Response of vegetative growth and chemical constituents of *Thuja orientalis* L. plant to foliar application of different amino acids at Nubaria.

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Abstract: A pot experiment was carried out during 2008 and 2009 seasons at Research and production Station, Nubaria of National of Research Centre, Dokki, Egypt to study the response of *Thuja orientalis* plants to foliar application of tyrosine, thiamine and tryptophan each at (0, 25, 50, and 100 ppm) on vegetative growth expressed as stem length, stem diameter, root length, fresh and dry weight of root and shoot and chemical constituents significantly were affected by the application of almost all of the three amino acids which were used in this study. Tyrosine, Thiamine and Tryptophan promoted all morphological characters. The comparison between the effect of tyrosine, thiamine and tryptophan revealed that the influence of tyrosine on increasing the growth parameters (especially at the rate of 10 ppm, which can be described as the most effective treatment) was superior to other amino acids (thiamine and tryptophan). The three amino acids increased total soluble sugar %, total free amino acid mg/g as well as essential oil %, essential oil yield / plant and N, P, K % and protein. Therefore, amino acid (Tyrosine, Thiamine and Tryptophan) at 100 ppm maybe recommended for promoted growth parameters and the best oil percentage in *Thuja orientalis* L. seedling. [Journal of American Science 2010;6(8):295-301]. (ISSN: 1545-1003).

Keywords: Tyrosine, Thiamine and Tryptophan, morphological characters.

1. Introduction

Thuja orientalis plants, Family Cupressaceae are an evergreen tree growing to 15m. at a slow rate. The seeds ripen from September to October. The flowers are monoecious (individual flowers are either male or female, but both sexes can be found on the same plant) and are pollinated by wind. It can grow in semi-shade (light woodland) or no shade. This plant is commonly used in Chinese herbalis, where it is considered to be one of the 50 fundamental herbs (Duke and Ayensu, 1985). The leaves are antibacterial, antipyretic, antitussive, astringent, diuretic, refrigerant and stomachic (Yeung, 1985).

The root bark is used in the treatment of burns and scalds (Duke and Ayensu, 1985). The stems are used in the treatment of coughs, colds, dysentery and parasitic skin-diseases.

Amino acids as organic nitrogenous compounds are the building blocks in the synthesis of proteins (Davies, 1982). Amino acids are particularly important for stimulation cell growth, they act as buffers which help to maintain favorable PH value within the plant cell, since they contain both acid and basic groups; they remove the ammonia from the cell.

This function is associated with amid formation, so they protect the plants from ammonia toxicity. They can serve as a source of carbon and energy, as well as protect the plants against pathogens.

Tyrosine is hydroxy phenyl amino acid that is used to build neurotransmitters and hormones. Hass (1975) stated that the biosyntheses of cinamic acids (which are the starting materials for the synthesis of phenols) are derived from phenylalanine and tyrosine.

The role of Tryptophan is well known: it has an indirect role on the growth via its
Influence on auxin synthesis. Phillips (1971) reported that alter native routes of IAA synthesis exist in plants, all starting from Tryptophan.
Thus, when Tryptophan was supplied to some plant tissues IAA was formed.

Thiamine (vitamin B1) could serve as coenzyme in decaboxylation of a-keto acids, such as Pyruvic acid and keto-glutamic acid which has its importance in the metabolism of carbohydrates and fats (Bidwell, 1979). Thiamine is an important cofactor for the transketolation reactions of the pentose phosphate cycle, which provides pentose phosphate for nucleotide synthesis and for the reduced NADP required or various synthetic pathways (Kawasaki, 1992).

Several authors indicated the promotive effect of amino acids on plants growth including, Mohamed and Khalil (1992) on Antirrhinum majus, Mathiola incana and Callistephus chinensis, Abou Dahab and Nahed (2006) on Philodendron erubescens, Nahed and Balbaa (2007) on Salvia farinacea and Nahed et al, (2009) on Antirrhinum majus .

Harridy (1986) on *Catharanthus reseus* L., Abou Dahab and Nahed (2006) on Philodendron erubescens, and Nahed et al. (2009) on Antirrhinum majus found that foliar application of different amino acid treatments caused a significant increase in the content of total free amino acids. Talaat and Youssef (2002) on Ocimum basilicum L., Wahba et al., (2002) on Antholyza aethiopica, Abou Dahab and Nahed (2006) on Philodendron erubescens and Nahed et al. (2009) on Antirrhinum majus stated that application of amino acids as a foliar spray caused an increase in the contents of total soluble sugars.

