

**"Comparison of Heavy Metals in Red pepper packed and loose"**

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**Abstract**

Chili Hot pepper is considered a staple spice in Libyan cuisine. In recent years, interest in food safety from contaminants such as the spread of heavy metal contamination in food has increased. This study notes the scarcity of previous studies that have examined and analyzed Libyan red pepper. This study was conducted to compare the percentage of heavy metals in ground packaged and bulk Libyan chili peppers available in local markets in the city of Al-jumayl. The metals (Pb, Cd, Cu, Ni, Mn, and Zn) were estimated using a plasma-coupled optical emission spectrometer, and the results showed the following, the average concentration of Cd was 0.0001 mg/kg, while the average concentration of Cu was 0.0397-0.0600 mg/kg, the average concentration of Pb was 0.0297-0.0300 mg/kg, and the average concentration of Ni was 1.600-0.001 mg/kg. The results showed that all samples under study were below the permissible limits. As for the two elements (Zn, Mn), they were greater than the permissible limits at a rate of (27.10-15.80) mg/kg and (31.7-14.5) mg/kg, respectively, in the packed and loose samples. We recommend encouraging the Libyan community to consume Libyan spices.

**Keyworded: Spice, Red peppers, Heavy Metal, Food Contamination.**

**1.1. Introduction**

Herbs and spices have been an essential part in human life for thousands of years that is used at a domestic and industrial level as flavoring, preservation, and coloring agent in nutraceutical, pharmaceutical, and cosmetics products. (Dini 2018). Spices are organic food additives that have been used to improve the sensory quality of foods for thousands of years (Oladeji et al. 2023). Different parts of spices such as barks, leaf, roots, seeds and fruits are mainly used as flavoring agents in cooking process for the aroma, taste and color of food and sometimes mask undesirable odors (Harangozo et al. 2018).

Red hot pepper: Pepper (*Capsicum annum* L.) is one of the oldest and most widely grown crops in the world. The capsicum species have been used as a spice since 5500 BCE in prehistoric records.

Red peppers have long been used as food additives and as decorative plants. In addition, it is used as dried spices (whole fruits and powder); fresh, as multicolored, green, and red whole fruits in sauces, pastes, canning, and pickling (Batiha et al. 2020). Due to its antimicrobial, antitumor, immunosuppressive, and antioxidant properties, it is also used as a medicinal herb for pain management (Banerji et al. 2016). In addition, it has ant atherosclerotic, ant diabetic, and antihypertensive drug properties and helps to control dyspepsia, oxidative stress, inflammation, and body weight. (Baenas et al. 2019).

Red pepper carotenoids are beneficial to humans in a variety of ways, such as providing provitamin A and a natural color of food (Villa-Rivera et al. 2020). Oleoresins and ground pepper also enhance the taste and color of soups, stews, sausages, cheese, snacks, salad dressings, sauces, pizza, candies, and drinks (Arimboor et al.2015). Furthermore, oleoresin

contains vitamin C, phenolic compounds, proteins, fats, carbohydrates, dietary fiber, sodium, potassium, calcium, magnesium, iron, zinc, copper, pungent compounds, and manganese (Dobon-Suárez et al. 2021). Due to its essential function as an artificial food coloring, it serves as a source of income for many countries that produce red hot peppers. (Melgar-Lalanne et al. 2017). The fading of the color of red pepper during processing and storage is a significant threat to the quality of red-hot pepper products during storage due to several intricate factor (Bekele.2022) (El-Hamzy. et al 2016) (Tesfasilassie. et al 2017) (Dessie. et al 2024)(Ibsa et al 2019)

Heavy metal refers to chemical elements that have relatively high density and they are toxic even at low concentrations (Islam et al. 2023 Rahman et al., 2022. Vetri et al., 2019). Metals can exhibit either positive or negative impact in human health (Manousi et al.2022).

Recent studies have raised concerns about heavy metal concentrations in culinary ingredients posing potential health risks. High dietary exposure to heavy metals is associated with a range of health issues, including decreased immune defenses, cardiac malfunction, fetal malformation, impaired psychosocial and neurological behavior, and gastrointestinal cancer (Dghaim et al.2015). Heavy metals trigger inflammatory processes by releasing pro-inflammatory cytokines (Anka et al. 2022). Their presence in the diet has been shown to decrease beneficial gut microbiota, affecting mood and cognitive capacity (Porru et al., 2024). Additionally, case studies indicate increased mortality risks from cardiovascular and cerebrovascular diseases due to heavy metal exposure (Guo et al. 2022).

The process of packaging in food manufacturing is an important part of the processes carried out on food to contain it, facilitate its transportation and distribution, and sell it conveniently, and to preserve the nature and specifications of the food throughout the period between the moment of manufacture and packaging until the sale and consumption locally or after export, without the food item being spoiled it remains preserving its specifications.(Abu wali 2018).

### **1.2. Study problem:**

In recent years, there has been an increasing interest in studying food contamination with heavy metals because they cause many diseases. The use of spices has increased dramatically in most parts of the world. This is largely due to their medicinal properties. As a result of the high prevalence of heavy metals in the environment, heavy metal residues may also reach spices. To avoid toxicity in humans, it is important to monitor the levels of toxic metals present at each step of the food chain.

