Effects of Auxin and Cytokinin on morphological and Physiological factors in stem and root in (*Avene sativa L*)

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Abstract: NAA and BA hormones are of plant growth regulators. These hormones interfere in different physiological and morphological, growth and germination, oat is important as a food for livestock and also for its economic and medicine value. In this research, the effect of NAA and BA hormones was surveyed in different concentrations 0, 0/5, 1/5and 2/5 mg/l for some growth and biochemical factors of oat. In treatment plants indifferent concentrationsof NAA and BA, the highest grow thin the root and stem length related to NAAhas been 1/5 mg/lit. So by increasing NAA concentration to 1/5 mg/lit concentration, the stem length was increased and then decreased. And by increasing BAconcentration, the stem growth was decreased. And by the effect of NAA and BAconcentration to 1/5 mg/lit was observed increasing in stem length and then to decrease. And by increasing BA concentration, the length growth of stem was decreased to 1/5 mg/litconcentration and then it was observed increasing in the length growth of stem. So, the length growth of stem and root has statistically been meaningful than control group in the level of 0/01 and 0/05, respectively. The highest effect of different concentrations NAA and BA on the dry weight of root and stem in 2/5 mg/lit concentrations of BA hormone was observed, the highest wet Wight of root was observed relating to 0/5 mg/lit of NAAconcentration, and then it was decreased by increasing in wet weight concentration. But, by increasing BA hormone concentration, wet weight of root was increased. For dry weight, also like as wet weight, it was observed increasing similar to wet weight. Statistically, the effects of thesehormones for wet weight of root and stem were meaningful in the 0/01 level, but for dry weight of root and stem has been in 0/05 level. In the other word, the highest amount of a and b chlorophyll with treatment of 2/5 mg/lit NAA and BA hormones and the highest amount carotenoid with 0/5 concentration were observed. So, statistically the effects of these hormones on the amount of a and b chlorophyll, totalchlorophyll and carotenoid have been meaningful in 0/05 level. The effect of different treatment of these two hormones on the amount of carbohydrate and protein did not show a meaningful effect in oat.

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Introduction

Oat with scientific name of Avene sativa is a plant from millet family (Poaceae) that has very tiny and indeterminate flowers. It is an annualgrassy plant that is planted as the food of livestock. The oat flowers are hyacinthine and its seeds have most amount of hydrate carbon such as starch and also fat acids such as palmetic acid, olnic acid and linoleic acid in its compositions. Oat has multicore root and segmented and hollow stem that each of them ended to a cluster. Its leaves are narrow and the bottom of them are surrounding as a sheath around the stem. Oat flour is a good source with the high nutritional value that is used for baking bread. Using oat bread causes to decrease sugar (saccahrose), cholesterol, HDL and LDL of blood. The founding of research indicates that using oat bread (25% total oat flour and 75% flour, 87% wheat) causes to decrease sugar. (saccahrose), cholesterol, HDL and LDL of consumers' blood. Different climatesand hormonal factors are enabled to affect on the trend of growth, the function level and also production quality. Cereal is the energy resource for human. In the developing country, this group of material includes the whole of diet. Cereal contains hydrocarbon, protein, fat, minerals and all kind of vitamins (1).Auxins are of plant hormones that have the axial role in regulating plant growth (18). Protein synthesis is accounted the main factors in cells growth. Any factor that prevents from proteinsynthesiswill undermine growth, too. Considering to nutritional value of oat in the diet and sufficient protein resource and reducing sugar (saccahrose), chlorophyll, HDL and LDL of blood and also as an adequate food for livestock, in this research was surveyed NAA and BA plant hormones, some physiological factors such as the amount of sugar (saccahrose), protein and chlorophyll and … in the oat plant.

Materials and methods

In order to implementing this research, the seed of (*Avene sativa*) has been provided from the agriculture office of karaj city. Expected survey were done in plant sciences laboratory of Islamic Azad University in 1391 and has been independently examined the effect of two hormones, auxin (NNA) and cytokinine (BA) and a compound of both of them with concentrations (0.5- 105- 2.5 mg/L)on and morphological photosynthesis pigments and amount of carbohydrate and protein of (*Avene sativa*) plant.