The aim of this work was to enhance plant growth and chemical constituents of *Thuja orientalis* by foliar application of tyrosine, thiamine and tryptophan.

2. Material and Methods

Two pot experiments were carried out at National Research Centre (Research and production Station Nubaria), during two successive seasons 2008 and 2009 to study the effect of some amino acids (tyrosine, thiamine and tryptophan) on growth, chemical constituents and essential oil of Thuja orientalis L. Six months old seedling of Thuja were obtained from nursery of Forestry Department, Horticulture Research Institute, the seedlings were planted on the last week of March at the two seasons 2008 and 2009, in plastic pot 30cm. in diameter, filled with 10kg of sand soil, one plant / pot, the average heights of seedlings were 15-20cm. The investigated soil characterized by sand 73.33 %, silt 4.43%, clay 17.24% with PH 7.81, EC 2.18ds/m. CaCo 22.5% OM 1.55%, Ca 11.61, Mg 4.80, Na 4.64, Cl 1.80, HCo 1.40, So 8.5 meq/L, N 19.60, P 65.80, K 165.64 meq/100g. The available commercially fertilizer used through this experimental work was Kristalon (NPK 19:19:19) produced by Phayzon Company, Holand. The fertilizers rates (5.0 gm/pot) used in four equal doses after 4,8,16 and 20 weeks from transplanting. The plants were sprayed with the three amino acids tyrosine, thiamine and tryptophan (Each concentration of 25, 50 or 100ppm), in addition to the untreated plants. The amino acids foliar spray treatments were applied two month after transplanting (on May 25^{th} in both seasons) and were repeated two times at one month intervals. At last week of November of 2008 and 2009, the following data were recorded: plant height (cm), stem diameter (mm), root length (cm), fresh and dry weights of shoots and roots (gm).

The experiment was set in a completely randomized design with ten treatments and six replicates of each treatment. The data were statistically analyzed according to Snedcor and Cochran (1980) using the least significant differences (LSD) at 5% level. The following chemical analyses were determined: total soluble sugar percentages were determined according to the method of Dubois *et al.*, (1956). Essential oil was determined in the shoots of each treatment according to Badawy *et al.* (1991), and essential oil yield / plant were determined by multiplying essential oil percentage X average of dry weight of shoots /plants. Free amino acid content was determined according to Rosen, H., (1957) Nitrogen, Phosphorus and Potassium were determined according to the method described by Cottenie *et al.*,(1982). Protein percentage in the root and shoot was determined by multiplying Nitrogen % x 6.25. (This is based in the assumption that plant proteins contain 16% nitrogen) according to the method as described by Ranganna (1978).

3. Results and Discussions

Effect o of Tyrosine, Thiamine and Tryptophan on vegetative growth:

Data presented in fig. (1, 2, 3, 4 and 5), indicated that foliar application of amino acid (tyrosine, thiamine and tryptophan) significantly promoted stem length, stem diameter, root length, fresh and dry weights of shoots and roots, as compared with the untreated plants, except the application of thiamine at 25ppm increased stem diameter root length and root fresh weight compared with control but it was non significant. Tyrosine at all level gave the greatest values of result compared with the other amino acids at the same level. The highest values of results were obtained in plants treated with Tyrosine followed by tryptophan and thiamine at 100ppm. The increase in fresh weight of shoots by 52.6%, 41.7% and 31.9%, respectively compared with control plants, as well as dry weight of shoot by 83.0%, 61.5% and 46.6% respectively. Generally, all growth parameters gradually increased by increasing amino acids concentration. The positive effect of amino acids on yield may be due to the vital effect of these amino acids stimulation on the growth of plant cells. The positive effect of amino acids on growth was stated by Goss (1973) who indicated that amino acids can serve as a source of carbon and energy when carbohydrates become deficient in the plant's amino acids are determinate, releasing the ammonia and organic acid from which the amino acid was originally formed. The organic acids then enter the Kerb's cycle, to be broken down to release energy through respiration. Waller and Nawacki (1978) reported that the regulatory effects of the amino acids could indirectly be explained since certain amino acids were suggested to affect plant development through their influence on gibberellins biosynthesis. Thon et al., (1981) pointed out that amino acids provide plant cells with an immediately available source of nitrogen, which generally can be taken by the cells more rapidly than inorganic nitrogen. Kawasaki (1992) mentioned that thiamine is a necessary ingredient for the biosynthesis of the coenzyme thiamine pyrophosphate; in this latter from it plays an important role in carbohydrate metabolism. It is an essential nutrient for both plant and animal. In plants, it is synthesized in the leaves and is transported to the roots where it controls growth.

