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THE EFFECT OF PUTERSCINE AND SAL SALIC ACID AND THEIR INTERACTION ON THE MULTIPLICATION OF VEGETATIVE BUDS AND THEIR CHARACTERISTICS FOR DATE PALM CULTIVARS AL-BARHI *In vitro*

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ABSTRACT

This study was conducted at the Date Palm Research Center University of Basrah for the period from 25/1/2018 to 3/2/2019 AD, with the aim of knowing the effect of polyamine puterscine (put) and salicylic acid (SA) on the multiplication and elongation of vegetative buds and the results obtained can be summarized come:

1 - Addition of Put. to MS medium at a concentration of 150 mg.L^{-1} recorded significant increase in number of vegetative buds and Chlorophyll pigment and Carotene concentration (21.23 buds, 0.98 and 0.079 mg.gm^{-1} fresh weight) respectively, while the same concentration decreased from The percentage of deformed buds and phenols was (15.55% and 30.86 mg.gm^{-1} fresh weight).

2- The addition of SA. in the concentration of 1 mg.L^{-1} exceeded the diameter of the buds and the leaf content of the chlorophyll, carotene and carbohydrate dye (0.65 mm, 6.66 and 0.099 mg.gm^{-1} Fresh weight and 23.05 mg.gm^{-1} Dry weight), respectively, in reducing the same concentration of deformed buds and phenols (20.16% and 15.19 mg.gm^{-1} fresh weight), respectively.

3- The interaction of the concentrations 150 mg L^{-1} Put. and 1 mg.L^{-1} SA. showed a significant increase than the other interactions of chlorophyll and carotene which were (7.25 and 0.113) mg.g^{-1} fresh weight respectively, while the same interactions decreased the Percentage of deformed buds and phenols (13.33% and 26.23 mg.gm^{-1} dry weight) respectively.

Keywords: Puterscine; salsalic acid; vegetative buds; *in vitro*.

INTRODUCTION

The date palm tree *Phoenix dactylifera* L. belongs to the Arecaceae palm tree family and is a monoexual dioecious plant and one of the monocotyledons plants scattered in sub-tropical regions between 10-30° north latitude and extends up to 20° latitude south of the equator [1,2].

The technique of plant tissue culture is a means of propagation and improvement of many different types of plants, including palm trees. The vegetative propagation in vitro is one of the major applications of great commercial importance to plant tissue culture technology as it is the best way to propagate vegetative plants, which are followed by different methods of differentiation and formal formation such as Formation of adventious buds, growth of axillary buds, and stimulation of somatic embryos [3,4].

The process of organ formation is defined as the process by which cells and tissues are forced to undergo changes that lead to the production of Unipolar structures called vegetative growth principles whose transporting vessels are associated with the vessels carrying the tissue from which they were formed [5].

The polyamine group has been used in recent years, including puterscine, in plant tissue culture technologies in order to form plant members because of its prominent and important role in the formation of vegetative buds, their growth and their multiplication. The researchers stated that adding multiple amines to the MS medium encourages the emergence of organs *in vitro* of many plant species [6,4]. [7,8] obtained buds directly from the embryos of date palm variety OF Medgol on the medium of MS supplied with 100 $mg.L^{-1}$ of Put.. [9] explained the use of several combinations of Put. added to the MS nutritional medium, these concentrations are (0, 0.5, 1, 2 and 3) mM molar for date palm Braim cultivar, and the result showed a superiority of (2 mM) concentration significantly over the other concentrations in the number of embryos. Vegetative and fresh and dry weight of somatic embryos. [10] studied the effect of interference between Put. in concentrations (0, 50 and 100) $mg.L^{-1}$ and adenine sulfate in concentrations (80 and 160) $mg.L^{-1}$ in the multiplication of adventiouse buds and the rate of growth in date palm khalas cultivar culture in vitro to the study showed Significant superiority of the interaction between Put. at a concentration of 50 $mg.L^{-1}$ and adenine sulfate at a concentration of 160 $mg.L^{-1}$ in response to growth, number of shoots and their fresh weight.

