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Investigating the variation of diameter and height of *Populus* sp. clone I-214 under various fertilization treatments in northern Greece – a case study

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Abstract

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The study of young poplar tree characteristics like initial diameter and height as well as mineral fertilization that promotes the rapid growth of trees for biomass production was the aim of this research. In two forest nurseries of northern Greece, the growth of *Populus* sp. clone I-214 under five fertilization treatments per dosage was studied, i.e. (*i*) 120 kg $(NH_4)_2SO_4 \cdot ha^{-1}$, (*ii*) 200 kg $(NH_4)_2SO_4 \cdot ha^{-1}$, (*iii*) 150 kg $(NH_4)_2SO_4 \cdot ha^{-1}$ and 300 kg mixed fertilizer 11-15-15 (i.e. 11 kg N, 15 kg P_2O_5 and 15 kg K_2O per 100 kg of fertilizer) per ha, (*iv*) 150 kg $(NH_4)_2SO_4 \cdot ha^{-1}$, 200 kg mixed fertilizer 16-20-0 (i.e. 16 kg N and 20 kg P_2O_5 per 100 kg of fertilizer) per ha and 100 kg $K_2SO_4 \cdot ha^{-1}$, (*v*) no fertilization. Results showed that initial diameter was the variable that had the greatest effect on growth, while the fertilization with 150 kg $(NH_4)_2SO_4 \cdot ha^{-1}$ and 300 kg mixed fertilizer 11-15-15 per ha improved significantly diameter growth. Nevertheless, no specific pattern was found statistically for the best fertilization treatment concerning the height growth of young poplar trees. These findings could be taken into account for the best management practices for rapid poplar tree growth and biomass production under similar edaphoclimatic Mediterranean conditions.

Keywords: biomass production; Entisols; fertilizers; general linear model multivariate procedure; young poplar trees

The saltation in the creation of various tachyauxetic poplar stands managed with small rotating intervals, combined with the use of poplar wood for the production of boxes, paper, sawn wood, plywood, chipboards but also as biomass for the production of bioenergy as an alternative means for the reduction of transmissions generated by the greenhouse effect (Cañellas et al. 2012), resulted in the cultivation of large areas with poplar trees, particularly in the Mediterranean countries (Fiedler et al. 1973).

The poplar (*Populus* sp.) tree is used on a large scale because it is tachyauxetic, it proliferates easily with asexual reproduction, has been improved genetically, exhibits plenty of adjustability to various soils and climates. Additionally, poplar wood is suitable for the production of various commodities, while being a lucrative alternative culture for redundant agricultural lands (Kostakis 2001; Dickmann 2006). A contributing factor for that last use was the tendency to promote agricultural policy combined with the use of agricultural or forest areas of small productivity, such as the river alluviums. In those kinds of forestry, an intensive culture with short rotations ranging from two to ten years, according to the intended use of the produced biomass, is used (Makeschin 1999; Weih 2004).

There is an increasing interest in using poplar plantations as sources of bioenergy in many countries (Hjelm et al. 2015). With the use of tachyauxetic poplar species on agricultural lands, a contribution to the protection of the environment is performed with a simultaneous and constant supply of bioenergy (Gurgel 2011). Nowadays the bioenergy deriving from the biomass of poplars, willows and eucalyptuses is one of the best options for rapid production of alternative forms of energy (Arias Navarro 2011). The density of plantation plays an important role in the production of large amounts of biomass (Cañellas et al. 2012). A range of density values, instead of a specific density value, is used for achieving the optimum biomass production and is depended on the quality of the planted material produced in the nursery garden, the kind of cultivating methods and on the ecological factors (Bockheim et al. 1986; Willebrand, Verwijst 1993; Klašnja et al. 2008).

The poplar tree was used intensively in Greece during the early 1950s, when the first tachyauxetic cv. I-214, cv. I-262, cv. I-455 and cv. I-154 stands, of Italian origin, were introduced. Those are the stands of the hybrid between American *Populus deltoides* Marshall and European *Populus nigra* Linnaeus, and its scientific name is *Populus* × *euramericana* (Dode) Guinier.