The results are characteristically accompanied by Mona and Talaat (2005) on Pelargonium graveolens plants , Nahed and Balbaa (2007) on Salvia farinea plants, Nahed et al., (2009) on Gladiolus grandflorum and Nahed et al., (2009)on Antirrhinum majus , they found that amino acids significantly increased vegetative growth.

Total soluble sugar contents:

Data in Table (1) showed that, the shoots and roots contents of total soluble sugar gradually increased by increasing amino acids concentration compared with the untreated plants. In addition data revealed that the amino acids treatments which were used in this study had significant effect on total soluble sugar contents.

Plant treated with thiamine at 100ppm had the highest total soluble sugar contents (7.18, 7.18%) in roots and shoots respectively. The comparison between the effect of thiamine, tyrosine, and tryptophan revealed that the influence of thiamine on increasing the total soluble sugar contents (especially at the rate of 100ppm, which can be described as the most effective treatment) was surperior to other amino acids (tyrosine, and tryptophan). The promotive affect of the amino acids on the total soluble sugars percentages may be due to their important role of the biosynthesis of chlorophyll molecules which in turn affected chlorophyll content.

In this concern, Devlin (1969) stated that there is agreement that succinyl CoA (Kerbs cycle amino acids glycin, initiate the biosynthetic leading to chlorophyll formation. Similar results have been reported in other plant species, Attoa *et al.*, (2002) on *Iberis amara* L., Yossef and Talaat (2003) on rosemary, Iman et al., (2005) on Catharanthus roseus L., Nahed and Balbaa (2007) on Salvia farinacea plants and Nahed et al., (2009) on Antirrhinum majus.

Total free amino acid: The results presented in Table (1) illustrated that total free amino acid contents were significantly increased as a result of different foliar application of amino acid. Application of the high tyrosine concentration (100ppm) was the most effective treatment in producing the highest values. The percentage of increase due to this treatment over the control reached 81.03 and 28.88% in the root and shoot respectively. The increase in the content of total free amino acids as a result of the different amino acid treatments may be attributed to amino acids as organic nitrogenous compound are the building blocks in the synthesis of proteins which formed by a process in which ribosome catalyze the polymerization of amino acids (Davies, 1982). Our results are in agreement with the findings of Abou Dahab and Nahed (2006) on Philodendron erubescens, Nahed and Balbaa, (2007) on Salvia farinacea.

Essential oil % and essential oil yield /plant (ml):

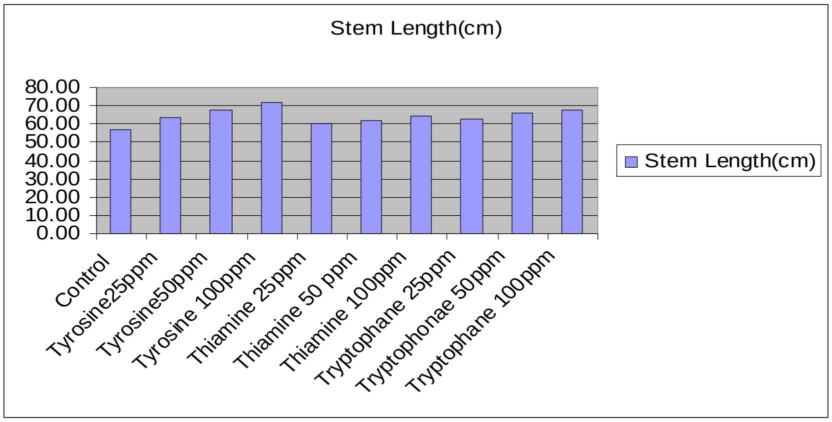
Data presented in Table (1) show that oil % and total oil yield (ml) / plant were significantly increased as a result of foliar spray of different amino acids. It is clear from the obtained data that the highest recorded value of essential oil was obtained in the herb of plants treated with 100ppm tyrosine. In this respect several investigators studied the effect of different amino acids on the total oil percentage and oil yield (ml / plant) Gamal El.Din et al., (1997) on Cymbopogon citratus hort, Talaat and Youssef (2002) on basil plant, Karima et al., (2005) on Matricaria chamomilla L.and Mona and Talaat (2005) on pelargonum graveolens L.