Salicylic acid (SA) is one of the plant hormones that modern research began to address its study and its effective role in regulating many physiological processes and regulating the absorption of ions and hormonal balance and movement of stomata and plays an important role in regulating the response of plants to environmental stresses [11]. Because of the many physiological roles of SA. acid in plant growth, development, and detection, this compound has been added to the list of well-known plant hormones such as oxins, gibberelins, and cytokinens, and at the present time it is considered one of A natural Plant Hormone [12]. Some studies have indicated that the external addition of SA has reduced the growth inhibition caused by abiotic stress and stimulated growth in many agricultural crops [13,14,15]. In a study by [16] culture of 250 mg of grape callus and growing under the influence of saline stress in the MS medium with concentrations (0, 75 and 150) mM sodium chloride respectively and by interfering with SA and concentrations (0, 0.5 and 1) mM, the study showed significant superiority of SA concentration (1) mM over other studied characteristics. A study [9] in which several combinations of SA was used in concentrations (0, 0.5, 1, 2 and 3) mM in the mean MS of Propagation by tissue culture of date palm braim cultivar and the results of the study showed that there were significant differences as it gave a concentration of (3) mM has the highest scores in studied traits. Despite the advances in the field of palm propagation by the technique of direct budding, many problems and constraints remained, including the many formed, weak and adherent vegetative branches with each other, therefore it is difficult to separate, which causes as a result of that, many of them have lost, and some of them suffer from wrapping their leaves, weakening, deformation and appearance Transparency Virology. Therefore, the research

aims to study the effect of Putrescine and Salicylic acid and their interaction in improving the growth and development of vegetative buds of the Barhi cultivar *in vitro*.

MATERIALS AND METHODS

The current study was carried out in the plant tissue culture laboratory of the Date Palm Research Center - University of Basrah for the period from 25/2/2018 until 3/2/2019. The vegetative bud prefixes for date palm cultivars were used in Al-Barhi cultivar with a six-month-old age (Plate 1) and induced from terminal buds. Apical buds cultivated in the medium [17] defined by MS salts manufactured by Phytotechnology Lab Company with the addition of the substances mentioned in Table 1.

Table 1. Concentrations of additives in the MS medium

Subject	Quantity mg.L $^{-1}$
Sucrose	30000
Sodium di hydrogen orthophosphates	170
Myoinositol	100
Adenine sulphate	30
Thiamine-Hcl	0.5
Glycine	2
Riboflavin	1
Naphthalene acetic acid NAA	1
Nophthyacitic acid NOA	1
2-ip Isopentenyl adenine	2
Benzyl adenine	2
Kinetin	2
Activated charcoal	1000
Agar	8000

Plate 1. The beginning of the emergence of vegetative shoots

The effect of four concentrations of Put (0, 50, 100, 150) mg.L^{-1} and three concentrations of SA (0, 0.5, 1) mg.L^{-1} and their interaction In the development of the vegetative sprouts of date palm, the cultivated Al-Barhi cultivar, Masses of vegetable buds were cultured in agricultural vessels (Jars), with an average of approximately 5 buds per jar were incubated at a temperature of 27 ± 2°C and lighting intensity of 2000 lux for 16 hours. The reculture was done once every 30 days and the experiment lasted 120 days. 10 replications were used for each concentration, the following characteristics were studied:

1- The fresh weight of vegetative buds 120 days after culture. It was used as an indicator of growth according to the fresh weight as follows [18].
2- The number of vegetative buds = the number of total buds at the end of the experiment - the number of buds at the beginning of the experiment, and Plate 2 represents the multiplication of vegetative buds.

Plate 2. Shows the multiplication of vegetative buds

3- Length of vegetative buds (cm)

Three replicates were chosen from each of the studied concentrations and their length was measured (the method of selecting the replicates was randomized). Plate 3 represents the elongation of vegetative buds.

Plate 3. Shows the elongation of the vegetative buds

4- The percentage of natural and deformed buds

The following was calculated: (deformed buds number) / (vegetative buds total number) x 100.

Plate 4. Shows deformed vegetative buds

5- Degree of separation

The degree of separation of the vegetable buds has been calculated, after splitting the vegetative buds when replanting and showing the ease of separating the buds. A symbol (+) has been added to indicate the best separation process. But if it is (+++) it indicates a good grade and if it is (++) it indicates an average degree, but (+) indicates that the degree to separate the buds is weak.

6- Leaves content of plant pigments

The leaf content was estimated from chlorophyll and carotene pigments based on the Holden method described by [19].

7- Determination of leaf content of total dissolved carbohydrates

The leaf content of dissolved carbohydrates was estimated according to the phenol-sulfuric acid method based on [20].

8- Estimate the total phenols

Phenolic substances were estimated according to the Folin-Denis method mentioned in [21].

