower rate of the enzyme among products' sellers in reference to consumers and to the physiological values of unexposed. A larger community-based study evaluating exposures to pesticides is highly recommended.

Introduction

Agriculture in Sudan is the most important national resource, as 60-80% of Sudanese households use it as their first income. Moreover, it accounted for about 80% of the exports of the country [1]. The current increasing industries and use of agricultural chemicals for the purpose of increasing production, have affected the environment negatively; and exerted adverse effects on human health as well. Workers in the agricultural sector use pesticides commonly, including a wide range of compounds such as plant growth regulators, insecticides, herbicides, fungicides, nematicides, rodenticides, and others [2]. Environmental and occupational exposure to these chemicals results in contamination and carries serious health risks through eating contaminated food and drinking water carrying chemical residues. Exposure to agricultural chemicals occurs in open agriculture fields, greenhouses, and pesticide industries. The adverse effect comes from pesticides when reaching a certain level of exposure through contact with large quantities of pesticide. People who are exposed to these chemicals have the greatest risks. People frequently come in contact with these chemicals at work, in their homes, or in gardens during the preparation and handling of pesticides. On the other hand, the general population acquired the exposure through eating contaminated vegetables, fruits, and drinking water that holds pesticide residues. Some, less expensive pesticides, such as dichlorodiphenyltrichloroethane (DDT) can stay for a long time in soil and water resulting in environmental pollution causing potential risk to humans [3]. Through the last three decades, there has been an increase in the use of chemicals to yield an increase in agricultural production. Farmers use chemicals to control the growth of pests and weeds, prevent plant diseases, reduce losses, and maintain a high level of production to meet global demand. This progress in agriculture, livestock farming, dairy farming products, and the animal protein industry, accompanied and resulted in more need for the use of agricultural chemicals, antibiotics, and anti-parasite drugs, which might leak into the environment due to improper management, affecting public health adversely. There are very strict regulation processes to control the production of pesticides in order to minimize the impact on the environment and human health, serious concerns have been raised about risks resulting from occupational exposure and environmental leaks resulting in residues in food and water. Farmers and farmworkers are at a higher risk of exposure to pesticides. Some of them had a lower level of knowledge regarding pesticide safety measures which increases their health risk. The poor management of residues of these chemicals could leak into the environment, affecting the ecosystem, soil life, and all-natural processes. The presence of chemical residues in milk and meat can affect the general health of the population. Moreover, the adverse effect of chemicals occurs frequently in countries with poor and insufficient food quality controls. These processes contribute to the growing global threat concerning the health of humans and animals [4]. The current industrialization of the agricultural and animal production sectors to match the agricultural and crossbreeding advancement for increasing productivity has resulted in the use of more agricultural chemicals increasing the chemical burden on natural ecosystems. A better and more sustainable ecological approach for a cleaner and safer environment, reforms, policies, and plans for increasing organic food production for human safety and consumers' health protection is needed [5]. Some farmers ignore the recommended concentration level of organophosphorus written on the label instructions during farming and pesticide use. Some of them do not follow the advised protective measures, including the pre-harvest intervals between pesticide application and crop harvesting. Many farmers are unaware of the pre-harvest interval for the pesticides they use and its importance for consumer health and safety. Both farmers and market vendors apply pesticides to commercially gain more time and protection of their products in the market. Moreover, pesticide residues can be found throughout the food chain, from fruits and vegetables on the farm, through transporters and market vendors, to the final consumers, noting that some consumers purchase directly from farmers or transporters. The level of pesticide residues on fruits and vegetables is measured from farmers' pesticide application practices close to harvest. Other factors that affect pesticide residue levels on fruits and vegetables along the supply chain include the handling and processing methods used by various stakeholders, such as washing, peeling, spraying with pesticides, blending, and wiping with a cloth. The findings of the study on the trends of agricultural chemical residues and their movement along the food chain in Uganda revealed a very crucial point as contamination might occur with the farmers' pesticide application practices near harvest, in addition to the handling and processing methods used by stakeholders [6].

In Sudan, the Food and Agriculture Organization (FAO) reported environmental contamination with 666 tons of outdated pesticides affecting 44 sites, leading to soil contamination. A total of over 200 active pesticide ingredients are registered in Sudan, either singly or combined in over 600 different formulations [7-9]. Mosquito control accounted for about one-fifth of these pesticides, the remaining four-fifths are chemicals used in controlling pests and some of them were chemicals used to prevent desert locusts, birds, and rodents. In Sudan, the environmental section faced the problem of using outdated and unauthorized pesticides. furthermore, the storage conditions of such pesticides are below the standards in general, sometimes the pesticides are stored in eroded or damaged containers which will open windows for leaking in the environment. No facilities for local disposal exist. Despite the lack of epidemiological data on research conducted and addressing this health problem in Sudan, it is probable that exposures to pesticides and herbicides contribute to the increasing rate of cancer in the country [10,11].

