*Mesopotamia Journal of Agriculture, Vol. 51, No. 3, 2023 (56-66)*

Mesopotamia Journal of Agriculture

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# EVALUATION OF SOME QUALITY CHARACTERISTICS OF FLAXSEED IN RESPONSE TO FOLIAR APPLICATION OF ZINC AND MANGANESE

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Article information

Article history:

Received: 14/ 06/2023

Accepted: 19/09/2023

Available: 30/09/2023

***Keywords***:

*Flaxseed oil, Zn, Mn, fatty acids, cluster analysis.*

DOI:

http://10.33899/mja.2023.141085.1250.

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**ABSTRACT**

This research was conducted at Grdarsha Field, College of Agricultural Engineering Sciences, Salahaddin University, Erbil with a GPS reading of (Latitude: 36° 4' N and Longitude: 44° 2' E- elevation 415 Meters above sea level). The factorial experiment based on a randomized complete block design using three replicates was done to study the influence of three levels (0, 200, and 400 mg L-1) of each of Zinc and Manganese as foliar application and their combination on their concentration, uptake and fatty acids profile in flaxseed. The results indicate that the Zinc application effect on Zn and Mn concentration with values (0.0032 and 0.0068) % in the treatment (Zn200 and Mn200) additionally caused an increase in the uptake for the two micronutrients in Zn400 and Mn200 with values (15.74 and 14.90) mg plant-1. The treatment combination Zn200 Mn400 caused an increase in palmitic, oleic, linoleic and linolenic acid while the treatment combination of Zn400 Mn200 gained the lowest value. Cluster analysis or dendrogram classified the combination of the two micronutrients Zn and Mn into four main clusters depending on Zn, Mn and the fatty acids profile. The Principal Component Analysis (PCA) shows the angle value between each of the fatty acids and between the two micronutrients is ≤ 90 ° which means there was a significant correlation between them and vice versa. There is a very high correlation between linoleic acid & linolenic and F6 while F7 correlated with oleic, stearic and palmitic acid for the vectors for studied traits.



## INTRODUCTION

Flaxseed is a major commercial crop that impacts regional policy via both domestic producers and exporting (Kaithwas and Majumdar, 2013). It is regarded as a vital and economical food supply as they are high in alpha-linolenic acid (ALA, omega-3 fatty acid), a beneficial fatty acid for a variety of health issues such as heart disease, inflammation and arthritis, among others (Goyal *et al.*, 2014). Flaxseed is used in a wide range of products and industries as it provides a variety of nutrients, including proteins, fiber and fatty acids. It contains 41% oil, 20% protein and 28% dietary fiber and fatty acids (50-60%) comprising both linoleic and alpha-linolenic acid (Oomah, 2001). Furthermore, oil offers specific properties for other sectors, such as the manufacture of different types of paints (Mahmood and Sarkees, 2014).

Khalifa *et al.* (2011), stated that flaxseed production has dropped owing to fierce competition from other crops such as wheat, barley and others. As a result, there is a significant shortfall between production and consumption, particularly in seed yield. Potentially, it can be narrowed by employing improved agricultural methods such as efficient and appropriate fertilization (Keram *et al.*, 2012). Flax is vulnerable to micronutrient deficits, particularly in calcic soils, because of the high pH and precipitation of these elements in carbonic and hydroxyl forms, resulting in limited solubility. Amberger, (1991), cleared that the availability of micronutrients such as Fe, Mn, Zn and Cu are much affected by pH, $CaCO_3$ concentration, organic matter and soil texture which means an increase in pH and $CaCO_3$ causing a decrease in their availability, while the increase in soil organic matter and clay concentration causing an increase in their availability. Numerous studies demonstrate that foliar application of certain micronutrients might alleviate this problem, as foliar treatment enhances flaxseed quality and yield (Esmail, 2018). Zinc is a cofactor in enzyme synthesis, and activation and influences electron transport, it is a critical element in glucose metabolism and protein formation in plants (Martin *et al*., 2006 and Brown *et al*., 1993). Manganese is necessary for several plant physiological and enzymatic functions such as Chlorophyll production, photosynthesis and cofactors of more than photosynthesis in addition to cell elongation and division and fatty acid formation (Diedrick, 2010 and Hakala *et al*., 2006) .

