Tikrit Journal for Agricultural Sciences (2023) 23 (3): 32-40
DOI: https://doi.org/10.25130/tjas.23.3.4

IRAQI Academic Scientific Journals
العراقية المجلات الأكاديمية العلمية

ISSN:1813-1646 (Print); 2664-0597 (Online)
Tikrit Journal for Agricultural Sciences
Journal Homepage: http://www.tjas.org
E-mail: tjas@tu.edu.iq

TJAS
Tikrit Journal for Agricultural Sciences

Effect of different levels of pruning and nitrogen fertilizer on vegetative traits and chemical content of *Rosmarinus officinalis* L.

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KEY WORDS:
rosemary, pruning, nitrogen fertilizer, chemical content, vegetative traits

Received: 25/08/2022
Accepted: 27/09/2022
Available online: 30/09/2023

ABSTRACT

The experiment was performed in a greenhouse belonging to the Department of Horticulture and Landscaping gardening, College of Agriculture, Tikrit University for the period from November 2021 - May 2022. The experiment included two factors: the level of pruning (without pruning, moderate pruning, and severe pruning); the addition of nitrogen fertilizer at two levels (73.4 and 146.7 g $N.L^{-1}$ in the form of urea (46% N) according to the randomized complete block design (RCBD), with three replicates and three plants for each experimental unit. The results showed that the treatment of severe pruning (H2) and moderate pruning (H1) were significantly superior in the trait of the branches number (26.65 and 24.61 plants.branches^{-1}), respectively. The treatment without pruning (H0)) was significantly superior in the leaves content of chlorophyll of (0.36 $mg.g^{-1}$). The nitrogen fertilization treatment of the first concentration (F1) had a significant effect on the trait of plant height, which amounted to 37.28 cm. As for the nitrogen fertilization treatment of the second concentration (F2), it was significantly superior in the percentage of nitrogen 1.31%, phosphorous 0.24%, and potassium 1.43%. As for the interaction, the treatment (S1H0) was significantly superior in the leaves content of chlorophyll, and the treatments (H0F1) and (H0F2) also gave a significant superiority in the leaves content of chlorophyll. The interference treatment (H2F2) also led to a significant superiority in the leaves content of phosphorous. As for potassium.

تأثير مستويات مختلفة من التقليم والسماد النتروجيني على الصفات الخضرية والمحتوى الكيميائي لبنات اكليل الجبل *Rosmarinus officinalis* L.

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الخلاصة

نفذت التجربة في البيت الزجاجي التابع لقسم البستنة وهندسة الحدائق في كلية الزراعة – جامعة تكريت للفترة من تشرين الثاني 2021 – أيار 2022. وتكونت التجربة من عاملين هما مستوى التقليم (بدون تقليمH0 وتقليم متوسطH1 وتقليم جائرH2)؛ واضافة السماد النيتروجيني بمستوين (F1 73.4 و F2 146.7 غم N لتر$^{-1}$على هيئة يوريا (46% N). نفذت التجربة وفق تصميم القطاعات العشوائية الكاملةRCBD وبثلاث مكررات وبواقع ثلاث نباتات لكل وحدة تجريبية. بينت النتائج التي تم التوصل اليها ان معاملة التقليم الجائر (H2) والتقليم المتوسط (H1) قد تفوقت بشكل معنوي في صفة عدد الأفرع 26.65 و24.61 فرع نبات$^{-1}$ على التوالي وتفوقت معاملة بدون تقليم (H0) في محتوى الأوراق من الكلوروفيل معنوياً 0.36ملغم غم$^{-1}$. أثرت معاملة التسميد النتروجيني للتركيز الاول(F1) معنوياً في صفة ارتفاع النبات بلغت 37.28سم . أما معاملة التسميد النتروجيني للتركيز الثاني (F2) فقد تفوقت معنوياً في صفة النتروجين 1.31% والفسفور 0.24% والبوتاسيوم 1.43%. اما التداخل فقد تفوقت المعاملة (S1H0) معنوياً في صفة محتوى الأوراق من الكلوروفيل وايضا أعطت معاملتي (H0F1) و(H0F2) تفوق معنوي في محتوى الأوراق من الكلوروفيل . كما ادت معاملة التداخل (H2F2) الى تفوق معنوي في محتوى الأوراق من الفسفور. أما فيما يخص البوتاسيوم بينت نتائج المعاملات (H0F2), (H1F2), (H2F2) تفوق معنوي.

الكلمات المفتاحية: اكليل الجبل, تقليم, سماد نتروجيني, المحتوى الكيميائي, الصفات الخضرية.

