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Response of Young grapevines (*Kishmishi cultivar*) to Spraying by Iron Chelated and the growth regulator benzyl adenine

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KEY WORDS:
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ABSTRACT

The research was conducted in the grape field of the University of Kirkuk / College of Agriculture in the Sayada region-Iraq, at latitude 35.33 N and 44.20 E, from 1/4/2023 to 1/8/2023 to study the effect of spraying by chelated iron in three concentrations (0, 0.5 and 1) g L^{-1} and Benzyl Adenine in three concentrations (0, 50 and 100) mg L^{-1} on the vegetative growth and chemical content of young grape vines of Kishmishi cultivar, where the spraying was done with three dates each 4/15, 5/1 and 5/15/2023. The experiment was carried out according to a completely RCBD with three replications and two vines for each experimental unit. Thus, the number of vines included in the experiment was 54. The data were analyzed according to the SAS system, the means were compared according to the Duncan test under the 5% probability level. The results showed that spraying the vines by iron chelated at concentration 1 g L^{-1} led to a significant increase in all the studied traits (Length and diameter of stem, leaf area, content of chlorophyll, percentage of NPK, content of Fe, and percentage of dry matter in branches) 139.56 cm, 182.78 mm, 76.04 cm^2, 40.53 CCI, 2.34%, 0.34%, 0.85%, 651.70 mg kg^{-1}, 41.28% respectively. On the other hand, spraying by Benzyl Adenine at concentration 100 mg L^{-1} gave significant superiority in length and diameter stem, leaf area, content of chlorophyll, percentage of P and K, content of Fe, and percentage of dry matter in branches reached to 125.91 cm, 175.78 mm, 72.35 cm^2, 39.51 CCI, 0.28%, 0.72%, 558.91 mg kg^{-1}, 41.28% respectively, Meanwhile the concentrations 50 and 100 mg L^{-1} of Benzyl Adenine significantly affected the percentage of nitrogen in leaves 2.09 and 2.11% respectively. At the same time, the interaction treatment between iron at the concentration 1 g L^{-1} Benzyl Adenine at the concentration 100 mg L^{-1} significantly effect on most of the characteristics.

استجابة كرمات العنب الفتية صنف كشمشي للرش بالحديد المخلبي ومنظم النمو بنزايل ادنين

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الخلاصة

تم اجراء البحث في حقل العنب التابع لجامعة كركوك/كلية الزراعة في منطقة الصيادة-العراق على خط عرض 35.33 شمالا وخط طول 44.20 شرقا، للفترة من 2023/4/1 ولغاية 2023/8/1 لدراسة تأثير الرش بالحديد المخلبي بثلاث تراكيز (0، 0.5، 1) غم لتر$^{-1}$ والبنزل ادنين بثلاث تراكيز (0، 50، 100) ملغم لتر$^{-1}$ على نمو الخضري والمحتوى الكيميائي لكرمات العنب الفتية صنف كشمشي، حيث تم الرش بثلاث مواعيد لكل منهما في 4/15 و5/1 و2023/5/15، ونفذت التجربة وفق تصميم القطاعات العشوائية الكاملة وبثلاث مكررات وبواقع كرمتين لكل وحدة تجريبية وبهذا كان عدد الكرمات الداخلة في التجربة 54 كرمة. وحللت البيانات وفق نظام SAS وقورنت المتوسطات وفق اختبار دنكن تحت مستوى احتمالية 5%. واوضحت النتائج التي تم الحصول عليها ان رش الكرمات بتركيز 1 غم لتر$^{-1}$ من الحديد المخلبي قد ادت الى زيادة معنوية في جميع الصفات المدروسة (طول وقطر الساق ومساحة الورقة ومحتواها من الكلوروفيل وNPK وFe ومحتوى الافرع من المادة الجافة) الذي بلغ (139.56 سم، 182.78 ملم، 76.04 سم^2، CCI 40.53، 2.34%، 0.34%، 0.85%، 651.70 ملغم.كغم$^{-1}$، 41.28%) على التوالي، وادى الرش بتركيز 100 ملغم لتر$^{-1}$ من منظم النمو بنزايل ادنين الى تفوق معنوي في الصفات (طول وقطر الساق ومساحة الورقة ومحتواها من الكلوروفيل وP وK وFe ومحتوى الافرع من المادة الجافة) الذي بلغ (125.91 سم، 175.78 ملم، 72.35 سم^2، CCI 39.51، 0.28%، 0.72%، 558.91 ملغم. كغم$^{-1}$، 41.28%) على التوالي، ولم يختلف معنويا مع التركيز 50 ملغم لتر$^{-1}$ في صفة النسبة المئوية للنتروجين في الاوراق اذا بلغ (2.09 و 2.11)% على التوالي لكلا التركيزين، وكان للتداخل الثنائي بين مستويات العوامل الداخلة في الدراسة اثر معنوي في اغلب الصفات حيث تفوقت المستويين (1 غم لتر$^{-1}$ من الحديد المخلبي متداخلا مع 100 ملغم لتر$^{-1}$ من بنزايل ادنين) و (1 غم لتر$^{-1}$ من الحديد المخلبي متداخلا مع 50 ملغم لتر$^{-1}$ من بنزايل ادنين) بصورة معنوية على بقية التداخلات.