### **1.3. Study importance:**

The importance of the study stems from the importance of maintaining consumer safety and product quality, as heavy metals cause food contamination and negatively impact public health. In addition, the use of heavy metals in the spice industry can help develop safe and effective metal extraction technologies to ensure the future of the spice industry without negative impact on health or the environment. It is essential to ensure the safety of food consumed by the consumer, and therefore the levels of heavy metals in spices must be monitored to ensure that safe limits are not exceeded.

### **1.4. Study Objectives:**

1. Determination and estimation of heavy metals concentrations in red packed and loose pepper samples.
2. Study the impact of discovered heavy metals on general health and their effect on the body.
3. Comparison of heavy metals in packaged and loose red pepper.

### **1.5. Study hypotheses:**

1. There are no statistically significant differences at the significance level of 0.05 in the mean concentrations of heavy metals that can be attributed to the type of red pepper under study.
2. There are no statistically significant differences at the 0.05 level in the concentrations of heavy metals between packaged and unpackaged red pepper.

### 1.8. Previous studies:

#### 1.first study: Mohammed Wafaa: et al (2021) investigation of the presence of some heavy metals and microbes in spices.

The study included collecting 11 types of spices sold in the local markets of Curcuma city, curry, turmeric (*Coriandrum sativum* L.) Tikrit, coriander (*Piper nigrum*), black pepper (*Zingiber officinale*), ginger (*Longa R. coriaria*), sumac (*cinnamomum ze ylanicum*), pickle spices, biryani spices, black seed (*Nigella sativa*), cinnamon and cumin (*Cuminum Cyminum*) to investigate the presence of some types of heavy elements and identify the microorganisms that contaminate them. The results showed that the concentration of some heavy elements was within the permissible limits according to the standard specifications set by the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) to ensure that the spices were free of cadmium and lead, while zinc, nickel and cobalt were found in different concentrations in the other different types of spices. The concentration of nickel was higher than the permissible limit in the samples of black pepper, cinnamon and turmeric, reaching (4.959, 3.850, 2.740 ppm respectively, while the concentration of nickel in the rest of the samples was less than 0.05 ppm, which is less than the permissible limit. The concentration of cobalt was also higher than the permissible limit in the samples of black pepper and curry, reaching (4.930 and 3.732 ppm respectively, while the rest of the spice samples were less than the permissible limit, while it was found that the zinc content of all spice samples was below the permissible limit (100) ppm according to (FAO) and (WHO). The values ranged between 1.794 ppm and 0.392 ppm. The total bacterial count in the spices ranged between (5) 510 and (95×510) colony forming units/g, while the presence of *Escherichia coli* was observed in all the tested samples in numbers ranging between (93×510 35×510) colony forming units/gm while the number of *Staphylococcus aureus* bacteria ranged from 1×510 to 17×510) colony forming units/gm, with the appearance of yeasts and molds observed in each of coriander, ginger, biryani spices, cinnamon, pickles, cumin, black pepper, curry and turmeric spices in numbers of 105×5, 105×14, 105×6, 105×12, 105×10, 105×12 (reaching 2×10<sup>5</sup>, 7×10<sup>5</sup> and 5106 colony forming units/gm respectively).

#### 2. The second study : Dessie Ezez: et al (2024) Bioaccumulation of heavy metals assessment of carcinogenic and non-carcinogenic health risk in various spices

Spices consumption is the main pathway for human exposure to certain environmental pollutants. This study was aimed to evaluate the concentration of heavy metals such as cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), manganese (Mn), lead (Pb) and zinc (Zn) in different parts of various spices samples in Arba Minch Ethiopia. The average levels of metals in white cumin, fenugreek, black cumin, turmeric, basil, cardamom, and coriander were varied in the range 0.35–1.8, 1.02–11.96, 1.69–32.19, 20.74–38.98, 7.38–26.01, 2.96–9.57 and 5.38–44.63 mg/kg for Cd, Cr, Cu, Fe, Mn, Pb and Zn, respectively. Principal component analysis (PCA) showed that the robust linkage between Zn, Cr, and Mn in PC1, Cu and Pb in PC2, and Cd and Fe in PC3 comprising 39.10 %, 25.72 % and 18.57 % of the total variance, respectively. The average daily intake values were calculated in the range  $5.22 \times 10^{-5}$ – $2.69 \times 10^{-4}$ ,  $1.52 \times 10^{-4}$ – $1.78 \times 10^{-3}$ ,  $2.53 \times 10^{-4}$ – $4.81 \times 10^{-3}$ ,  $3.09 \times 10^{-3}$ – $5.82 \times 10^{-3}$ ,  $1.1 \times 10^{-3}$ – $3.88 \times 10^{-3}$ ,  $4.42 \times 10^{-4}$ – $1.43 \times 10^{-3}$ , and  $8.18 \times 10^{-4}$ – $6.66 \times 10^{-3}$ , for Cd, Cr, Cu, Fe, Mn, Pb and Zn, respectively. The target hazard quotient (THQ) and hazard index (except in white cumin and fenugreek) for heavy metals in various spices were lower than the acceptable limit of unity (THQ and HI 1). The carcinogenic risk values were in the acceptable threshold limit ( $1 \times 10^{-6}$ – $1 \times 10^{-4}$ ), indicating that consuming different spices in the studied area will not pose a potential health risk to human health.