INTRODUCTION

Aromatic plants are among the oldest plants known and used by humans for the purpose of food and medicine throughout the ages. Studies have shown that a large proportion of the population of developing countries depends heavily on these plants to meet their basic medical care needs, and despite the availability of modern medicine, herbal medicine was the most prevalent (El-gabaly, 2007). One of the most important aromatic plants that are widely used in the food and pharmaceutical industries is the rosemary plant (*Rosmarinus officinalis* L.), which is a perennial plant belonging to the Lamiaceae family, which is often known as the mint family (Celiktas *et al*., 2007). The rosemary plant is used to preserve many foods, such as meat and fish, because of its properties that prevent fat rancidity (Azzazi, 2009).

It is also used in treating many diseases. The importance of the many active substances made it an important plant, and it is necessary to take care of it, develop its growth, improve its qualities, and increase the oils produced from it (Wiedenhoeft, 2006). Pruning is considered one of the most important economic techniques where it not only controls plant growth but also improves vegetative growth traits (Saffari *et al*., 2004). Pruning also increases the commercial value in terms of controlling the amount of both vegetative growth and controlling the yield during the different seasons of the year. The importance of nitrogen fertilization comes from the importance of nitrogen,

which plays a major role in increasing plant growth where it is considered one of the necessary elements for its growth, it is a synthetic part of many plant materials and compounds such as proteins and nucleic acids, which are one of the components of the plant cell. It also enters into the construction of the chlorophyll molecule and activates the processes and reactions associated with the protoplasm, enzymatic reactions, and the process of photosynthesis to greatly encourage vegetative growth. Hassan *et al*., (1990) showed that nitrogen contributes to increasing the vegetative growth in general of plants as a result of its entry into the important structures of plant tissues, as well as leads to an increase in the lateral buds of the plant. For these reasons, this study was developed, which aims to: Study the different levels of pruning and how nitrogen fertilizer affects in improving vegetative growth and chemical content.

MATERIALS AND METHODS

The experiment was performed in a greenhouse belonging to the Department of Horticulture and Landscap , College of Agriculture, Tikrit University for the period from November 2021 - May 2022. The experiment included studying the effect of pruning and adding nitrogen fertilizer on plant growth and production from the quantity and quality of the essential oil of *Rosmarinus officinalis* L.. Rosemary seedlings were selected from specialized nurseries in Baghdad province in mid-November 2021, A year and a half old and length 50cm, homogeneous in their growth as possible in terms of branches and size. The plants were transferred from plastic bags to large plastic pots with a capacity of 8 liters and dimensions of 23 cm in diameter and 26 cm in height. Which were filled with gypsum soil from the fields of the College of Agriculture, Department of Horticulture. and landscape, after removing the surface layer at a depth of 30 cm and with a rate of 3 replications per treatment. It was acclimatized for a week inside the greenhouse to provide the appropriate environmental conditions for all plants. Before starting the experiment, the gypsum soil was analyzed and all chemical and physical tests were conducted for it as shown in Table (1).

Table 1: Physical and chemical properties of the soil used in the experiment.

Traits		Units	Values
pH		---	7.43
EC		$mmhos.cm^{-3}$	1.97
O.M		$g.kg^{-1}$	11.5
Texture		---	Loamy sandy
Sand		$g.kg^{-1}$	750
Silt		$g.kg^{-1}$	230
Clay		$g.kg^{-1}$	20
N availability		$gm.kg^{-1}$	15.69
P availability		$gm.kg^{-1}$	5.6
K	exchangeable	$gm.kg^{-1}$	72.6
	dissolved		35
	availability		107.6

* All analyzes were conducted in the laboratory of the Department of Soil Sciences and Water Resources, Tikrit University, College of Agriculture.

Study factors:

First factor: pruning:

Seedlings were pruned in three levels Without pruning, which is symbolized by the symbol (H0), moderate pruning (the pruning is about 15 cm) which is symbolized by the symbol (H1), severe pruning (the pruning is about 25 cm) and symbolized by the symbol (H2). Taking into account that the average length of plants is about 50.33 cm. The pruning process was conducted a week after the transfer of seedlings to the pots on 6/12/2021 according to the treatments and their random distribution in the greenhouse by means of manual pruning scissors.

The second factor: the addition of nitrogen fertilizer at two levels:

Nitrogen fertilizer (urea) was added at an amount of (73.4 g urea) and (146.7 g urea) according to Studies Previous for the first and second levels, respectively, and this amount was divided into two batches for each level, this means (36.7 and 74.4 g urea) for the first and second levels, respectively, where both fertilizers (F1 and F2) are dissolved in 22 L of water, after which the fertilizer was added in liquid form and with an amount of 400 ml through the soil for one pot and for all batches. Batches were added on 12/08/2021, and between each batch and another, is one week.