Recent studies in soils with alluvium have demonstrated that applying some micronutrients, particularly Zn and Mn, as foliar applications have a significant impact on plants (Al-Doori, 2021). Foliar fertilizer is therefore the most efficient way to use fertilizer, particularly micronutrients. Because of the rapid deficiency treatment, relatively reduced cost and effective usage (Gul *et al*., 2011) and the olvability of micronutrient is less and cause decline uptake micro elements, so finally the plants requirement to this element will increase (Mousavi, 2011). The effect of available Mn and Zn concentration in plants and the correlation between them in plants is rarely studied, so this study aimed to evaluate the oil quality in response to some micronutrients applied as a foliar spray application of flaxseed (*Linum usitatissimum L*.).

## MATERIALS AND METHODS

A field experiment was conducted at Grdarasha Field, Agricultural Engineering Sciences College/ Salahaddin University- Erbil, Kurdistan Region/Iraq, to investigate the effects of three levels (0, 200, and 400) mg L-1 for each of Zn and Mn as a foliar application on some micronutrients, uptake by seeds and fatty acids of flaxseed oil extracted. A factorial experiment based on Randomized Complete Block Design (RCBD) using three replications. Soil samples were taken randomly from the upper 30 cm and analyzed table (1).

Nutrient uptake = concentration *weight of seeds

The experiment unit area was 4m2 each replicate consisted of nine experimental units, recommended fertilizer urea (46% N) at a rate of 100 kg N ha-1 and triple superphosphate (46% P2O5) at a rate of 80 kg ha- 1 was applied at sowing time (Esmail et al. ,2017).

Table (1): Some physical and chemical properties of soil at depth (0 - 30 cm)

| Soil Properties | Particle size distribution | | | Texture | pH | EC | O.M | Total N | P | K | Fe | Zn |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Sand | Silt | Clay | Clay loam | 7.43 | dS m-1 | % | | ppm | | | |
| | % | | | | | | | | | | | |
| 0-30 cm | 28.3 | 32.5 | 39.2 | | | 0.80 | 0.90 | 0.18 | 9.50 | 240 | 2.10 | 3.00 |

Flaxseed (Lider) genotypes obtained from Agricultural Research Center in Erbil - Iraq were manually sown on 1st November 2019 at a row spacing of 10 cm and plant spacing of 5 cm, at a depth of 3 cm. The thinning practice was done to obtain 200 plants m-2, and foliar application of Zn and Mn was done twice, 30 days from sowing and at the flowering stage.

Furthermore, the seeds of five plants in each experimental unit were randomly selected to study the chemical concentration and the fatty acids estimation was done. The oil was estimated using the Soxhlet apparatus for oil extraction as mentioned by (A.O.A.C, 1995).

The fatty acid compounds were analyzed using a gas chromatography device (GC-2010), a Japanese Shimadzu model. With the GC a Flame Ionized Detector (FID) was used and a capillary separation column type (SE-30) with lengths of (30m * 0.25 mm) was used (Zhang *et al.*, 2015) according to the following conditions in the table (2).

Table (2): Conditions of fatty acid analysis using GC

| S | Section Name | Temperature |
|---|---|---|
| 1 | Temperature of the injection area | 280C |
| 2 | The detector temperature | 310C |
| 3 | Temperature of separation column | 120 - 290 (10C/min) |
| 4 | Gas flow rate | 100 Kpa |

**Statistical Analysis Using Agglomerative Hierarchical Clustering (AHC) and Principal Components Analysis (PCA)**

Statistical charts and spider charts were drawn using Excel computer software package. cluster analysis was conducted between chemical components in flax cultivars for grouping them to different classes using XLSTATE- premium program to obtain homogenous groups by Agglomerative Hierarchical Clustering (AHC) and Principal Components Analysis (PCA), to show the similarity between them (Urdan, 2016).