Populus spp. are known for their high demands on nutritive elements, moisture, light, ventilation and also for their small tolerance to severe alkaline conditions (Berthelot et al. 2000). According to Alexandris (1971) the best growth of poplar trees in the Mediterranean countries compared to the mid-European ones is justified by their high demands on light and higher temperature. In Greece, poplars (black or hybrids) are widely spaced on arable lands with fertile soils and are sometimes irrigated with water from artificial channels coming from a local river. However, the most common practice is the establishment of poplars along watercourses or around arable fields, cultivated with vegetables or other crops resulting in traditional silvoarable systems. Poplars are used for timber production and they also serve as boundary markers and windbreaks (Papanastasis et al. 2009; Mantzanas et al. 2015).

For the growth of poplar plantlets in nurseries, favourable moisture conditions and good nutrient status of soil are needed, expressed by the pH value and the supply of nutritive elements to the soil (Vukorep 1970). Fertilization is a common practice for forest nurseries and for tachyauxetic species plantations. N and P deficiencies are observed many times in nurseries and plantations, as well as deficiencies of S, K and Mg to a lesser extent (Cole et al. 1990). The variation in the percentage of nutritive elements absorbed after the application of fertilizers depends on how the added nutritive element was sufficient for plant growth, on the amount and frequency of fertilization and on the amount of fertilizer lost due to leaching (Bockheim et al. 1986). The fertilization programs should be designed in such a way as to achieve maximum absorption of the elements contained in fertilizers and minimize losses due to leaching (Nambiar, Bowen 1986).

The purpose of this research is the study of the growth of I-214 poplar stands under various fertilization treatments in the nurseries of Thessaloniki's Forest Service (Northern Greece). Taking into account the initial diameter and height of young poplar trees as well as the application of four fertilization treatments, the goal was to find out the best options for rapid biomass production under the edaphoclimatic conditions of similar areas.

MATERIAL AND METHODS

Experimental sites and treatments. The fertilization experiment was conducted in two forest nurseries (Chalkidona and Lagadas areas, respectively) of the Department of Reforestation of Central Macedonia (Northern Greece).

The Chalkidona forest nursery is located about 30 km northwest of the city of Thessaloniki and close to the Axios River. The soil is of alluvial river origin and is classified as Entisol (Stefanou, Papazafeiriou 2014). The Lagadas forest nursery is located about 15 km northeast of Thessaloniki city and its soil is also classified as Entisol. The climate of the region is of Mediterranean type.

The experimental design included one-year saplings and the following fertilization treatments:

(*i*) 120 kg $(NH_4)_2SO_4$ (21-0-0, i.e. 21 kg N per 100 kg of fertilizer) per ha per fertilization, a total of 8 fertilizations per 15 days;

(*ii*) 200 kg $(NH_4)_2SO_4$ (21-0-0) per ha per fertilization, a total of 8 fertilizations per 15 days;

(*iii*) 150 kg $(NH_4)_2SO_4$ (21-0-0) per ha and 300 kg mixed fertilizer 11-15-15 (i.e. 11 kg N, 15 kg P_2O_5

and 15 kg K_2O per 100 kg of fertilizer) per ha per fertilization, a total of 8 fertilizations per 15 days;

(*iv*) 150 kg $(NH_4)_2SO_4$ (21-0-0) per ha, 200 kg mixed fertilizer 16-20-0 (i.e. 16 kg N and 20 kg P_2O_5 per 100 kg of fertilizer) per ha and 100 kg K_2SO_4 (0-0-50, i.e. 50 kg K_2O per 100 kg of fertilizer) per ha per fertilization, a total of 8 fertilizations per 15 days;

(*v*) No fertilization (control).