#### 3.Third study: AL mariami mohmoud: et al (2019) Estimation of some heavy metals in spices available in local markets in Wadi Al-Shati area, southern Libya.

In recent years, there has been increased interest in the safety of food products from harmful pollutants, including food contamination with heavy metals. Spices are substances added to food to improve its sensory properties. These spices may contain some harmful metals, while others, if their concentration exceeds certain limits, cause many health problems for humans. Therefore, this study was conducted on some samples of spices sold in the markets in the Wadi al-Shati area in southern Libya, in order to determine their concentration of some heavy metals. The following spices included ginger, turmeric, caraway, black pepper, black cumin, red pepper, cumin, cinnamon, and coriander. The study showed a variation in the concentration of metals in the spice samples, as the highest concentration of iron was recorded in the caraway sample at 255 mg/kg, while the highest concentration of zinc was in the cumin sample at 78 mg/kg. As for cadmium, the highest concentration was in the cumin sample at 3.2 mg/kg, while the highest concentration of lead was recorded in the coriander sample at 32 mg/kg, and the highest concentration of manganese was recorded in the coriander sample at 76 mg/kg. kg, and some samples exceeded the maximum permissible limit according to the standard specifications of the World Health Organization and the Food and Agriculture Organization.

## **2. Methodological Procedures:**

### **2.1. Study Methodology:**

Since the aim of the study is measurement of heavy metals in red pepper packaged and loose in the city of Al-jumayl the researchers relied on a descriptive analytical approach due to its suitability for the nature and objectives of the study. The following data were utilized:

**2.2. Chemical Analysis:** The necessary chemical analyses to fulfill the study requirements, which allow for the identification of heavy metal concentrations in the selected red pepper for this study, include the following elements: Cd, Cu, Mn, Ni, Pb, Zn.

### **2.3. Study Sample:**

The study sample consisted of a group of samples of three types of packed red pepper and samples of two groups of loose red peppers from the markets of Al-jumayl city, this study extended over the November 2024 to May 2025

## **3. Materials and Methods:**

### **3.1. Laboratory Analyses:**

The following devices were used to conduct all necessary tests for the study in the petroleum study center (in Tripoli).

### **3.2. Devices Used:**

- Sensitive balance picture
- High performance microwave digestion system picture
- Inductively coupled plasma optical emission spectrometer picture

### **3.3. Samples Used**

#### **1. Sampling preparation: (Red pepper)**

- Code the samples.
- Take a quantity of samples to facilitate the analysis procedure.
- Wash the spoon with soap and water, then rinse it with diluted nitric acid followed by distilled water, to avoid contaminating the sample with metallic impurities which would produce inconsistent results. This step is repeated for all samples.

#### **2. Sample Digestion Stage: sample weight to 0.5g**

- This method involves acid digestion of the sample in a closed vessel apparatus using microwave heating to control the temperature for Inductively coupled plasma optical emission spectrometer identification of the metal. (Farrell, et al. 2010).

Procedure:

1. Place Tetrafluoromethoxylated (TFM) vessel on the balance plate, tare it and weigh of the sample.
2. Introduce the TFM vessel into the High Temperature Combustion (HTC) safety shield.

3. Add the acids in part of the sample stays on the inner wall of the TFM vessel, wet it by adding acids drop, then gently swirl the solution to homogenize the sample with the acids.
4. Close the vessel and introduce it into the rotor segment, then tighten by using the torque wrench.
5. Insert the segment into the microwave cavity and connect the temperature sensor.
6. Run the microwave program to completion.
7. Cool the rotor by air or by water until the solution reaches room temperature.
8. Open the vessel and transfer the solution to a marked flask.

### 3. Heavy Metal Estimation Stage:

Extensive analyses using inductively coupled plasma emission spectrometry are being conducted at oil research center in Tripoli

### 3.4. Statistical processing methods used:

The statistical methods that are consistent with the nature of the study, which are as follows:

- Arithmetic means and percentage differences for chemical analyses.
- Test one way a nova
- Post hoc test and turkey test
- Test (independent T-test)

## 4. Results and Discussion

All results obtained through the chemical analyses related to the study topic will be presented, discussed, and analyzed in order to answer the study hypotheses:

### 1. The extent to which the concentrations of heavy metals in the pepper samples compliance with the specified standards.

→ Element Cadmium (Cd)

**Table (4.1):** Descriptive statistics for the element (Cd)

Variable	Red Pepper. Type	Mean ± St.d
Cd	A	0.001± 0.0001
	B	0.00009± 0.000015
	C	0.000011± 0.000006
	D	0.0001± 0.00000
	E	0.0001± 0.00003

Based on Table (4.1) above, the following can be observed:

The average concentration of the element (Cd) in all types of red pepper under study is below the permissible limit, which is set at 0.032 mg/kg to Based on the Libyan specifications and standards. The following figure illustrates this.