The aim of this study was to identify the occupational hazard for sellers and consumers resulting from exposure to organophosphorus (OP), through measuring blood (AChE) enzyme activity in Khartoum State, Sudan, 2020.

Methods

This study was an observational cross-sectional community-based study conducted in Central Market for Vegetables and Fruits, Khartoum City in the capital of Sudan. We included sellers and consumers who were presented at Khartoum City Central Market for Vegetables and Fruits, Sudan during the study period in 2020. We enrolled two hundred and nineteen participants who consented to participate voluntarily in this study. Due to the situation in the study area (Khartoum Central market for vegetables and fruits), we applied a nonprobability sampling technique to suit the busy and crowded nature of the market, hence it was convenience sampling. Therefore, all the available pool of respondents (sellers and consumers) who attended the market on the research day were recruited consecutively; first invited to participate, those who agreed consented then interviewed and tested for blood enzyme level, making a total sample of 219 participants.

We used a structured, pretested questionnaire as a tool comprised variables of gender, age, educational level, original residency, and occupational exposure (agricultural products' sellers and consumers).

Blood test for AChE activity

Preparation of Reagents and Blood Test Methodology

We prepared the required reagents bromothymol blue and acetylcholine perchlorate as well as the blood samples for testing by a skilled toxicology specialist through the following steps: step one preparation of reagents started with water preparation as the distilled water was boiled, cooled, and kept secure. We used a fresh solution of 0.25 mg of acetylcholine perchlorate in water on daily base. Then, we prepared a solution of bromothymol blue with 112 mg in 250 ml of water. The ultimate solution had a pH range of 5.8 - 6.6. Step two comprised of preparation and testing of blood samples by adding one ml of water mixed with 0.01 ml of the blood putting them into the comparator, and acting as the storage point for the samples to be taken of the samples daily. After that, we added 0.5 ml of indicator solution to 10 µl of blood taken from a control subject and we added 0.5 ml of the substrate solution. Then, we promptly transferred the given solution to another clean 2.5 mm cell and put it on the right side of the compartment. We read the results accordingly and documented the cholinesterase activity. The reagent solutions were deemed correct if readings were between 0 and 12.5 on the scale. Step three consisted of testing individual blood samples, and for each blood sample, a clean test tube was used. The final step was the categorization of the results as we quantified the enzymatic activity of acetylcholinesterase (AChE) levels according to the manufacturer's standards, which categorized the readings into three groups: normal range (100-75%), overexposure (74.9-50%), and serious overexposure (49.9-25%).

AChE Blood Test Using the Lovibond Color Instrument AF267 Kit: Quantitative assessment of AChE activity in blood was conducted using the Lovibond Color Instrument AF267 kit, which employs simple equipment and a straightforward colorimetric test for rapid and accurate results. The kits, supplied in a sturdy wooden case, included all required reagents, apparatus for field testing, and detailed instructions. A blood sample (one drop) from a finger stick was taken, and after adding the reagent, which caused the sample to develop a color ranging from green to yellow based on cholinesterase activity levels, the samples were visually matched with glass color standards in the Lovibond Comparator 2000+. Each glass standard corresponded to a percentage of cholinesterase activity, ranging from 0% to 100% in increments of 12.5% [12, 13].

Statistical analysis

A Statistical Package for Social Sciences (SPSS) version 25 (IBM Corp., Armonk, NY, USA) was applied as standard statistical procedure. The collected data was cleaned and validated for accuracy and completeness before conducting the statistical analysis. A group of descriptive and inferential statistical tests were performed. In descriptive analysis, the socio-demographic variables were analyzed and reported as frequencies and means ± standard deviation (SD) to summarize continuous variables, and in inferential statistics, Chi-square and t-tests were applied. Participants were categorized into two groups exposed and unexposed, while differences between groups and associations were considered statistically significant for all two-sided p-values was ≤ 0.05 and 95% confidence interval (CI).

Ethical aspect: Ethical approval was obtained from the concerned authority, and informed consent was obtained from all participants in the study.

Results

The majority of the participants 139 (63.5%) were males with a mean age of 41.6 ± 12.9 years. Most of them 83 (37.9%) completed only elementary schooling, 52 (23.8%) had middle and high school and 54 (24.6%) had a graduate and postgraduate level of education, while 30 (13.7%) of them were illiterate. Most of the respondents 136 (62.1%) were residents of Gezira State, Sudan, followed by 29 (13.2%) from the Northern Region of Sudan, 21 (9.6%) from Khartoum, 17 (7.3%) from the Western Region and only 16 (7.3%) from Eastern Region of Sudan. Results showed that more than half 129 (58.9%) of the participants were consumers and the remaining 90 (41.1%) were sellers (Table 1).