## RESULTS & DISCUSSION

**Effect of Zn foliar application on Zn and Mn concentration and uptake**

Figure (1) Shows that the application of Zn caused a (0.0029) % increase in Zn this in the same figure Mn concentration in flax oil recorded the highest value of 0.0068 % in Mn200. The uptake values (15.74 and 14.90) mg plant-1 were obtained from the application of Zn400 and Mn200 respectively. Abdullah et al., (2022) obtained that Zn and Mn application effects on most of flax growth and yield.

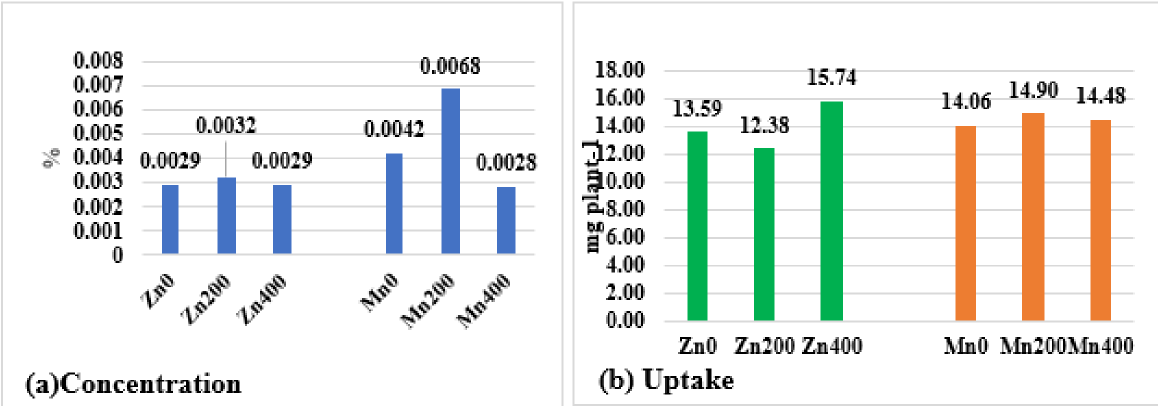



Figure (1): Effect of Zn foliar application on (a) Zn and Mn concentration (b) on their uptake in flax

**Effect of Mn foliar application on Zn and Mn concentration and uptake**

The application of Mn increases the Zn % the highest and lowest value (0.0066 and 0.0056) was recorded for control and Mn200, which equivalent to 66 and 56 ppm.This was in harmony with the finding of Hawf and Schmid (1967) who clarified that different concentrations of Mn had an effect on Zn uptake at high concentrations only. While Mn caused an increase in Mn concentration with the highest value of 0.0122 % in the application of Mn400. Furthermore, the application of Mn decreases the uptake of Zn, the highest uptake was 6.80 mg seeds -1 in control treatments while Mn uptake was highly 7.69 mg seeds -1 in Mn200. This may be due to the balance of the two microelements in growth media. Soltangheisi et al. (2014) indicated that the translocation of Mn in the upper part of corn showed a highly positive correlation with Mn concentration in the nutrient solution.