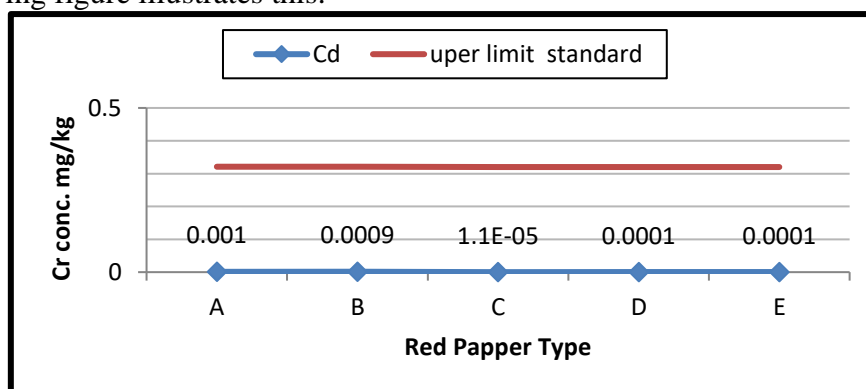


Figure (4.1): Comparison of (Cd) concentration with the permissible limits

This is consistent with a previous study by (Amrimi Mahmoud et al. 2019)

→ Element Cooper (Cu)

Table (4.2): Descriptive Statistics for the Element (Cu)

Variable	Red Pepper. type	Mean $\pm$ St.d
<u>Cu</u>	<u>A</u>	<u>0.0397 <math>\pm</math> 0.00896</u>
	<u>B</u>	<u>0.0400 <math>\pm</math> 0.01000</u>
	<u>C</u>	<u>0.0400 <math>\pm</math> 0.00000</u>
	<u>D</u>	<u>0.0010.0600 <math>\pm</math> 0.0</u>
	<u>E</u>	<u>0.0500 <math>\pm</math> 0.01113</u>

Based on Table (4.2) above, the following can be observed:

The average concentration of the element (Cu) in all types of red pepper under study is below the permissible limits, which range from 3.8 to 35.4 mg/kg, Based on what is stated according to Libyan standards. The following figure illustrates this.

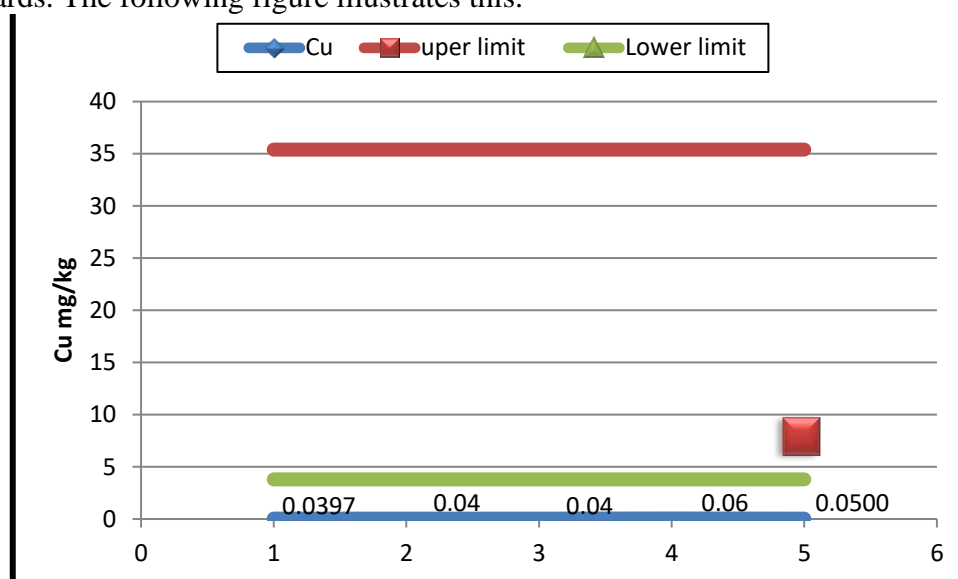


Figure 4. (2): Comparison of (Cu) concentration with the permissible limits

→ Element Manganese (Mn)

Table (4.3): Descriptive Statistics for the Element (Mn)

Variable	Red Pepper. type	Mean $\pm$ St.d
<u>Mn</u>	<u>A</u>	<u>14.500 <math>\pm</math> 0.2000</u>
	<u>B</u>	<u>31.700 <math>\pm</math> 0.2000</u>
	<u>C</u>	<u>15.800 <math>\pm</math> 0.1000</u>
	<u>D</u>	<u>16.900 <math>\pm</math> 0.1000</u>
	<u>E</u>	<u>25.700 <math>\pm</math> 0.2000</u>

Based on Table (4.3) above, the following can be observed:

The average concentration of the element (Mn) in all types of red pepper under study is above the permissible limits, which are set at 3.0 to 3.5 mg/kg, Based on what is stated according to Libyan standards. The following figure illustrates this.