كلمات المفتاحية: Fe–EDDHA، بنزايل ادنين، كشمشي، *Vitis vinifera* L.

INTROUCTION:

The grape, *Vitis vinifera* L., belongs to the Vitaceae family, which includes 14 genera and more than 1000 species, grapes are among the plants of the subtropical, warm temperate and cold temperate regions, due to the spread of its cultivation between latitudes 20-50°N and 20-40°S (Jules and Mooer, 1996). And that the original home of European grapes is the region located in Central Asia between the south of the Black Sea and the Caspian Sea, and all other grape varieties originated from it before the discovery of the North American continent, grapes are of great economic it growths in various types of lands, as sandy, low-fertile and shallow lands. It is also important in stabilizing the soil and preventing erosion (Hassan and Salman, 1989; Algarb *et al*., 2021). Its fruits have a high nutritional value because contain a percentage of monosaccharides (glucose and fructose), vitamins, organic acids such as malic, citric, and tartaric, and mineral salts such as potassium, calcium, magnesium, proteins, fats, and others (Al-Saeedi, 2000). In addition to its importance in medical uses in the treatment of many diseases as a tonic for brain cells and heart muscles, as fresh grapes contain a compound Resveratrol that inhibits the growth of prostate cancer cells (Hudson *et al*., 2007). The Kishmishi (or the Sultanin) grape variety, or what is called the Sultanin, is one of the Thompson seedless varieties, mainly intended for drying, and it is also used as table grapes,

characterized by its conical clusters, oval grains, juicy fleshy pulp, and regular hermaphrodite flowers (Al-Saeedi, 2000).

Spraying nutrients on the vegetative system can provide the plant with 85% of its need for nutrients (Al-Sahhaf, 1989). Iron is one of the micronutrients that plays a role in the growth and development of plants and is the driving force for many of the vital activities carried out by plants, and one of the most widely used forms of iron is chelated iron. Which is included in composition and formation of Cytochromes, which are of great importance in the process of respiration and photosynthesis, Fe-EDDHA and Fe-EDTA are common chelated iron compounds in many plants (Al-Qayyim, 1999; Focus, 2003; Al-Shalt, 2006). In this regard, Jalab and Al-Sallom (2016) showed in their study the foliar spraying of grape vines by iron chelating compounds EDTA, chelated iron 20%, at a rate of 1.25 g L^{-1}, to significantly improve their characteristics. And in the study of Al-Obeidi (2017) when spraying grape seedlings with iron at a concentration of 100 mg L^{-1}, a significant increase in the length and diameter of the main stem, the area of one leaf and its content of chlorophyll, nitrogen, phosphorus and potassium for both seasons reached (120.31, 407.00 cm and 1.48, 2.67 cm and 46.72, 143.40 cm^2 and 46.28, 43.11 SPAD unit and 1.84, 2.45% and 0.195, 0.295% and 1.90, 1.60%) respectively compared with comparison treatment.

