



## Response of parsley plant to foliar application of ferrous sulfate combined with amino acid

El Sayed S.A.A., M. A. Abou Seeda and H. I. El Eila

National Research Center, Plant Nutrition, Dept., Dokki, Cairo, Egypt

**Abstract:** Pots experiments were conducted to fulfill the effects of application of Fe with and without amino acids on the parsley plants. Data observed that application of Fe alone stimulate the fresh and dry material of parsley plant, results also revealed that increasing rate of Fe addition increasing the fresh and dry weight of plant particularly in the first cut. Maximum increases noticed at rate of 75ppm, similar trends were observed in the second cut. Foliar application of Fe at a rate of 100ppm gradually increase (NPK) uptake particularly in the first cut as compared with the second one. Application of amino acids alone has a positive effect on the nutrients uptake. Results observed that the combination of ferrous sulfate with amino acids gradually increased of the nutrients uptake of NPK by the tested plants, maximum of increase obtained at rate of (Fe75ppm+1cm/l) dose were 87, 84, 95, 94, 95, 95 % for first and second cut respectively. Data observed that ferrous sulfate at different doses on the chl. a, band carotene were gradually, increase by increasing rate of FeSO<sub>4</sub>. Amino acids added by spraying stimulate the chlorophyll a, b, and carotene. Data revealed that combination of Fe with amino acid together gradually increase the chlorophyll content as compared with FeSO<sub>4</sub> alone. Maximum results of chlorophyll content particularly at rate of 75 ppm combined with amino acid. The effect of amino acids on the uptake of iron nutrient by parsley plants reflect the ability of amino acid to act as nature organic chelators which may enhanced the absorption through the efficient carriers theory of Fe into plant.

**Key words:** foliar application, ferrous sulfate, amino acids, parsley plant, agronomy parameter.

### Introduction

Amino acids as natural organic chelation can be consider as the best solution to increase as well as avoid the micronutrients efficiency in plants and considered as the most efficient agricultural product. These are expensive products, which needed more comprehensive experimental particularly at different stages of the growth in order to be effective and applicable. Application of these products in soil may cause environmental risks due to the weak degradability of these products<sup>1</sup> as well as their mobility throughout the soil profile<sup>2</sup>.

Recently, alterations in the production have been enlargement regarding the low-cost, as well as environmentally friendly system to be ensure the quantity and quality of the yield.<sup>3,4</sup> stated that using of bio stimulators in plant nutrition as growth, and metabolic enhancers, such as hormones, humic substances or amino acids has been reported in this concern.

The application of these products in small amounts containing amino acids usually induce the amendements of plants metabolism containing essential elements such as nitrogen and carbon metabolism, and the defiance of plants to a biotic and biotic emphasis<sup>5,6,7</sup> However, few available research to judge the bio analeptic action of these products on nutritional status of iron plants.

Several researchers stated that application of Fe-EDDHA combined with natural organic material e.g. Amino acids stimulate the absorption of iron and consequently increased leaf Fe concentration and ameliorate some characteristics criterion of citrus and tomato fruits.<sup>8,9,10</sup> Reported that, in asparagus plants studies, application of amino acids in combination with different synthetic chelates of iron, gradually enhance the growth parameters root, shoot and chlorophyll contents. They also stated that absorption and translocation of iron nutrient particularly inside the plant is easier. This phenomenon due to chelating action of amino acids with micronutrients<sup>11</sup>.

Deficiency of iron is abounding aspect of plant growth. However, high quantities of iron in soils, and due to its low solubility products resulted in low availability as well as translocation to the plant; therefore, iron deficiency is a common problem. Plant required natural organic substances such as amino acids for synthesis of most important enzymes and hormones, which is essential for completing metabolic activities inside plant. Amino acids are responsible for most of protein syntheses, which are known as building units that play an important role in plant life.

The aim of this research is to study the effect of FeSO<sub>4</sub> in combination with natural organic materials such as amino acid applied as foliar application on growth parameters of parsley plant.

