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Influence of Hormones of IAA, IBA, and NAA on Improvement of Rooting and Early Growth of Tilia rubra subsp. caucasica Form Angulata (Rupr.) V. Engler

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ABSTRACT

Aims So far there has not been any research on propagation of Tilia rubra subsp. caucasica form angulata (Rupr.) V. Engler in the world. In this research, for the first time, the rooting and early growth in cuttings of Tilia rubra subsp. caucasica form angulata (Rupr.) V. Engler was studied by applying various hormones of IAA, IBA, and NAA.

Materials and Methods In the present study, experiments were conducted as completely randomized block design and three replications with indole-acetic acid (IAA), Indole-Butyric Acid (IBA) and naphthalene-acetic acid (NAA) with concentrations of 0 (Control), 50, 100 and 200mg/l for 115 days.

Findings Based on the findings, compared to control, rooting percentage was enhanced about 37.5% in cuttings imbibed with 100mg/l IBA. Root number and root length respectively showed an increase of 28.6% and 22.3% in cuttings imbibed with 200mg/l IAA. The greatest root mass was perceived in IBA 200mg/l, which was about 2.5 times larger than that in control. In IBA 200mg/l, the increase in leaf mass and total plant mass was 51.4% and 36.2%, respectively. The hormone of 100 mg/l IBA performed an advancement of 37.5% in rooting, 42% in shoot length and 37.5% in seedling survival.

Conclusion In total, it can be accepted that although hormones promoted rooting as well as early growth of seedling in Tilia; however, the 100mg/l IBA can be favored as a superior choice for seedling production of this species.

Keywords Cutting; Indole-Butyric Acid; Leaf Mass; Linden; Survival

CITATION LINKS

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Introduction

In relation with the propagation of forest species, in most researches the use of sexual propagation (with seed) is considered, this is while that propagation with seed has a particular problem. Of course, in some fast-growing species, such as willows and poplars, due to the simplicity of rooting, propagation and mast seedling production are commonly done through non-sexual reproduction (with cuttings) [1, 2].

Lindens (Tilia spp.) are tree species that due to the difficulty of seed dormancy breaking as well as the lack of seedling production efficiency in the nursery, their growth through cuttings is also considered so that in most researches for rooting and propagation with different species cuttings are used by hormones. As in the cuttings of *T. argentea,* Naphthalene Acetic Acid (NAA) and Indole-Butyric Acid (IBA) increased its rooting, but etiolation (Plant growth, or part of it in the shade) caused the largeness of branches, undeveloped and necrosis leaves. IBA hormone (For 5 seconds at a concentration of 2500mg/l) increased rooting from 5 to 75%, however, the seedlings in NAA treatment had no rooting [3].

The rooting results of the semi-woody cuttings *T. cordata*, *T. platyphyllos* and *T. tomentosa* affected by IBA, IAA and NAA were evaluated after three months [4]. In *T. cordata* the most rooting (69%) and the highest coarse root number (17) were affected by IBA 50mg/l, and the longest root (4.8mm) did by IAA 150 mg/l. In *T. platyphyllos,* the most rooting (79.8%) and the longest root (4.8mm) allocated to IBA 40mg/l and the highest root number (21.45) to a concentration of 50mg/l IBA. In *T. tomentosa*, the highest root percentage (94.2%) was treated by NAA 40 mg/l, the highest root number (29.6%) did by IBA 50mg/l and the longest root (7.2mm) did by NAA 30mg/l. In general, high concentrations of IBA, IAA, and NAA caused defects of the root system and reduced the chance of establishment of the plant. In *T. amurensis*, the best rooting (63.3%) was obtained by using 3-4-year-old semi-wood cuttings (Prepared in April), 16 hours immersed in IAA hormone (Concentration of 0.0001mg/l) grown in a sandy bed (Mixed with river sand, perlite and vermiculite) [5]. In some researches on linden's cuttings, the best treatment was to improve the rooting rate of IBA. In research on cuttings of several species of lindens, using a 10000mg/l IBA in *T. tomentosa* caused 31.8% rooting, 5.8 root number and 26mm root length, and in *T. rubra*, 34.6% rooting, 3/3 root number and 30mm root length. In *T. cordata*, 60.9% rooting and 32mm root length and In *T. platyphyllos*, 27.7% rooting, 4.7 root number and 31mm root length [6]. In *T. mandshurica*, the concentration of 2000mg/l increased rooting to more than 80% [7] and in *T. mongolica*, the concentration of 500mg/l resulted in an increase of 82% rooting and a decrease in root period (5 days less than control) [8]. In *T. miqueliana*, using a concentration of 5000mg/l, rooting (43%), the semi-lignin cutting was better than woody cutting [9]. In woody cutting of *T. miqueliana*, the concentration of 1000mg/l produced the highest rooting (70%) and the concentration of 1500mg/l the best root quality (10 numbers, 2.5cm length, and 2.4mm thickness) [10]. In non-wood cuttings of *T. amurensis*, the concentration of 100mg/l (8 hours) created the highest rooting (63%) [11].