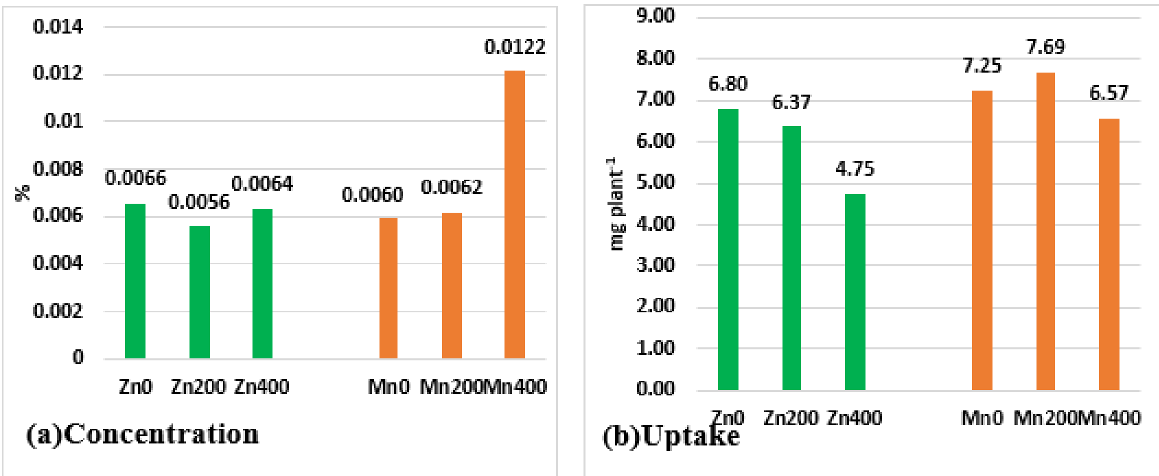



Figure (2): Effect of Mn foliar application on (a) Zn and Mn concentration (b) on Zn and Mn uptake in flax

**Interaction effect of Zn and Mn on Zn and Mn concentration and uptake**

The radar chart in (Figure 3, a) shows Zn and Mn concentrations which affected by the interaction between the two microelements, the highest and lowest value recorded from F1 (Zn0Mn0) and F3 (Zn0 Mn400) for Zn concentration with values (35 and 22) (mg kg-1). Furthermore, the highest and lowest value for Mn concentration was (84 and 36) (mg kg-1) which obtained for F2 (Zn0 Mn200) and F3(Zn0 Mn400) respectively.

The treatment combination Zn200 Mn400 caused the highest uptake of Zn 7.69 mg plant-1 and the Zn0 Mn200 recorded the highest uptake value of 17.85 mg plant -1 while the lowest uptake value obtained from Zn0 and Mn400 with values (4.75 and 6.28) mg plant seeds-1 as shown in (Figure 4). This may be due to the ionic competition between Zn and Mn during the process of absorption led to these results.


Figure (3): The interaction effect of Zn and Mn on Zn and Mn concentration (mg kg[1])

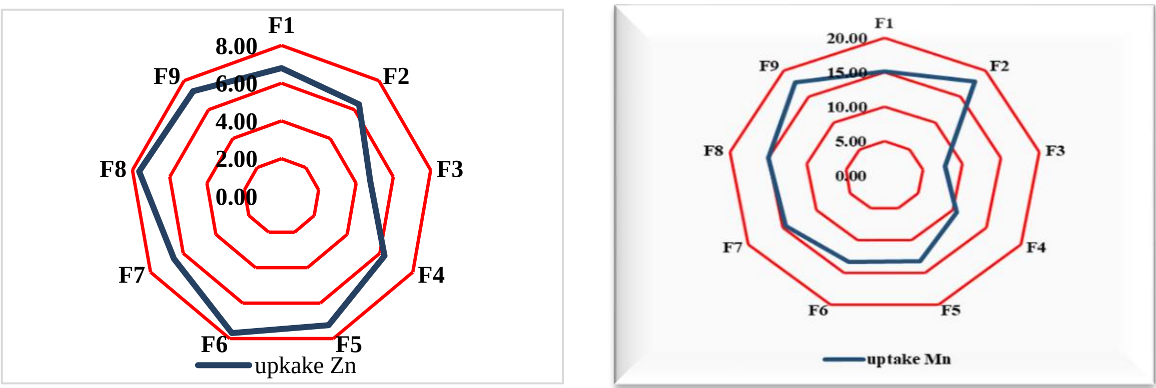



Figure (4): The interaction effect of Zinc and Manganese on Zn and Mn uptake

## Fatty Acids (%) Profile

Data in table (3) pointed out that all treatments of Zn element especially high doses gave a positive effect on saturated fatty acids percentage, it seems that Zn has a beneficial effect on the formation of fatty acids.Similler results was recorded by(Nofal and Bakry, 2011).