The first fertilization was applied on June 1st and the last on September 15th of 2013. The total amount (8 fertilizations) of the basic nutritive elements in kg·ha^{-1} administered to the saplings per treatment was as follows: (*i*) N: 201.6, (*ii*) N: 336.0, (*iii*) N: 258.0, P_2O_5: 180.0, K_2O: 180.0, (*iv*) N: 254.0, P_2O_5: 160.0, K_2O: 204.0, (*v*) N: 0, P_2O_5: 0, K_2O: 0.

Each treatment was applied onto a surface area of 55 m^2 in a plot of 7 rows with 20 saplings each, amounting to 140. During the entire germination period, the herbaceous plants between the rows were removed because the poplar growth is significantly affected by their presence (Cañellas et al. 2012). Growth measurements were performed at 18 saplings in the three middle rows of each treatment; 54 saplings in total. These measurements included the diameter at the 5 cm mark above the main bud, as well as the sapling height above that bud. The first (initial) measurement was performed before the first fertilization and the second one (final) at the end of the germination period and after the fall of the leaves. Each treatment was repeated three times in each nursery. A total of 5 treatments × 3 replications × 2 nurseries = 30 plots was set up.

Soil analysis. Soil sampling took place before fertilization in both experimental sites. Soil samples were collected from three depths (0–15, 15–30 and 30–90 cm) a few days before the first fertilization. All soil samples were air dried and sieved through 2-mm mesh screens. Particle size distribution of mineral soil was determined according to Bouyoucos (1962) and pH was determined in a soil and water suspension (1:1, by weight) using a pH meter (Crison, Spain). Electrical conductivity was measured in a saturated extract (Rhoades 1996) and free $CaCO_3$ using a Scheibler calcimeter (Neubert-Glas, Germany). Soil organic matter was determined by means of wet oxidation (Nelson, Sommers 1982). Organic N was determined by the Kjeldahl method (Stevenson 1982). Available P was extracted with 0.5N $NaHCO_3$ pH 8.5 and was measured spectrophotometrically (Olsen, Sommers 1982). Exchangeable Ca, Mg, K and Na were extracted with CH_3COONH_4 pH 7 (Grant 1982) and they were measured by atomic absorption spectrophotometry.

Statistical analysis. The general linear model (GLM) multivariate procedure of IBM SPSS software (Version 20, 2012) (IBM 2012) was used. The GLM multivariate procedure allows modelling the values of multiple dependent scale variables based on their relationships to categorical and scale predictors. It is based on the general linear model, in which factors and covariates are assumed to have linear relationships to the dependent variables.

Two dependent variables were included in the analysis: final diameter (FD, mm) and final height (FH, cm). Two categorical variables, "treatments" per fertilization described above (*i*–*v*) and "area" (1: Chalkidona, 2: Lagadas), were used as factors in the model. Each level of the factor can have a different linear effect on the value of dependent variables.

Two scale predictors, initial diameter (ID, mm) and initial height (IH, cm), were selected as covariates in the model. Within combinations of the factor levels, values of covariates are assumed to be linearly correlated with the values of dependent variables. Interactions were also included in the GLM multivariate procedure.

RESULTS

The results of soil analyses before fertilization in both experimental sites are presented in Table 1.

Soils in both sites are characterized as loamy sands and are permeable for water. They are al-

Table 1. Soil analysis before fertilization in the two forest nurseries

Area	Depth (cm)	Texture	pH	EC (dS·m^{-1})	OM (%)	$CaCO_3$ (%)	N (%)	P (mg·kg^{-1})	K	Na	Ca	Mg
									(cmol·kg^{-1})			
Chalkidona	0–15	LS	7.62	0.351	2.69	2.93	0.158	28.0	1.45	1.12	14.83	4.61
	15–30	LS	7.90	0.498	1.64	5.45	0.145	17.5	1.21	1.24	12.23	4.42
	30–90	LS	8.01	0.648	0.71	3.66	0.091	11.3	0.54	1.20	7.92	3.54
Lagadas	0–15	LS	7.65	0.745	2.93	5.06	0.163	22.8	1.02	1.89	12.70	6.39
	15–30	LS	7.70	0.464	1.84	5.08	0.132	19.3	0.93	1.15	7.62	6.13
	30–90	SL	7.81	0.294	1.16	6.19	0.079	16.7	0.77	1.09	3.93	4.34