Characteristics		N (%)
Indicators	**Age**	**41.6 ± 12.9 years**
Site of recruitment/ occupation (Occupational exposure)	Agricultural products' sellers	90 (41.1)
	Consumers	129 (58.9)
Gender	Male	139 (63.5)
	Female	80 (36.5)
Educational status	Illiterate	30 (13.7)
	Elementary school	83 (37.9)
	High school	52 (23.8)
	College and above	54 (24.6)
Residence	North Sudan	29 (13.2)
	Khartoum	21 (9.6)
	West Sudan	17 (7.8)
	East Sudan	16 (7.3)
	North Gezira	39 (17.8)
	East Gezira	26 (11.9)
	Central Gezira	25 (11.4)
	West Gezira	24 (11.0)
	South Gezira	22 (10.0)

Table 1: Demographic Characteristics, Organophosphorus Pesticide Residues Contaminating Vegetables and Fruits in Sudan are a Major Public Health Hazard; 2020; (n=219).

Univariate analysis revealed sellers have significantly more proportion with impaired (AChE) activity than consumers, [33 (36.6%) versus 9 (7%), p-value 0.001] (Table 2).

However, the mean (AChE) activity for whole respondents was lower than the physiologic mean for unexposed (82.3% versus 87.5%) participants. Independent t-test revealed significantly impaired (AChE) activity among sellers (74.9% versus 87.5%), (P-value = 0.001) (Table 3).

AChE activity	Sellers	Consumers	Total	P-value
100%	18 (20.0)	52 (40.3)	70 (32.0)	0.001
87.5%	15 (16.7)	36 (27.9)	51 (23.3)	
75%	24 (26.7)	32 (24.8)	56 (25.6)	
62.5	18 (20.0)	07 (5.4)	25 (11.4)	
50%	11 (12.2)	2 (1.6)	13 (5.9)	
37.5%	4 (4.4)	0 (0.0)	4 (1.8)	
Total	90	129	219	

*Normal physiological mean (AChE) in the population is 87.5% (normal range of 75% - 100%)

Table 2: Univariate analysis reporting Chi-square test, Organophosphorus Pesticide Residues Contaminating Vegetables and Fruits in Sudan are a Major Public Health Hazard; 2020; (n=219).

Respondent's Group	Mean AChE activity	t-test	P-value
Whole respondents	82.3% ± 16.3%	5.7	0.001
Vegetables and fruits' sellers	74.9 % ± 9.6%		
Vegetable consumers	87.5% ± 11.4%		

Table 3: Univariate analysis reporting independent t-test, Organophosphorus Pesticide Residues Contaminating Vegetables and Fruits in Sudan are a Major Public Health Hazard; 2020; (n=219).

Discussion

In this study, we identified the level of impaired (AChE) enzymatic blood activity among people exposed to vegetables and fruits contaminated with pesticides (agricultural products' sellers and consumers). The results detected impaired enzyme activities among most of the participants in the study sample compared to the mean physiological level of the enzyme in normally unexposed people.

In Sudan, the harmful effect of contaminated food by OP pesticide residues might extend beyond the agricultural products to contaminate the drinking water. Research on the drinking water of the Nile revealed the presence of 22 commonly used pesticides in 54 water samples gathered from six locations along the Blue Nile, White Nile, and River Nile. These samples were analyzed using gas chromatography. The findings showed that nearly half of the Blue Nile water samples contained organochlorine (OC) pesticides, 11% contained organophosphates (OP), and over 20% contained pyrethroids, with heptachlor and DDTs being the most frequently detected, followed by pyrethroids. In the main Nile, OC pesticides were found in 22% of the samples, while pyrethroids were present in 4%; DDT was the most common, followed by heptachlor. The overall concentration of pesticides in the main River Nile was higher than in the Blue Nile, possibly due to agricultural runoff from central Sudan [14].

In Sudan, the negative impact of food contaminated with organophosphate (OP) pesticide residues may extend beyond agricultural products, potentially affecting drinking water contamination. Studies on Nile drinking water detected 22 frequently used pesticides in 54 samples taken from six locations along the Blue Nile, White Nile, and River Nile. These samples were analyzed using gas chromatography. The Blue Nile water samples contained organochlorine (OC) pesticides in almost half of the samples, OP pesticides in 11%, and pyrethroids in more than one-fifth of the samples, with heptachlor and DDTs being the most common, followed by pyrethroids. In the main Nile, OC pesticides were found in one-fifth of the samples, and pyrethroids were present in 4%, with DDT being the most common, followed by heptachlor. Overall, the total concentration of contaminants in the main River Nile was higher than in the Blue Nile, likely due to agricultural runoff from central Sudan.

