Effect of BA and NAA on growth and multiplication indicators of tulip bulbs in vitro

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Abstract

This study was conducted in the plant tissue culture laboratory of the Department of Plant Production Techniques, Musayyib Technical College, during the year 2020-2021. The experiment included the use of plant tissue culture technology in the vegetative propagation of tulip bulbs. The experiment included studying the effect of five concentrations of BA (0, 0.5, 1, 1.5, 2) mg/L and five concentrations of NAA (0, 0.5, 1, 1.5, 2) mg/L in the stage of growing and multiplication tulip bulbs. The results showed where the concentration of 1.5 mg.L-¹ excelled and gave the highest differentiation average of 56.4%, while the concentration of 2 mg.L-¹ gave the highest rate in the number of shoots and the length of shoots reached 4.447 and 3.044, respectively. As for the treatment with NAA, there were no significant differences in the percentage of differentiation , while the concentration 1.5 mg.L^{-1} excelled and gave the average number of shoots 4.16 and the concentration of 2 mg.L-¹ recorded the highest average in shoots length 43.30. As for the interaction between BA and NAA, it was noted that the treatment (2 mg.l^{-1} BA with 1.5 mg.L^{-1} NAA) in the differentiation average was 70%. While treatment (0 mg.L^{-1} BA with 2 mg.L^{-1} NAA) recorded the highest average in the number of shoots 5.55, and the treatment (2 mg.L^{-1} BA with 0.5 mg.L^{-1} NAA) recorded the highest average in shoots length 3.820 compared to the control treatment

Introduction

The Tulipa plant belongs to the Liliacea family, and its origin country is Turkey. It is also called the turban flower because it consists of several layers of colored petals. The cultivation of these flowers moved to Europe 400 years ago and spread in it in a very large way, and it found special care in its cultivation, especially in the Netherlands, which has become A symbol and a blanket for her great income, It produces a billion flowers annually and is exported to all countries of the world. The genus of tulips includes about 120 species, some of which are perennial and others are perennial or winter (Khattab Wassfi 1988 and Khader, 2001).The tulips are found in the north in a wild way that extends from Zakho to Haji Omran and Sulaymaniyah province(Al-Sultan and others 1992). The tulip flowers are carried on a long strand, and also the tulip bulb is true, which is a disc stem surrounded by the bases of the merging juicy leaves covered from the outside with brown scaly leaves, (Hay and Beckeet,1978 ; Ress, 1985 and Alkatib 2000) The tulips are multi-colored, including white, yellow, blue, dark purple, and purple, yellow-white-yellow tinged with a light purple color, and they are important in landscaping gardens and flower galaxies.They are grown in ponds and used as determinants and can also be grown in rock gardens and can be used as picking plants (Naglaa and Kandeel 2012, Tawajen 1987, Khader 2001, Abu Dahab 1992 and Abu Zaid 2002).Growth regulators are one of the most important factors in tissue culture, and these materials have an impact on the success of laboratory breeding of bulbs (Chanteloube et al., 1995).Propagation of plants using the tissue culture technique is also one of the scientific applications of this technique due to its unique characteristics over traditional methods.Auxins and cytokinins are among the most widely used regulators for this purpose (Ramawat, 2004).

research aims

1- Knowing the appropriate concentrations of growth regulators to detect tulips

2- The effect of growth regulators on the formation of the number of shoots

3- Effect of growth regulators on the length of shoots

Materials and methods

The experiment was conducted in the Plant Tissue Culture Laboratory, Department of Plant Production Techniques, Musayyib Technical College, to study the effect of BA and NAA in vitro growth and multiplication of tulip bulbs. The study was conducted according to the Completely Randomized Design (CRD)with two factors and five replicates. In this