Studied traits:

Traits of vegetative growth

1- **Plant height (cm.plant^{-1}):** The plant height was measured at the end of the experiment on the date2022/5/1 by means of a tape measure from the area of contact of the stem with the soil to the highest growing top of the three plants for each treatment and its average was taken.

2- **Number of branches (branch.plant^{-1}):** The number of branches connected to the main stem was calculated at the end of the experiment on the date2022/5/1 from the first branch near the surface of the soil to the top and for the three plants in each treatment and their average was taken.

3- **The leaves content of Total chlorophyll (mg.g^{-1}):** The content of chlorophyll in leaves was estimated according to the method of (Knudson *et al*., 1977), on the date2022/5/6 By taking 1.5 g of the fresh weight of the leaves and placing them in containers and adding 10 ml of ethanol to them every day for four days, and after the chlorophyll dissolved, it was placed in glass bottles and then the light absorption was read for the sample device spectrophotometer At two wavelengths (649 and 665) nanometers.

4- **Percentage of dry matter for the total vegetative (%).** The fresh weight of the total vegetative on the date2022/5/7 was estimated after being separated from the root system of the plant using a sensitive scale and then dried in the electric oven at a temperature of 65-70 C for 48-72 hours until the weight was stable, and weighed to take the dry weight (Al-

Sahhaf, 1989), then the percentage was extracted using the following equation: percentage of dry matter = dry weight / fresh weight x 100.

5- **Estimating nitrogen concentration (N%):** the percentage of nitrogen was estimated by the semi-micro Kjeldahl method, according to the method described by (AOAC, 1980).
6- **Estimating the phosphorous concentration (P%):** it was estimated according to the method mentioned by (AOAC, 1980).
7- **Estimating potassium concentration (K%):** according to the method described by (AOAC, 1980).

RESULTS AND DISCUSSION

Table (2) indicates that the levels of pruning and the bi-interaction between the levels of pruning and nitrogen fertilization did not significantly affect the plant height, While nitrogen fertilization (F1) led to a significant increase in this trait with an increase of 37.28 cm compared to the treatment of nitrogen fertilization (F2), which was 27.74 cm.

Table 2: The effect of pruning and levels of nitrogen fertilization and their interactions on the plant height for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	34.88 a	26.33 a	30.61 a
H1	39.74 a	25.52 a	32.63 a
H2	37.22 a	31.36 a	34.29 a
Nitrogen fertilization (F)	37.28 a	27.74 b	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Table (3) indicates that the levels of moderate and severe pruning did not differ from each other significantly, but treatment (H2) gave the highest percentage of the number of branches amounted to (26.65 branches.plant^{-1}) compared to treatment (H0), which differed significantly from the two pruning treatments, which gave The lowest number of branches was (18.18 branches.plant^{-1}). The effect of nitrogen fertilization did not significantly affect the number of branches. As for the treatment of bi-interaction between pruning and nitrogen fertilization, it did not significantly affect the traits of the number of branches.

Table 3: The effect of pruning and levels of nitrogen fertilization and their interactions on the Number of branches (branches.plant^{-1})for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	18.27 a	18.08 a	18.18 b
H1	26.58 a	22.63 a	24.61 a
H2	28.33 a	24.97 a	26.65 a
Nitrogen fertilization (F)	24.39 a	21.89 a	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Table (4) indicates significant differences for treatment (H0) in chlorophyll content amounted to (0.36 $mg.g^{-1}$) compared with treatment (H2), which gave the lowest percentage of chlorophyll content amounted to (0.28 $mg.g^{-1}$). There was no significant effect on the effect of nitrogen fertilization. From the interaction treatment between pruning and nitrogen fertilization, we note that treatments (H0F1) and (H0F2) were significantly superior by giving the highest percentage of (0.36 $mg.g^{-1}$), which did not differ significantly from treatments (H1F1) and (H1F2) compared with treatments (H2F1) and (H2F2) which gave the lowest value of (0.27 and 0.29 $mg.g^{-1}$), respectively.

Table (5) shows that the pruning levels and the effect of nitrogen fertilization and their bi-interactions did not significantly affect this trait.

Table 4: The effect of pruning and levels of nitrogen fertilization and their interactions on the leaves content of total chlorophyll ($mg.g^{-1}$) for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	0.36 a	0.36 a	0.36 a
H1	0.33 a	0.33 a	0.33 b
H2	0.27 b	0.29 b	0.28 c
Nitrogen fertilization (F)	0.32 a	0.33 a	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Table 5: The effect of pruning and levels of nitrogen fertilization and their interactions on the Percentage of dry matter in the total vegetative (%) for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	30.60 a	30.02 a	30.31 a
H1	31.80 a	31.52 a	31.66 a
H2	30.99 a	32.00 a	31.50 a
Nitrogen fertilization (F)	31.13 a	31.18 a	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Table (6) shows that pruning levels and their bi-interactions did not significantly affect this trait, but the effect of nitrogen fertilization (F2) led to an increase in the percentage of nitrogen that amounted to 1.31% compared to the treatment of nitrogen fertilization (F1), which gave the lowest percentage of nitrogen 1.24%.