Statistical Analysis

The present study experiment was designed as a global experiment using Completely Randomized Design (CRD), it included two factors and the results were analyzed using the statistical program Genestat (8.1) for the year 2005 and averages were compared according to the (Least Significant Differences LSD test) and blindness level of 5% [22].

RESULTS

The Effect of Put. and SA and Their Interaction on

1-Fresh weight of vegetative buds (g)

It is clear from Table 2 that the addition of put. to the food medium resulted in a significant increase in the soft weight of the vegetative buds and exceeded the concentration of 100 $mg.L^{-1}$ was significant over the other studied concentrations, as the average soft weight reached 23.361 gm, followed by the concentration effect of 150 $mg.L^{-1}$ with an average of 18,774 gm, while the concentration was 50 $mg.L^{-1}$, the mean recorded 12.560 gm, while the concentration was 0 $mg.L^{-1}$ of the potassin (control). The lowest mean weight of the weight was 10.980 gm, recording a significant decrease from the other concentrations.

It is also noted from the table that the addition of SA to the dietary medium showed a significant superiority over the control treatment and the

concentration was recorded at 0.5 mg.L^{-1} SA the highest mean in the fresh weight of buds as it reached 16.731 gm followed by effect 1mg.L^{-1} concentration with an average of 16,680 gm was a soft weight, with no significant difference between them. The concentration was 0 mg.L^{-1} L-1 of salicylic scored a significant decrease from the two studied concentrations, as the average soft weight reached 16.020 gm.

Table results show that the interference was significant and the interference exceeded (concentration 100 mg.L^{-1} Put. with 0.5 mg.L^{-1} SA), and a significant difference over other interventions, where the average fresh weight of vegetative buds reached 25.03 gm, while interference was recorded (Concentration 0 mg.L^{-1} Put. With concentration 0.5 mg.L^{-1} SA. The lowest fresh weight was 10.92 gm.

2- The number of vegetative bud**s**

From Table 3, it is clear that the addition of Put. to the medium resulted in a significant increase in the number of vegetative buds formed and exceeded the concentration of 150 mg.L^{-1} Put. the concentration of the comparison is 0 mg.L^{-1} from it, with a significant difference, as the average number of vegetative shoots in them (21,233 and 12,133) buds respectively, while the two concentrations recorded 100 and 50 mg.L^{-1} Put. the average number of buds formed was (20,500 and 18.943) buds, respectively, and with a significant difference from the control treatment.

It is also noted from the table that adding SA to the medium showed significant superiority in the multiplication of vegetative buds and recorded the two concentrations (0.5 and 1) mg.L^{-1} of it significantly superior to the control treatment as the average number of buds formed in them (19.21518.458 and 16.920) buds, respectively.

It is also noted from the table that the interference between Put. and SA was significant, as the overlap of the two concentrations was recorded (150 mg.L^{-1} Put. with a concentration of 0.5 mg.L^{-1} SA) 23.801 buds, with a vegetative bud registered significantly superiority of interference (150 and 100 mg.L^{-1} Put. with a concentration of zero mg.L^{-1} SA) and interference concentration 50 mg.L^{-1} Put. with two concentrations zero and 0.5 mg.L^{-1} SA and interference (concentration 0 mg.L^{-1} Put. with zero concentration 0.5 and 1 mg.L^{-1} SA) and gave an intervention (comparison zero mg.L^{-1} Put. with a concentration of zero mg.L^{-1} SA), the results were the lowest recorded 10.660 buds with no significant difference with the same (concentration) interference of Put. with 0.5 and 1 mg.L-1 SA) as the count two buds 12.560 and 13.121, respectively, bud, representing plate 5 doubled vegetative buds.

Table 2. Effect of Put. and SA and their interaction on the fresh weight of buds (g)

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	10.920	10.960	11.060	10.980
50	12.510	11.760	13.410	12.560
100	21.320	25.030	24.440	23.596
150	19.311	19.190	17.820	18.774
Means of SA	16.015	16.735	16.680	
	LSD(0.05) SA= 0.05	**Put.=0.60**	**Interaction =0.65**	

Table 3. Effect of Put. and SA and their interaction on the number of vegetative buds formed

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	10.660	12.560	13.121	12.113
50	18.120	18.800	19.911	18.943
100	19.501	21.700	20.301	20.500
150	19.400	23.801	20.500	21.233
Means of SA	16.920	19.215	18.458	
	LSD(0.05) SA= 1.31	**Put.=2.70**	**Interaction =4.02**	