Measuring the Parameter of Growth

Measuring the length of stem and root was done with millimeterruler. The wet and dry weight was weighted by digital scale with 0/0001precision.

Measuring Chlorophylls and Carotenoids

For measuring the degree of chlorophyll and carotenoid were used the method of (Lichtenthaler). For accounting the amount of chlorophylland carotenoid, attraction of extractions was read in the wave length 663 nm for a chlorophyll and the wave length 645 for b chlorophyll and 470 nm for carotenoid by spectrophotometer.

Measuring the Soluble Sugar (Saccahrose)

For measuring carbohydrate, it was used the method of Nelson (1943). The degree of attracting extractions was read in the wave length 600 nanometer by spectrophotometer. The concentration of carbohydrate sampled was accounted by standard curve base on mg/gr dry weight (mg.g- DW).

Measuring Protein

For measuring protein, it was used the method of (Lowry 1951). The degree of extractions was read in the wave length660 nanometer by spectrophotometer. And the amount of protein of each sample was accounted by standard curve base on wet weight mg/gr ($mg.g^{-1}$ DW).

Statistical Analysis

The tests were implemented with three replications in a full-accidental plan form. The amounts of three replicate were obtained and calculated variance and the standard deviation of means and the difference among means by variance analysis. Surveying the result of tests and drawing curves were implemented according to comparing means and standard deviation (Mean $\pm$ SE). Grouping treatments in the level of 5% ($p \leq 0.05$) was done by Tukey test. Statistical analysis of data was done by SPSS software and for drawing diagrams, it was used EXCEL software

Results and Discussion

Alterations of aerial organs and root

By applying BA and NAA treatment sindifferent concentrations, the highest stem length related to 1/5 mg/lit NAA and the lowest amount in the control level were observed (diagram 1-1). And also it was observed the highest amount root length in NAAtreatment, 1/5 mg/lit and the lowest amount in control (2-1). Regarding to stem length between control and NAA treatments, 1/5 mg/lit and for root between control and the treatments of level of 1/5 NAA and 0/5 mg/lit BA and 1/5 mg/lit BA and NAA their difference statistically is meaningful in the level of ($p<0/01$) and ($p<0/05$), respectively.

Changes of Wet and Dry Weight of Plant Steam and Root

By applying different BA and NAA treatments on wet weight of root, it was observed the highest amount related to 1/5 mg/lit and it's the lowest amount related to 0/5 mg/litBA and NAA. Statistically, it was observed a meaningful between control level and 2/5 mg/lit BA, so. Their difference was meaningful in the statistically level, 0/01(1-3).For the dry weight, by increasing in applied NAA treatment, dry weight of root was decreased. But by increasing treatment BA, the dry weight (DW) was increased, so by the effect of BA and NA treatments, the highest and the lowest amounts have been related to 1/5 mg/lit and 0/5 mg/lit concentrations. And its difference is meaningful in the statistically level 0/05.

By applying different level BA and NAA in stem, it was observed the highest amount of wet weight related to treatment 2/5 mg/lit and the lowestamount related to BA and NAA 0/5 mg/lit. But, there is not a meaningful difference in the other BA and NAA treatments levels (diagram 5-1).Relating to the effect of above hormones, the dry weight (DW) of stem was statistically increased by applying different treatments BA and NAA ($p<0/05$)

The content changes of soluble sugar (Saccahrose) in aerial organs

The content changes of protein in aerial organs

Relating to the amount of leaf protein, it was observed the highest amount of protein concentration in the control level with 1/24 mg/litconcentration on the dry weight gram and the lowest protein concentration in the BA and NAA level, 1/5 mg/lit with 0/09 mg/lit concentration on the dry weight. Of course, by the effect of different treatments BA and

