

Minimizing nutrient leaching and improving nutrient use efficiency of *Liriodendron tulipifera* and *Larix leptolepis* in a container nursery system

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Abstract Fertilization is essential to seedling production in nursery culture, but excessive fertilization can contaminate surface and ground water around the nursery. The optimal fertilization practice is that which maximizes seedling growth and minimizes nutrient loss. We tested three fertilization strategies: (1) constant fertilization (2) a three-stage rate, and (3) exponential fertilization on *Liriodendron tulipifera* and *Larix leptolepis* containerized seedlings. Growth performance, nutrient uptake, and nutrient loss in leaching were measured. Height, root collar diameter, and dry weight of both species were not significantly different among treatments even though the nutrient supply of the exponential treatment was half that of the constant and three-stage treatments. Generally, nutrient losses in leached solutions were higher in constant and three-stage than the exponential treatment. Nutrient use efficiency was calculated as the ratio of the nutrient content of the seedlings to the amount of nutrient applied to the containers. The nitrogen use efficiency in the constant, three-stage, and exponential treatments was 63, 61, and 85% for yellow poplar,

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respectively, and 35, 30, and 53% for larch. Similar results were obtained for phosphorus and potassium. Thus, the exponential treatment had the highest nutrient use efficiency as well as the least nutrient loss. Adjusting fertilization rates can reduce soil and water contamination around the nursery without compromising growth performance, which reduces both producer's investments and environmental impacts.

Keywords Biomass · Exponential fertilization · Leached solution · Nitrogen use efficiency · Phosphorus use efficiency · Potassium use efficiency

Introduction

Providing proper mineral nutrition is essential to seedling production in nursery culture, but excessive fertilization can contaminate surface and ground water around the nursery (Broschat 1995; Andersen and Hansen 2000). For example, nitrogen leaching in a greenhouse with a fixed overhead irrigation system was 50 kg ha^{-1} (Dumroese et al. 2006). In a Finnish container nursery, nitrogen leaching was $19\text{--}42 \text{ kg ha}^{-1} \text{ year}^{-1}$ and phosphorus leaching was $11\text{--}56 \text{ kg ha}^{-1} \text{ year}^{-1}$ (Juntunen et al. 2002). Because large volumes of water are discharged with nutrients from the nursery during production of container seedlings (Bumgarner et al. 2008), increased levels of nitrate and phosphate in surface water can cause serious problems in water quality. Improving fertilizer use efficiency and reducing nutrient discharge would reduce the environmental impacts of nursery operations.

In principle, the optimal application of nutrients is synchronized with seedling demand (Ingestad and Lund 1986; Timmer 1996). Three-stage fertilization may offer an improvement over a constant supply (Landis et al. 1989). Three-stage fertilization recognizes three growth stages during seedling development: the establishment phase, which is from emergence to the cotyledon stage; the rapid growth phase, when seedlings exponentially grow in height; and the hardening phase, when the seedlings set their bud and shoot growth ceases. Exponential fertilization uses a continuous increasing supply of nutrients, calculated based on an exponential accumulation of nutrients in the plants (Timmer 1996; Timmer and Armstrong 1987). By inducing luxury consumption of nutrients, seedlings should have greater nutrient reserves at the time of outplanting (Timmer 1996). Exponential fertilization has been studied in a variety of forest tree seedlings (Quoreshi and Timmer 2000; Salifu and Timmer 2003). Exponential fertilization of white pine (*Pinus monticola* Dougl. Ex D. Don) reduced fertilizer application by 45% while producing seedlings with similar morphological characteristics and nutrient status as those produced with conventional fertilization (Dumroese et al. 2005). To fully evaluate the efficiency of nutrient use requires measuring fertilizer losses as well as nutrient accumulation in the plants.

The purpose of this study was to measure fertilization effects on growth and nutrient use efficiency of *Liriodendron tulipifera* (yellow poplar) and *Larix leptolepis* (larch) in a container nursery system. We tested three fertilization strategies: (1) constant fertilization (2) a three-stage rate, and (3) exponential fertilization on yellow poplar and larch containerized seedlings, which are the most planted species in Korea (KFS 2009). We measured applied nutrients, leached nutrients, nutrients accumulated in the growth medium, and nutrient accumulation in the plants. Our goal was to find the fertilization practice that would maximize seedling growth and minimize nutrient loss.

Materials and methods

Study material

This study was conducted in a greenhouse at the Forest Practice Research Center, Korea Forest Research Institute, Kyeonggi-do, Korea (37° 45' N, 127° 10' E) from March to August, 2009. The containers for yellow poplar were 6.8 cm in diameter and 15 cm tall (400 ml). The containers for larch were 5.0 cm in diameter and 16 cm tall (240 ml) (Kukil Chem., Korea). Each container was filled with a mixture of equal volumes of peat moss, vermiculite, and perlite. Yellow poplar was germinated on the mixed growth medium and then transplanted to the containers. Larch seeds germinated in the containers and were thinned to one seedling per container. Until the fertilization treatment began, all containers were watered daily by overhead irrigation for 7 weeks. After that, the containers were manually irrigated with fertilizer solution three times a week, as described below. The air temperature ranged from 14.6°C to 29.0°C. The relative humidity ranged from 61 to 77%.

