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Determination of Copper and Arsenic in Some Vegetables Planted in Damascus and Assessment of Their Potential Health Risk

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Abstract:

Background & Objective: Vegetables are essential source of a wide range of vital nutrients for the people and constituted the lowest cost of main food for the majority of people in developing countries. Nonetheless, worries about its heavy metals contamination have escalated in recent years. The objective of this study was to investigate copper and arsenic accumulation in some vegetables grown in Damascus and to assess the human health risks from consuming these vegetables.

Materials and Methods: Samples were digested with wet oxidation method using concentrated nitric acid and hydrogen peroxide. Metals determination was carried out using atomic absorption spectrophotometer.

Results: The results showed that the highest copper average content of 20.52 mg/kg was found in watercress and the lowest level was in maize (6.86 mg/kg). The concentration of arsenic in most studied vegetable was not detectable. HQ values to the heavy metal due to the consumption of lettuce and watercress were> 1, for Cu, while the HQ value was lower than 1 in maize.

Conclusions: All studied vegetable samples contained copper at different levels and the HQ values of adults through consumption of lettuce and watercress were more than 1, suggesting that the consumers may be facing health risks due to its consumption.

Keywords: Vegetables, Copper, Arsenic, Atomic Absorption Spectrophotometer (AAS), Health Risk

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تحديد النحاس والزرنيخ في بعض الخضروات المزروعة في دمشق وتقييم مخاطرها الصحية المحتملة

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الملخص:

خلفية البحث وهدفه: تعَد الخضراوات مصدراً أساسياً لمجموعة واسعة من العناصر الغذائية الهامة للناس وتشكل الغذاء الرئيس الأقل كلفة لغالبية الناس في البلدان النامية. مع ذلك تصاعدت المخاوف بشأن تلوثها بالمعادن الثقيلة في السنوات الأخيرة.

هدفت هذه الدراسة إلى التحري عن تراكم النحاس والزرنيخ في بعض الخضراوات المزروعة في دمشق وتقييم المخاطر الصحية الناتجة عن استهلاك تلك الخضراوات.

مواد البحث وطرائقه: تم هضم العينات بطريقة الأكسدة الرطبة باستخدام حمض النتريك المركز والماء الأوكسجيني. تم تحديد المعادن بواسطة مقياس الامتصاص الذري.

النتائج: أظهرت النتائج أن متوسط أعلى تركيز من النحاس كان في نبات الجرجير ٢٠.٥٢ (mg/kg) بينما متوسط أقل تركيز كان في الذرة ٦.٨٦ .(mg/kg) كانت تراكيز الزرنيخ في معظم عينات الخضار المدروسة غير قابلة للكشف. كانت قيم HQ الناتجة عن استهلاك الخس والجرجير 1 □ بينما كانت أقل من ١ في الذرة.

الاستنتاجات: احتوت جميع عينات الخضار على النحاس بتراكيز مختلفة وكانت قيم HQ للبالغين الناتجة عن استهلاك الخس والجرجير أكبر من الواحد، مما يشير إلى أن المستهلكين قد يواجهون خطراً من استهلاكها.

الكلمات المفتاحية: الخضراوات، النحاس، الزرنيخ، مقياس الامتصاص الذري، المخاطر الصحية.

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1. Introduction:

Vegetables are essential components of a healthy diet, rich in vitamins, minerals, fibers, and antioxidants that support overall health. They play a crucial role in maintaining a balanced diet and proper bodily functions. However, despite their numerous health benefits, vegetables can serve as an intermediate reservoir, transferring trace elements from soil, water and air to humans and animals. **(Alegbe, *et al,* 2025)**. Thus, the uptake of metallic and metalloid elements by vegetables from the aforementioned media is a primary route for toxic elements to enter the food chain, bio-accumalate, and lead to deleterious effect to human health **(Feyisa, *et al,* 2025).** Metals such as lead (pb), Cadmium (Cd), arsenic (As) and copper (Cu) are cumulative poisons. These metals cause environmental hazards and are reported to be exceptionally toxic.