LS – loamy sand, SL – sandy loam, EC – electrical conductivity, OM – organic matter

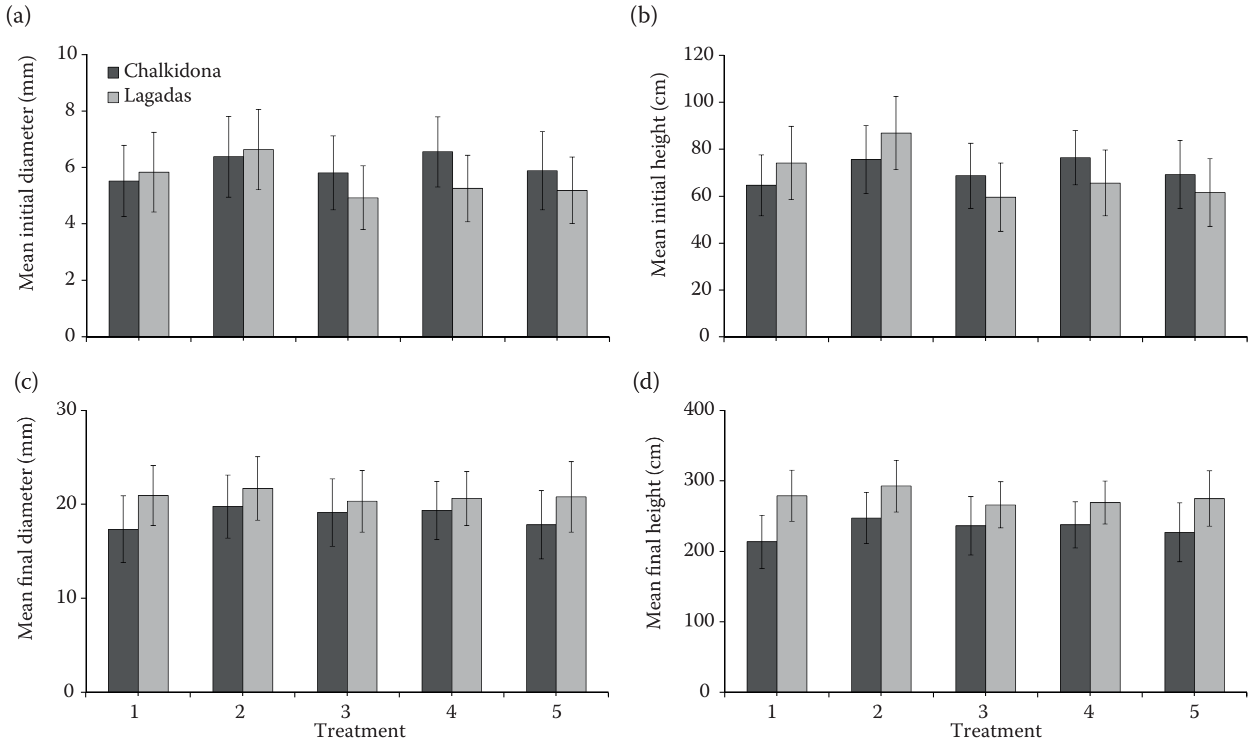

Fig. 1. Means ($N = 54 \times 3$ replications) and standard deviations of initial diameter (a), initial height (b), final diameter (c), final height (d) for the five treatments in the two experimental sites

kaline, not saline, and organic matter content although reduced by depth is on satisfactory levels in the rhizosphere. $CaCO_3$ content ranges from 2.93 to 6.19%. There is no macro-nutrient deficiency; N, P and K levels are characterized as adequate.

Means and standard deviations of ID, IH, FD and FH are presented in Figs 1a–d.

