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Study of the effect of cutting type and growth regulator indole butyric acid on the rooting and growth of mulberry cuttings planted in open and greenhouse cultivation.

[*1]Suzan Ali Hussein **[2]Mohammed Muntaz Ibrahim** **[3]Mine Pakyurek**

[1]*Department of Horticulture, College of Agriculture, Kirkuk University, IRAQ.*
[2]*Department of Forests Sciences, College of Agriculture, Kirkuk University, IRAQ.*
[3]*Department of Horticulture, Faculty of Agriculture, Siirt University, Turkey.*

*Corresponding Author: suzanali8@uokirkuk.edu.iq .

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ABSTRACT

The study was conducted in the field of the College of Agriculture, University of Kirkuk, located in Al-Sayada area, Iraq, during the *2024* growing season, to study the effect of three types of mulberry cuttings (Basal or Hardwood, Meddle and Terminal) by immersion in the growth regulator IBA at three concentrations (0, 1000 and 2000) mg L^{-1} at depth of 3cm for 10 seconds, and planted in two ways (Open and Greenhouse). The experiment was designed according to a RCBD design with two independent experiments. The number of cuttings for each cultivation method was 270 cuttings, and the total number of cuttings included in the study was 540 cuttings. The results showed that the basal (Hardwood) cuttings were significantly superior in all the studied traits of vegetative and root growth, except for the seedling height, for both the open and greenhouse cultivation methods, compared to the meddle and terminal cuttings. The treatment with a concentration of 2000 mg L^{-1} of auxin IBA was superior in the percentage of rooted cuttings, root length and the average number of new leaves formed on the cuttings, while the concentrations of 1000 and 2000 mg L^{-1} were superior in the number of roots and the average number of new branches compared to open cultivation. As for the greenhouse cultivation method, the concentration of 2000 mg L^{-1} of IBA had a significant improvement in all studied traits except for seedling height, in which the concentrations of 1000 and 2000 mg L^{-1} had a significant effect compared to the control treatment. The two-way interactions between the two factors a significant effect on all studied traits for both cultivation methods.

Keywords: *Type mulberry cutting, Indole butyric acid, Cultivation method.*

INTRODUCTION

The mulberry *Morus nigra* belongs to the Moracea family and the genus Morus, which contains approximately 16 species distributed in the northern temperate regions and extending to the tropical regions of Africa and America, among them, approximately 11 species are widely distributed in China, where it was cultivated since 400 years BC. From there, it spread to the continent of Asia and reached Europe in the 12th century [1, 2]. The mulberry tree has great economic importance as it is the main and only food for silkworms, its leaves are used as animal and poultry feed, as a diuretic, regulates blood sugar levels, and treats liver problems, its fruits have a high nutritional value and are rich in vitamins and minerals such as potassium, magnesium, copper, zinc and calcium, and are a source of iron for the human body, blackberries also have a stimulating effect due to the presence of anthocyanins, which act as a preventative and treatment for the mouth, throat, and esophagus, and increase immunity [3]. One of the methods of propagating mulberry is stem hardwood cutting, which is used in many countries, resulting plants are genetically similar to the mother plant from which the cuttings were taken, and have faster and more uniform growth, and in large numbers [4]. Propagation by cuttings is a type of vegetative propagation and is one of the most important means of reproductive renewal for many horticultural crops, the formation of lateral roots is considered a basic condition for the success of the cuttings, cuttings types are classified according to the type, age and freshness of the plant part used, which differ in their success rate according to these factors [5]. The location of the cuttings is of great importance for the success of rooting the cuttings, as it is due to the amount of nutrients stored in them and the percentage of wood, the cuttings are divided according to their location into hardwood (basal) cuttings, which are a year or more old, as they are characterized by being more woody and their rooting percentage increases due to the abundance of nutrients compared to the other parts, especially the terminal ones, which are characterized by the lack of nutritional storage in them, as well as the intermediate cuttings that are taken from the middle of the branches [4, 6]. [7] indicated that the rooting of hardwood cuttings of blackberry by the bunch-planting method recorded the highest percentage of rooted cuttings and number of roots compared to other planting methods, reaching (60% and 2.2 roots) respectively. [8] showed in their study of rooting two types of fig cutting that hard wood cuttings recorded the highest percentage of rooting, number of roots per cutting and root length