## Materials and Methods

A greenhouse experiment was conducted at the Plant Nutrition Dept. of the National Research Centre. Two kg of air-dried soil were packed in plastic pots 15cm height, 10 cm diameter. The characterization of the investigated soil illustrated in table 1

**Table (1): Initial physical and chemical soil properties of the experimental soil site.**

Particle size distribution (%)			Texture class	pH (1: 2.5)	EC dS m <sup>-1</sup> (1: 5)	O.M (%)	CaCO <sub>3</sub> (%)
Sand	Silt	Clay					
67.52	8.00	24.48	Sand clay loam	7.15	0.80	1.20	0.47
Soluble cations (me L <sup>-1</sup> )				Soluble anions (me L <sup>-1</sup> )			
Na <sup>+</sup>	K <sup>+</sup>	Ca <sup>++</sup>	Mg <sup>++</sup>	CO <sub>3</sub> <sup>--</sup>	HCO <sub>3</sub> <sup>-</sup>	Cl <sup>-</sup>	SO <sub>4</sub> <sup>--</sup>
3.25	0.82	1.15	3.82	0.00	2.41	2.57	3.96
Available Macronutrients (mg Kg <sup>-1</sup> )			CEC	Available Micronutrients (ppm)			
N	P	K	Meq/100gsoil	Fe	Zn	Mn	Cu
26.10	1.18	143	21.29	5.02	0.79	2.46	1.08

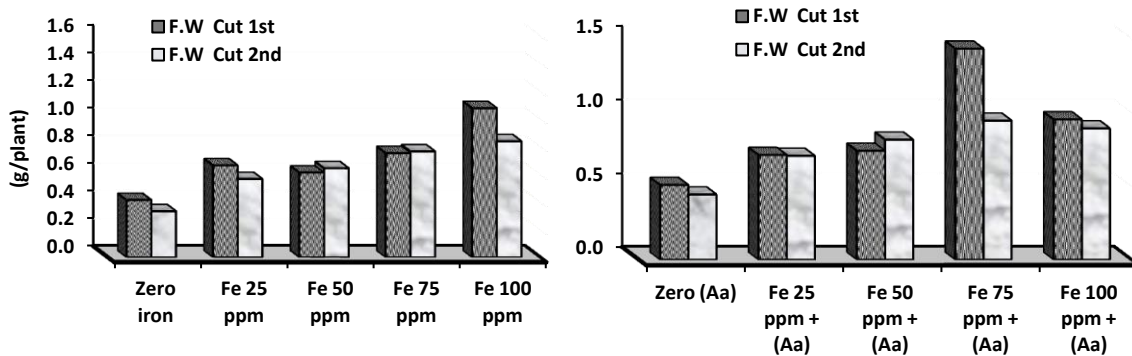
Treatments Foliar application of iron added at 0, 25, 50,75 and100 ppm also amino acid added as foliar application with and without amino acid at a rate of 1cm/l. whereas urea was applied at rates of(3.0 g/pot) calcium super phosphate was applied at a rate of 50 mg P<sub>2</sub>O<sub>5</sub>/kg soil. Potassium sulfate applied at a rate of 40 mg K<sub>2</sub>O/kg soil for treatments. Parsley plant was used as a test plant. Ferrous sulfate was applied as foliar spray after 20 & 30 days amino acid added as foliar spray (rate of spraying every week). The experiment laid out in a completely randomized block design with three replicates. Samples of plants were taken after30 and 50 days of planting. Fresh weight was recorded, then samples were dried at 70°C, weight and ground to pass through a 1-mm sieve. Nitrogen, phosphorous, potassium, sodium, calcium, chlorophyll a, b and carotene were determined according to the methods described by<sup>12</sup>. All data were statistical analyzed according to the technique of analysis of variance (ANOVA) for the completely Randomized design (CRD) using MSTATC software package according to<sup>13</sup>. Least Significant Difference (LSD) was used to compare the differences between treatments means at the level 0.05%.