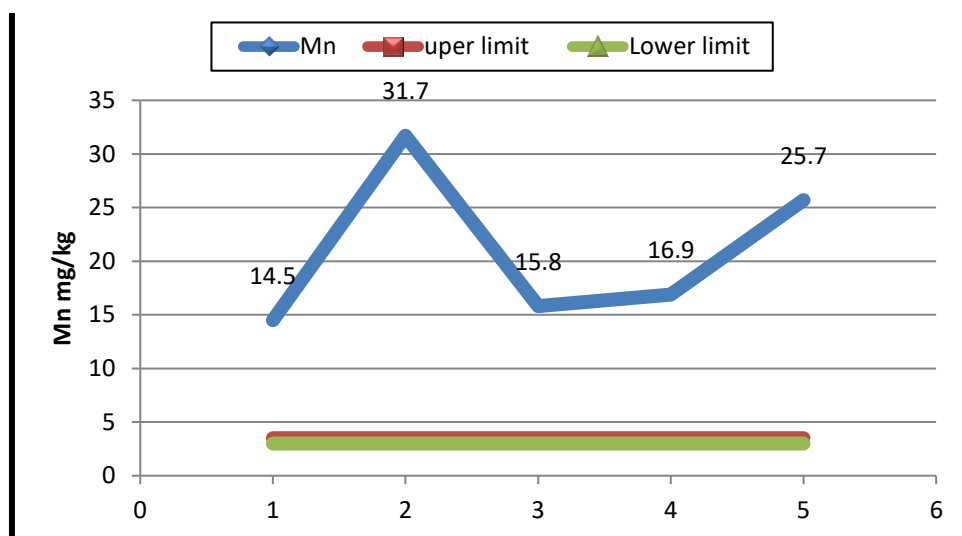


Figure (4.3): Comparison of (Mn) concentration with the permissible limits  
 The results of this study do not agree with the previous study by Al-Mariami Mahmoud et al. (2019), where the concentration of the element was 64 mg/kg.

→Element Nical (Ni)

Table (4.4): Descriptive Statistics for the Element (Ni)

Variable	Red Pepper. type	Mean ± St.d
Ni	A	1.6000± 0.10000
	B	0.001± 0.0001
	C	1.5000 ± 0.1000
	D	0.0001± 0.0001
	E	0.001± 0.0001

Based on Table (4.4) above, the following can be observed:

The average concentration of the element (Ni) in the studied samples indicates that all samples were below the permissible limits, except for samples (A) and (C), which exceeded the lower threshold of the allowable range set at 1.4 to 11.3 mg/kg, according to Codex standards. The following figure illustrates this

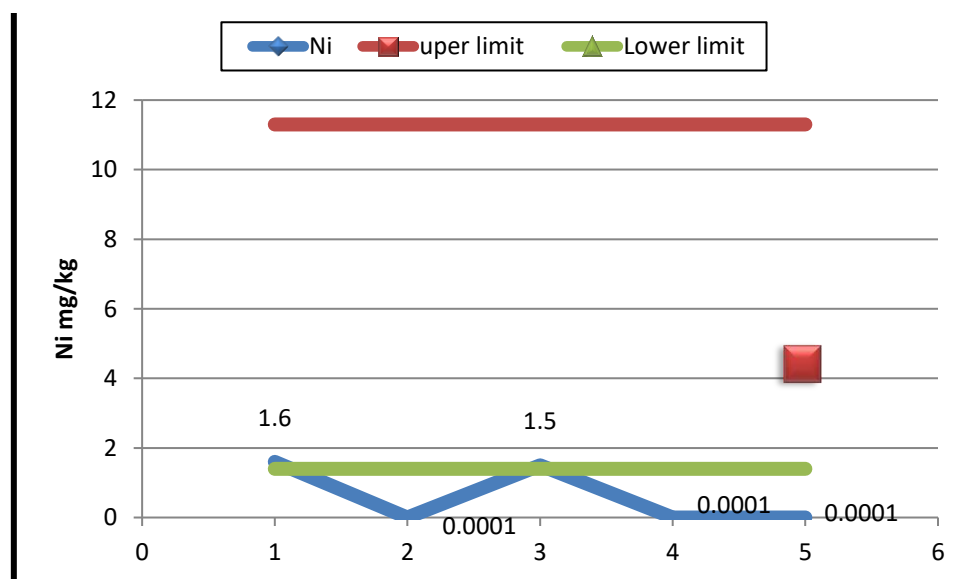


Figure (4.4): Comparison of (Ni) concentration with the permissible limits  
 → Element Lead (Pb)

Table (4.5): Descriptive Statistics for the Element (Pb)

Variable	Red Pepper. type	Mean ± St.d
Pb	<u>A</u>	<u>0.0260± 0.0053</u>
	<u>B</u>	<u>0.025 0.0277± 0.</u>
	<u>C</u>	<u>0.0297± 0.0058</u>
	<u>D</u>	<u>0.0300± 0.0100</u>
	<u>E</u>	<u>0.0247± 0.0050</u>

Based on Table (4.5) above, the following can be observed:  
 The average concentration of the element (Pb) in the studied samples indicates that all samples were below the permissible limit, which is set at 0.96 mg/kg according to Libyan standards.  
 The following figure illustrates this.