compared to semi hard wood cuttings. [9] also concluded that the type of cutting has a direct effect on the rooting and growth rate of Leconte pear seedlings, as the hard wood cuttings recorded the highest fresh and dry weight of the root vegetative system, number of leaves, total chlorophyll and leaf area compared to the soft cuttings. [10] explained in their study on increasing the rooting rate of soft and hard wood olive cuttings that the hard wood cuttings recorded the highest rate of rooting rate, average number and length of roots, number of new growths and dry weight of the vegetative and root system compared to the soft cuttings.

The use of growth regulators is one of the ways to enhance the success rate of cuttings, some regulators encourage the formation of adventitious roots on cuttings, such as auxins, especially indole butyric acid and indole acetic acid and some inhibit root formation, such as abscisic acid and ethylene [5]. Indole butyric acid is considered one of the synthetic auxins, its salts are in the form of sodium, potassium or ammonium salts, it is characterized by the stability of its chemical composition and lack of movement and transfer within plant tissues, it helps in producing small, strong and extremely long lateral roots [11]. Study by [12] on the effect of auxins on the rooting of mulberry stem cuttings, it was found that treatment with a concentration of 2000 mg L^{-1} of indole butyric acid significantly increased the percentage of rooted cuttings, the number of primary and secondary roots, root length and dry and fresh weight of root system, reaching (70%, 10.77, 12.77 root cutting^{-1}, 10.22 cm, 3.43 g and 1.88 g) respectively, compared to the concentration of 1000 mg L^{-1} and control treatment. [13] showed that treating hard wood cuttings of mulberry with a concentration of 1000 mg L^{-1} of indole butyric acid increased the percentage of rooted cuttings, number of roots, average number, length and diameter of branches and the number of leaves in both seasons compared to a concentration of 1500 mg L^{-1} and treatment. In the study of [14], treating mulberry cuttings with a concentration of 3000 mg L^{-1} of indole butyric acid significantly improved the number of roots produced by the cutting, root length and fresh and dry weight of the root system, reaching (9 root cutting^{-1}, 25.06 cm, 7.41 g and 2.46 g) respectively, compared to the rest of the concentrations. [15] found that when grape cuttings oh Halwani and Kamali cultivars were treated with 1000 mg L^{-1} of indole butyric acid, there was a significant increase in the percentage of rooted cuttings and the root and vegetative growth characteristics of the resulting seedlings. Thus, the study aims to determine the best location for taking cuttings and the appropriate concentration of indole butyric acid for mulberry propagation and the appropriate planting method to obtain good specifications for the root and vegetative growth of the resulting seedlings.

Materials and Methods:

The study was carried out in the field of the College of Agriculture/ University of Kirkuk-Iraq, located in the Sayada area, during the *2024* growing season, from February to June, the cuttings were selected from 12-year-old mulberry trees of three types (Basal or Hardwood, Meddle and Terminal) with a length of 20-25 cm and a diameter of 1-1.5 cm. All leaves on the cuttings were removed and their bases were immersed in three concentrations of indole butyric acid (0, 1000 and 2000) mg L^{-1} at a depth of 3cm for 10 seconds. Then they were planted in black polyethylene bags with a capacity of 5 kg of soil consisting of a growing medium (mixture: peat moss in a ratio of 2:1) and the cuttings were planted in two locations, the first inside the open field and the second in the greenhouse cultivation. The experiment was designed according to a randomized complete block design (RCBD) with three replicates and 10 cuttings per experimental unit. Thus, the number of cuttings in each replicate was 90 cuttings and the total number of cuttings included in the experiment was 540 cuttings for both locations (open field and greenhouse cultivation). The data were statistically analyzed according to the analysis of variance table using the [16] program for analyzing agricultural experiments, and the means were compared using Duncan's multiple range test at a probability level of 0.05 [17].

Studied characteristics: Measurement were taken at the end of the experiment, 4 months after the date of planting the cuttings.