Table 2 displays four tests of significance for each model effect, i.e. Pillai's trace, Wilks' lambda, Hotelling's trace and Roy's largest root (Rencher, Christensen 2012). Pillai's trace is a positive-valued statistic in which increasing values indicate effects that contribute more to the model. Wilks' lambda is a positive-valued statistic that ranges from 0 to 1. Decreasing values of the statistic indicate effects that contribute more to the model. Hotelling's trace is a positive-valued statistic for which increasing values indicate effects that contribute more to the model. Hotelling's trace is always larger than Pillai's trace, but in case that these two statistics are nearly equal, this indicates that the effect does not probably contribute much to the model. The results of the present study show that these values are far from equal, indicating a significant contribution of variables to the model. Roy's largest root is a positive-valued statistic for which increasing values indicate effects that contribute more to the model. Roy's largest root is always less than or equal to Hotelling's trace. When these two statistics are equal, the effect is predominantly associated with just one of the dependent variables, there is a strong correlation between the dependent variables, or the effect does not contribute much to the model. In our study, this phenomenon can be attributed to the fact that the dependent variables (diameter and height) are strongly correlated. From all four tests, ID seems to be the variable that has the greatest effect on growth.

Each multivariate statistic is transformed into a test statistic with an approximate or exact *F* distribution. The significance values of the main effects, ID, IH, treatment and area, are less than 0.05, indicating that the effects contribute to the model. Moreover, the "treatment × area" interaction effect contributes to the model. The partial eta squared statistic reports the practical significance of each term. Larger values of partial eta squared indicate a greater amount of variation accounted for by the model effect, to a maximum of 1. Since partial eta squared is very small for the "treatment × area" interaction effect, it does not contribute very much to the model. By comparison, partial eta squared for the initial diameter is quite large, which is to be expected.

Table 3 displays results for each contrast. Simple contrasts using the last level of "treatment" (*v*: no fertilization) as the reference category were specified. The contrast estimates show that on average the trees given treatment 1 had 0.596 mm smaller

Table 2. Multivariate tests (design: intercept + initial diameter + initial height + area + treatment + area × treatment)

Effect		Value	*F*-value	Hypothesis *df*	Error *df*	Partial eta squared
Intercept	Pillai's trace	0.500	770.795[b]	2.000	1,541.000	0.500
	Wilks' lambda	0.500	770.795[b]	2.000	1,541.000	0.500
	Hotelling's trace	1.000	770.795[b]	2.000	1,541.000	0.500
	Roy's largest root	1.000	770.795[b]	2.000	1,541.000	0.500
ID	Pillai's trace	0.092	77.647[b]	2.000	1,541.000	0.092
	Wilks' lambda	0.908	77.647[b]	2.000	1,541.000	0.092
	Hotelling's trace	0.101	77.647[b]	2.000	1,541.000	0.092
	Roy's largest root	0.101	77.647[b]	2.000	1,541.000	0.092
IH	Pillai's trace	0.043	34.895[b]	2.000	1,541.000	0.043
	Wilks' lambda	0.957	34.895[b]	2.000	1,541.000	0.043
	Hotelling's trace	0.045	34.895[b]	2.000	1,541.000	0.043
	Roy's largest root	0.045	34.895[b]	2.000	1,541.000	0.043
Area	Pillai's trace	0.416	549.610[b]	2.000	1,541.000	0.416
	Wilks' lambda	0.584	549.610[b]	2.000	1,541.000	0.416
	Hotelling's trace	0.713	549.610[b]	2.000	1,541.000	0.416
	Roy's largest root	0.713	549.610[b]	2.000	1,541.000	0.416
Treatment	Pillai's trace	0.069	13.688	8.000	3,084.000	0.034
	Wilks' lambda	0.932	13.734[b]	8.000	3,082.000	0.034
	Hotelling's trace	0.072	13.781	8.000	3,080.000	0.035
	Roy's largest root	0.053	20.458[c]	4.000	1,542.000	0.050
Area × treatment	Pillai's trace	0.065	12.993	8.000	3,084.000	0.033
	Wilks' lambda	0.935	13.097[b]	8.000	3,082.000	0.033
	Hotelling's trace	0.069	13.201	8.000	3,080.000	0.033
	Roy's largest root	0.059	22.703[c]	4.000	1,542.000	0.056