Plate 5. Shows the multiplication of vegetative buds

3- Length of vegetative buds (cm)

It is noted from Table 4 that the addition of Put. in the medium recorded significant differences from the control treatment. Concentration exceeded 100 mg.L^{-1} of Put. morally on the two concentrations (50 and 0) mg.L^{-1} in the average length of vegetative buds in which they reached (12.507, 9.019 and 6.915) cm respectively, while the average length of vegetative buds in the concentration reached 150 mg.L^{-1} as it averaged vegetable buds have 11.813 cm, with no significant difference, with a concentration of 100 mg.L^{-1} put.

It is also noted from the table the significant effect of SA on the length of vegetative buds and the superiority of the concentration is 0.5 mg.L^{-1} SA in the mean length of the vegetative bud which reached 11.094 cm and a significant difference from the concentration control which reached 10.793 cm, in While the average length of vegetative buds was 8.304 cm, with a concentration of 1 mg.L^{-1} SA with no significant difference with a concentration of 0.5 mg. L^{-1}.

It is also observed from the table that the interference was significant and the treatment interference was greater (100 mg.L^{-1} Put. with 0.5 mg.L^{-1} SA), with a significant difference from the other interventions except for the interference (the two concentrations 150 mg.L^{-1} Put. with 0.5 mg.L^{-1} SA and overlap concentration 100 mg.L^{-1} Put. with 1 mg.L^{-1} SA) as the average length of vegetative buds in them reached 14.501, 13.412 and 12.610 cm respectively, control treatment was recorded (0 mg.L^{-1} Put. and 0 mg.L^{-1} SA) the lowest results, as the average length of vegetative buds reached 5.303 cm with no significant difference with the two concentrations (50 mg.L^{-1} Put. And 0 mg.L^{-1} SA) and (0 mg.L^{-1} Put. And 0.5 mg.L^{-1} SA) whose average length of vegetative buds was 7,275 and 7,110 cm respectively. Plate No. 6 shows the length of vegetative buds.

4- The degree of separation of vegetative buds

It is noted from Table 5 and through the reculture processes of vegetative buds, that the separation degree was good when interaction (concentration 100 mg.L^{-1} Put. with 0.5 and 1 SA), followed by the effect of interaction (concentrations 50 and 100 mg.L^{-1} Put. with 0.5 and 1 mg.L^{-1} SA) as the degree of separation of the vegetative buds was moderate while (comparison interaction showed 0 mg.L^{-1} Put. with all concentrations of SA) and (SA overlap in concentration 0 with all Put. concentrations). Results if the vegetative buds are attached to each other and are difficult to separate.

5-The percentage of deformed buds

From Table 6, it is clear that the addition of Put. to the medium resulted in a significant decrease in the percentage of deformed plants and showed a concentration of 150 mg.L^{-1} Put. significant decrease from all other concentrations, as the ratio was 15.550%, followed by the two concentrations (100 and 50) mg.L^{-1}, as the ratio recorded (19.990 and 23.550)%, respectively, while the concentration recorded 0 mg.L^{-1} Put. the highest

value in the percentage of deformed plants reached 28.550%.

It is noticed from the table that adding SA to the medium showed a significant decrease in the percentage of deformed plants and the concentration was recorded as 1 mg.L^{-1} of which is the lowest value of the deformed plant percentage as it reached 20.160% while the other two concentrations recorded (0.5 and 0) mg.L^{-1} ratio of (21,490 and 24,080)%, respectively.

Table 4. Effect of Put. and SA and their interaction on the length of vegetative buds (cm)

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	5.303	7.110	8.33	6.915
50	7.275	9.450	10.430	9.019
100	10.410	14.501	12.610	12.507
150	10.228	13.412	11.800	11.813
Means of SA	8.304	11.094	10.793	
	LSD(0.05) SA= 0.82		**Put.=1.74**	**Interaction =2.57**

Plate 6. Showing the length of the vegetative buds under the influence of (Put and SA)

Table 5. Effect of Put. and SA acid and their interference on the degree of separation of vegetative buds

Put. mg.L^{-1}	SA mg.L^{-1}		
	0	0.5	1
0	+	+	+
50	+	++	++
100	+	+++	+++
150	+	++	++

Table 6. Effect of Put. and SA and their interference in estimating the percentage of distorted plants (%)

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	30.330	28.000	27.330	28.550
50	25.000	23.660	22.000	23.550
100	22.330	19.660	18.000	19.990
150	18.660	14.660	13.330	15.550
Means of SA	24.080	21.490	20.160	
	LSD(0.05) SA= 0.05		**Put.=15**	**Interaction =0.20**

It is also noted from the table that the interference between Put. and SA showed a significant decrease, as it gave interference (concentration 150 mg.L-1 comparison: Concentration 0 mg.L-1 Put. with concentration 0 mg.L-1 SA the highest value in the percentage of deformed plants was 30.330%.