experiment, certified Dutch tulip chromatids, purissuma, were used. The good bulbs free from wounds and injuries were selected and placed in the refrigerator at a temperature of 4°C for three months in the laboratory.Then the brown scaly leaves were removed from it, washed well with tap water and liquid soap, and then placed under running water for 24 hours to ensure the disposal of dust and dirt stuck between the scaly leaves, and the bulbs were then transferred to the stratified air flow cabin.It was superficially sterilized with sodium hypochlorite NaOcl at concentrations (5, 6, 7) for 25 minutes, then washed with distilled water three times for 5 minutes each time. The affected parts were removed from sterilization, then the bulbs were divided in a sterile Petri dish into plant parts.It included the lower part consisting of the disc stem, axillary bud and the apical bud. In this experiment, prepared MS medium was used in addition to sucrose in the amount of (30 g) and agar in the amount of (7 g), and the experiment agents were provided with five concentrations of BA (0, 0.5, 1, 1.5, 2) mg /L and five concentrations of NAA (0, 0.5, 1, 1.5, 2) mg/L,The medium was then placed on a thermo-magnetic mixing device to dissolve the agar and homogenize the nutrient medium. Then the medium was distributed directly into the culture glass at an average of (10 mm) ml and sterilized in the Autoclave at a temperature of 121 °C and a pressure of 1.04 kg / cm2 for 20 minutes. Cultivation of the parts inside the cabin air flow stratified.The cultures were incubated at temperatures of 25 ± 1 and 16 hours of light and 8 hours of darkness for 30 days to study the effect of BA and NAA interaction between them on the percentage of buds to shoots, the average number of shoots and the length of the plant.

1- The experiment was conducted to find out

Effect of NAA and BA (mg L^{-1}) and their interaction in culture of tulip plant parts in the nutrient media The plant parts were grown on the sterile media (Murashige, and Skoog, 1962) equipped with different concentrations of NAA growth regulators (0.0, 0.5, 1.0, 1.5, 2) mg L^{-1} and BA (0.0, 0.5, 1.0, 1.5, 2) mg L^{-1} and five replicates for each plant part and concentration to study the effect of three indicators:

1- The first indicator

Knowing the percentage of revealing the plant parts of the tulip bulbs after planting the plant parts on the MS agricultural medium after two weeks.

2- The second indicator

Knowing and studying the number of shoots resulting from the culture of the plant parts of tulip bulbs on the MS food medium after 30 days of culture

3- The third indicator

Study of the length of shoots resulting from the culture of the plant parts of tulip bulbs on the MS food medium after 30 days of culture

Results and discussion

1:- Effect of BA and NAA (mg/L^{-1}) and the interaction between them on the percentage of buds differentiation to shoots after 30 days of culture for tulips in MS food medium.The results in Table (1) indicate that there are significant differences for BA in the percentage of buds differentiation , where the concentration of (1.5) excelled by giving it the highest average of 56.4%.While the control treatment gave the lowest percentage of 32%, and the reason may be due to the fact that cytokinin stimulates cell division and encourages the formation of transverse and axillary branches (Grewal et al., 1995). It is also noted that there is a significant effect of NAA, which gave the highest rate of concentration 1 is 52%, while the control treatment gave the lowest average of 43.6%.It was also noticed from the interaction rates that there were significant differences for BA with NAA, as the concentration of (2 mg.L^{-1} of BA with 1.5 mg.L^{-1} of NAA) surpassed it by giving it the highest average of 70%.While the lowest differentiation average was 22% in the control treatment for factors , and the reason for the high differentiation average of sprouts may be due to the availability of appropriate ratios between cytokinin and auxins. This is consistent with (Ghaffoor et al., 2004) using two concentrations of NAA, which are (2 and 1) mg L^{-1} and also 1 BA was used and it gave the best percentage of differentiation buds and fresh growth of the culture tulip plant.

Table 1:- BA and NAA (mg/L) and the interaction between them in the percentage of buds differentiation to shoots after 30 days of culture for tulips in MS food medium

NAA	BA					average NAA
	0	0.5	1	1.5	2	
0	22	40	46	60	50	43.6
0.5	30	46	52	46	66	48
1	32	64	52	64	48	52
1.5	38	48	38	48	70	48.4
2	38	46	62	64	48	51.6
Average BA	32	48.8	50	56.4	56.4	
L.S.D	BA=12.44 NAA= 12.44 BA*NAA=14.02					

Figure 2: Tulip buds differentiation a week after culture on MS medium

2:- Effect of concentrations of NAA and BA (mg/L) and the interaction between them on the average number of shoots after 30 days of culture the explants of the tulip plant in the MS food medium.