Table (3): Interaction effect of Zn and Mn foliar application on fatty acids of flaxseed

| Treatment | Palmitic | Stearic | Oleic | Linoleic | Linolenic |
|---|---|---|---|---|---|
| | (%) | | | | |
| Zn$_0$ Mn$_0$ (F1) | 5.47 | 4.74 | 17.01 | 13.99 | 50.66 |
| Zn$_0$ Mn$_{200}$ (F2) | 5.20 | 4.33 | 16.74 | 13.65 | 50.30 |
| Zn$_0$ Mn$_{400}$ (F3) | 5.62 | 5.00 | 17.32 | 14.24 | 50.98 |
| Zn$_{200}$ Mn$_0$ (F4) | 5.59 | 4.90 | 17.18 | 14.11 | 50.80 |
| Zn$_{200}$ Mn$_{200}$ (F5) | 5.33 | 4.52 | 16.90 | 13.84 | 50.48 |
| Zn$_{200}$ Mn$_{400}$ (F6) | 5.78 | 5.12 | 17.50 | 14.35 | 51.15 |
| Zn$_{400}$ Mn$_0$ (F7) | 5.89 | 5.29 | 17.66 | 14.59 | 51.30 |
| Zn$_{400}$ Mn$_{200}$ (F8) | 5.98 | 5.40 | 17.78 | 17.77 | 51.45 |
| Zn$_{400}$ Mn$_{400}$ (F9) | 5.11 | 4.19 | 16.58 | 13.55 | 50.12 |

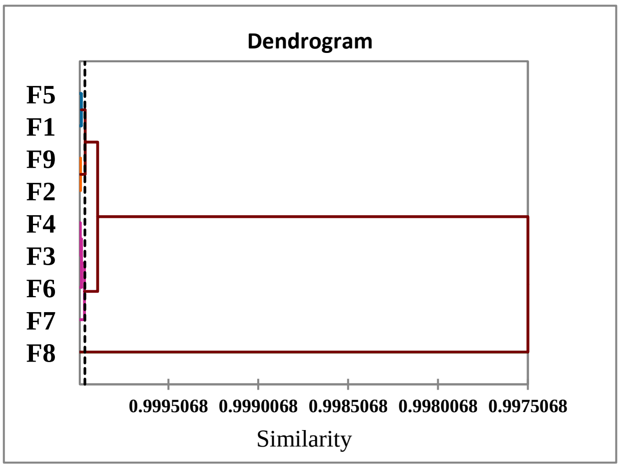
Movahhedy-Dehnavy *et al*., (2009) indicated that Zn and Mn application can improve the seed quality of safflower. The improved physiological performance in tested plants was due to more nutrient uptake by treated flax plants from soil table (3).

## CLUSTER ANALYSIS

Table (4) and Figure (4) show the cluster analysis of the studied treatment combination into four clusters depending on similarity. The clusters were C1, C2, C3 and C4. The first class included F1 and F5 the second cluster was (F2 and F5), the third cluster included (F3, F4, F6 and F7) and finally the fourth cluster relative to F8. Dendrogram was sketched from cluster analysis of the fatty acids and the micronutrients based on the 9 treatments (Figure 5). Palmitic, stearic, oleic, linoleic and linolenic acids, zinc and manganese mean value was (5.55, 4.83,17.19, 14.45, 50.80, 0.0030 and 0.0062) % respectively. The vertical line is responsible for limiting the similarity between groups or dividing the studied characteristic.

Table (4): Classifying the treatment combination into clusters

| Obse. | $F_1$ $Zn_0$ $Mn_0$ | $F_2$ $Zn_0$ $Zn_{200}$ | $F_3$ $Zn_0$ $Mn_{400}$ | $F_4$ $Zn_{200}$ $Mn_0$ | $F_5$ $Zn_{200}$ $Mn_{200}$ | $F_6$ $Zn_{200}$ $Mn_{400}$ | $F_7$ $Zn_{400}$ $Mn_0$ | $F_8$ $Zn_{400}$ $Mn_{200}$ | $F_9$ $Zn_{400}$ $Mn_{200}$ |
|---|---|---|---|---|---|---|---|---|---|
| Class | 1 | 2 | 3 | 3 | 1 | 3 | 3 | 4 | 2 |