**Results and Discussion**

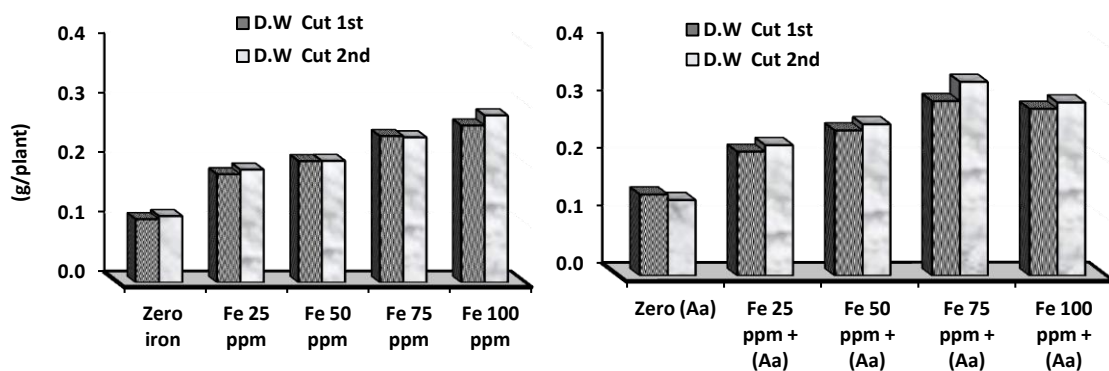
**Fresh and Dry weight**

Fresh and Dry matter The effect of different rates of FeSO<sub>4</sub> with and or/ without amino acid applied as foliar on fresh and dry matter content of parsley are illustrated Fig 1, data revealed that addition of FeSO<sub>4</sub> at different rates ( 25 up to 100 ppm) gradually stimulate the fresh and dry matter of parsley plant. It observed that maximum increase of fresh and dry weight of parsley were noticed at rate of Fe 100 ppm particularly in the first cut, similar with slightly increase observed in the second cut , explaining the role of Ferrous sulfate for improving and enhancing the root system. Data also indicated that the addition of ferrous sulfate combined with amino acid stimulate both fresh and dry weight of two cuts. Fig1 represent the effect of foliar application of Ferrous sulfate at different doses and amino acids with similar doses of Ferrous sulfate on the Fresh and Dry weight of Parsley. Results indicated that foliar application of amino acids alone stimulates the fresh and dry weight of parsley plant at the first and second cuts. Maximum value of fresh and dry weights was noticed in the rate of (Fe<sup>++</sup> 75 ppm+ amino acids) explaining that role of amino acids for enhancing the mode of action of Fe – nutritional status.

This phenomenon may relate to the relationship between these synthesized acids and growth process. Amino acids are responsible for production of new cells and energy production, creating suitable circumstances for enhancing both fresh and dry weight of Parsley plants. Similarly were noticed by<sup>14,1</sup>. Several researchers illustrated that application of amino acids with Fe stimulate the plant growth and gradually increase the long life mobility of iron in the soil and implies risks of environment due to low mobility and bio availability of micronutrients<sup>2</sup>. Foliar application of iron combination with amino acids decreases the degradability of amino-iron compound throughout the soil profile<sup>15</sup>.



**Treatments of ferrous sulfate with and without amino acid**



**Treatments of ferrous sulfate with and without amino acid**

**Fig (1): Fresh and dry weight of parsley as affected by foliar application of Ferrous sulfate at different doses with amino acid.**

Fig (2) represented that nutrients uptake of Nitrogen, Phosphorous and Potassium in parsley plants. Results observed that foliar application of ferrous sulfate gradually increase the uptake of (NPK) particularly in the first cut. Data indicated that application of ferrous sulfate stimulate nitrogen; phosphorous and potassium uptake particularly in the first cut, however slightly increase were noticed in the second cut.

Application of amino acids alone has positive effects on the uptake of the nutrients under investigation. Data noticed that the effect of amino acid alone was more pronounced effect than the none added ferrous sulfate.<sup>5,6,4,7</sup> They found that the application of amino acids particularly in small amounts mostly improve the plant metabolism, which include nitrogen and carbon metabolism, and struggle of plants to biotic and a biotic stress.

Maximum results were obtained particularly at rate Fe75ppm+1cm/l.<sup>16</sup> stated that amino acids application stimulate the plant content of macro and micronutrients in addition to that carotenoids and carbohydrates.<sup>17,18</sup> They stated that the importance of amino acids for increasing growth parameter, yield and chemical composition of some economic plants.