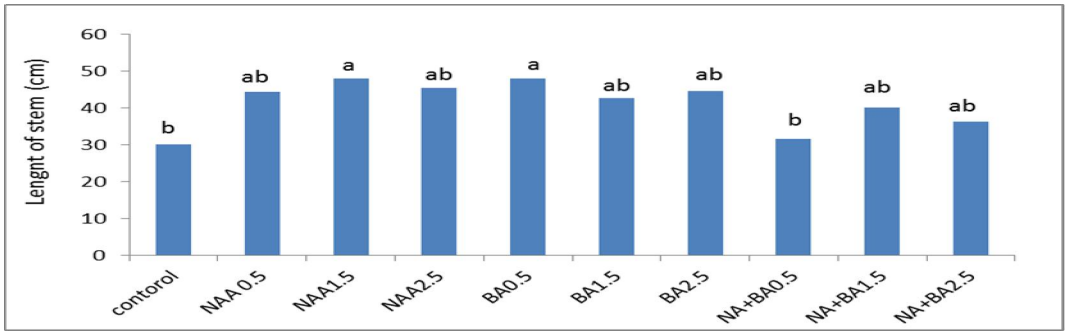

Figure 1: The effect of difference treatments (NAA; BA; NAA+BA) on the length of stem (cm)

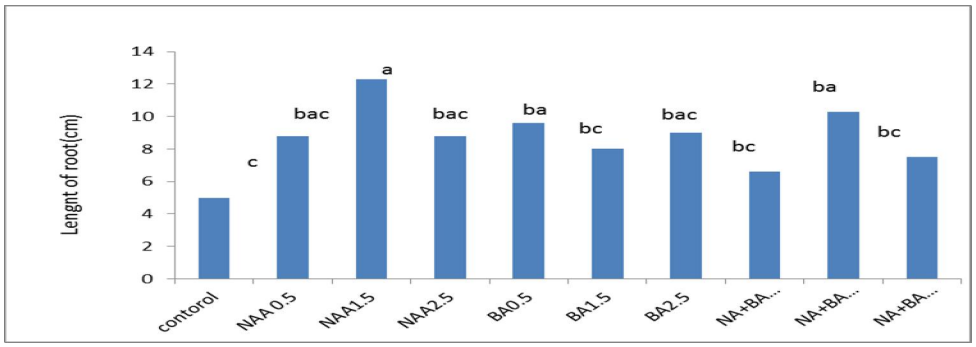

Figure 2: The effect of difference treatments (NAA; BA; NAA+BA) on the length of stem (cm)

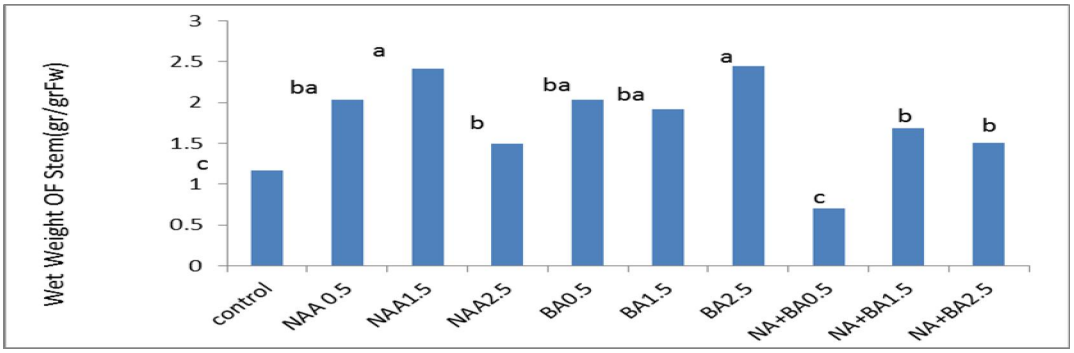

Figure 3:The effect of difference treatments (NAA; BA; NAA+BA) on the wet weight of root (g)