Contamination of vegetables with heavy metal may be due to irrigation with contaminated water, the addition of fertilizers and metal- based pesticides, transportation or as a result of industrial activities **(Aydinalp, C. and Marinova, S. 2012).**

Arsenic is a toxic element, known as class (1) human carcinogen and widely distributed in the environment **(Arya *et al,* 2019)**. Chronic arsenic exposure has been associated with circulatory disorders, neurological complications, diabetes, hepatic and renal dysfunction and it has been shown to cause skin and lung cancer **(Fatoki,J. and Badmus, J, 2022).** Certain fruits and vegetables including leafy greens have been found to contain elevated arsenic levels **(Shandana *et al,* 2024).**

Copper is the vital micronutrient for the plant growth but its concentration beyond critical limits affects the plant growth, promotes leaf chlorosis and causes cytotoxicity **(Kumar, *et al,* 2021**). Copper is an essential heavy metal for human metabolism at the trace level, but it is toxic at high quantities. The excessive intake of copper would lead to accumulation of the metal in liver cells and haemolytic crisis and Wilson's disease **(Isa, *et al,* 2015)**. Furthermore, high concentrations of copper have been linked to Alzheimer's disease (AD), as Cu ions seem to be a central cationic metal in the formation of plaque and have an essential role in the AD pathology **(Ejaz, *et al* 2020).**

Several analytical methods for the determination of heavy metals have been reported that include inductively coupled plasma-mass spectrometry (ICP-MS), potentiometry, ion chromatography, as well as voltammetry. However, atomic absorption spectrometry (ASS) is still the most cited technique **(Mariela *et al,* 2006).**

The aim of this research was to determine the arsenic and Copper contents of some vegetables lettuce, watercress and maize (as corn is considered as starchy vegetables) grown in Damascus by using ASS. Since the presence of contaminants in vegetables, especially toxic metals, can cause acute and chronic health effects in humans, this study aimed also to calculate the health risk posed by As and Cu for vegetable consumers.

2. Materials and Methods:

2.1 Reagents

Concentrated nitric acid 65% Panreac, Spain, hydrogen peroxide 30% Avonchem, UK, copper standard solution (1000 ppm) Merck, Germany and arsenic standard solution (1000 ppm) Merck, Germany were used in the current study. Double deionized water was used for preparation and dilution of standards and reagents. All Plastic and glassware were cleaned by soaking in dilute nitric acid 2% for 24 hours and were rinsed with distilled water prior to use.

2.2 Sample collection

The vegetables chosen for this study are lettuce, watercress and Maize. Ten samples of each of lettuce leaves, lettuce stems, maize and watercress were collected randomly from Damascus (Ibn Al- Nafis orchards). The total number of samples was 40. Details of vegetables samples are given in Table (1).

Table (1): Details of vegetables samples

S.No	Commen name	Botanical name	Edible part	Family
1	Lettuce	*Lactuca sativa L.*	Leaves & stems	Asteraceae
2	Watercress	*Nasturtium officinale R.Br.*	Leaves	Brassicaceae
3	Maize	*Zea mays*	Kernels	Graminae

2.3 Sample preparation

The collected fresh vegetables samples were washed first with tap water then distilled water to remove the attached soil particles and impurities. After they dried at room temperature, the samples were ground into fine powder using an electric mixer and stored in polyethylene containers.

2.4 Sample digestion

Digestion was achieved by using the open wet oxidation method **(Ali, z. et al 2012)**. A mixture of nitric acid 65% and hydrogen peroxide 30% (3:1) was added to a 1 g vegetable sample, then this mixture was heated up to 130 C̊ for 1 h. The samples were diluted with deionized water to 100 ml and filtered through whatman filter paper No 41.

2.5 Preparation of standards

Standard solution (1 ppm) was prepared and diluted to (0.5, 1, 2.5, 5, 10 and 20 ppm for copper) and (1, 2, 5, 10, 20 and 30 ppb for arsenic) to build the calibration curve. The standard solution and blank were analyzed by a flame Atomic Absorption Spectrophotometer (Model: HITACHI Z-2000) for copper and graphite furnace Atomic Absorption Spectrophotometer (Model: HITACHI Z-2000) for arsenic. The working conditions of AAS are shown in Table (2).

Table (2): Instrument operating conditions for the determination of Cu and As in vegetables samples by AAS.