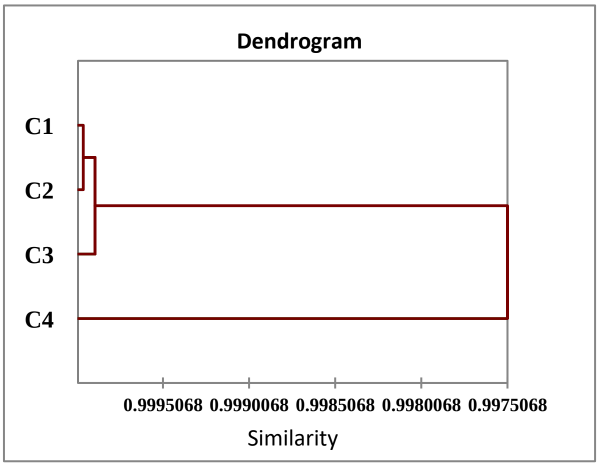




Figure (5): Dendrogram for the cluster of flax fatty acids (%) and two heavy metals

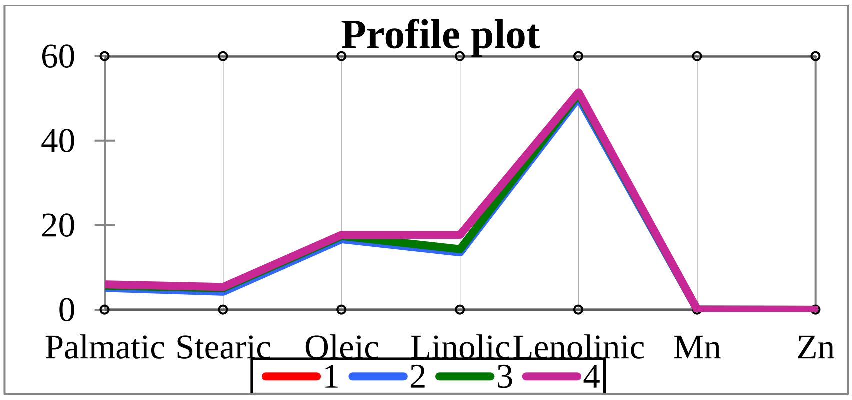



Figure (6): Shows the profile plot fatty acids, Zn and Mn

Table (5) shows the proximity matrix which refers to similarity and dissimilarity, the highest value (1.000) refers to the higher dissimilarity between $F_1$ and all fertilizer combinations except $F_8$. While, the lowest values refer to the similar relation between them such as the value 0.997 refers to similar relations between $F_2$ & $F_8$, $F_5$ & $F_8$ and $F_8$ & $F_9$.

Table (5): Proximity matrix (Euclidean Distance)

| | F1 | F2 | F3 | F4 | F5 | F6 | F7 | F8 | F9 |
|---|---|---|---|---|---|---|---|---|---|
| F1 | 1 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 0.998 | 1.000 |
| F2 | 1.000 | 1 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 0.997 | 1.000 |
| F3 | 1.000 | 1.000 | 1 | 1.000 | 1.000 | 1.000 | 1.000 | 0.998 | 1.000 |
| F4 | 1.000 | 1.000 | 1.000 | 1 | 1.000 | 1.000 | 1.000 | 0.998 | 1.000 |
| F5 | 1.000 | 1.000 | 1.000 | 1.000 | 1 | 1.000 | 1.000 | 0.997 | 1.000 |
| F6 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 1 | 1.000 | 0.998 | 1.000 |
| F7 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 1 | 0.998 | 1.000 |
| F8 | 0.998 | 0.997 | 0.998 | 0.998 | 0.997 | 0.998 | 0.998 | 1 | 0.997 |
| F9 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 | 0.997 | 1 |

Table (6) and Figure (7) explain that the eigenvalue for $F_1$ and $F_2$ had a great influence on variability, since their values were higher than one which was 4.79 and 1.51 respectively, and the cumulative variability was 68.45 and 89.95 % while the eigenvalues for other fertilizer were less than one or can be neglected. After $F_2$ the slope of the scree plot will decrease then an approach zero (Figure 7), which means the effects of $F_3$ to $F_9$ are very low and can be neglected.