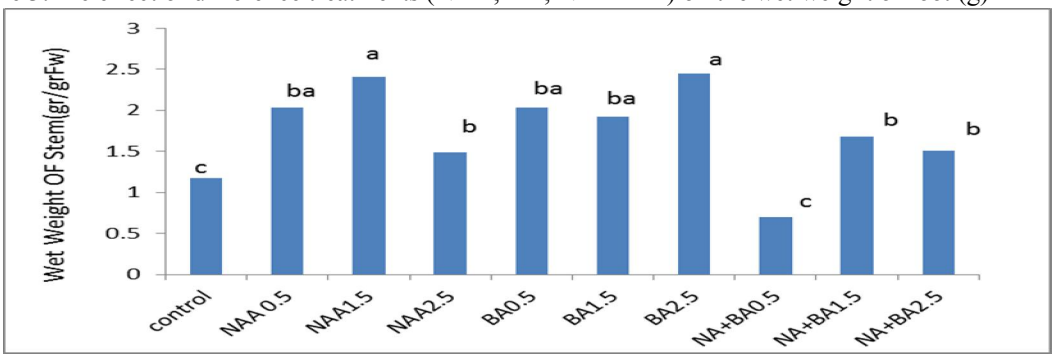

Figure 4: The effect of difference treatments (NAA; BA; NAA+BA) on the wet weight of stem (g)

NAA, increasing in BA and NAA concentration causes to increase protein amount. But, by increasing in BA and NAA concentration, the amount of protein was decreased. And there is not statistically a meaningful difference for the amount of protein among the control and different levels of BA and NAA and among the control and 2/5 mg/lit BA and NAA.

Changesin the pigment composition of leaves(ChlorophyllsandCarotenoids)

bytheeffectofdifferentconcentrations BA and NAAon the amount of Chlorophyll a, it was observed the highest amount related to 2/5 mg/lit BA and NAA with 12/78 (mg/gFW) and the lowest amount related to control with 6/1 (mg/gFW). There is not statistically a meaningful difference among the control and 2/5 mg/lit BA and NAA and 1/5 mg/lit NAA in the level of 0/05 ($p<0/05$). By applying different treatments BA and NAA, the amount of Chlorophyll b was not meaningful, statistically. So, the highest amount Chlorophyll b was related to 2/5 mg/lit BA and NAA by amount of 12/66 (mg/gFW) and the lowest amount was related to 0/5 mg/lit BA and NAA by amount of 5/82 (mg/gFW). Relating to the total Chlorophyll, it was observed the highest amount related to 2/5 mg/lit by amount of 33/63 (mg/gFW) and the lowest amount related to the control by amount of 12/87 (mg/gFW). That statistically, it is meaningful in the level of 0/01. By the effect of applied BA and NAA different concentrationon the concentration of Carotenoid, it was observed a meaningful difference in 0/01 level. the lowest amount of Carotenoid was related to 2/5 mg.g BA and NAA and the highest amount of Carotenoid was related to 0/5 mg/lit BA, so they have 1/73 wet weight mg.g and 7/41 dry weight mg.g. There is a meaningful difference in ($P<0/01$) level, statistically. The effect of different BA and NAA treatments on root and stem length:

In the research, it was shown that the response of growth by inducing IAA in the stem of seed or oat cleopetil in the different concentration of exogenous IAA, the effect of auxin will be decreased in high concentration (10^{-5}) and it is inhibitor in the concentration more than 10^{-4}, so in such concentration, the growth threshold will be severely decreased by the lack of IAA. Auxins will increase the stems growth and the stem sheath, so auxin willberapidly increased expansibility of cell wall by releasing proton and acidifying wall. Cytokines are used for inducing growth and distinguishing in the cell culture.The effect of cytokines is more particularly when is added along. Auxin to medium.

It was observedthe highest amount of carbohydrate in the control with the concentration 0/28 (mg/gDW) and the lowest amount of carbohydrate in the level of (BA and NAA) 2/5 mg/lit with concentration 0/077 (mg/gDW) (diagram 1-7). There is not a statistically meaningful difference for the amount of carbohydrate among

Cytokine in high concentration is caused lateral growth, while naturally preventing the root growth. In the most root merytisms, increasing in extra cytokine is caused to stop mitosis. Rafeekher et al (2001) showed that NAA increases the length of between nodes. Surveying the different concentrations effect of cytokine on micro propagation of zinnia in 1388 has been showed that the most number of leaves was by BA hormone with 3micromolarconcentration, and thehighestconcentrationof leafelongation was with 2 micro molar concentrations and the higheststem elongation was by BA with1micro molar concentration. Consideringtoresults from surveying different hormonal concentrations on rooting, it was observed the highest meanroot length related to 1/5 mg/lit.