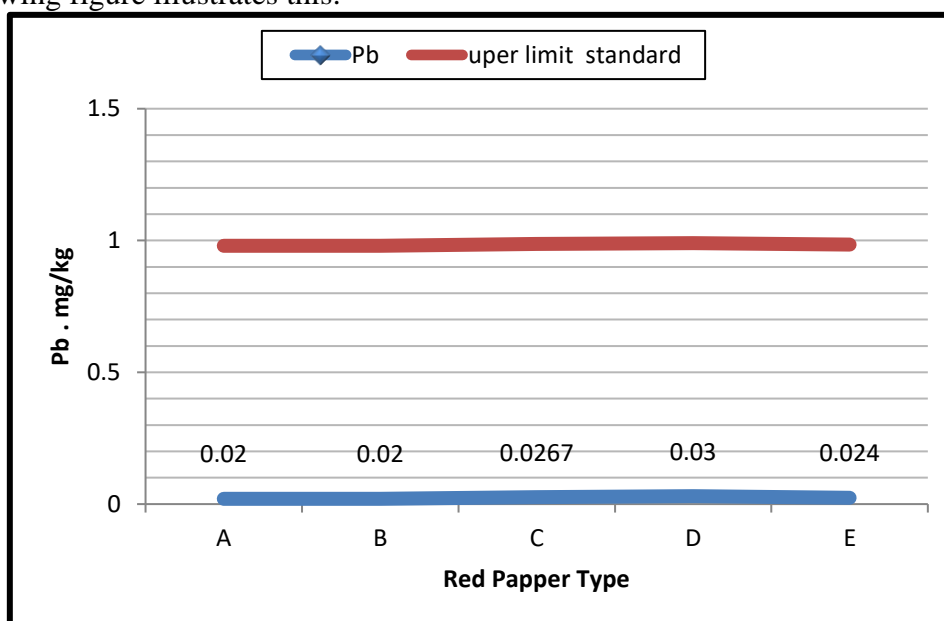


Figure (4.5): Comparison of (Pb) concentration with the permissible limits.  
 The results of this study were consistent with the previous study by Amrimi Mahmoud et al. (2019), where the concentration of lead was below the permissible limits:  
 → Element Zinc (Zn)

Table (4.6): Descriptive Statistics for the Element (Zn)

Variable	Red Pepper. type	Mean ± St.d
Zn	<u>A</u>	<u>19.40 ± 0.200</u>
	<u>B</u>	<u>27.10 ± 0.100</u>
	<u>C</u>	<u>15.80± 0.200</u>
	<u>D</u>	<u>16.90 ± 0.100</u>
	<u>E</u>	<u>25.70 ± 4.761</u>

Based on Table (4.6) above, the following can be observed:  
 The average concentration of the element (Zn) in the studied samples indicates that all samples exceeded the permissible limits, which are set at 0.056 to 0.895 mg/kg according to Codex standards.  
 The following figure illustrates this.

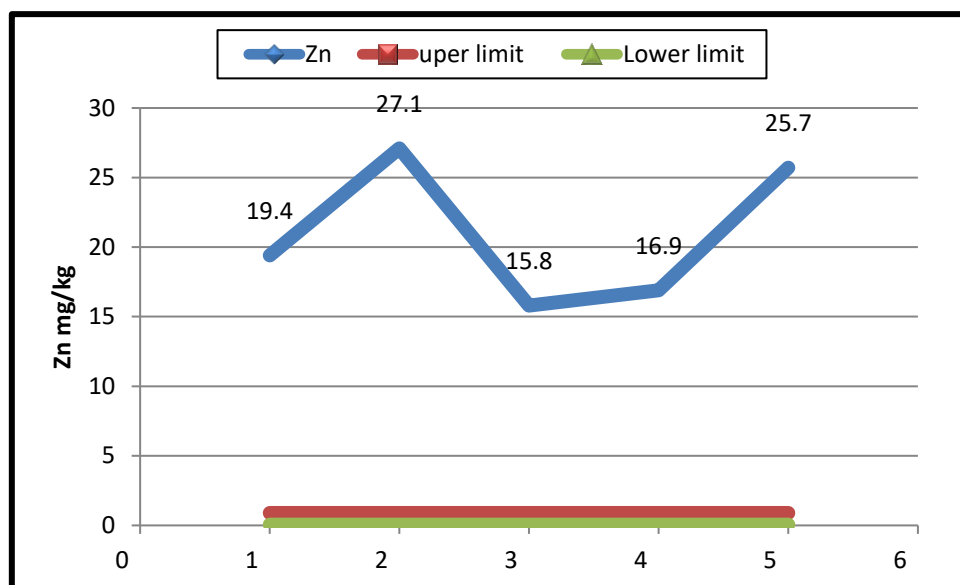


Figure (4.6): Comparison of (Zn) concentration with the permissible limits

This is consistent with a previous study by Amrimi Mahmoud et al. (2019) in types B and E, where the level of this element was (28.4mg/kg).

Thus, the variation in the content of these heavy metals in spices can be attributed to the location of the heavy metal storage in the parent plant. Some plants have storage capacity in the roots, while others allow the element to move through their tissues to the green parts of the plant, where it may reach the seeds and be stored, or be stored in the stem and leaves. For example, (Chu et al. 2018) indicated that copper and cadmium are poorly mobile throughout the plant, and therefore are stored in greater quantities in the roots. This, of course, does not apply to all plants, as (Kozlov et al. 2000) indicated. Zinc, on the other hand, behaves differently, as it has the ability to move easily from the roots to the leaves and flowers of the plant, where it is stored (Kiekens, 1990).