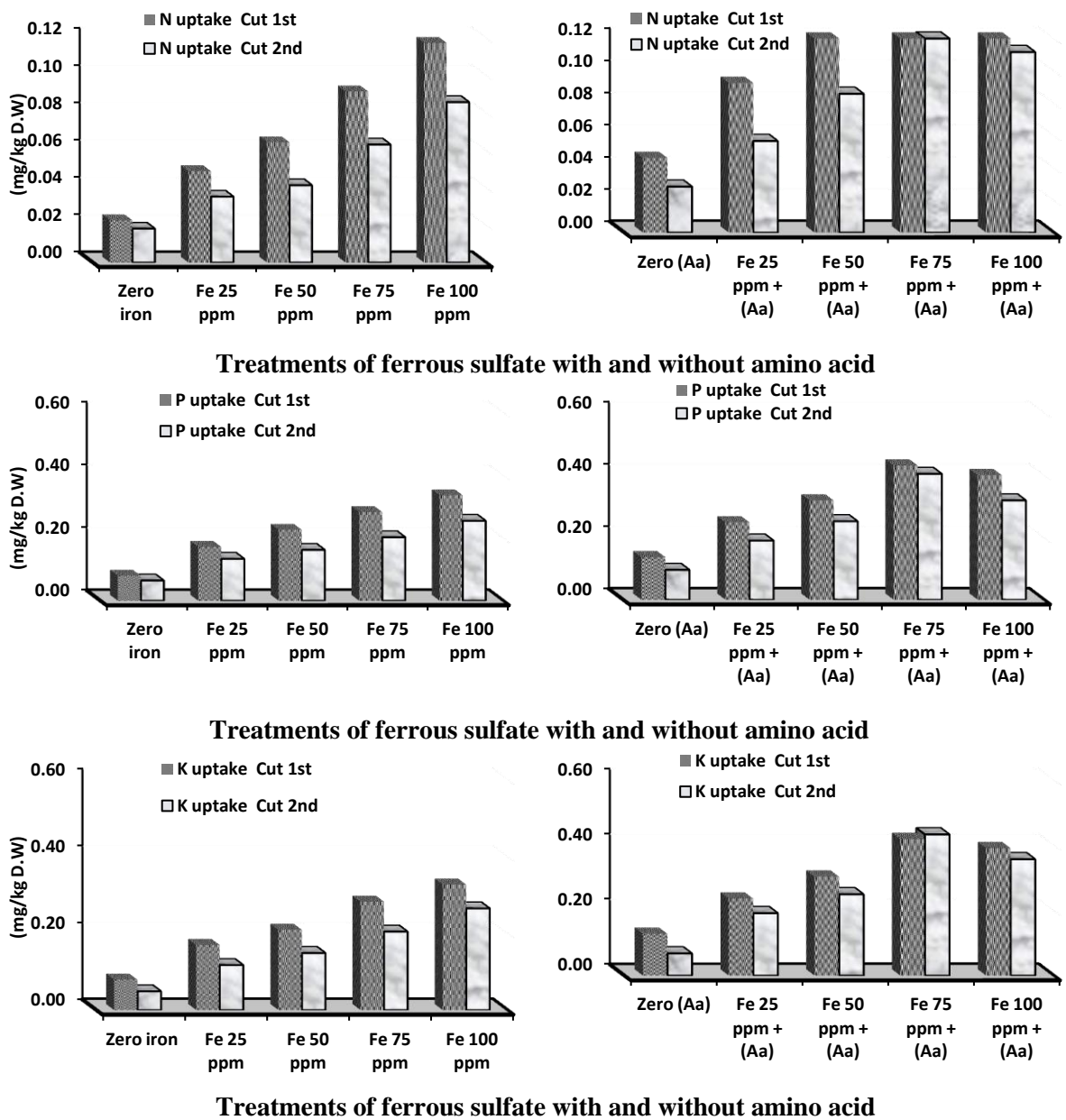


Fig (2): Macronutrient uptake in parsley as affected by foliar application of Ferrous sulfate at different doses with amino acid.

Table 2 represent foliar applications of Ferrous sulfate at different combined with and without amino acids on the Nutrient indices in Parsley plant. Interactions of nutrients are conventionally amplitude in terms of growth parameter. The response and change in concentration of nutrients, upon addition of two nutrients as well as organic materials can led to improve the nutritional status particularly iron in alkaline soil e.g.(amino acids) , an increase in crop yield that is more than adding only one, the interaction is positive (synergistic). Similarly, if adding the two nutrients together produced less yield as compared to individual ones, the interactions is negative (antagonistic).When there is no change, there is no interaction.Table3 illustrated that the interaction between Iron - amino acids were calculated in Table 3. Results noticed that the Nutrient Biotic Index (NBI) was used to determine the beneficial effect of the interaction between ferrous ions and the amino acid the site was strongly responsive to Fe and moderately responsive to amino acids. Thus in the treatment that received neither Fe nor amino acids, Fe was much more deficient than amino acids (Fe >>amino acids), as shown by the large DM responses to Fe. However, overall DM response to amino acids was also evident.