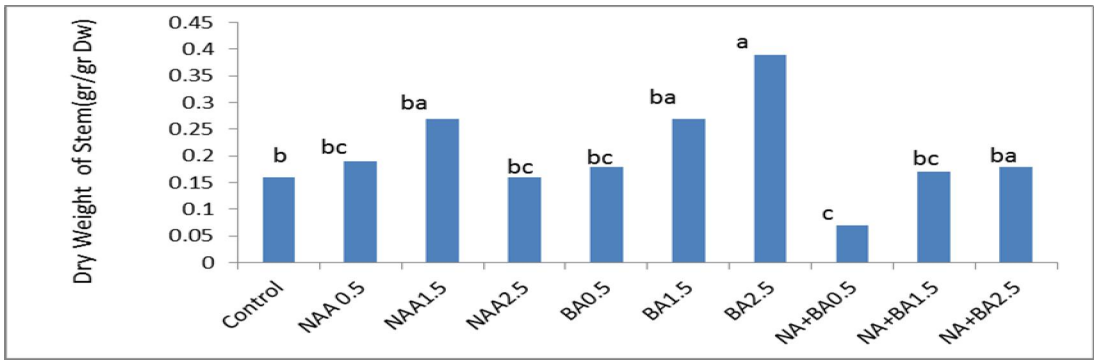

Figure 5: The effect of difference treatments NAA;BA; NAA+BA) on the dry weight of root (g)

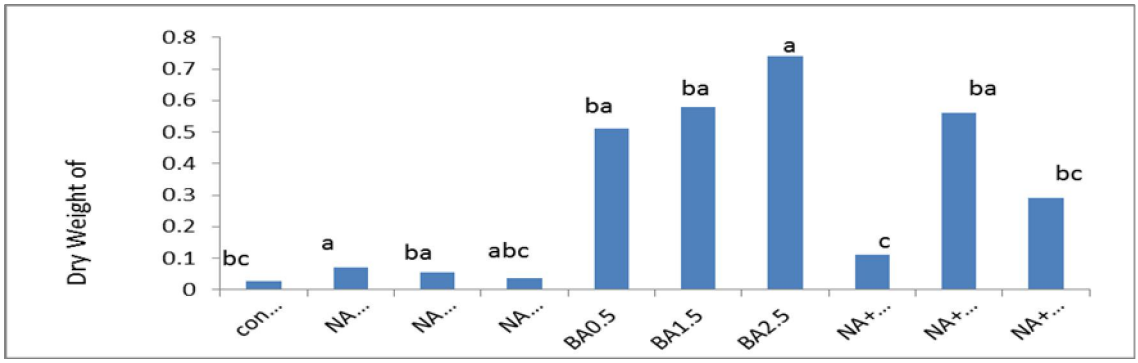

Figure 6: The effect of difference treatments (NAA; BA; NAA+BA) on the dry weight of stem (g)