6-Leaves content of plant pigments

Total chlorophyll: The results shown in Table 7 show that the concentration exceeded 150 mg.L^{-1} Put. in the amount of chlorophyll accumulated in the tissues of vegetative buds and a significant difference from the other concentrations studied as the average amount of chlorophyll in the leaves reached 6.28 mg.100 gm^{-1} fresh weight and the concentration record 0 mg.L^{-1} Put. the lowest results were in the amount of chlorophyll that amounted to 5.09 mg.100 gm^{-1} fresh weight.

It is noted from table, that the addition of SA showed a significant increase in the leaf content of chlorophyll and exceeded the concentration of 1 mg.L^{-1} for each over the other two concentrations (0.5 and 0) mg.L^{-1} and a significant difference as the average amount of chlorophyll in it (6.66, 5.44, and 4.700) mg.100 gm^{-1} fresh weight, respectively.

It is noted from Fig. 1(c) that the interference was significant and the concentration interaction exceeded 150 mg.L^{-1} Put. with 1 mg.L^{-1} SA as the amount of chlorophyll in it amounted to 7.23 mg.100 gm^{-1} fresh weight, while control treatment recorded the lowest amount of Chlorophyll of 4.13 mg.100 gm^{-1} fresh weight.

Carotine pigment: Table 8 shows that adding polyamine Put. to the dietary mean scored significantly in carotene pigment for cotrol treatment and exceeded the concentration of 150 mg.L^{-1} Put. with a significant difference from the two concentrations (50 and 0) mg.L^{-1} when it reached Average dye in them (0.076, 0.043, and 0.032) mg.100 gm^{-1} soft weight, respectively, and the concentration was recorded at 100 mg.L^{-1} 0.068 mg.100 gm^{-1} fresh weight of carotene tincture with no significant difference with concentration 150 mg.L^{-1} Put.

It is noted from Table 8 that the addition of SA to the medium recorded a significant increase in the leaf content of the carotene dye compared to the control treatment and exceeded the concentration of 1 mg.L-1 significantly over the other two concentrations (0.5 and 0) mg.L-1 as it reached Carotene dye in it (0.099, 0.057 and 0.038) mg. 100 gm-1 fresh weight.

Table 7. Effect of Put. and SA and their interaction on the leaves content of total chlorophyll mg.100 gm^{-1} fresh weight

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	4.13	4.81	6.32	5.09
50	4.39	5.50	6.55	5.48
100	4.53	5.57	6.58	5.66
150	5.72	5.88	7.23	6.28
Means of SA	4.70	5.44	6.67	
	LSD(0.05) SA= 0.004		Put.=0.046	Interaction =0.08

Table 8. Effect of Put. and SA and their interaction on the leaves content of carotene pigment mg.100 gm^{-1} fresh weight

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	0.030	0.033	0.069	0.044
50	0.033	0.040	0.108	0.060
100	0.045	0.078	0.109	0.077
150	0.046	0.079	0.113	0.079
Means of SA	0.038	0.057	0.099	
	LSD(0.05) SA= 0.002		Put.=0.003	Interaction =0.004

The data in table show that the interference was significant and the interference exceeded (concentration 150 mg.L^{-1} Put. with 1 mg.L^{-1} SA) and with a significant difference from other interventions as the carotene dye reached 0.113 mg.100 gm^{-1} fresh weight while the interference (concentration 0 mg.L^{-1} Put. with 0 mg.L^{-1} SA) recorded the lowest results in the amount of carotene that reached 0.030 mg.100 gm $^{-1}$ fresh weight.