ID – initial diameter, IH – initial height, [b] exact statistic, [c] the statistic is an upper bound on *F* that yields a lower bound on the significance level, *df* – degree of freedom, significance = 0.000

diameter and 9.178 cm smaller height than the trees given treatment 5. In addition, only treatment 3 improved diameter and height growth, compared to treatment 5. However, since the significance value for "final height" is greater than 0.10, this difference may be entirely due to chance variation. Thus, the model suggests that treatment 3 should be used, if we are interested in diameter growth.

Table 3. Contrast results* (K matrix)

Treatment simple contrast[a]		Dependent variable	
		FD (mm)	FH (cm)
Level 1 vs. 5	contrast estimate	–0.596	–9.178
	SE	0.187	2.263
	significance	0.001	0.000
Level 2 vs. 5	contrast estimate	–0.790	–2.944
	SE	0.199	2.403
	significance	0.000	0.221
Level 3 vs. 5	contrast estimate	0.735	3.255
	SE	0.187	2.264
	significance	0.000	0.151
Level 4 vs. 5	contrast estimate	–0.124	–5.557
	SE	0.188	2.273
	significance	0.511	0.015

*hypothesized value, difference (estimate – hypothesized) and 95% confidence interval for difference are not presented in the table, [a] reference category = *v* (no fertilization), SE – standard error, FD – final diameter, FH – final height

The profile plots (Figs 2a, b) provide a visual representation of the model-estimated mean final diameter and final height for each of the five treatments. The profile plots show that Lagadas area tends to have poplar trees of larger diameters and heights in all treatments. Additionally, treatment 3 seems to have the most profound effect on diameter growth especially in Chalkidona area. Across treatments, there is a different pattern between areas, even though diameter and height follow the same pattern.

DISCUSSION

One important goal of nurseries today is maintaining an adequate level of soil fertility to produce high quality seedlings. The complex relationship between fertilization, site conditions, and other cultural practices makes fertilization decisions some of the most

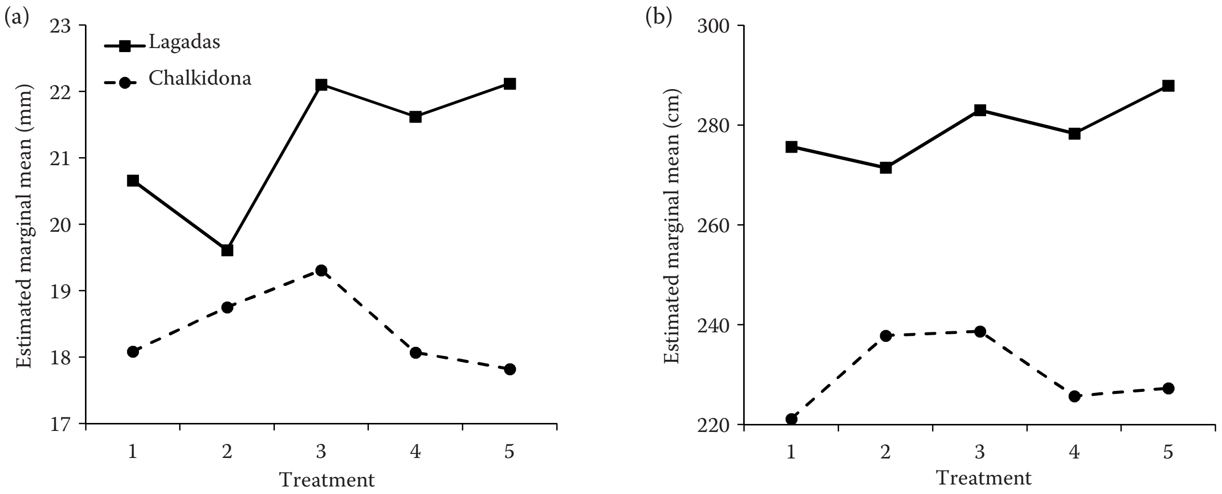

Fig. 2. Estimated marginal means of final diameter (a), final height (b) for the five treatments in the two experimental sites