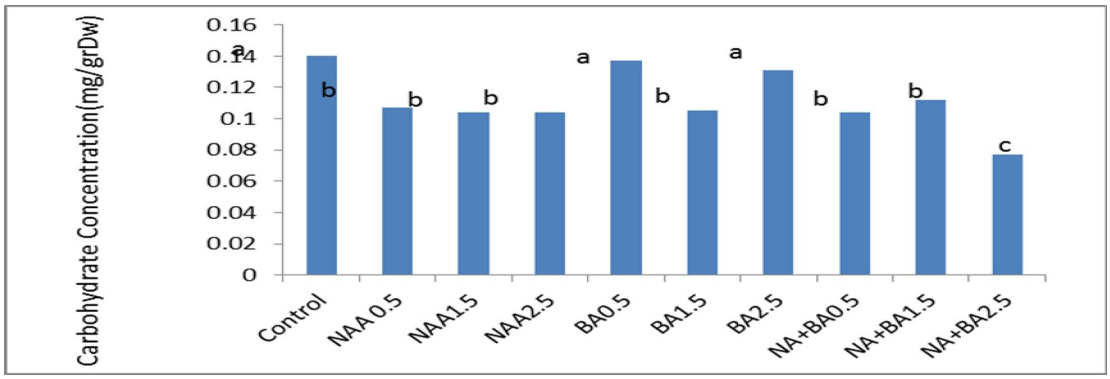

Figure7: The effect of difference treatments (NAA; BA; NAA+BA) on the amount of carbohydrate (mg/mfw)

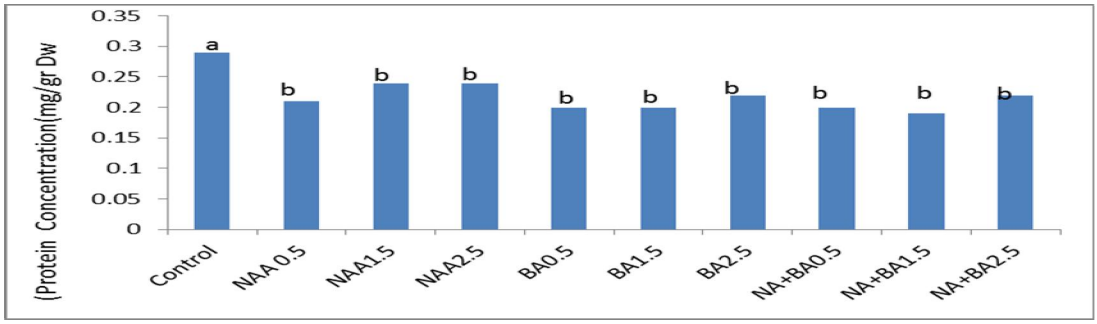

Figure8: The effect of difference treatments (NAA; BA; NAA+BA) on the amount of protein (mg/gFw)

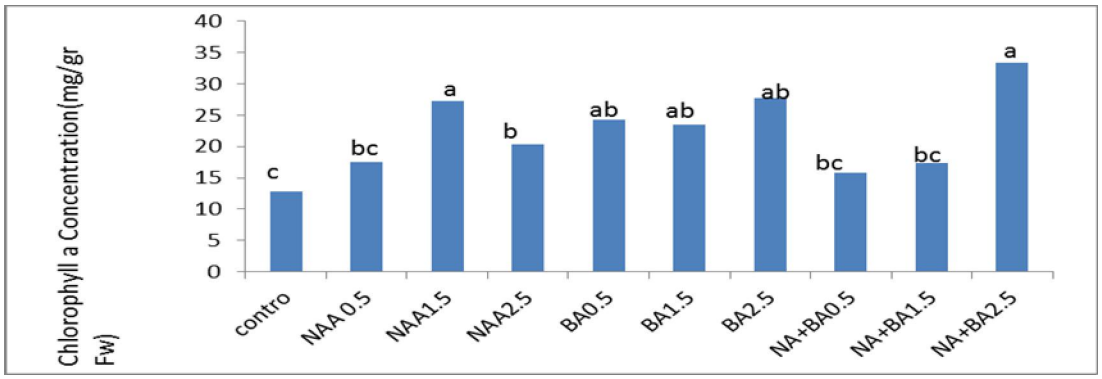

Figure 9: The effect of difference treatments (NAA; BA; NAA+BA) on the amount of chlorophyll a (mg/gFW)