**2. There are no statistically significant differences at the significance level of 0.05 in the mean concentrations of heavy metals that can be attributed to the type of red pepper under study.**

based on the One-Way ANOVA statistical analysis tool. The hypothesis can be rejected if ( $P\text{-Value} \leq \alpha$ ), where  $\alpha = 5\%$ , as shown below:

- ANOVA Analysis Table

Table (7): Results of the ANOVA Statistical Test

ANOVA						
		Sum of Squares	Df	Mean Square	F	P-value.
Cd	Between Groups	.000	4	.000	.313	.863
	Within Groups	.000	10	.000		
	Total	.000	14			
Cu	Between Groups	.001	4	.000	3.195	.062
	Within Groups	.001	10	.000		

	Total	.002	14			
Mn	Between Groups	667.944	4	166.986	5963.786	.0001**
	Within Groups	.280	10	.028		
	Total	668.224	14			
Ni	Between Groups	8.663	4	2.166	541.432	.0001**
	Within Groups	.040	10	.004		
	Total	8.703	14			
Pb	Between Groups	.000	4	.000	.773	.567
	Within Groups	.001	10	.000		
	Total	.001	14			
Zn	Between Groups	317.124	4	79.281	3603.682	.0001**
	Within Groups	.220	10	.022		
	Total	317.344	14			

\*\*Significant at 1%

Based on the previous table, it is evident that the P-values for the elements (Mn, Ni, Zn) are 0.000, which are less than 0.05. This indicates that there are statistically significant differences at the 0.05 significance level in the mean concentrations of the heavy metals (Mn, Ni, Zn) that can be attributed to the variation in the types of red pepper under study, and that each element has a distinct effect on the others.

- Using the Post Hoc Test

The Tukey statistical tool was used in order to identify the most significant pairwise differences among the means, and to determine which comparisons show the strongest associations and effects. The following tables illustrate this:

→ Element (Mn)

**Table (8): Results of the Tukey Post Hoc Test for the element (Mn) in the samples under study**

(I) type	(J) type	Mean Difference (I-J)	Std. Error	P-value.
A	B	-17.2000*	0.1366	0.000
	C	-1.3000*	0.1366	0.000
	D	-2.4000*	0.1366	0.000
	E	-11.2000*	0.1366	0.000
B	A	17.2000*	0.1366	0.000
	C	15.9000*	0.1366	0.000
	D	14.8000*	0.1366	0.000
	E	6.0000*	0.1366	0.000
C	A	1.3000*	0.1366	0.000

	B	-15.9000*	0.1366	0.000
	D	-1.1000*	0.1366	0.000
	E	-9.9000*	0.1366	0.000
D	A	2.4000*	0.1366	0.000
	B	-14.8000*	0.1366	0.000
	C	1.1000*	0.1366	0.000
	E	-8.8000*	0.1366	0.000
E	A	11.2000*	0.1366	0.000
	B	-6.0000*	0.1366	0.000
	C	9.9000*	0.1366	0.000
	D	8.8000*	0.1366	0.000

\*Significant at 5%

Using the Tukey Post Hoc Test for multiple comparisons, it was found that all pairwise differences between the groups were statistically significant ( $p = 0.000$ ). This indicates that each group differs significantly from the others in terms of Mn concentration.

Upon analyzing the direction of the differences based on the means derived from the comparisons, the order of the groups from lowest to highest manganese concentration was as follows:

$$A < C < D < E < B$$

This means that Group B had the highest Mn concentration, while Group A had the lowest. However, it is important to note that all values exceeded the acceptable health limit.

According to the standard specifications (which define the acceptable Mn concentration in pepper as 3.0–3.5 mg/kg), all samples exceeded this limit, which constitutes a violation of safety standards and represents a potential health risk, especially with frequent or heavy consumption.

→ Element (Ni)

**Table (9): Results of the Tukey Post Hoc Test for the element (Ni) in the samples under study**

(I) type	(J) type	Mean Difference (I-J)	Std. Error	P-value.
A	B	1.59990667*	0.05163978	0.000
	C	.10000000	0.05163978	0.359
	D	1.59990000*	0.05163978	0.000
	E	1.59990000*	0.05163978	0.000
B	A	-1.59990667*	0.05163978	0.000
	C	-1.49990667*	0.05163978	0.000
	D	-.00000667	0.05163978	1.000
	E	-.00000667	0.05163978	1.000
C	A	-.10000000	0.05163978	0.359
	B	1.49990667*	0.05163978	0.000
	D	1.49990000*	0.05163978	0.000
	E	1.49990000*	0.05163978	0.000
D	A	-1.59990000*	0.05163978	0.000

	B	.00000667	0.05163978	1.000
	C	-1.49990000*	0.05163978	0.000
	E	.00000000	0.05163978	1.000
E	A	-1.59990000*	0.05163978	0.000
	B	.00000667	0.05163978	1.000
	C	-1.49990000*	0.05163978	0.000
	D	.00000000	0.05163978	1.000

\*Significant at 5%

The ANOVA test results indicated that there are significant differences in the mean concentration of nickel (Ni) among the different types of ground red pepper ( $p < 0.001$ ).

Using the Tukey HSD Post Hoc Test for pairwise comparisons, it was found that Groups A and B had higher mean concentrations than the other groups, with statistically significant differences ( $p < 0.001$ ) when compared to Groups C, D, and E.

However, Groups C, D, and E showed no significant differences among each other ( $p > 0.05$ ), indicating a similar level of nickel concentration within these samples. Analyzing the direction of the differences based on the derived means, the ranking of groups from lowest to highest nickel concentration was as follows:

$$A \approx B > C \approx D \approx E$$

According to the permissible health limits for nickel concentrations in ground red pepper (1.4 – 11.3 mg/kg):

- Groups A and B fall within the acceptable range, indicating they are safe for consumption.
- Meanwhile, Groups C, D, and E fall below the minimum recommended level (*less than 1.4 mg/kg*), which may suggest a deficiency of nickel in these samples, possibly due to soil conditions or processing factors.