Minerals and total protein percentage:

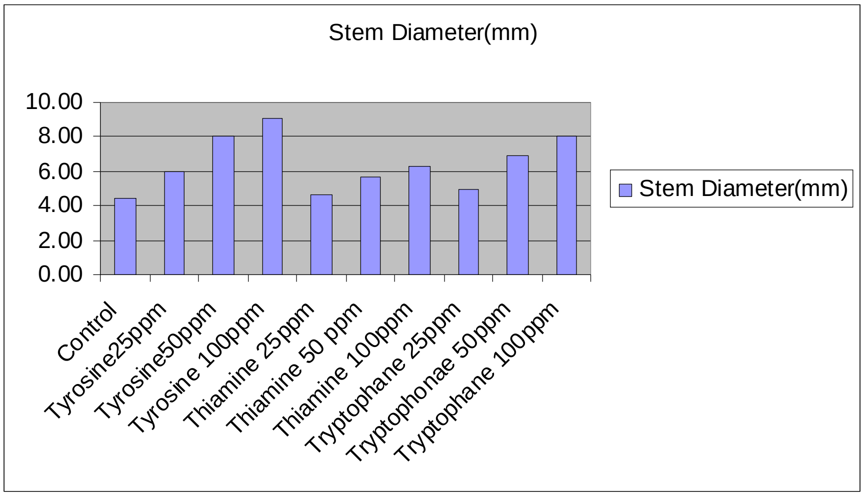
As regards the effect of different amino acids application on macronutrients (N, P and K) and total protein, it is to be from the results in Table (2) tyrosine, thiamine and tryptophan at 100ppm respectively gave the highest values in most cases (shoots, roots) as compared with untreated plants.

These results are in agreement with those obtained by Youssef and Talaat (2003) who reported that folia application of thiamine increased the total nitrogen percentage, total phosphorus % and total potassium % on rosemary plants. These increments led to quantitive changes in amino acids and specific proteins which acted positively in cell division and cell elongation (Bekheta and Mahgoub, 2005) on Carnation plants.

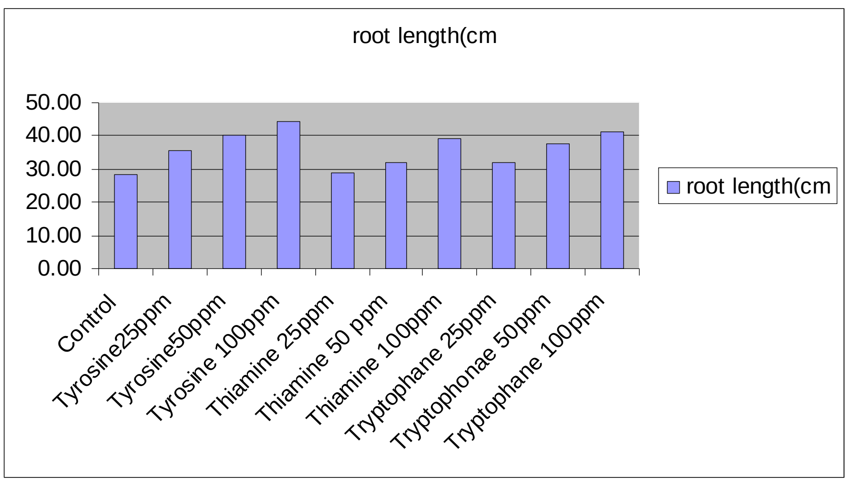
Therefore, amino acid (thiamine, tyrosine, and tryptophan) at 100 ppm maybe recommended for promoted growth parameters and best oil percentage in Thuja orientalis L., seedlings.



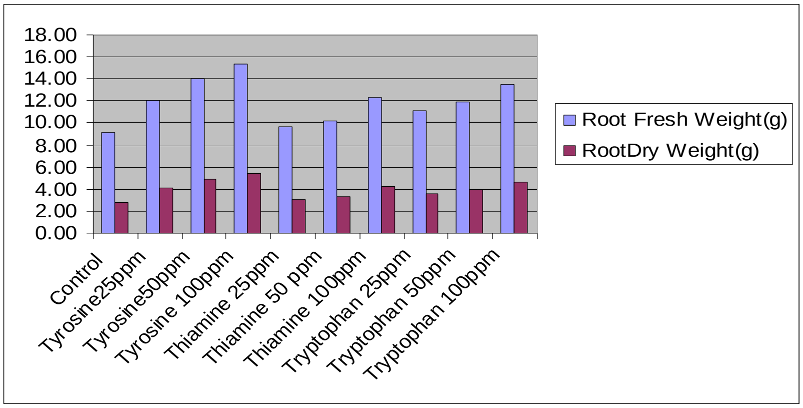
Figure(1): Effect of tyrosine, thiamine and tryptophan on Stem length(cm) of *Thuja orientalis* seedlings