Our study indicated that market activities and handling practices, such as unloading, packing, washing, and drying agricultural products, can increase exposure to agricultural chemicals for both sellers and consumers, consequently reducing their blood acetylcholinesterase (AChE) activity. A comparable study in Zambia highlighted the levels of agricultural chemical residues in fruits and vegetables and revealed that more than 50% of the samples contained residues that exceeded the standards set by the Food and Drug Act (Zambia) [15]. Another recent study conducted by Eltoum et al. in the main Central Markets of Khartoum, Sudan, showed high levels of pesticide residues present on vegetables which were detected by gas chromatography. These findings indicated that most of the farmers lacked training in pesticide handling, storage, application, and awareness of the environmental and health risks. Laboratory results showed chemical contamination, with tomatoes, eggplants, and okra being the most affected crops, and all samples exceeding the maximum residue limits (MRLs) established by Codex Alimentarius, with tomatoes showing the highest levels of Dimethoate residues [16].

Regional studies showed extensive usage of pesticides which in turn brings farmers and consumers to be vulnerable to the risk of pesticide exposure. In the central region of Uganda, the excessive usage of pesticides is obviously detected in the tomato crop. It was observed that all the samples analyzed had detectable levels of mancozeb pesticide with higher concentrations enough to put the farmers and final consumers at risk of exposure to pesticides [17]. Another African study conducted in Uganda assessed 160 samples and detected occurrences of pesticides in more than ninety percent of consumed fruits and vegetables in the country [18]. One Egyptian study identified high levels of pesticide residues in food exceeding the allowed (MRLs), they detected 40 types of pesticides in different vegetables and fruits [19].

A group of investigators in Chile who studied environmental exposure (EE) and occupational exposure (OE) indicated that people under exposure to OP pesticides showed impairment (AChE) in agricultural workers, their results demonstrated a higher rate of impairment compared to the results of the current study. Moreover, their study revealed acute intoxications of agricultural workers occurred with frequent use of OP, the inhibition of (AChE) in the occupationally exposed participants was observed among 55% of them, which was higher than the rate of our study (36.7%) [20].

Pesticides are developed to facilitate agricultural requirements and to be applied with high quality and minimal risk to applicators' health. However, many concerns are now raised regarding adverse health effects from occupational and environmental populations, and exposure to residues found on vegetables, fruits, and drinking water. Pesticide contamination among people who work in agricultural products shown in this study, added to the results reported by Taha et al of contaminated food items during the spring season, and Azhari et al findings which revealed 4 pesticides out of 11 contaminated eggplants are a major public health hazard [21, 22]. These three national studies raise the awareness of pesticide contamination about the unclear etiology of the increasing rates of cancer and renal failure in Sudan. An international study of pooled data from more than 40 thousand different countries provided evidence of altered kidney function in people exposed to organophosphate pesticides, another hazard is the role of pesticides as a major trigger of the development of cancer [23,24].

Our current study has provided findings of impaired (AChE) among occupationally exposed people. We have shown in this study that farmers and product sellers were under unprotected exposure to a wide variety of OP pesticides. This occupational exposure jeopardizes the health of farmers, sellers, and consumers in Sudan and places them at high risk for short- and long-term intoxication consequences. In conclusion, the presence of pesticide residues in consumed fruits and vegetables is an alarm for the urgent need to develop comprehensive interventional measures to reduce the potential health risk to agricultural workers, product sellers, and consumers. There is a need for the regular monitoring of pesticide residues and screening of exposed individuals regularly, taking the necessary precautions during handling of pesticides, with a focus on adherence to optimal spray time pre-harvest periods that is sufficient to clear pesticides from agricultural products, besides health education on cautious handing and washing of vegetables and fruits. Our findings need further exploration of this field for detailed evidence that might give clues to the steadily increasing rates of some diseases in Sudan including but not limited to cancer and renal failure.

The study has the strength of being one of the first studies conducted in the country to provide data on occupationally exposed agricultural products' sellers and consumers, besides reporting quantitative measures of (AChE) in the blood of exposed people. However, it is also important to acknowledge some limitations of the study. A fundamental limitation is having more dominant males in total, and none among sellers, besides being a cross-sectional observational study, and not reporting the maximum residual limits (MRLs) for safe consumption. Moreover, further limitations can be highlighted including the use of the old technique for testing (AChE) activity by applying the Lovibond machine.

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Author Contributions

GA and AA did the methodology. Reagent preparedness, blood tests, and data curation were done by GA and AM. GA and AA conducted the original draft preparation, writing, and reviewing. WMA reviews, responds, and edits the publisher's feedback and rewrites the final version. All authors have read and agreed to the published version of the manuscript.

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

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