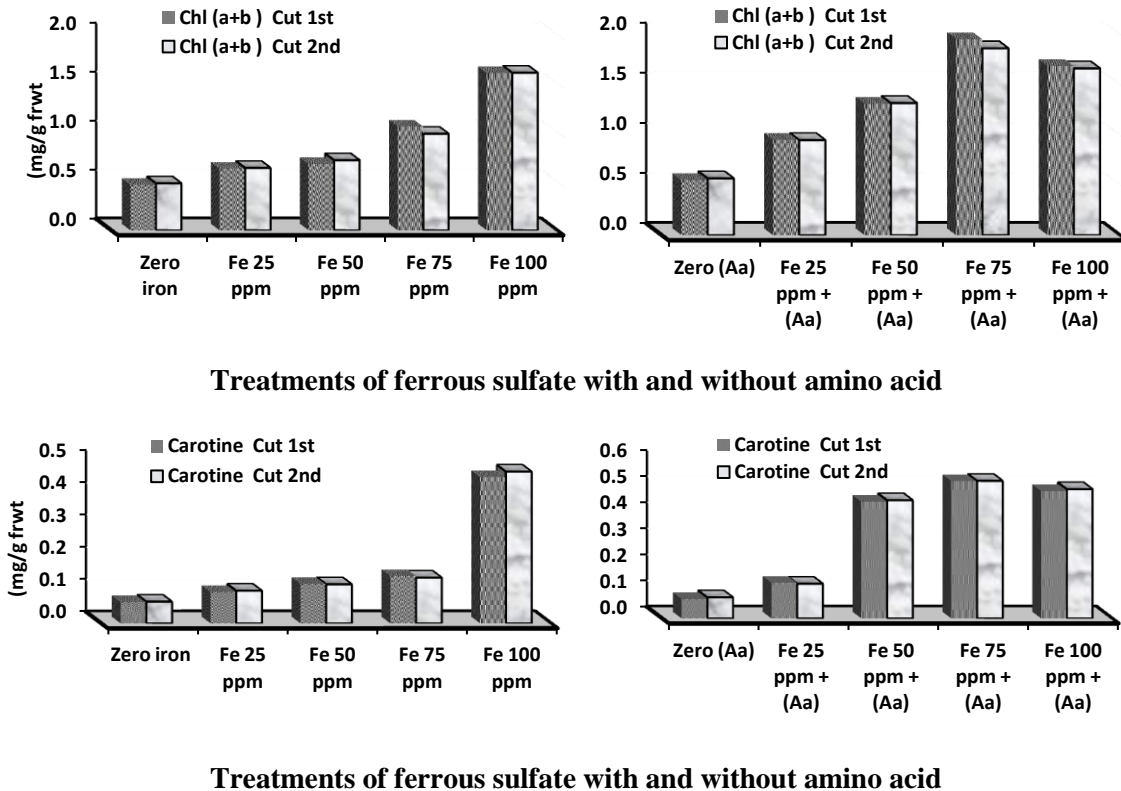
Therefore amino acids was, also deemed deficient in this treatment (amino acids >Nil) particularly in the first cut. However, in the second cuts the results reflect the adequate nutrition than the first one. Fig 4 represents the application of Ferrous sulfate alone or combined with amino acids on the chl. a, b and carotene content of parsley plant.

**Table (2): Nutrient indices and balance Index as affected by foliar application of Ferrous sulfate at different doses with amino acid.**

Treatment	Nutrient indices						NBI*	
	N		P		K		Cut 1 <sup>st</sup>	Cut 2 <sup>nd</sup>
	Cut 1 <sup>st</sup>	Cut 2 <sup>nd</sup>	Cut 1 <sup>st</sup>	Cut 2 <sup>nd</sup>	Cut 1 <sup>st</sup>	Cut 2 <sup>nd</sup>		
Iron Fe <sup>++</sup> (zero)	2.24	1.72	0.35	0.24	0.64	0.52	3.23	2.48
Fe 25 ppm	2.23	2.10	0.35	0.31	0.64	0.57	3.21	2.98
Fe 50 ppm	2.09	2.25	0.32	0.33	0.60	0.59	3.01	3.17
Fe 75 ppm	2.04	2.12	0.34	0.35	0.66	0.66	3.04	3.13
Fe 100 ppm	1.88	2.06	0.31	0.35	0.66	0.69	2.85	3.10
Amino acid (Aa) zero	1.86	1.55	0.30	0.21	0.64	0.50	2.80	2.26
Fe 25 ppm + 1cm/l (Aa)	1.75	2.19	0.29	0.36	0.66	0.65	2.70	3.20
Fe 50 ppm + 1cm/l (Aa)	1.65	1.95	0.27	0.32	0.68	0.67	2.60	2.94
Fe 75 ppm +1cm/l (Aa)	1.66	2.01	0.28	0.35	0.69	0.72	2.64	3.08
Fe 100 ppm + 1cm/l (Aa)	1.68	2.16	0.28	0.38	0.69	0.74	2.66	3.28

NBI\* : Nutrient Balance Index

Application of ferrous sulfate at different doses ( 25ppm up to 100ppm) on the chl. a, b and carotene are illustrated in Fig 4 from which results observed that foliar application with ferrous sulfate stimulate the total chl. a, b and carotene in the leaves of parsley. Increasing the application rates of ferrous sulfate .gradually increase the chl. a, b and the carotene for all cuts. Data revealed that in the second cuts the application were more pronounced effect than the first cut. Amino acids application stimulates the chlorophyll a, b, and carotene as compared with the control (iron F<sup>++</sup> zero).<sup>19</sup> stated that application of amino acids increasing the chlorophyll content in the plant, governing to higher degree of photosynthesis, makes crops lush green. <sup>20,21</sup> they reported that the plant can assimilate the amino acids when applied by foliar application through the stomata tool. This performance is controlled by the environmental ambience such as temperature, wind, humidity. They also reported that such process could directly or indirectly influence the growth parameters of plants. The indirect effect may be explained by stimulating plant cells division. <sup>22</sup>also emphasize that phenomenon as amino acids could regulate the physiological activities through hormone levels in plant, improving the growth parameter, and enhancing the stress tolerance.