7- Leaves content of total soluble carbohydrates (mg.gm^{-1} dry weight)

From Table 9, it is clear that the addition of Put. to the medium mean scored significant differences from the control treatment 0 mg.L^{-1} and the concentration exceeded 100 mg.L^{-1} with a significant difference from the other studied concentrations as the average total soluble carbohydrates in it reached 29.430 mg.gm^{-1} dry weight while concentration was recorded 0 mg.L^{-1} Put. the lowest average total soluble carbohydrate is 13,140 mg. gm^{-1} dry weight.

The table shows that the concentration of SA acid (0.5 and 1) mg.L^{-1} record significantly superior in the amount of total soluble carbohydrates for the control treatment as the quantity in them (22.840 and 23.50 and 017.08) mg.gm^{-1} dry weight, while there was no significant difference between the two concentrations (0.5 and 1) mg.L^{-1} SA.

The results of the table note that the interference between Put. and SA was significant and the interference (concentration 100 mg.L^{-1} and 0.5 mg.L^{-1} SA) was significant over all interventions, as the total carbohydrate amount was 37.00 mg.gm^{-1} dry weight, while the control treatment 0 mg.L^{-1} Put. With 0 mg.L^{-1} SA) was recorded Less results, as the amount of carbohydrates reached 10.00 mg.gm^{-1} dry weight which did not differ significantly from the interference (concentration 0 mg.L^{-1} Put. and 0.5 mg.L^{-1} SA), which recorded 13.160 mg.gm^{-1} dry weight.

8-Leaves content of total phenols (mg.gm^{-1} fresh weight)

The data in Table 10 indicate that the use of Put. in the medium showed a significant reduction in the leaf content of total phenols and the concentration was recorded 150 mg.L^{-1} Put. the lowest amount of total phenols in the tissue of the leaves, which amounted to 30.865 mg.gm^{-1} fresh weight and with a difference Significant from other concentrations, followed by effect in the two concentrations (100 and 50) mg.L^{-1} Put. as the leaf content in them reached total phenols (65.822 and 69.512) mg.gm^{-1} fresh weight with a significant difference between them, while the control treatment recorded the highest content of the total phenols which amounted to 74,079 mg.gm $^{-1}$ fresh weight.

It is also noted from the table that adding SA to the medium showed a significant reduction in the leaf content of the total phenols and scored the concentration of 1 mg.L^{-1} SA the smallest amount of phenols as it reached 56.674 mg.gm^{-1} fresh weight and a significant difference from the other two concentrations (0.5 And 0) mg.L^{-1} SA as the average total amount of phenols in them (60,486 and 63,049) mg.gm^{-1} fresh weight, respectively, with no significant difference between them.

Table 9. Effect of Put. and SA and their interaction on total carbohydrate levels (mg.gm^{-1} dry weight)

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	10.000	13.160	16.260	13.140
50	16.000	16.060	26.060	19.370
100	19.030	37.000	32.260	29.430
150	21.300	25.160	27.260	25.240
Means of SA	17.080	22.840	23.050	
	LSD(0.05) SA= 1.75		Put.=1.90	Interaction =3.65

Table 10. Effect of Put. and SA and their interaction in estimating total phenols

Put. mg.L^{-1}	SA mg.L^{-1}			Means of Put.
	0	0.5	1	
0	79.102	73.032	70.105	74.079
50	70.406	70.001	68.130	69.512
100	67.630	67.607	62.231	65.822
150	35.060	31.304	26.232	30.865
Means of SA	63.049	60.486	56.674	
	LSD(0.05) SA= 2.67		Put.=2.75	Interaction =5.42

The data in the table also showed that the interference between the Put. and the SA recorded significant differences in the total amount of phenols in the leaves and the superiority of the interference (concentration 150 mg.L^{-1} and 1 mg.L^{-1} SA) in reducing the amount of phenols in the leaves of plants that reached 26.232 mg-gm^{-1} is a fresh weight, with a significant difference from other interventions except the interference (same concentration from Put and 0.5 mg.l-1 SA), as the total amount of phenols in it reached 31,304 mg.gm^{-1} fresh weight, while interaction control treatment was recorded (0 mg.L^{-1} Put. and 0 mg.L^{-1} SA the lowest results were 79.102 mg.gm^{-1} fresh weight.

DISCUSSION

Many sources point to the role of growth regulators in the formation of vegetative buds, as they have been documented by many researchers [23,8,24,25]. Put is characterized by having similar effects to auxine, cytokinine and gibbereline, which leads to an increase in the volume of tissue separated and grown on sterile media [26,27,28,29,30,31].