Element	Wavelength (nm)	Lamp current (mA)	Slit width (nm)	Flame gases
As	193.7	7.0 mA	1	-
Cu	324.8	7.5 mA	1.3	Acetylene/ Air

2.6 Sample analysis

The digestion solutions were subsequently analyzed for determination of Cu and As by using AAS. All standards and samples were analyzed in triplicates. The absorbance measured by atomic absorption spectrophotometer was converted to concentrations using standard calibration curves. Heavy metal concentrations in vegetables samples were calculated according to Equation 1

$$\mathbf{mg/kg = (C \times A) / W} \quad \mathbf{(1)}$$

Where, C = Heavy metal concentration in vegetables (mg/L Cu, µg/ L As); A = Final volume of diluted sample; W = weight of sample (g). The results were converted to mg/kg for copper.

2.7 Health risk assessment

To determine the health risk assessment, the Estimated Daily Intake (EDI) (mg/kg bw/day) of Metals was calculated according to the Equation 2

$$\mathbf{EDI = Concentration\ of\ metal\ in\ vegetables\ mg/Kg} \times \frac{\textit{daily food consumption}}{\textit{body weight}} \quad \mathbf{(2)}$$

According to the report of Food and Agriculture Organization of the United Nations (2024), average per capita vegetables consumption, measured in Kilogram per person per year, the average Syrian individual's consumption is approximately 0.220 kg of vegetables per day. The average body weight is 70 kg for adults based on the standard assumption for an integrated USEPA risk analysis.

The hazard quotient (HQ) for noncarcinogenic risk of metal was evaluated using the Equation 3

$$\mathbf{HQ = EDI / RfD} \quad \mathbf{(3)}$$

Where RfD is the oral reference dose (mg/Kg/day). According to FAO/WHO, the RfD Values are 0.3 and 40 µg/Kg for As and Cu respectively **(Rostamzadeh et al 2025)**. If the value of HQ is < 1, It is generally presumed to be safe for the risk of noncarcinogenic effects and if it is > 1, it is supposed that there is a chance of noncarcinogenic effects with an increasing probability as a value upsurges.

2.8 Statistical Evaluations

The statistical analysis was performed by computer program of Excel. Student's t test was used to assess the significance of the differences between concentrations of elements in vegetables.

3. Results:

3.1 Linearity

The relationship between concentration of each element (As and Cu) and the corresponding absorbance values is represented in Figure 1 (A & B) respectively. The R^2 values obtained were higher than 0.995 as shown in the (Figure 1).

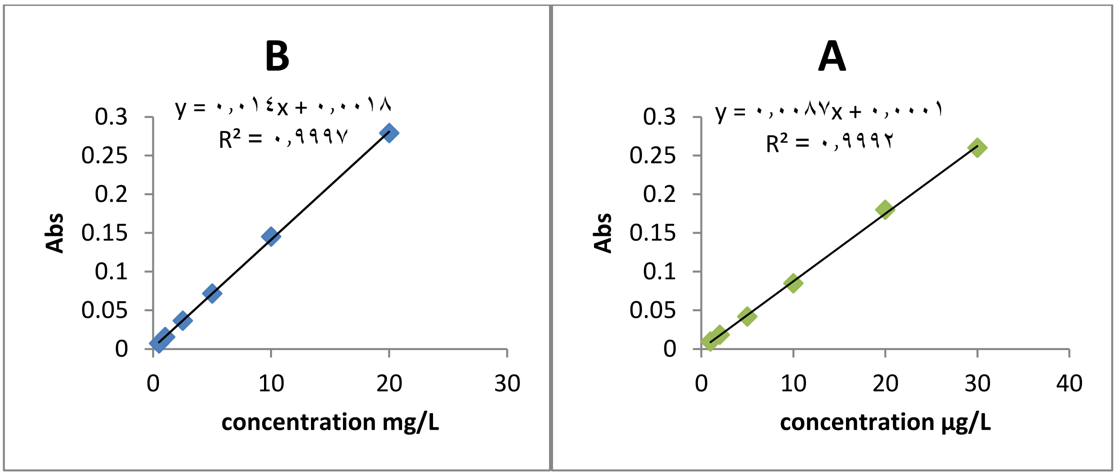

Figure (1): Calibration curve of Arsenic (A), and Copper (B)

3.2 Copper and Arsenic concentrations in vegetables samples

Tables (3) and (4) show the copper and arsenic content of vegetables samples, where the results expressed as the average of the three replicate analyses. Figure (2) shows the average of copper concentrations (mg/kg) in the studied vegetables samples.