Table (6): The eigenvalue and the variability among the genotypes

| | F1 | F2 | F3 | F4 | F5 | F6 | F7 |
|---|---|---|---|---|---|---|---|
| Eigenvalue | 4.79 | 1.51 | 0.44 | 0.25 | 0.01 | 0.00 | 0.00 |
| Variability (%) | 68.45 | 21.50 | 6.31 | 3.61 | 0.09 | 0.03 | 0.00 |
| Cumulative % | 68.45 | 89.95 | 96.27 | 99.88 | 99.97 | 99.99 | 100 |

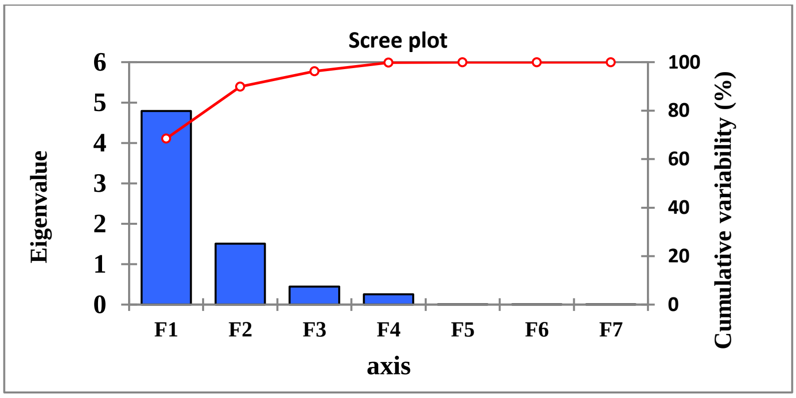



Figure (7): Shows the decrease in slope for the scree plot after $F_2$

Figure (8**)** explains the vectors for the studied traits. The angles between vectors explain the correlation between them. The decrease in angle value to < 90◦ means an increase in positive correlation coefficient value and if the angle approached zero the correlation coefficient value approaches 1.00; if the angle is °90 that means the correlation coefficient equals zero. If it is °180, it means the correlation (r = -1) or open-angle between two variables decreases to a negative correlation between them. However, if the angle between two variables is less than °90, it means a positive correlation coefficient. The length of the vector explains the value of loading factors.

The combination treatments in Figure (8) are merged and give a different explanation. For the variables close to the center there will not be significant differences between them for example in $F_6$ linoleic acids is the closest variable. Oleic with $F_7$ is closer. If any of the variables are close meaning there are positive reactions between them and the other side has a negative reaction to the first one. The first two axes for four clusters were significant (eigenvalues ≤ 90) and contributed to 89.95 % of total variance, and were mainly correlated to $F_1$, $F_2$.