difficult in nursery management (Duryea 1984). Research is mainly focused on studying the influence of various fertilization treatments on growth parameters of poplar species in nurseries. However, inadequate data exist on the growth of young poplar trees (clone I-214) under Mediterranean conditions (semi-arid conditions and alkaline soils) and, specifically, on the possible influence of their initial diameter and height on their growth. There is also a lack of references to the influence of fertilization using inorganic fertilizers and to the short-term responses of growth, i.e. increase of diameter and height of young poplar trees (clone I-214). According to Nakos (1979), experiments with four poplar clones and various chemical fertilizers in a nursery in southern Greece showed that at the end of the first growth period, of the fertilizer nutrients N, P, K and Mg only N improved heights of all clones significantly and especially of the clone I-214. The addition of 100 or 200 kg of P fertilizer per hectare had a minimal or negative effect on height increment of all clones. Ammonium sulphate, ammonium nitrate and potassium nitrate were found equally effective in improving the height growth of clone I-214, but ammonium nitrate was the N fertilizer of choice by its higher N content and relatively lower price. The addition of ammonium nitrate at 200 kg N·ha^{-1}, in two or three equal dosages during the first growth period (June-July), gave the maximum height increment for two consecutive years. In a study of Lazdiņa et al. (2014) in Latvia, among various treatments the mineral fertilizer NPK (12:5:14) was the most effective on the first three-year development of ALASIA poplar clones AF2, AF6, AF7, AF8. Also, Singh (2001) reported that the application of NPK fertilizer ($N_{100}P_{50}K_{25}$, $N_{200}P_{100}K_{50}$ and $N_{400}P_{200}K_{100}$ in kg·ha^{-1}) significantly increased the collar diameter, height, shoot biomass and nutrient uptake by *P. deltoides* seedlings in nursery. Our results are in accordance with Lazdiņa et al. (2014) as well as Singh (2001) in that N, P and K had a positive effect on the one-year sapling growth as opposed to Nakos (1979), who reported that only N affected their growth. A combination of ammonium sulphate (21-0-0), which is the only N fertilizer of choice and mixed fertilizer 11-15-15, in eight dosages, was the most effective in improving the diameter growth of clone I-214 and especially in Chalkidona area during the fertilization period (June through mid-September). Byrd (2013) measured *Populus balsamifera* Linnaeus growth and yield rates in an established stand in Alaska under two-year rotation and fertilizer application of 112 kg N·ha^{-1}, 25 kg P·ha^{-1} and 60 kg K·ha^{-1} and demonstrated no effect on growth. On the other hand, our results showed that total fertilization with 258 kg N·ha^{-1}, 180 kg P_2O_5·ha^{-1} and 180 kg K_2O·ha^{-1} (treatment 3) offered the optimum conditions for the diameter growth of saplings, which was more evident in Chalkidona nursery, while fertilization with N, P and K (treatments 3 and 4) in Lagadas nursery seemed to have a greater effect on sapling height than just N fertilization (treatments 1 and 2). Generally, local edaphoclimatic conditions should be taken in account in order to have the optimum poplar biomass for bioenergy production.

CONCLUSIONS

The study of the growth of *Populus* sp. clone I-214 one-year saplings in two forest nurseries of northern Greece under various fertilization treatments showed that initial diameter was the parameter that had the greatest effect on growth, while fertilization treatment with 150 kg 21-0-0 per ha and 300 kg mixed fertilizer 11-15-15 per ha improved significantly the diameter growth in both study areas and more clearly in Chalkidona area. On the other hand, no specific pattern was found statistically for the best fertilization treatment concerning the height growth of young poplar trees. These results could be taken into account for the best management practices with regard to rapid poplar tree growth and biomass production under the edaphoclimatic Mediterranean conditions of the study areas or similar areas.

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