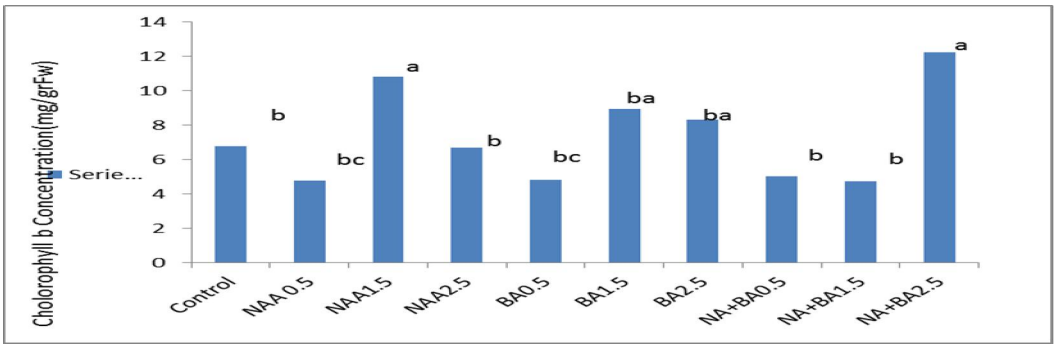

Figure10: The effect of difference treatments (NAA; BA; NAA+BA) on the amount of chlorophyll b(mg/g FW)

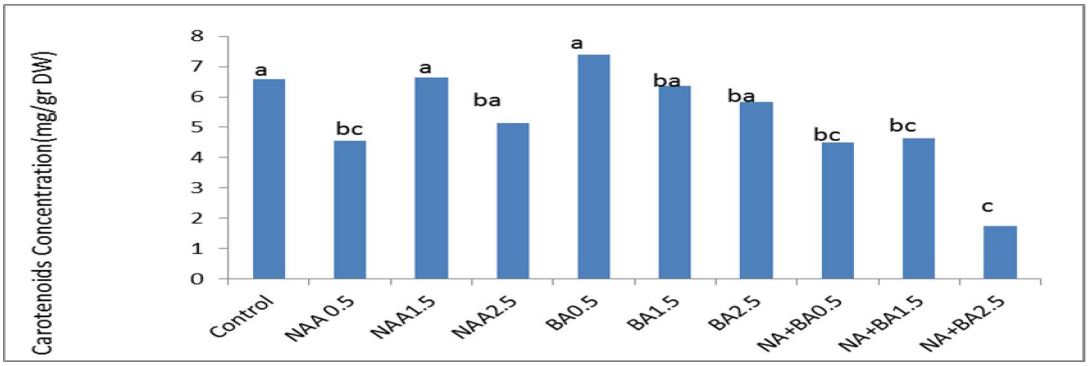

Figure11: The effect of difference treatments (NAA; BA; NAA+BA) on the amount of carotenoid (mg

The effect of NAA and BA treatments on the wet and dry weight of root and stem

In this research, the wet weight of root and stem was increased by increasingBA,so in the concentration of 2/5 mg/lit; it shows the highest wet weight that was similar to Mont's observations in 1986. Mont saw that the concentration of 2/5 mg/lit in Benzyl Amino Poorin-6 has most effect on increasing primary wet weight with concentration 0-4 mg/lit. (5) And also in combination of BA and NAA along with increasing concentration, the wet weight is increased. But by increasing NAA concentration, the trend of increasing to 1/5 mg/lit treatment, it is observed increasing in wet weight and then is decreased. In this research, it is observed that the inverse effect of NAA on aerial organs and root is consistent with the implemented researches. Cytokine causes to increase the movement of foodin leaves and their accumulation and increasing in dry weight. In the other hand, the main cells weight is related to wall cellulose and water in it. Therefore, it is logical that by increasing BA to the concentration of 2/5 mg/lit, wet and dry weight of stemis increased (Lesani- Mojtahedi, 1367) the accumulation of Estatolytes by NAA and Phenomenon ofgeotropism causes to increase the dry root.