Growth regulators play an important role in the growth and productivity of fruit trees. They are a group of non-nutritive organic compounds that are naturally produced in the endogenous plant or produced outside the plant industrially, these compounds play a vital role in the plant in low concentrations by controlling many physiological processes in the plant, as they stimulate, inhibit or modify the vital and metabolic processes necessary for the growth and development of the plant (Al-Khafaji, 2014). Cytokinins are compounds derived from Adenine that were isolated from Kinetin in 1955 by the scientist Miller. Cytokinins encourage cell division and it was found that they work to remove apical dominance, encourage lateral branching, and regulate the emergence of buds (Letham, 1969). Benzyl adenine is considered one of the most widely used cytokinins due to its cheapness and availability in the market, as well as its effectiveness because it contains three double bonds in its side chain (Krishnamorthy, 1981). It was found from the study of Al-ahbaby (2016) that spraying grape seedling by KT-30 cytokinin at concentration 10 mg L^{-1} led to a significant increase in the leaf area, chlorophyll index, dry weight of the plant, and the plant content of the macronutrient NPK, reached (17.92 leaf $Seedling^{-1}$, 704.29 cm^2, 48.40 Spad unit, 2.71 g, 1.42%, 0.132%, 6.46%) respectively. Hameed and Abbas (2017) showed that spraying grape vines by the growth regulator cytokinin KT-30 at a concentration 0.50 ml L^{-1} led to a significant increase in the area of one leaf, the leaf area of the vine, and the leaf content of nitrogen and potassium, compared with a concentration of 0.25 ml L^{-1} and treatment comparison. Due to the lack of studies on the subject for the grape cultivar Kishmishi, the research was conducted with the aim of encouraging vegetative growth of vines and knowing their chemical content through spraying by chelated iron and the growth regulator benzyl adenine, determining the best concentrations used between them individually or in combination. In addition to accelerating its growth and bringing it to the fruiting stage during the maximum period.

MATERIAL AND METHODS:

Location of the Experiment:

The research was conducted in the grape field of the University of Kirkuk / College of Agriculture in the Sayada region-Iraq, at latitude 35.33°N and 44.20°E, from 1/4/2023 to 1/8/2023. On vegetative growth and chemical content of young grape vines of Kishmishi cultivar, homogeneous in size and growth, planted in parallel lines, the distance between one line and another is 2.5 m, and between one vine and another on the same line is 2.5 m, at the age 7 years and pruned in the middle of February of each year, with all service operations for the vines such as irrigation, hoeing and weeding being carried out during the study period for all vines included in the experiment. The experiment was carried out in a randomized complete block design (RCBD) with three replications and two vines for each experimental unit. Thus, the number of vines in one replicate was 18 vines and the number of vines in the total experiment was 54 vines.

Treatments: Spraying with chelated iron with three concentrations (0, 0.5 and 1) g L^{-1} and Benzyl Adenine (6-benzyl amino purine) after preparing it by dissolving it with a little ethyl alcohol and completing it to the required volume with distilled water and with three concentrations (0, 50, 100) mg L^{-1}, where the factors were sprayed with three dates each 15/4, 1/5 and 15/5/2023 until complete wetness in the early morning.