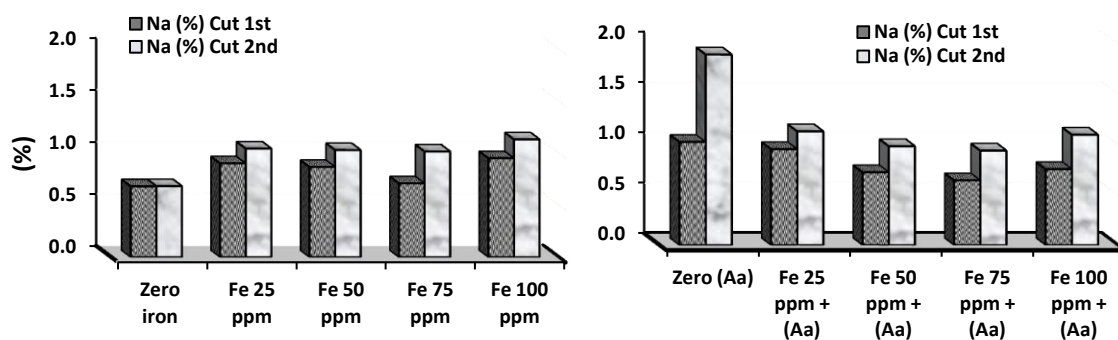
**Fig (3): chlorophyll (a+b) and carotene as affected by foliar application of Ferrous sulfate at different doses with amino acid.**

Data observed that ferrous sulfate combined with amino acids gradually increase the chlorophyll content on parsley plants as compared to the ferrous sulfate alone. Amino Acids are also reached to the soil through foliar application and can help the biological fertility of the soil through improving the micro flora of the soil thereby assisting the progress of the assimilation of nutrient elements particularly micronutrients (Fe). Maximum chlorophyll content obtained at the rates of Fe 75 ppm combined with amino acids in both cuts. <sup>15</sup>they stated that using of iron chelates such as Fe- EDDAH or Fe- EDTA is the additional proficient agricultural discipline to escape or comfort relieve Fe deficiency in plants. Similar conclusion were reported by <sup>23,15</sup> they also reported that amino acids in combination with micronutrients stimulating some micronutrient more available to plant.

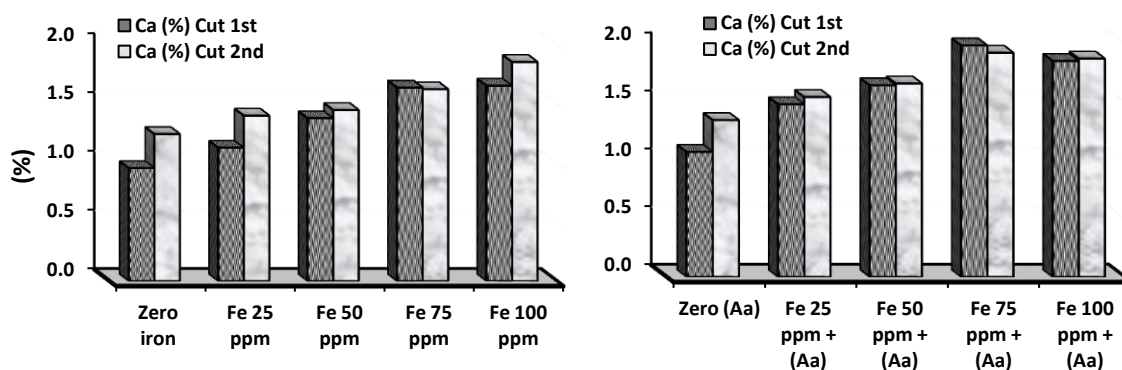
Fig 4 represents the effect of spraying of ferrous sulfate with and without the amino acids on the sodium, calcium and Ca/Na ratio of parsley plant. Data indicated that application of FeSO<sub>4</sub> increase the percentage of total sodium content in leaf plant in both cuts, slightly increases were observed in the second one. Application of ferrous sulfate gradually stimulates the percentage of calcium content than the sodium in leaf of parsley plant for both cuts. However, amino acids added alone, increase both sodium and calcium content. The combination of Ferrous sulfate with the amino acids gradually decrease the sodium content in parsley plants, and was more pronounced effect in first cut than the second one, This phenomenon probably due to the effective of amino acids on the physiological activities of the plant. Which produce a new plant cells, produce energy and play an important position in plant resistance<sup>24</sup>.