1- Rooted cuttings percentage (%): Calculated according to the following equation:

$$\text{Percentage rooted cuttings} = \frac{\text{number of rooted cittings}}{\text{number of planted cuttings}} \times 100$$

2- Number of roots (root cutting^{-1})
3- Root length (cm)
4- Transplant height (cm): The length of the new growths formed on the cutting was measured
5- Average number of leaves (leaf cutting^{-1}): The number of new formed leaves on the cuttings was calculated for all transplants and then divided by their number
6- Average number of branches (branch cutting^{-1}): The number of new branches formed on the cuttings for all transplants was calculated and then divided by their number.

Results and Discussion:

1_ Open field planted: The results shown in Table (1) show that the type of cutting has a direct effect on the rooting rate of the cutting and the characteristics of vegetative and root growth of the resulting transplants of mulberry planted in the open field. The basal cuttings were significantly superior in the percentage of rooted cuttings, number of roots, root length and average number of leaves and new branches, reaching (67.07%, 3.78 root cutting^{-1}, 1.94 cm, 6.33 leaf cutting^{-1} and 3.56 branch cutting^{-1}) respectively, compared to the meddle and terminal cuttings, while the highest rate of transplant height was recorded in the basal cuttings, reaching 23.52 cm, which did not differ significantly from the meddle and terminal cuttings, which recorded (23.38 and 22.92) cm, respectively.

The effect of treating the cuttings with auxin IBA was significant in improving the root and vegetative characteristics, as the concentration of 2000 mg L^{-1} significantly exceeded the percentage of rooted cuttings, root length and average number of new leaves, which reached (71.44%, 1.98 cm and 6.56 leaf $cutting^{-1}$) respectively, with an increase over the control treatment of (27.57, 20.73 and 28.37)% respectively, while the concentrations of 1000 and 2000 mg L^{-1} were significantly superior in the characteristics of the number of roots and the average number of new branches, which reached (3.44, 3.78 root $cutting^{-1}$ and 3.22, 3.33 branch $cutting^{-1}$) respectively, compared to the control treatment, which recorded the lowest rate for both characteristics, which reached (2.56 root $cutting^{-1}$ and 2.22 branch $cutting^{-1}$) respectively. The concentration of 2000 mg L^{-1} of indole butyric acid recorded the highest rate in transplant height, reaching 23.46 cm, and it did not differ significantly with the concentration of 1000 and control treatment, which reached (23.28 and 23.09) cm, respectively.

The results of Table (2) show that the interference between the type of cutting and the concentrations of the auxin indole butyric acid had a clear effect in improving the studied characteristics of the mulberry cuttings planted in a open field, as the interaction at basal cuttings with a concentration of 2000 mg L^{-1} of indole butyric acid and recorded the highest rate in the percentage of rooted cuttings, root length and the average number of leaves and new branches, which reached (80.00%, 2.13 cm, 7.67 leaf $cutting^{-1}$ and 4.33 branch $cutting^{-1}$) respectively, compared to the rest of the interactions, while the interaction at basal cuttings with a concentration of 1000 mg L^{-1} of indole butyric acid, recorded the highest rate of transplant height, reaching 24.13 cm, while the lowest rate was in the interaction at terminal cuttings with a concentration of 1000 mg L^{-1} of indole butyric acid, which reached 22.00 cm, as for the number of roots formed on the cuttings, the interaction at basal cuttings with a concentration of 1000 and 2000 mg L^{-1} of indole butyric acid, recorded the highest rate, reaching 4.33 root $cutting^{-1}$ for both treatments, while the interaction terminal cuttings with the control treatment, recorded the lowest rate, reaching 2.33 root $cutting^{-1}$.