Studied characteristics:

Measurements were taken at the end of the experiment on 1/8/2023, and the following characteristics were studied:

a. Stem length (cm)
b. Stem diameter (mm)
c. Leaf area (cm^2): It was measured according to (Saieed, 1990) as in the following equation: leaf area = large leaf area × weight of cut part / weight of large leaf.
d. Leaf content of chlorophyll (CCI): It was estimated using a CCM-200 Chlorophyll meter (Biber, 2007) by taking 10 leaf readings from each vine and then calculating their average.
e. Percentage of dry matter branches (%): Branches with a length of 20 cm were taken from each vine in the experimental unit and weighed, then dried in an oven at 70°C until the weight was stable, then the dry weight was weight was found according to the following equation: dry weigh of the branches = dry weight / wet weight × 100
f. Percentage of Nitrogen in leaf (%): Estimated using the micro_Kjeldahl apparatus as reported by (A.O.A.C., 1980).
g. Percentage of Phosphorous in leaf (%): It was estimated using a Spectrophotometer (EMC lab v-1 100) according to (Estefan *et al*., 2013).
h. Percentage of Potassium in leaf (%): It was estimated using a Flame Photometer (Elico CL-378) according to (Estefan et al., 2013)

i. Leaf content of Iron (mg km^{-1}): Estimated using an Atomic Absorption machine as reported by (Bhargava and Raghupathi, 1999).

Statistical analysis

The data were analyzed statistically using the SAS V9.0 program (SAS, 2001), and the averages were compared according to the Duncan test under a probability level of 0.05 (Roger Mead and Hasted, 2003).

RESULTS AND DISSCUSION:

It appears from the results shown in Table (1) that spraying by iron chelated at a concentration of 1 g L^{-1} had a significant effect on improving the vegetative growth characteristics of grape vines (Length and diameter of stem, leaf area, leaf content of chlorophyll, and percentage of dry matter in branches) which amounted (139.56 cm, 182.78 mm, 76.04 cm^2, 40.53 CCI and 41.28%) respectively, compare with the control treatment (102.82 cm, 159.89 mm, 61.86 cm^2, 35.60 CCI and 37.96%) respectively. The reason for this is due to the role of iron in the vital activities of the plant, where it enters as a catalyst in the construction of chlorophyll and also participates in the processes of oxidation and reduction and the building of important cytochromes in the processes of respiration and photosynthesis as the enzyme Cytochrome Oxidase participates in the process of transferring electrons to the respiratory chain and participates in helping the formation of chlorophyll, although it does not enter into its composition (Al-Sahhaf, 1989; Focus, 2003). The percentage of nitrogen, phosphorus, potassium and content of iron in the leaves of grapevines were significantly affected at a concentration of 1 g L^{-1} of iron chelated (2.34%, 0.34%, 0.85% and 651.70 mg kg^{-1}) respectively. The reason for this is due to the effect of iron on vegetative growth due to its participation in the manufacture of chlorophyll and the increase in the number of crana lamellae in chloroplasts and its participation in the formation of protein and the reduction of nitrates (Guller and Kruka, 1993). It is also involved in the synthesis of porphyrin and activates chlorophyll assimilation enzymes and in the synthesis of cytochromes, which play a major role in the process of transferring electrons in mitochondria(Abdel-Aal *et al*., 2013), which is positively reflected in the increase in the content of the leaves of nutrients, and that the increase in the content of the leaves of iron may be a result of the direct spread of the spray solution into the plant when feeding with iron (Al-Tahafi, 2004) These results agree with Jalab and Al-Sallom (2016), Azeez *et al*. (2017), Al-Obeidi (2017), Hussein *et al*. (2021), Muhammad and Latif (2022) and Ajboory and Al-Douri (2023)

Table (1) Effect of spraying with Fe-EDDHA on some characteristics of vegetative growth and chemical content of young Grape Vines Cv.Kishmishi

Fe-EDDHA g L^{-1}	Stem length (cm)	Stem diameter (mm)	Leaf area (cm^2)	Leaf content of chlorophyll (CCI)	Percentage of dry matter branches (%)
0	102.82 c	159.89 c	61.86 c	35.60 c	37.96 b
0.5	117.17 b	167.78 b	67.71 b	36.95 b	38.14 b
1	139.56 a	182.78 a	76.04 a	40.53 a	41.28 a