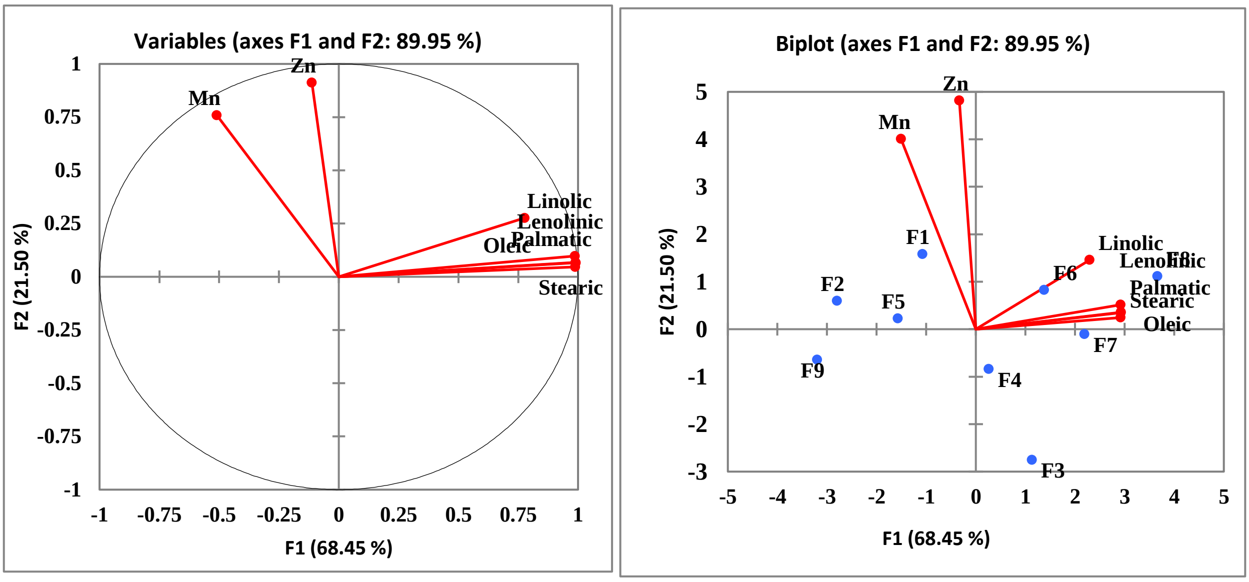



Figure (8): a. The vectors for the studied traits b. The vectors for the studied traits and fertilizers

## CONCLUSIONS

The most outstanding conclusions that can be drawn from this study are: the application of Zn caused an increase in Zn and Mn concentration in flax oil because of an increase in their uptake, in the same aspect Mn caused an increase in Mn concentration the application of Mn and a decrease in Zn uptake due to the ionic competition between them and increase levels of Zn and Mn caused a decrease in all studied fatty acids

## ACKNOWLEDGMENT

The researchers thanked the Salahaddin University for their assistance, which contributed to the completion of this research.

## CONFLICT OF INTEREST

The researcher supports the idea that this work does not conflict with the interests of others.

## تقييم بعض خصائص الجودة لبذور الكتان استجابة للتطبيق الورقي للزنك والمنغنيز

جوان جودت هاشم[1], اريان مصطفى عبدالله[2], بهار جلال محمود[3]
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**الخلاصة**

تم اجراء البحث في حقل كردرشة، كلية علوم الهندسة الزراعية / جامعة صلاح الدين – اربيل (٣٦،٣٦° ٤ شمالا، ٤٤° ٢ شرقا، ارتفاع ٤١٥ مترا فوق سطح البحر). تم إجراء التجربة العاملية بناءً على تصميم القطاعات الكاملة العشوائية باستخدام ثلاث مكررات لدراسة تأثير ثلاثة مستويات (٠،٢٠٠،٤٠٠) ملجم لتر-١ لكل من الزنك والمنغنيز كتطبيق ورقي وتدخلهما يضاف بالرش على الاجزاء الخضرية. تشير النتائج إلى أن تأثير إضافة الزنك والمنغنيز بقيم (٠,٠٠٣٢ و ٠,٠٠٦٨) ٪ في المعاملة الزنك ٢٠٠ملجم لتر-١ و المنغنيز ٢٠٠ملجم لتر-١ تسبب بالاضافة الى ذلك فى زيادة امتصاص اثنين من العناصر الدقيقة في الزنك ٢٠٠ملجم لتر-١ و المنغنيز ٢٠٠ملجم لتر-١ بقيم (١٥,٧٤ و ١٤,٩٠) ملجم نبات -١. تسببت استخدام كلا العنصرين الزنك ٤٠٠ملجم لتر-١ والمنغنيز ٢٠٠ملجم لتر- ١ فى زيادة حمض البالمتيك والأوليك واللينوليك بينما استخدام الزنك ٢٠٠ملجم لتر-١ و المنغنيز ٤٠٠ملجم لتر- ١ اكتسبت اقل قيمة. صنف تحليل العنقودي او الديندروكرام مزيج العناصر الدقيقة الزنك و المنغنيز الى اربع مجموعات رئيسية اعتمادا على الزنك و المنغنيز و ملف تعريف الاحماض الدهنية. يوضح تحليل المكون الرئيسي PCA ان قيمة الزاوية بين كل من الاحماض وبين العناصر الدقيقة ٩٠ درجة، وهذا يعنى ان هناك ارتباطا كبيرا بينهما والعكس صحيح. توجد علاقة ارتباطية عالية بين حمض اللينوليك واللينولينيك وF6 بينما يرتبط F7 مع حمض الأوليك، الدهني وحمض البالمتيك.

الكلمات المفتاحية: زيت بذور الكتان، الزنك، المنغنيز ، الأحماض الدهنية، التحليل العنقودي

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