Table (1): Effect of cutting type and auxin IBA on some root and vegetative growth characteristics of mulberry cuttings planted in the open field

Treatments	Studied characteristics					
	Rooted cuttings (%)	Number of roots (root $cutting^{-1}$)	Root length (cm)	Transplant height (cm)	Number of leaves (leaf $cutting^{-1}$)	Number of branches (branch $cutting^{-1}$)
Cuttings type						
Basal	67.07 a	3.78 a	1.94 a	23.52 a	6.33 a	3.56 a
Meddle	60.67 b	2.89 b	1.71 ab	23.38 a	5.11 b	2.44 b
Terminal	58.44 b	3.11 b	1.62 b	22.92 a	5.67 ab	2.78 b
Auxin IBA (mg L^{-1})						
0	56.00 b	2.56 b	1.64 b	23.09 a	5.11 b	2.22 b
1000	58.73 b	3.44 a	1.66 b	23.28 a	5.44 b	3.22 a
2000	71.44 a	3.78 a	1.98 a	23.46 a	6.56 a	3.33 a

Similar letters for the same trait mean that there are no significant differences between the levels, Duncan's test at 0.05

Table (2): Effect of interference between the cutting type and auxin IBA on some root and vegetative growth characteristics of mulberry cuttings planted in the open field

Treatments		Studied characteristics					
Cuttings type	IBA (mg L^{-1})	Rooted cuttings (%)	Number of roots (root $cutting^{-1}$)	Root length (cm)	Transplant height (cm)	Number of leaves (leaf $cutting^{-1}$)	Number of branches (branch $cutting^{-1}$)
Basal	0	60.33 cd	2.67 bc	1.63 bc	23.50 ab	5.33 bc	2.33 c
	1000	60.87 cd	4.33 a	2.07 ab	24.13 a	6.00 bc	4.00 ab
	2000	80.00 a	4.33 a	2.13 a	22.93 abc	7.67 a	4.33 a
Meddle	0	53.00 d	2.67 bc	1.60 c	22.50 bc	4.67 c	2.00 c
	1000	57.67 cd	2.67 bc	1.47 c	23.70 ab	5.00 bc	3.00 abc

	2000	71.33 b	3.33 abc	2.07 ab	23.93 ab	6.67 bc	2.33 c
	0	54.67 cd	2.33 c	1.70 abc	23.27 abc	5.33 bc	2.33 c
Terminal	1000	57.67 cd	3.33 abc	1.43 c	22.00 c	5.33 bc	2.67 bc
	2000	63.00 c	3.67 ab	1.73 abc	23.50 ab	6.33 ab	3.33 abc

Similar letters for the same trait mean that there are no significant differences between the levels, Duncan's test at 0.05

2_ Greenhouse cultivation: The results shown in Table (3) show that the basal cuttings of mulberry significantly outperformed the two types of meddle and terminal cuttings by the greenhouse cultivation method in the percentage of root cuttings, number and length of roots, and average number of leaves and new branches, which reached (83.78%, 4.89 root cutting^{-1}, 2.12 cm, 7.89 leaf cutting^{-1} and 3.89 branch cutting^{-1}) respectively, the terminal cuttings recorded the lowest rate in the percentage of rooted cuttings and the number and length of roots, reaching (74.89%, 3.11 root cutting^{-1} and 1.67 cm), respectively, while the meddle cuttings recorded the lowest rate in the number of leaves and new branches, reaching (6.56 leaf cutting^{-1} and 2.11 branch cutting^{-1}) respectively, while the type of the cuttings did not significantly affect the transplant height trait. The effect of treating the cuttings with auxin Indole butyric acid was significant on the studied traits, as the treatment with a concentration of 2000 mg L^{-1} was superior in the percentage of rooted cuttings, number and length of roots, transplant height, and the average number of leaves and new branches, which reached (87.22%, 5.00 root cutting^{-1}, 2.07 cm, 24.64 cm, 8.11 leaf cutting^{-1} and 4.00 branch cutting^{-1}) respectively, with an increase rate of (22.08, 66.6, 35.71, 5.29, 30.38 and 125.9)% respectively compared to the control treatment.

The two-way interaction between the type of cuttings and auxin IBA concentration had an effect on the studied characteristics of the mulberry cuttings planted in a greenhouse cultivation, as shown in Table (4), as the interaction at basal cuttings with a concentration of 2000 mg L^{-1} of auxin indole butyric acid, were significantly superior in the percentage of rooted cuttings, number and length of roots, and the average number of leaves and new branches formed on the cuttings, which reached (94.67%, 6.33 root cutting-1, 2.57 cm, 9.67 leaf cutting^{-1} and 5.67 branch cutting^{-1}) respectively, compared to the rest of the interactions. While the interaction at meddle cuttings with a concentration of 2000 mg L^{-1} of indole butyric acid recorded the highest rate of transplant height, reaching 25.53 cm, compared to interaction the basal cuttings with control treatment of indole butyric acid concentration, which recorded the lowest rate, reaching 23.03 cm.