→ Element (Zn)

**Table (10): Results of the Tukey Post Hoc Test for the element (Zn) in the samples under study**

(I) type	(J) type	Mean Difference (I-J)	Std. Error	P-value.
A	B	-7.70000*	0.12111	0.000
	C	3.60000*	0.12111	0.000
	D	2.50000*	0.12111	0.000
	E	-6.30000*	0.12111	0.000
B	A	7.70000*	0.12111	0.000
	C	11.30000*	0.12111	0.000
	D	10.20000*	0.12111	0.000
	E	1.40000*	0.12111	0.000
C	A	-3.60000*	0.12111	0.000
	B	-11.30000*	0.12111	0.000
	D	-1.10000*	0.12111	0.000
	E	-9.90000*	0.12111	0.000
D	A	-2.50000*	0.12111	0.000

	B	-10.20000*	0.12111	0.000
	C	1.10000*	0.12111	0.000
	E	-8.80000*	0.12111	0.000
E	A	6.30000*	0.12111	0.000
	B	-1.40000*	0.12111	0.000
	C	9.90000*	0.12111	0.000
	D	8.80000*	0.12111	0.000

Using the Tukey Post Hoc Test for pairwise comparisons, it was found that all differences between group pairs were statistically significant ( $p = 0.000$ ). This indicates that each group significantly differs from the others in terms of Zn concentration.

Upon analyzing the direction of the differences based on the derived means, the order of the groups from lowest to highest zinc concentration was as follows:

$$C < D < A < E < B$$

This means that Group B had the highest Zinc concentration, while Group C had the lowest. However, all values exceeded the acceptable health limits.

According to the standard specifications, which define the acceptable Zn concentration in pepper as (0.056–0.895) mg/kg, all samples exceeded this limit, constituting a violation of the specifications and a potential health risk, particularly with frequent or heavy consumption.

### 3. There are no statistically significant differences at the 0.05 level in the concentrations of heavy metals between packaged and unpackaged red pepper.

Using the Independent Sample T-Test, the null hypothesis can be rejected if ( $P\text{-Value} \leq \alpha$ ), where  $\alpha = 5\%$ , as follows:

**Table (11): Results of the Independent Sample T-Test Statistical Analysis**

		F	Sig.	T	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference
Cd	Equal variances assumed	.053	0.821	0.000	13	1.000	.00000000	0.00000716
	Equal variances not assumed			0.000	7.977	1.000	.00000000	0.00000778
Cu	Equal variances assumed	2.151	0.166	3.425	13	0.005	.01511	0.00441
	Equal variances not assumed			3.127	7.746	0.015	.01511	0.00483
Mn	Equal variances assumed	6.420	0.025	0.168	13	0.869	.6333	3.7746
	Equal variances not assumed			0.187	12.873	0.855	.6333	3.3943
Ni	Equal variances assumed	37.714	0.000	-3.207	13	0.007	-1.03326444	0.32223251
	Equal variances not assumed			-3.977	8.000	0.004	-1.03326444	0.25979215
Pb	Equal variances assumed	.015	0.905	1.020	13	0.326	.00444	0.00436
	Equal variances not assumed			1.024	11.031	0.328	.00444	0.00434
Zn	Equal variances assumed	.037	0.850	.205	13	0.841	.53333	2.59980
	Equal variances not assumed			.207	11.159	0.840	.53333	2.57930

Based on the previous table, the results showed that there were statistically significant differences at the 0.05 significance level in the concentrations of nickel (Ni) and copper (Cu) attributable to the type of packaging (packaged vs. unpackaged), as the P-values were less than 0.05. However, no significant differences were observed for the other elements. This suggests the following:

- The difference in nickel and copper concentrations may indicate that the type of packaging, storage conditions, or the source of raw material has a clear influence on the accumulation of these two elements.
- The absence of significant differences for the other elements may suggest that the sources of the pepper are similar, or that these elements are stable under different packaging conditions.

### 5.1. Conclusions:

Through this study, it was observed that previous study on Libyan red pepper is scarce, highlighting the importance of this study in bridging this knowledge gap and providing precise scientific data that can be utilized in the fields of agriculture, food industries, and genetic studies of local plants.

1. The concentration of heavy metals in the study sample was lower than the permissible limits, except for manganese and zinc, which were higher than the permissible limits.
2. There were statistically significant differences at the significance level (0.05) between the average concentrations of heavy metals (Mn, Ni, and Zn) attributed to the different types of peppers under study, and that each had a different effect on the other.
3. Statistically significant differences at the significance level (0.05) in the concentrations of nickel (Ni) and copper (Cu) were attributed to the type of packaging (packaged or loose), with a of (p-value > 0.05), while no significant differences were recorded for the remaining elements.

### 5.2. Recommendations:

1. Encourage the Libyan community to consume Libyan spices.
2. Hold community awareness seminars on the health risks associated with increased metal concentrations in imported spices.
3. Conduct further study on the percentage of heavy metals in Libyan spices and compare them with imported spices.
4. Contact the Libyan national center for standards and metrology with the search results.

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