Table 6: The effect of pruning and levels of nitrogen fertilization and their interactions on the Nitrogen content in leaves (%)for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	1.24 b	1.31 a	1.28 a
H1	1.25 b	1.31 a	1.28 a
H2	1.24 b	1.32 a	1.28 a
Nitrogen fertilization (F)	1.24 b	1.31 a	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Table (7) indicates that pruning levels did not have a significant effect on increasing the percentage of phosphorous. The effect of nitrogen fertilization (F2) was significantly superior with

an increase of the phosphorous percentage of 0.24% compared with the effect of nitrogen fertilization (F1) which gave the lowest percentage of 0.20%.As for the interaction treatment between pruning and nitrogen fertilization, the results showed that (H2F1) treatment was significantly superior by giving the highest percentage of phosphorous, which amounted to 0.25%, which did not differ significantly from (H0F2) and (H1F2) treatments compared with the treatment between severe pruning and nitrogen fertilization (F1), which did not Significantly different from other treatments by giving the lowest value of 0.20% which did not differ from (H0F1) and (H1F1) treatments.

Table 7: The effect of pruning and levels of nitrogen fertilization and their interactions on the Percentage of phosphorous in leaves (%)for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	0.21 b	0.24 a	0.22 a
H1	0.21 b	0.24 a	0.22 a
H2	0.20 b	0.25 a	0.22 a
Nitrogen fertilization (F)	0.20 b	0.24 a	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Table (8) shows that pruning levels did not significantly affect this trait, while the effect of nitrogen fertilization (F2) led to an increase in potassium that amounted to 1.43% compared with nitrogen fertilization (F1), which gave the lowest value of 1.38%.As for the interaction between pruning and nitrogen fertilization, we note that the interaction treatments (H0F2), (H1F2), and (H2F2) were significantly superior to an increase in potassium that amounted to 1.43% and did not differ significantly from each other compared with the treatments of (H0F1) and (H2F1), which gave the lowest value of 1.38%, which did not differ significantly from (H1F1) treatment.

Table 8: The effect of pruning and levels of nitrogen fertilization and their interactions on the leaves content of Potassium (%)for rosemary leaves.

Nitrogen fertilization / Pruning	**F1**	**F2**	Pruning levels (H)
H0	1.38 b	1.43 a	1.40 a
H1	1.39 b	1.43 a	1.41 a
H2	1.38 b	1.43 a	1.40 a
Nitrogen fertilization (F)	1.38 b	1.43 a	

* Values with similar letters for each factor or their interactions do not differ significantly according to Duncan's new multiple range test under the 5% probability level.

Through the results shown in the previous results tables. Pruning is an important factor to push the plant to form a total vegetative as a result of the influence of growth regulators, light, and temperature, and pruning led to a significant increase in the number of branches and the leaves content of chlorophyll, This is because the greater the intensity of pruning, the less wood left behind, and thus the reduction in the number of nodes containing the buds or the lack of buds left on

the wood left during pruning. Pruning has a role in removing the developing apical meristem of the branches, thus breaking the apical dominance of the terminal bud and encouraging the growth of lateral buds (Jindia, 2003). The pruning also leads to an increase in the permeability of light to the heart of the tree and its entry into the process of photosynthesis, thus increasing processed nutrients (including carbohydrates) inside the tree and improving vegetative growth. Nitrogen has an important role in increasing plant height, where nitrogen is the main component of protein and nucleic acids, which is useful in plant growth (Haque *et al*., 2001). Nitrogen also stimulates the production of auxins, which may encourage the process of cell division and elongation of cells, so the height of the plant increases (Arslan, 1974). As for the nutrients nitrogen, phosphorous, and potassium, the increase in nitrogen in the leaves is attributed to the direct addition of nitrogen, which increases its absorption from the plant tissue (Al-Sahhaf, 1989). Nitrogen fertilization also helps in the formation of a good total vegetative and root system, which increases the process of absorption and accumulation of this element in the plant tissues. The increase resulting from the addition of urea may be attributed to the role of nitrogen in the formation of a strong total vegetative and root system, through its effect on increasing the activity of gibberellins within the plant tissues, which leads to cell division and elongation (Rajagopal and Roa, 1974). As for the increase in phosphorus, it is due to the fact that nitrogen encouraged its absorption from the agricultural medium, and this was confirmed by (Quraysh, 1984).

CONFLICT OF INTEREST

The authors declare no conflicts of interest associated with this manuscript.

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