Due to the fact that in the north of Iran, the propagation of linden's seeds in nurseries is difficult [12, 13], the production of its seedlings through cutting requires comprehensive research. So far there has not been any research on propagation of *Tilia rubra* subsp. *caucasica* form *angulata* (Rupr.) V. Engler in the world. Thus, to resolve part of this problem, the study on rooting and seedling production through the cuttings can be imperative. In fact, in this study, for the first time, the rooting of cuttings and the performance of early growth of seedlings of this species using hormones of IAA, NAA and IBA at different concentrations are investigated. It is hypothesized that the hormones with medium concentration improve the rooting and survival in seedlings of *T. rubra*.

Materials and Methods

Cuttings of *Tilia rubra* subsp. *caucasica* form *angulata* Rupr. [14] with length of 20cm [15], and the diameter of 0.5cm were prepared from annual seedlings in Amol forest nursery. Experiment was carried out in randomized complete blocks with IBA hormones, IAA NAA with concentrations 0 (Control), 50, 100 and 200mg/l for 8 hours) [11] in three replicates. The examination was performed at greenhouse with a temperature of 25°C [16] and a fogging system [17] (With a stable moisture content of up to 70%) in the city of Nashtarood (Tonekabon

County). To prepare the plant bedding of cuttings in plastic pots [18] (15×15×30cm), the sand was first washed with fresh water and disinfected with a fungicide of copper oxychloride [19] 35% (2mg/l). Then, the cuttings were disinfected for 10min. in a copper oxychloride (2mg/l) fungicide solution and dried for 30min. in free air. The cuttings were planted in the bed in late March, and after 20 days the new emerged plants disinfected with copper oxychloride fungicide (copper oxychloride, 2g/l).

Measurements: After 115 days of the experiment, shoot length, diameter, root length, root number, rooting percentage, survival, root dry mass, leaf dry mass, and the total plant mass were measured. The rooting percentage was calculated by dividing the number of cuttings rooted in each treatment to total number of cuttings in that treatment multiple 100. For determination of dry mass, a seedling was selected from each replicate. The root and leaf as well as the whole seedlings were dried in an oven for 24 hours at 70°C and then weighed [1].

Data analysis: Statistical analysis was conducted by SPSS 22 software. First, the normality of data was accomplished by Kolmogrov-Smirnov's test, and homogeneity of variance by Levene' test. Then, one-way ANOVA was used to determine the difference between the data and Duncan's test was used to compare the means. Drawing of diagrams was performed by Excel software.

Findings

The results of the study showed that the effect of treatments on all variables was significant (Diagram 1). The rooting percentage increased about 37.5% in cuttings imbibed with 100mg/l IBA in comparison with control. In cuttings imbibed with 200mg/l IAA root number and root length in the order showed an increase of 28.6% and 22.3% compared to control. The highest root mass was observed in IBA 200mg/l, which was about 2.5 times greater than that in control. In high concentrations of IBA (200mg/l), the increase in leaf mass and total plant mass was 51.4% and 36.2%, respectively. Hormone of 100mg/l IBA appeared a promotion of 37.5% in rooting, 42% in shoot length and 37.5% in seedling survival. In total, it can be accepted that hormones promoted rooting as well as seedling growth; nevertheless, the non-imbibed cuttings were able to play a rather good role for rooting and seedling production, too (Diagram 1).