Fe-EDDHA g L^{-1}	Percentage of N in leaf (%)	Percentage of P in leaf (%)	Percentage of K in leaf (%)	Leaf content of Fe (mg kgm^{-1})
0	1.66 c	0.16 c	0.49 c	354.80 c
0.5	2.12 b	0.24 b	0.69 b	521.46 b
1	2.34 a	0.34 a	0.85 a	651.70 a

Values with similar letters are not significantly different according to Duncan multiple testing with 5% probability

The result in Table (2) show that the characteristics of the vegetative growth of young grape vines were significantly affected when spraying with different concentrations of the growth regulator Benzyl Adenine, where the concentration of 100 mg L^{-1} was superior and gave the highest rate in the studied traits (Length and diameter of stem, leaf area, its chlorophyll content and percentage of dry matter branches) with an increase rate of (11.25, 5.96, 12.54, 8.30 and 6.70)%, respectively, compared to the control treatment. This is attributed to the role of cytokinins in increasing cell division and widening, breaking apical dominance, stimulating the formation of lateral buds, and regulating the distribution of photosynthetic products towards the growth regions of the plant, as well as its role in delaying the loss and decomposition of chlorophyll, due to its inhibitory effect on the formation of degradation enzymes such as the enzyme Nuclease and Protease ant the inhibition of the enzyme chlorophyllase responsible for the decomposition of chlorophyll pigment (Abu Zeid, 2000; Jundia, 2003), which is reflected in a significant increase in vegetative traits. Spraying by a concentration of 50 and 100 mg L^{-1} of Benzyl Adenine also resulted in a significant increase in the leaf percentage of nitrogen (2.09 and 2.11)%, respectively, compared to the control treatment, which amounted to 1.92%, while the percentage of phosphorous and potassium, content of iron had a significant increase at a concentration of 100 mg L^{-1} compared with the rest of the concentrations (0.28%, 2.72% and 558.91 mg km^{-1}), respectively. The reason for this is due to the role of cytokinins in increasing the width of the stomata of the leaves by about 50% which causes an increase in the absorption and transfer of nutrients that enter through the wood, as well as its role in the transmission and reception of dissolved nitrogen compounds and the of sinks formation or places to attract and accumulate nutrients in the leaves and the regulation of the absorption of potassium ions K^+ (Davies, 2004). Also, the increase in vegetative growth in the plant is reflected in the ability of plants to absorb nutrients and thus increase their concentration in the leaves, and this agrees with Al-ahbaby (2016), Hameed and Abbas (2017), Rumman (2018), Al-Doori and Hussein (2023), Abbas and Tawfeeiq (2023) and Hana *et al*. (2024).

Table (2) Effect of spraying with Benzyl Adenine on some characteristics of vegetative growth and chemical content of young Grape Vines Cv.Kishmishi

Fe-EDDHA g L^{-1}	Stem length (cm)	Stem diameter (mm)	Leaf area (cm^2)	Leaf content of chlorophyll (CCI)	Percentage of dry matter branches (%)
0	113.18 c	165.89 c	64.29 c	36.48 b	38.49 b
50	120.46 b	168.78 b	68.96 b	37.09 b	37.82 b
100	125.91 a	175.78 a	72.35 a	39.51 a	41.28 a

Fe-EDDHA g L^{-1}	Percentage of N in leaf (%)	Percentage of P in leaf (%)	Percentage of K in leaf (%)	Leaf content of Fe (mg kgm^{-1})
0	1.92 b	0.22 b	0.61 c	470.37 b
50	2.09 a	0.24 b	0.69 b	498.68 b
100	2.11 a	0.28 a	0.73 a	558.91 a

Values with similar letters are not significantly different according to Duncan multiple testing with 5% probability

It is clear from the results of Table (3) that interaction treatment between spraying by chelated iron at concentration of 1 g L^{-1} of with benzyl adenine at concentration 100 mg L^{-1} of gave the highest values in all traits (Length of stem, content of chlorophyll, percentage of nitrogen and phosphorus and percentage of dry matter in branches), reached (144.04 cm, 41.91 CCI, 2.38%, 0.35% and 43.02%), respectively, significantly outperforming the rest of the overlaps. While the comparison treatment gave 0 g L^{-1} of chelated iron and 0 mg L^{-1} of benzyl adenine the lowest rate (96.76 cm, 34.09 CCI, 1.35%, 0.13%, 35.89%), respectively, for all the mentioned characteristics.