Table (3): The average concentration of copper and arsenic (Average ± SD, n = 10, mg Kg^{-1} dry weight) in Lettuce leaves and stems

Lettuce leaves	Cu	As	Lettuce stems	Cu	As
1	16.9±0.82	ND	1	14.8±0.15	ND
2	14.8±0.19	ND	2	15.5±0.43	ND
3	16.9±0.31	0.002±0.001	3	21.7±0.56	ND
4	14.1±0.13	ND	4	18.9±0.55	0.034
5	16.9±0.22	ND	5	16.2±0.33	ND
6	12.1±0.28	ND	6	15.5±0.41	0.002
7	15.5±0.31	ND	7	22.4±0.82	ND
8	21.7±0.76	ND	8	18.9±0.76	0.015
9	14.1±0.11	0.002±0.001	9	19.6±0.54	ND
10	18.9±0.66	ND	10	19.6±0.47	ND

ND, not detected.

Data are not significantly different as determined by sample t- test at p> 0.05

Table (4): The average concentration of copper and arsenic (Average ± SD, n = 10, mg Kg^{-1} dry weight) in Watercress and Maize

Watercress	Cu	As	Maize	Cu	AS
1	10±0.12	ND	1	6.6±0.09	ND
2	16.2±0.22	ND	2	5.9±0.13	ND
3	36.1±0.87	ND	3	5.2±0.06	ND
4	14.1±0.12	ND	4	4.5±0.02	ND
5	21±0.19	ND	5	6.6±0.05	ND
6	15.5±0.23	ND	6	3.8±0.01	ND
7	25.8±0.66	ND	7	6.6±0.12	ND
8	22.4±0.30	ND	8	7.3±0.08	ND
9	27.9±0.28	ND	9	8±0.06	0.021±0.009
10	16.2±0.14	ND	10	14.1±0.11	ND

ND, not detected

Data are significantly different as determined by sample t- test at p> 0.05

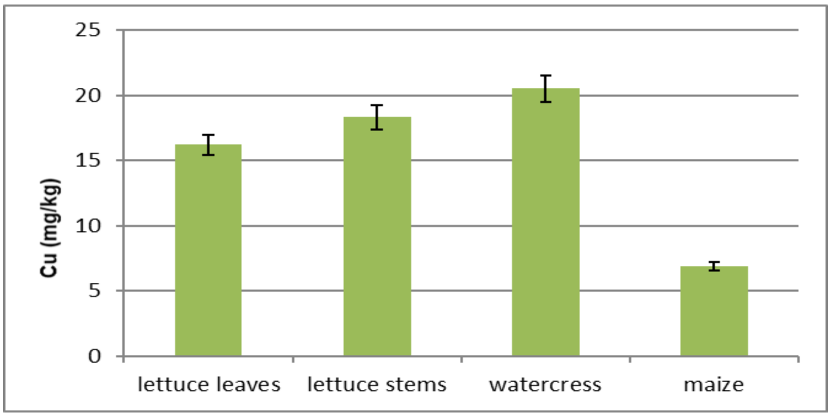

Figure (2): The average of copper concentrations in the studied vegetables samples

3.3 Health risk assessment of vegetable consumption

Chronic low-level intake of toxic metal elements has a negative effect on human health, and the detrimental impact becomes apparent after several years of exposure. The Estimated Daily Intake (EDI) is commonly used method which helps to identify the number of daily consumed pollutants. The hazard quotient (HQ) is the ratio between EDI and the reference oral dose (RfD).

The EDI (mg/kg bw/day) and HQ values of copper and arsenic due to vegetables consumption for residents of the study area are listed in Table (5)

Table (5): Health risk assessment for consuming lettuce, watercress and maize

Vegetable	Metal	Mean (mg/kg)	EDI (mg/kg bw/day)	HQ
Lettuce leaves	Cu	16.19	0.050	1.25
	As	0.0004	0.12×10^{-5}	0.004
Lettuce stems	Cu	18.31	0.057	1.42
	As	0.0051	0.16×10^{-4}	0.053
Watercress	Cu	20.52	0.064	1.61
	As	-	-	-
Maize	Cu	6.86	0.022	0.53
	As	0.0027	0.8×10^{-5}	0.026

4. Discussion:

In the light of our results, it is clearly shown that all vegetables samples contained copper which may be due to modern agricultural practices that involve the use of chemical fertilizers, pesticides, and industrial waste, all of which can contribute to copper accumulation in the soil. Additionally, irrigation water can be a major source of copper contamination. Most of agriculture lands located near urban are irrigated by wastewater because of the low availability of fresh water for crop production. The waste waters contain high levels of heavy metals which may cause remarkable increases in soil heavy metal content **(Eissa, M.,and Negim, O., 2018)**. All vegetables in this study were collected from Damascus ((Ibn Al- Nafis orchards) from lands irrigated by the yazid River which may be contaminated with heavy metals.