The role of proline production in plant may be influence on the stress tolerance in different approach, to protect protein integrity and can augmented the enterprise of different enzymes, amino acids also used to regulate water potential when subjected to draught or salinity stress in the root zone. <sup>25,26,27,28,29,30,31</sup>.





#### Treatments of ferrous sulfate with and without amino acid



#### Treatments of ferrous sulfate with and without amino acid

**Fig (4): Sodium and calcium content as affected by foliar application of Ferrous sulfate at different doses and amino acid.**

## References

- Hyvömen, H., Orama, M., Saarinen, H., Aksela, R. (2003). Studies on biodegradable chelating agents: complexation of iminodisuccinic acid (ISA) with Cu (II), Zn (II), Mn (II) and Fe(III) ions in aqueous solution. *Green Chem.* 5, 410–414.
- Rombolà, A. D., Cremonini, M. A., Lucchi, A., Sorrenti, G., Placucci, G., Marangoni, B. (2002). Leaching of soil-applied synthetic Fe chelates (Fe-EDDHA) in orchard ecosystem. *Book of Abstracts of the XI International Symposium on Iron Nutrition and Interaction in Plants.* Udine, Italy.
- Filippini, L., Bonfiglioli, M. (2005). L'impiego di biostimolanti per la difesa delle colture dagli stress abiotici. *Phytomagazine* 36, 27–31.
- Schiavon M, Ertani A and Nardi S. (2008). Effect of an alfalfa protein hydrolysate on the gene expression and activity of enzymes of tri-carboxylic acid (TCA) cycle and nitrogen metabolism in *Zea mays* L. *J. Agric. Food Chem.* 11800-11808.
- Maini, P. (2006). The experience of the first bio stimulant, based on Amino acids and peptides: a short retrospective review on the laboratory researches and the practical results. *Fertilitas Agrorum.* 1, 29-43.
- Kauffman III, G. L., Kneivel, D. P., Watschke, T. L. (2007). Effects of a Bio stimulant on the heat tolerance associated with photosynthetic Capacity, membrane thermo stability and polyphenol production of Perennial ryegrass. *Crop Sci.* 47, 261–267.
- Paradiković, N., Vinković, T., Vinković Vrc'ek, I., Žuntar, I., Bojčić, M. Medić-Šarić, M. (2011). Effect of natural bio stimulants on yield and nutritional quality: an example of sweet yellow pepper (*Capsicum Annuum* L.) *Plants. J. Sci. Food Agric.* 12, 2146–2152.
- Sánchez-Sánchez, A., Sánchez-Andreu, J., Juárez, M., Jordá, J. Bermúdez, D. (2002). Humic substances and amino acids improve effectiveness of chelate FeEDDHA in lemon trees. *J. Plant Nutr.* 25;2433-2442