Table (3): Effect of cutting type and auxin IBA on some root and vegetative growth characteristics of mulberry cuttings planted in the greenhouse cultivation

Treatments	Studied characteristics					
	Rooted cuttings (%)	Number of roots (root cutting^{-1})	Root length (cm)	Transplant height (cm)	Number of leaves (leaf cutting^{-1})	Number of branches (branch cutting^{-1})
			Cuttings type			
Basal	83.78 a	4.89 a	2.12 a	23.89 a	7.89 a	3.89 a
Meddle	77.78 b	3.78 b	1.74 b	24.68 a	6.56 b	2.11 b
Terminal	74.89 b	3.11 b	1.67 b	23.96 a	7.00 b	2.67 b
			Auxin IBA (mg L^{-1})			
0	71.44 c	3.00 c	1.62 c	23.40 b	6.22 c	1.77 c
1000	77.78 b	3.77 b	1.84 b	24.48 ab	7.11 b	2.89 b
2000	87.22 a	5.00 a	2.07 a	24.64 a	8.11 a	4.00 a

Similar letters for the same trait mean that there are no significant differences between the levels, Duncan's test at 0.05

Table (4): Effect of interference between the cutting type and auxin IBA on some root and vegetative growth characteristics of mulberry cuttings planted in the greenhouse cultivation

Treatments		Studied characteristics					
Cuttings type	IBA (mg L^{-1})	Rooted cuttings (%)	Number of roots (root cutting^{-1})	Root length (cm)	Transplant height (cm)	Number of leaves (leaf cutting^{-1})	Number of branches (branch cutting^{-1})
Basal	*0*	74.00 cd	3.67 bcd	1.67 c	23.03 c	6.00 c	2.00 d
	1000	82.67 bc	4.67 b	2.13 b	23.43 abc	8.00 b	4.00 b

	2000	94.67 a	6.33 a	2.57 a	25.20 ab	9.67 a	5.67 a
	0	71.67 d	3.00 cd	1.60 c	23.50 abc	6.33 c	1.33 d
Meddle	1000	76.67 bcd	3.67 bcd	1.73 c	25.00 abc	6.67 bc	2.33 cd
	2000	85.00 b	4.67 b	1.90 bc	25.53 a	6.67 bc	2.67 bcd
	0	68.67 d	2.33 d	1.60 c	23.67 abc	6.33 c	2.00 d
Terminal	1000	74.00 cd	3.00 cd	1.67 c	25.00 abc	6.67 bc	2.33 cd
	2000	82.00 bc	4.00 bc	1.73 c	23.20 bc	8.00 b	3.67 bc

Similar letters for the same trait mean that there are no significant differences between the levels, Duncan s test at 0.05

The results obtained from this study can be interpreted as the superiority of basal (hardwood) mulberry cuttings compared meddle and terminal cuttings in most of the root and vegetative growth characteristics in open field (Tables 1 and 2) and greenhouse cultivation (Tables 3 and 4), this may be due to the fact that the basal cuttings contain stored nutrients such as carbohydrates in higher concentrations than the terminal and meddle cuttings, which play an important role in the rooting process, especially in the early stages of root formation, it is also a source of energy needed for vital processes in various plants, including the formation, growth and development of root primordia [18] In addition to the buds in the basal (hardwood) cuttings being more mature and in a physiological state that makes them more prepared for opening and growth, these factors lead to the formation of a strong root group and a high rooting rate, which is positively reflected in the growth and opening of the buds and improving the vegetative growth of the transplants [19]. These results conform with [7, 8, 20].