Copper and arsenic concentrations varied among different vegetables as indicated in Tables (3), (4). The observed ranges in the concentrations of Cu in the edible parts of chosen vegetables were 12.1- 21.7 mg/kg in lettuce leaves, 14.8- 22.4 mg/kg in lettuce stems, 10- 36.1 mg/Kg in watercress, and 3.8- 14.1 mg/kg in maize, with mean concentrations of 16.19, 18.31, 20.52 and 7.9 mg/kg respectively.

The concentration of arsenic in all samples of watercress and maize was not detectable, except for one maize sample that had arsenic content of 0.021 mg/kg. While the majority of lettuce samples had no detectable arsenic a few (two of leaves and three stems) contained low levels, ranging between 0.002- 0.034 mg/kg.

Among the three analyzed vegetables species, maize contained the lowest copper concentrations in its edible portions, while watercress, belonging to the Brassicaceae family, exhibited the highest Cu levels.

Most of heavy metals that enter the plant are retained in root cells, where they bind with amino acids, organic acids or metal binding peptides and are subsequently stored in vacuoles. This strongly inhibits transfer of metals to the shoots and leaves. Nevertheless, a number of hypertolerant species, defined as «hyperaccumulators» exhibit an opposite behavior **(Rasico, N. and Navri-Izzo, F., 2011).**

Several Brassica species are known metal accumulators and have been evaluated as potential phytoextraction plants **(Mourato, M. *et al* 2015)**. By releasing protons from their roots, hyperaccumulator plants enhance soil acidity, promoting metal ion mobility and improving metal uptake into roots and shoots **(Zunaidi, A. *et al* 2024).**

The maximum permissible concentration of Cu and As in the edible parts of vegetables for human consumption is 40 mg/kg, 0.1 mg/kg dry weight respectively **(WHO/FAO 2007, EU 2006)**

The Cu and As levels in the vegetables presented in this study were below the maximum limits established by FAO/WHO.

By applying the independent samples t- test to compare the mean of two unrelated samples, we compared the average copper value between green lettuce leaves and lettuce stems. There were no statistically significant differences in the average copper value between the two studied categories, according to the probability value resulting from the test (p- value = 0.08), which is greater than the significance level (0.05). In the same way, by comparing the copper content of lettuce and watercress samples, there were no significant difference between mean copper concentrations in the two kinds of vegetables (p- value = 0.11). On the contrary, there was a significant difference between mean copper concentrations of both lettuce and watercress compared to maize (p- value< 0.05). This means there is evidence to suggest that lettuce and watercress can accumulate more levels of copper on average than maize.

In order to assess the intake of heavy metals and the extent of the risk from its consumption, the EDI and HQ calculation was adopted. As presented in table 5 EDI values of copper for lettuce leaves and lettuce stems were close, reaching 0.05 and 0.054 respectively, and the hazard quotient (HQ) of 1.25 and 1.45 exceeded the permissible limits. The EDI value of copper for watercress was 0.06 (mg/kg bw/day) and the HQ value was higher than 1 which suggest that consumption of watercress grown in this area may pose a significant health risk.

The EDI value of copper for maize was 0.0022 (mg/kg bw/day) and the HQ value was lower than 1 indicating that there is no risk to the health of the consumers of maize.

All HQ values for arsenic were less than one for all studied vegetables since the concentration of As in most vegetable samples was not detectable.

5. Conclusion:

The results obtained in this study show that most of the evaluated vegetables samples were contaminated by copper. The concentrations of copper in lettuce leaves, lettuce stems and watercress were generally higher than maize, which could indicate that maize doesn't accumulate heavy metals. The concentration of arsenic in all studied vegetable was either very low or not detectable.

It was found that the HQ to the heavy metal due to the consumption of lettuce and watercress were>1, for Cu, indicating a health risk over a life time exposure. This suggests the need for strict regulation and continuous monitoring of heavy metals in vegetables to ensure food safety and consumer health protection.

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