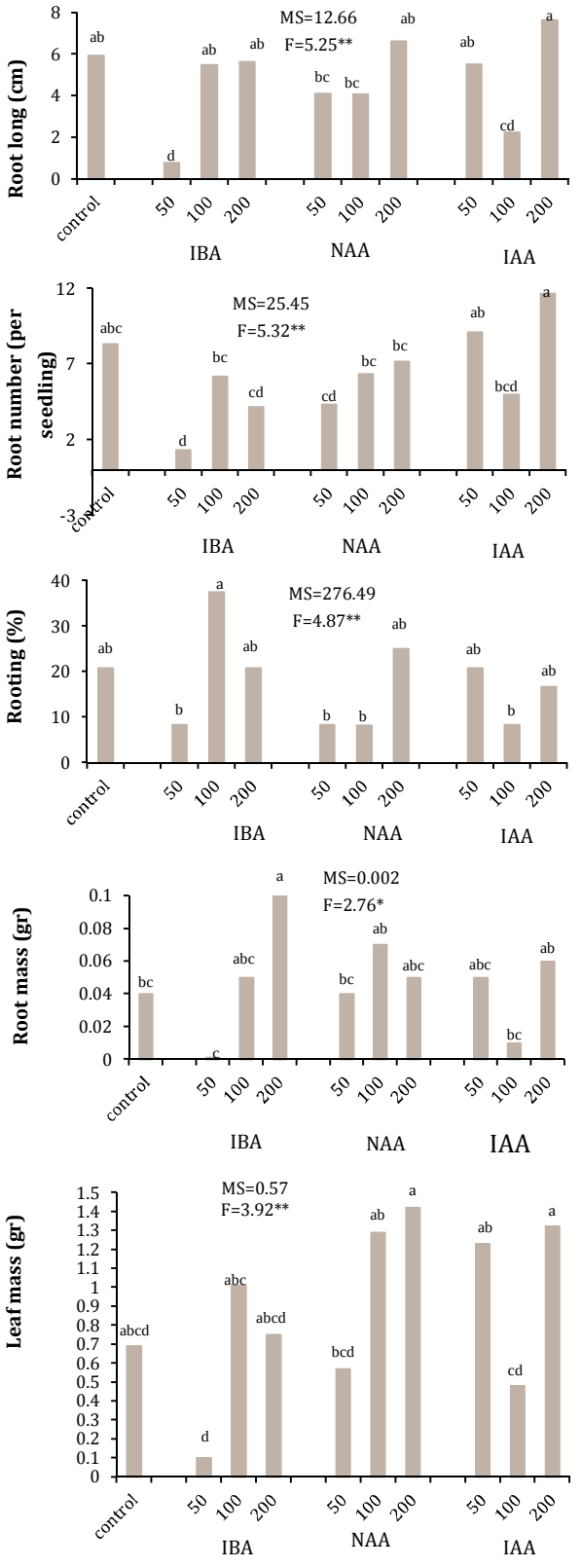

Diagram 1) Mean comparison of parameters as affected by treatments in *Tilia rubra* subsp. *caucasica* form *angulata* Rupr. (Unit of hormones in mg/l); * and ** significant difference among the means at the level of 0.05 and 0.01, respectively.

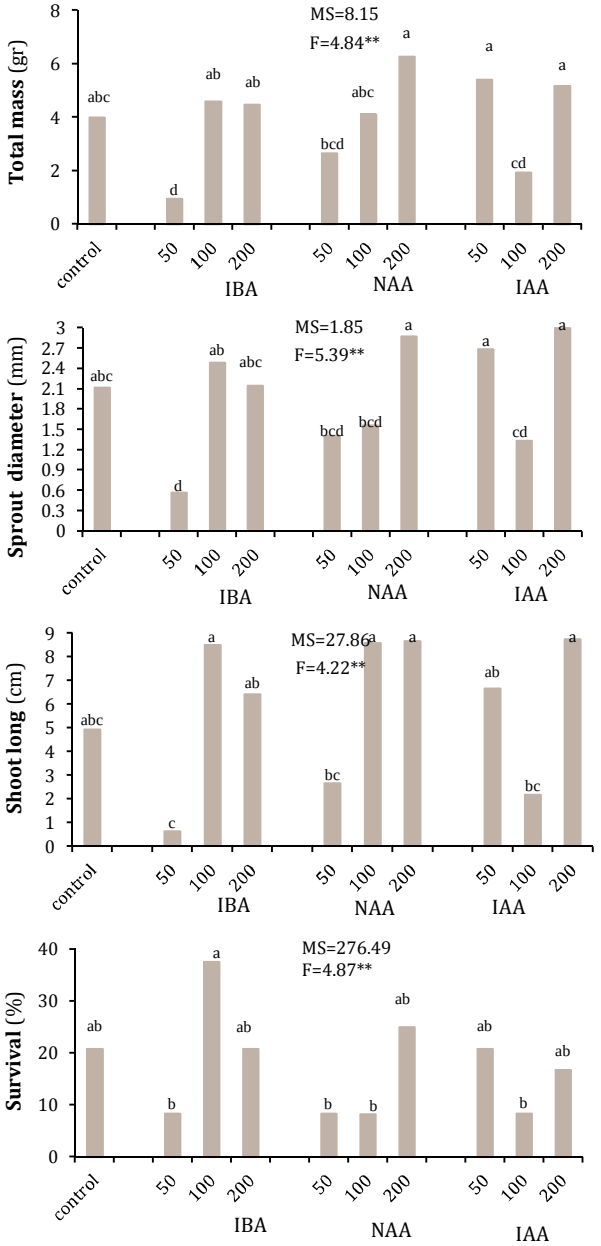

Diagram 1- Continued) Mean comparison of parameters as affected by treatments in *Tilia rubra* subsp. *caucasica* form *angulata* Rupr. (Unit of hormones in mg/l); * and ** significant difference among the means at the level of 0.05 and 0.01, respectively.