9. Sánchez-Sánchez, A., Juárez, M., Sánchez-Andreu, J., Jordá, J., Bermúdez, D. (2005). Use of humic substances and amino acids to enhance iron availability for tomato plants from applications of the chelate FeEDDHA. *J. Plant Nutr.* 28, 1877–1886.
10. Gámiz, R., Espejo, J. A., Tejada, M., Dobao, M. M., González, J. L. (1998). Evolución de los contenidos de clorofilas en plantas de espárrago verde (*Asparagus officinalis*, L.) tras la adición de aminoácidos y ácidos fúlvicos. *Actas del VII Simposio Nacional-III Ibérico sobre Nutrición Mineral de las plantas*, Madrid, Spain, pp. 173–178.
11. Rombolà, A. D., Brüggemann, W., Tagliavini, M., Marangoni, B., Moog, P. R. (2000). Iron source affects iron reduction and greening of kiwifruit (*Actinidia deliciosa*) leaves. *J. Plant Nutr.* 23, 1751–1765.
12. Black, C.A., D.D. Evans, L.E. Ensminger, (1982). *Methods of soil analysis*. Agronomy J. Amer. Soc. Agron. Inc. Publ., Madison, Wisconsin, U.S.A.
13. Gomez K.A. and Gomez K.A. (1976). *Statistical procedures for Agricultural research with emphasis of rice*, Los Bans, Philippines', International Rice Research Institute.
14. Aberg, B., (1961). Nucleic acids and proteins in plants. *Encycl. Plant Physiol.*, Vol. 14, Springer Verlag, Berlin.
15. Mar Cerdan, Antoniosanchez- sanchez, juana D, jorda, Margarita Juarez and Sanchez-Andreu (2013). Effect of commercial amino acids on iron nutrition of tomato plants grown under lime-induced iron deficiency. *J. Plant Nutr. Soil Sci* 2013, 176, 859-866.
16. Khalil, M.Y., N.Y. Naguib, S.E. El-Sherbeny, (2002). Effect of (*Tagetes erecta*, L.) to some foliar application under compost levels. *Arab Univ. J. Agric. Sci.*, Ain Shams Univ., Cairo, 10(3): 939-964.
17. El-Shabasi MSS, Mohamed SMA, Mahfouz SA (2005). Effect of foliar spray with some amino acids on growth, yield and chemical composition of garlic plants. *Proc. the 6th Arabian Conference for Horticulture*, March 20-22, Faculty of Agric., Suez Canal University, Ismailia, Egypt.
18. Rawia, A.E, S.T. Lobna, M.M.I. Soad, (2001). Alleviation of Adverse Effects of Salinity on Growth, and Chemical Constituents of Marigold Plants by Using Glutathione and Ascorbate. *Journal of Applied Sciences Research*, 7(5): 714-721.
19. Ali, E.F. and Hassan, F.A.S. (2013). Impact of Foliar Application of Commercial Amino Acids Nutrition on the Growth and Flowering of *Tagetes Erecta*, L. *Plant Journal of Applied Sciences Research*, 9(1): 652-657.
20. Atiyeh, R.M., S. Lee, C.A. Edwards, N.Q. Arancon, J.D. Metzger, (2002). The influence of humic acids derived from earthworm processed organic wastes on plant growth. *Bioresource Technology*, 84: 7-14.
21. Chen, Y., M.D. Nobili, T. Aviad, (2004). Stimulatory effect of humic substances on plant growth. In "Soil organic matter in sustainable agriculture". (Eds F. Magdoff, R.R. Weil). 103-130.
22. Piccolo, A., S. Nardi, G. Concheri, (1992). Structural characteristics of humic substances as regulated to nitrate uptake and growth regulation in plant systems. *Soil Biochem.* 24: 373-380.
23. Lucena, J. J. (2006). Synthetic iron chelates to correct iron deficiency in plants, in Barton L. L., Abadía J. (eds.): *Iron Nutrition in Plants and Rhizospheric Microorganisms*. Springer, Dordrecht, pp. 103–127.
24. Bush, D.R., (1993). Proton coupled sugar and amino acid transporters in plants. *Annual Review of Plant Physiology and Plant Molecular Biology*, 44: 513-542.
25. Rajendrakumar, C.S., (1994). Proline-protein interactions: protection of structural and functional integrity of M4 lactate dehydrogenase. *Biochem. Biophys. Res. Commun.* 201: 957-963.
26. Mansour, M.M., (2000). Nitrogen containing compounds and adaptation of plants to salinity stress. *Biol. Plant.* 43:491-500.
27. Doaa M.R. Abo Basha and H.I. El-Aila (2015). Response of Foliar Spraying with Amino Acids and Integrated use of Nitrogen Fertilizer on Radish (*Raphanus sativus* L.) Plant. *International Journal of ChemTech Research*. Vol.8, No.11, pp 135-140.
28. Gaikwad S.A, Asha Kale, Kavita Mundhe, N.R. Deshpande, J. P. Salvekar (2010). Detection of Amino Acids Present in the Leaves of *Cassia auriculata* L. *International Journal of ChemTech Research*, Vol.2, No.2, pp 1092-1094.
29. Bhadane B.S. and T.J. Patil (2009). Influence of amino acids on the micellar behaviour of nonionic surfactant in aqueous medium. *International Journal of ChemTech Research*, Vol.1, No.2, pp 121-125.



30. Shafeek, M.R.; Ali, Aisha H. and Asmaa R. Mahmoud (2016). Foliar application of amino acids and bio fertilizer promote execution of broad bean plant (*Vicia faba* L) under newly reclaimed land conditions. International Journal of ChemTech Research, Vol.9, No.5, pp 100-109.
31. Thanaa Sh. M., Fatma K. M. Shaaban, Morsey M. M. and El-Nagger Y. I. (2016). Study on the Effect of Pre-harvest Treatments by Seaweed Extract and Amino Acids on Anna Apple Growth, Leaf Mineral Content, Yield, Fruit Quality at Harvest and Storability. International Journal of ChemTech Research, Vol.9, No.05 pp 161-171.

\*\*\*\*\*