The results in Table (2) indicate that there is a significant effect of the growth regulator NAA in increasing the number of shoots, where the concentration gave 1.5 mg L^{-1} a significant increase in the number of shoots, reaching 4.168, while the lowest average was given by the control treatment 82.15.Concerning the cytokinin BA, the concentration of 2 mg L^{-1} gave the highest average of 4.447, and the lowest average given by the comparison treatment was 0.156. As for the interaction between the growth regulators NAA and BA, the interaction (0 mg L^{-1} of BA with 2 mg L^{-1} of NAA) gave the highest average in the number of shoots. Where it gave the highest average of 25.55 and the lowest average it gave was an interaction between (0 mg L^{-1} BA with 0.5 mg L^{-1} NAA) and treatment (0 mg.l BA with 1 mg L^{-1} NAA). The reason for the increase in the number of shoots may be due to the role of auxins in cell division, where they work to increase the meristematic areas (Jain and Rout, 2004),This may be due to the role of cytokinins in their stimulating effect on cell division, transport of nutrients, breaking of apical

dominance and formation of active peripheral meristems (Taiz and E. Zeiger, 2002).Also, the results agree with (Ghaffor et al., 2004) when culture axillary and apical buds, which gave 2-8 vegetative growths of axillary buds and 2-6 growths of apical buds in the presence of 1 mg L^{-1} NAA and 4 mg L^{-1} BA.

Table 2:- Concentrations of NAA and BA (mg/L) and the interaction between them in the average number of shoots after 30 days of culture the explants of the tulip plant in the MS food medium.

NAA	BA					average NAA
	0	0.5	1	1.5	2	
0	0.438	4.412	1.474	2.500	1.968	2.158
0.5	0.156	2.584	2.478	4.462	4.224	2.781
1	0.152	4.704	4.924	4.352	4.998	3.826
1.5	0.748	5.116	4.968	4.518	5.492	4.168
2	1.088	4.620	4.568	4.960	5.552	4.158
Average BA	0.516	4.287	3.682	4.158	4.447	
L.S.D	BA=0.499 NAA= 0.499 BA*NAA=1.117					

Figure 3: shoots of tulip culture on MS . medium

3 :- Effect of NAA and BA (mg L^{-1}) and the interaction between them on the average length of shoots (cm) after 30 days of culture the explants of tulips in MS food medium

The results in Table (3) indicate the treatment of 2 mg L^{-1} of NAA, excelled and gave the highest average in the length of shoots reached 3.304 cm, which differed significantly from the rest of the treatments.While the concentration treatment of 2 mg L^{-1} was excelled on BA by giving it the highest rate of 3.044 cm, while the control treatment gave the lowest average of 02.16 cm.As for the interaction between growth regulators, the interaction treatment 0.5 mg L^{-1} NAA and 2 mg L^{-1}BA was significantly excelled by giving it the highest rate of branch length, which was 3.820 cm compared to the rest of the interactions. Auxins are one of the main factors that help the activity of the cambium inside the plant and work to increase the cell division of meristematic cells in a large and rapid way (Coartney et al., 1967). This did not agree with what was found (Amin et al., 2014) when adding BA to the nutrient medium led to a reduction in the number of shoots and shoots lengths.

Table 3: NAA and BA (mg L^{-1}) and the interaction between them in the average length of shoots (cm) after 30 days of culture the explants of tulips in MS food medium

NAA	BA					average NAA
	0	0.5	1	1.5	2	
0	0.0	2.440	2.400	2.560	3.680	2.216
0.5	2.480	2.6680	3.080	3.550	3.820	3.124
1	2.520	2.040	2.040	2.220	2.120	2.188
1.5	2.240	2.060	2.320	2.360	2.240	2.244
2	3.560	3.440	3.240	2.920	3.360	3.304
Average BA	2.160	2.532	2.616	2.724	3.044	
L.S.D	BA=0.302		NAA= 0.302		BA*NAA=0.675	

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