Many sources indicated that the polyamine group, including Put., is a growth organization that regulates physiological responses such as cell division and phenotypic formation and stimulates the formation of adventiouse buds. It is believed to protect cell membranes, as it is strongly associated with negative charges of cellular components such as acids. Nuclei, proteins and phospholipids are related to the activity of antioxidant enzymes such as Peroxidase and Catalase [32,33]. Also, Put. or polyamines can have an effect as growth regulators as their low molecular weights and cationic charges are useful for their rapid transmission between plant parts and their inclusion in growth regulation [34].

From the following Tables 2, 3, 4, 7, 8 and 9 and Figs. 1 and 2, we notice that the increase in my concentration (100 and 150) mg.L^{-1} Put in the average fresh weight, the number and length of the green shoots, the amount of total soluble carbohydrates and the total chlorophyll and carotene pigments is due to the effect of the quantity Put added to the medium have a constant level of growth regulators in encouraging cells to divide and widen [35,36,37,38].

The conclusions reached on the effects of Put on the formation of adventiouse buds, their number and their lengths are consistent with their findings [39,40,41]. who found that polyamine has a major role in formation morphogenesis for buds, cell growth and division, differentiation and multiplication in plant tissue culture, [42] and [3] indicated that adding growth-promoting substances, including polyamine, Put to the medium, may encourage vegetative growth by stimulating cell division and differentiation and attracting nutrients to the plant parts of the medium, in addition stimulating photosynthesis enzymes whose effect is reflected in increasing the size of the cell and encouraging the process of division and morphological differentiation also plays a role in building RNA and proteins and enzymes within the cell as well.

From Table 10, it is clear that Put reduced the leaf content of total phenolic materials and the result was consistent with what [43] that Put plays an important role in addition to cell division in increasing plants' tolerance to stress due to high concentrations of elements in the media and thereby reducing the effect of phenolic substances in the plant.

That the effect of SA as one of the plant hormones can encourage the building of the hormones auxine and gibberelin, and it may also encourage

the process of gene expression to build many chemical compounds, including auxins and gibberelins, in addition to encouraging it to build chlorophyll and carbohydrates resulting from increased growth, [44] and the reason for the increase in the soft weight of the vegetative buds, their number and length is due to the role of SA in regulating growth and physiological processes, nutrient absorption, protein synthesis, inhibiting ethylene and respiration and protecting the functions of some cellular organelles, which in turn encouraged this increase, [12] and the reason for the decrease The result in the studied traits is attributed to the fact that SA is one of the plant hormones and its physiological effects depend on the concentration used, the study is consistent with the findings of [14] who found that SA caused a significant increase in the fresh weight of maize seedlings.

As for the increase in carbohydrates, the reason may be that the SA, under the condition of tension, has inhibited the activity of the enzymatic system that analyzes sugars or accelerates the conversion of monosaccharides into multiple sugars, which contributes to increasing the concentration of carbohydrates in the plant [45] or the reason is due to SA works to reduce plant stresses and to regulate growth and all physiological processes, which in turn increases the concentration of carbohydrates to provide the energy needed for the biological processes of the plant [46].

The superiority of salicylic acid in the studied traits is consistent with its physiological role as it works to accelerate the formation of chlorophyll and carotene pigments and accelerate the process of photosynthesis and increase the activity of some important enzymes and reduce proline and phenolic substances, and this is consistent with what [5,12,47,48,46,49].

The superiority of SA in the studied traits is in line with its physiological role as it works to accelerate the formation of chlorophyll and carotene pigments and accelerate the process of photosynthesis and increase the activity of some important enzymes and reduce phenolic substances, and this is consistent with what [5,12,47,48,46,49].

CONCLUSION

From the study, we conclude that adding polyamines Put and SA to the nutritional medium of multiple the vegetative buds of the Date palm propagation *in vitro*. Well the studied characteristics, including the fresh weight of the vegetative buds, their number and length, led to a decrease in the roasted buds, an increase in the chlorophyll and carotene pigments, an increase in the concentration of carbohydrates and phenol, and a decrease in the quantity of amino acid Proline accumulated in the leaves, so the study recommends adding Put and SA to the nutritional media to propagation of Date palm cv. Barhi and conducting many studies on other varieties to find out their effect and doubt Accurate.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. Author KAM designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author EAH managed the analyses of the study. Author MAD managed the literature searches. All authors read and approved the final manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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