Discussion

The growth regulating agents improve plant growth, and auxins such as IBA, IAA and NAA are the most important compounds that influence root growth. In recent years influence of hormones on the improvement of rooting and seedling performance has been reported by some researchers. Among the investigations carried out on cuttings of and particularly forest trees it can be referred to *Populus caspica* [20], *Conocarpus erectus* [21], *Callistemon viminalis* [22, 23], *Cerasus mahaleb* [24], *olea europaea* [25], *Salix nigra* [26], *Ficus* spp. [27]. In most of above researches, following the hormone application, the parameters measured have been often improved. Some findings have been also published on lindens (*Tilia* spp.). According to literature, different species of lindens root hardly and auxins help the rooting ability [4, 6].

In the present study, among the hormones and the concentrations used on the *Tilia rubra* subsp. *caucasica* form *angulate*, IBA (100mg/l) helped an increase by about two times in rooting. Similarly, in a research on cuttings of *T. rubra* by applying IBA, IAA and NAA hormones, although the type of hormone was effective on root development the cuttings treated with IBA (100mg/l) produced the highest rooting percentage (33.33%) [6]. Rooting under the influence of IBA in cutting of *T. amurensis* was 63.1% [11] and in the cutting of *T. argentea* was 75% [3]. In most reports, IBA provided more rooting in cuttings than other hormones, so that in cuttings of *T. mandshurica* [7], *T. miqueliana* [9] and *T. mongolica* [8], rooting was 82%, 80% and 43%, respectively. It can be stated that like the reported results [4] on vegetative propagation in semi-hardwoods of *T. cordata, T. platyphyllos* and *T. tomentosa*, in our findings high doses of IBA, IAA and NAA provided weak, brittle root system, which reduces the chance of successful adaptation in replanting.

In the present study, the concentration of 200mg/l IAA with root length of 6.7cm was approximately three times greater than that in control. This increase is similar to the results obtained on *Tilia* sp. [4], and also more than the results obtained for the semi-lignin and woody cuttings of *T. miqueliana* [9]. Of course, in the present research, IBA did not improve root length compared to control, this is while that the positive effect of this hormone (With the concentration of 10000mg/l) on *T. cordata* and *T. platyphyllos* was reported [6].

The maximum root number in the present research (11.67) belonged to the cuttings applied to the IAA (200mg/l), with an increase of about 28.6% compared to the control. Root number of produced from the cuttings was more than those of the semi-lignin (2.6) and woody cuttings (9.1) of *T. miqueliana* [9]. Of course, in some similar studies, including woody cuttings of *T. platyphyllos*, the IBA produced a considerable root number (31) [6]. In another study, the number of roots affected by this hormone was 21 [4].

In the present study, the greatest root mass belonged to the cuttings applied to IBA (200mg/l), 2.5 times greater than that of control. The biggest leaf mass (42.1gr) was affected by NAA (200mg/l) and the maximum total mass appeared with NAA and IAA (High concentrations). In high concentrations of IBA (200mg/l), root mass was about 2.5 times larger than that in control, besides that the increase in leaf mass and total plant mass over the control was near two times.

The shoot length at a concentration of 200mg/l NAA and IAA improved about 43%, respectively. The concentration of 100mg/l IBA produced an increase of 37.5% in rooting, 42% in shoot length and 37.5% in survival compared to control.

Conclusion

From the findings of the present study, it can be stated that the used hormones improved rooting and increased seedling production. However, the concentration of 100 mg/l IBA produced the favorable results, especially in terms of rooting, survival, and shoot length of seedlings in the greenhouse (with fogging conditions) which can be considered as a best for seedling production of Tilia. It is also may be mentioned that with considering the planting of cuttings in the winter and their rather satisfactory rooting in the spring, with preparation and planting of winter woody cuttings of Tilia, proper emergence and growth of the roots and the effective efficiency of seedlings production is expectable. In order to the farther enhancement of the measured parameters, diversity in doses of hormones can be examined.

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