As for the effect of the auxin , Indole butyric acid in improving the percentage of rooted cuttings and the characteristics of root and vegetative growth, the reason for this may be due to the fact that auxins play an important role in the loss of differentiation, the transformation of meristematic cells, the formation of root primordia, and then the formation of lateral roots on the bases of the cuttings [21]. It is also noted that there are significant differences between the concentrations of auxin used in the experiment, Tables (1 and 2), for planted in the open field and Tables (3 and 4) for planted in the greenhouse cultivation, as the percentage of rooted cuttings increases as the concentration of auxin increases, as a result of the increased accumulation of nutrients, such as sugars, necessary for the formation of lateral roots in the treatment area, the association of auxins with some naturally occurring substances (Auxin Synergist) in the presence of some specialized enzymes (Poly phenol oxidase) to form auxin conjugates, which are highly effective in stimulating, forming and developing root principles [22, 21]. As a result of the strength of the root system, represented by the increase in the number and length of the roots formed on the cuttings, which work to absorb water and nutrients, in addition to the production of the growing tips of these roots of growth hormones such as auxins and cytokinins, which have a major role in the process of cell division and increasing the number of leaves formed [23] which improves the characteristics of vegetative growth in a significant way. These results are consistent with [12, 24, 14, 25, 26, 27].

Conclusion:

From the above, we conclude that propagating mulberry by basal (hardwood) cuttings and treating them with a concentration of 2000 mg L^{-1} of auxin Indole butyric acid, individually and in combination, gave the best results and improved the characteristics of root and vegetative growth open field and greenhouse cultivation.

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دراسة تأثير نوع العقلة ومنظم النمو إندول بيوترك اسد في نسبة تجذير ونمو عقل التوت المزروعة بطريقة مكشوفة ومحمية.

سوزان علي حسين[1] محمد منتاز ابراهيم[2] مينا باكيورك[3]
[1]قسم البستنة وهندسة الحدائق، كلية الزراعة، جامعة كركوك، العراق
[2]قسم علوم الغابات، كلية الزراعة، جامعة كركوك، العراق
[3]قسم البستنة، كلية الزراعة، جامعة سيرت، تركيا

الخلاصة
اجريت الدراسة في الحقل التابع لكلية الزراعة، جامعة كركوك، الواقعة في منطقة الصيادة، العراق خلال موسم النمو 2024، لدراسة تأثير ثلاث انواع من عقل التوت (قاعدية، وسطية، طرفية) بالغمر بمنظم النمو إندول بيوترك اسد بثلاث تراكيز (0، 1000، 2000) ملغم لتر$^{-1}$ بعمق 3سم ولمدة 10 ثواني والمزروعة بطريقتين (مكشوفة ومحمية)، صممت التجربة وفق تصميم القطاعات العشوائية الكاملة وبتجربتين مستقلة وكانت عدد العقل لكل طريقة من طرق الزراعة 270 عقلة وعدد العقل الكلية الداخلة في الدراسة 540 عقلة. واوضحت النتائج بان العقل القاعدية قد تفوقت معنويا بجميع الصفات المدروسة للنمو الخضري والجذري ما عدا صفة ارتفاع الشتلة ولكلا طريقتي الزراعة المكشوفة والمحمية مقارنة بالعقل الطرفية والوسطية، وتفوق المعاملة بتركيز 2000 ملغم لتر$^{-1}$ من الأوكسين إندول بيوترك اسد في نسبة العقل المجذرة وطول الجذور ومعدل عدد الاوراق الحديثة المتكونة على العقل، في حين تفوق التركيزين 1000 و2000 ملغم لتر$^{-1}$ في صفة عدد الجذور ومعدل عدد الافرع الحديثة بالنسبة للزراعة المكشوفة، اما في طريقة الزراعة المحمية فقد كان لتركيز 2000 ملغم لتر$^{-1}$ من إندول بيوترك اسد تحسن معنوي في جميع الصفات المدروسة عدا ارتفاع الشتلة الذي كان للتركيزين 1000 و 2000 ملغم لتر$^{-1}$ اثر معنوي مقارنة مع معاملة المقارنة، وكانت التداخلات

الثنائية بين العاملين تأثير معنوي واضح في جميع الصفات المدروسة ولكلا طريقتي الزراعة.

الكلمات المفتاحية: نوع عقل التوت، إندول بيوترك اسد، طريقة الزراعة.