The interaction treatment between spraying by chelated iron at concentration of (1 g L^{-1} of with benzyl adenine 100 mg L^{-1}) and (1 g L^{-1} of with benzyl adenine 50 mg L^{-1}) resulted in a significant superiority in diameter of stem, leaf area, percentage of potassium and content of iron compared with the rest of the interferences and gave the highest average (186.33 and 184.67 mm, 79.73 and 78.39 cm^2, 0.88 and 0.86 %, 675.89 and 668.17 mg km^{-1}), respectively, the comparison treatment gave chelated iron at concentration of 0 g L^{-1} with benzyl adenine 0 mg L^{-1}, the lowest rate for the mentioned characteristics (157.33 mm, 58.88 cm^2, 0.38%, 309.74 mg km^{-1}), respectively. The interpretation of the overlapping effects may be due to the physiological factors represented in the absorption and transfer of nutrients within the plant, and the production of growth-promoting substances, As well as the length of the growth period, which reflects positively on the nutritional status of the vine and improve the characteristics of vegetative growth (Salman, 1988).

Table (3) Effect of interference Fe-EDDHA and Benzyl Adenine on some characteristics of vegetative growth and chemical content of young Grape Vines Cv.Kishmishi

Fe-EDDHA g L^{-1}	Benzyl Adenine mg L^{-1}	Stem length (cm)	Stem diameter (mm)	Leaf area (cm^2)	Leaf content of chlorophyll (CCI)	Percentage of dry matter branches (%)
0	0	96.76 h	157.33 e	58.88 g	34.09 f	35.89 d
	50	98.66 h	160.00 d	61.52 f	34.16 f	36.95 d
	100	113.03 f	162.33 cd	65.17 e	38.55 c	41.04 b
0.5	0	107.31 g	163.00 c	63.99 e	35.99 e	38.69 c
	50	123.55 d	161.67 cd	66.97 d	36.77 de	36.59 d
	100	120.67 e	178.67 b	72.16 b	38.08 cd	39.14 c
1	0	135.47 c	177.33 b	70.01 c	39.35 bc	40.88 b
	50	139.17 b	184.67 a	78.39 a	40.33 b	39.93 bc
	100	144.04 a	186.33 a	79.73 a	41.91 a	43.02 a

Fe-EDDHA g L^{-1}	Benzyl Adenine mg L^{-1}	Percentage of N in leaf (%)	Percentage of P in leaf (%)	Percentage of K in leaf (%)	Leaf content of Fe (mg kgm^{-1})
0	0	1.35 d	0.13 e	0.38 g	309.74 e
	50	1.86 c	0.15 e	0.53 f	335.67 e
	100	1.76 c	0.20 d	0.55 f	419.00 d
0.5	0	2.10 b	0.23 d	0.63 e	490.33 c
	50	2.08 b	0.21 d	0.69 d	492.19 c
	100	2.19 ab	0.29 c	0.75 c	581.85 b
1	0	2.31 ab	0.31 bc	0.81 b	611.04 b
	50	2.33 ab	0.35 ab	0.86 a	668.17 a
	100	2.38 a	0.35 a	0.88 a	675.89 a

Values with similar letters are not significantly different according to Duncan multiple testing with 5% probability

CONCLUSION

Conclude from the results of the Tables (1, 2 and 3) that spraying young grape vines of Kishmishi variety by chelated iron at concentration 1 g L^{-1} and the growth regulator Benzyl Adenine at concentration 100 mg L^{-1} significantly improved the growth of the vines and increased the content of its leaves of mineral elements.

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