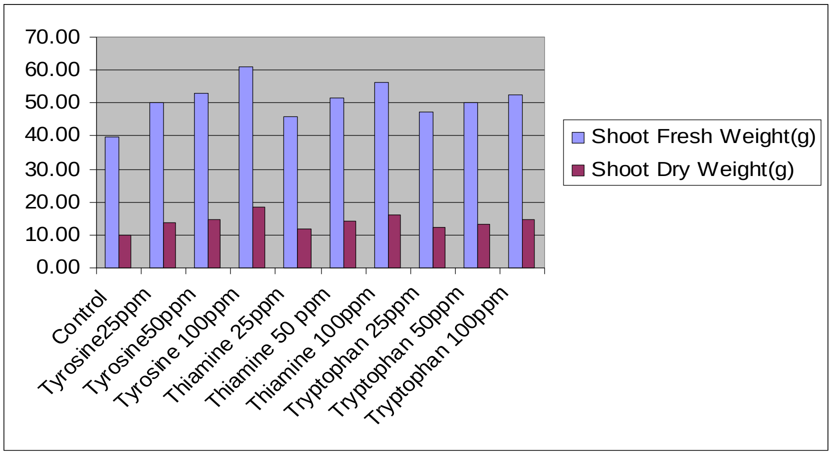
Figure(2): Effect of tyrosine, thiamine and tryptophan on Stem diameter(mm)of *Thuja orientalis* seedlings



Figure(3): Effect of tyrosine, thiamine and tryptophan on root length(cm) of *Thuja orientalis* seedlings



Figure(4): Effect of tyrosine, thiamine and tryptophan on root fresh and dry weights (g) of *Thuja orientalis* seedlings



Figure(5): Effect of tyrosine, thiamine and tryptophan on shoot fresh and dry weights of *Thuja orientalis* seedlings

Table(1) Effect of tyrosine, thiamine and tryptophan on total soluble sugar percentage in root and shoot, total free amino acid mg/g in root and shoot, essential oil percentage in shoot and essential oil yield/plant m/l. of *Thuja orientalis* seedlings(average of two seasons 2008 and 2009)

characters/treatments	Total soluble sugars %		Total free acid mg/g		Essential oil %	Essential oil yield/plant (ml)
	root	Shoot	Root	Shoot		
Control	4.10	3.59	3.11	4.46	0.259	0.026
Tyrosine25ppm	4.85	4.85	5.40	5.53	1.367	0.186
Tyrosine50ppm	5.36	5.36	5.54	5.69	1.497	0.223
Tyrosine100ppm	5.97	5.97	5.63	5.98	1.693	0.309
Thiamine 25ppm	5.89	5.93	3.73	4.83	0.447	0.053
Thiamine 50ppm	6.38	6.65	3.9	4.99	0.473	0.067
Thiamine 100ppm	7.18	7.18	4.24	5.39	0.537	0.087
Tryptophan 25ppm	4.37	4.37	4.34	4.96	0.766	0.096
Tryptophan 50ppm	4.67	4.67	4.59	5.18	0.879	0.118
Tryptophan 100ppm	5.17	5.17	4.91	5.61	0.996	0.146
L.S.D at 5%	0.06	0.05	0.14	0.08	0.11	

Table(2): Effect of tyrosine, thiamine and tryptophan on Nitrogen, phosphorus, potassium, and protein percentage in (Root and Shoot)of *Thuja orientalis* seedlings (average for two seasons 2008 and 2009)

characters	Shoot				Root			
treatments	N%	P%	K%	Protein%	N%	P%	K%	Protein%
Control	0.89	0.26	0.24	5.56	1.08	0.24	0.43	6.75
Tyrosine 25ppm	1.08	0.32	0.59	6.75	1.18	0.29	0.58	7.38
Tyrosine 50ppm	1.45	0.39	0.68	9.06	1.22	0.35	0.6	7.63
Tyrosine 100ppm	1.61	0.43	0.86	10.06	1.26	0.39	0.66	7.88
Thiamine 25ppm	1.98	0.42	0.86	12.38	1.24	0.33	0.68	7.75
Thiamine 50ppm	2.04	0.52	0.98	12.75	1.39	0.39	0.72	8.69
Thiamine 100ppm	2.26	0.58	1.12	14.13	1.67	0.53	0.88	10.44
Tryptophan 25ppm	1.65	0.35	0.76	10.31	1.2	0.31	0.61	7.5
Tryptophan 50ppm	1.72	0.44	0.86	10.75	1.29	0.38	0.63	8.06
Tryptophan 100ppm	1.88	0.49	0.93	11.75	1.36	0.43	0.69	8.5
L.S.D at5%	0.07	0.06	0.05	0.44	0.03	0.07	0.04	0.2

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