The effect of NAA and BA treatments on photosynthesis pigments

By increasing NAA concentration, the amount of a chlorophyll has been shown increasing to 1/5 mg/lit then its amount was decreased. By implying BA treatment, it was seen decreasing and then increasing a chlorophyllbyincreasing concentration to 1/5 mg/lit. But, by applyingcombination of (NAA+BA) increasing in concentration of a chlorophyllamountshowsto increase. Relating to the effect of these hormones on the b chlorophyll amount, it was increased b chlorophyll to 1/5 mg/lit and then decreased by increasing BA+ANN concentration, but the amount of this pigment was increased by increasing (NAA+BA). By applying the different concentration of NAA, it was increased the total chlorophyll to 1/5 mg/lit by increasing concentration and then decreased. Chlorophyll in plants has primarily role in photosynthesis for attraction and using light energy. Therefore, the regulators of plant growth directly affect on biosynthesis and chlorophyll

degradation directly affects on photosynthesis.Various reports describe about decreasing chlorophyllsand Carotenoids contents under stress, so it can be referred to some reports for soybean (Sheteawi,2001), bean (Kaymakanova & Stoeva) and on Withania somnifera (Jaleel & Azooz.

The results show that Cytokinis along the other factors, such as light, food materials and growth regulate forming photosynthesis pigments and proteins. The researches show that the highest chlorophyll amount related to NAA+BA has been 2/5 mg/lit that are consistent with the above findings.

In the present research, in the treatment that affected by NAA and BA, it was observed the incremental effect on the chlorophyll and Carotenoid content of oat. So, there are consistent with the numerous reports based on improving chlorophyll and Carotenoid content in the plants which was treated than control plants, so it can be referred to Khaya sengalensis (Abd El-Aziz), soybean (Sheteawi, 2007) and on the chickpeas (Beltagi, 2008).

The effect of NAA and BA treatments on protein content

Auxin is effective in biosynthesis some enzymes such as cellulose and peroxidase. Auxin interferes from the transcription stage and makes active RNA polymerase enzyme to make DNA, RNA from it and in real, Auxin does not interfere in biosynthesis DNA, but it affects on the transcription stage.There are some documents that show Cytokines play role in synthesis protein. Cytokines not only are able to increase synthesis protein, but also they can alter the made portions spectrum by plant tissues. This subject is proved by marking proteins of cultured soybean cells and separating it by gel poly acryl amid electrophoresis. Auto-radiograms of gel showed that treatment with Cytokines increase some proteins, soprevent to synthesis another protein (Fosket and Tifer, 1978). The tissues of treated tobacoo with Cytokine also indicates the alteration of protein synthesis pattern, so it shows hormone is necessary for synthesis of some proteins and preventing the other proteins (Eik Holes et al. 1983).

IAA and Benzyl adenine both are two inhibitor materials of advancing activity in peptidase in the intact grains,Cucurbita maxima. Cytokines is increased the ribonuclease activity of tobacco leaves, tomato leaves and oat and wheat leaves and chickpea Epicotyls. So, it can be decreased the degree of protein synthesis in the next stages. The protease activity is decreased in tobacco leaves by Cytokine, but kinas protein activity is increased by Cytokine, but it is decreased in the cut leaves. The inhibitors made of protein stops making stimulated Ethylene by Auxin that indicates making ethylene by Auxin has been caused to significantly increase in ethylene. Some genes are indentified that their transcription is increased following than using exterior Auxin, this case indicates that the transcription advancement is almost the cause of increasing ethylene in response to Auxin. In protein includes NAA and BA, increasing the hormone concentration shows that it has been decreased in Control, but it was increased under hormone treatment than the plants treated by protein.

The effect of NAA and BA treatments on Carbohydrate content

Many enzymes interfere in carbohydrate metabolism; one of them is Alfa Amylase that affects on starch hydrolysis and the other is Invertase that degrades sucrose. Auxins increase Invertase activity amongnodesand restoring Paranshim of Cane and Chicory root slices and decreaseInvertaseactivityin Jerusalem artichoke tubers. Metabolytes accumulation is one of plant probable general responses to exterior potential alteration and metabolites accumulation such as sugar (saccahrose)s were seen in plants under inadequate situations (Sotiropoulos). Various reports express increasing in soluble sugar (saccahrose) in plant cells under this stress that it can be referred to the reports on sunflower (Ashraf & Tufail), on soybean (Sheteawi, 2007) and on oat (Khosravinejad et al, 2009). The results of this research showed that soluble sugar (saccahrose) content is not increased in treatment affected by NAA and BA in comparison with control. Therefore the above treatments have not been caused to improve photosynthesis and carbon productions.

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