The seedlings were clustered in 48 trays for each species (40 cm × 32 cm for yellow poplar and 44 cm × 30 cm for larch) each holding 20 yellow poplar or 35 larch containers. Each cluster of 4 trays was randomly assigned to one of three fertilization treatments, described below. There were 4 replicates of each treatment, for a total of 16 trays in each treatment for each species. The 4 trays in a cluster were the experimental unit. The observational unit for growth measurements was one seedling randomly selected on a tray. The observational unit for nutrient solution losses was a set of 4 trays.

Fertilization treatment

Fertilization treatments were initiated on June 1st 2009 and continued for 11 weeks. All treatments were applied three times per week. We applied a water soluble fertilizer, MultiFeed19 (Haifa Chemicals, Israel), composed of 19N:19P₂O₅:19K₂O plus micronutrients. All fertilization treatments were manually sprayed as top dressing and seedlings were rinsed with water after each spray to avoid fertilizer burn. The volume of solution applied was 20 l for each set of 4 trays at each application.

The constant fertilization treatment was typical for containerized seedling production in Korea (KFRI 2009). In this treatment, yellow poplar received consistent applications of 25.6 mg tree⁻¹ and larch received 11.8 mg tree⁻¹, three times per week.

In the three-stage treatment, yellow poplar received 8.5 mg tree⁻¹ during the establishment stage (8 applications), 37.7 mg tree⁻¹ during the accelerated growth phase (17 applications), and 17.1 mg tree⁻¹ during the hardening phase (8 applications). In this treatment, larch received 3.9, 17.4, and 7.9 mg tree⁻¹ for the establishment stages, accelerated growth phase, and hardening phase, respectively.

In the exponential fertilization treatment, we obtained r , the relative addition rate, from the initial nutrient content of the plant, N_i , the expected final nutrient content of the plant ($N_f + N_i$), and the number of additions, t , assuming that all the N added (N_f) would be retained, in which case $N_f = N_i \times (e^{rt} - 1)$ (Dumroese et al. 2005). In this study, N_i and N_f were 6.8 and 74.8 mg tree⁻¹ for yellow poplar and 2.3 and 16.6 mg tree⁻¹ for larch, respectively, based on their N contents (unpublished data). Using $t = 33$, we obtained relative addition rates (r) of 0.0753 for yellow poplar and 0.0642 for larch. The total amount of fertilizer added was half as much as in the constant and three-stage rate fertilization treatments. Specifically, yellow poplar received 846 mg tree⁻¹ in both the constant and the three-stage rate fertilization treatments but only 423 mg tree⁻¹ in the

exponential fertilization treatment. Similarly, larch received 389 mg tree^{-1} in the constant and the three-stage rate fertilization treatments but only 195 mg tree^{-1} in the exponential fertilization treatment.

Growth measurement and tissue analyses

During the experiments, the height and root collar diameter (rcd) of 16 seedlings from each treatment (one from each tray) were measured about every 3 weeks. On August 28th 2009, these 16 seedlings were harvested and divided into shoots and roots; yellow poplar shoots were further divided into leaves and stems. Roots were washed with tap water. All components were dried to constant weight at 65°C . For larch, the shoots and roots were ground to pass a 1 mm screen. For yellow poplar, the stems and fine roots were ground, the leaves were subsampled and only the blades were ground, and the root collar was not ground. This procedure results in a bias towards higher nutrient contents, because petioles have lower nutrient concentrations than leaf blades (Auchmoody 1974) and fine roots have lower concentrations than coarse roots (Gordon and Jackson 2000).

Tissue samples were composited, four per set of trays, and analyzed for total N, P, and K. Nitrogen concentration was determined by the micro-Kjeldahl method with 1 g dry tissue (Bremner 1965). Phosphorus and K were extracted from 0.5 g of sample by wet oxidation with concentrated $\text{HNO}_3\text{--HClO}_4$ in a microwave digester. Phosphorus concentration was determined colorimetrically with vanadate-molybdate (Wilde et al. 1972) and K concentration was determined by atomic absorption spectrophotometry (AA280FS, Varian Inc., USA).

Solution collection and analyses

The nutrient solution applied was collected outside of the trays, under the trays, and in the containers. Solution sprayed outside of the trays was collected in a plastic box placed under four tray clusters of each species and the volume was measured after every fertilization treatment. The fraction of solution discharged was 87% for yellow poplar and 88% for larch.

Leachate from the containers was collected weekly using a plastic box 15 cm deep attached to the bottom of four tray clusters of each species for the whole study period. The volume of solution was measured and 50 ml of leachate was filtered with $0.45 \mu\text{m}$ membrane filter (Millipore Inc.) and then frozen until chemical analysis. Concentrations of NO_3 , NH_4 , PO_4 , and K in the leachate were determined by ion chromatography (IC25A, Dionex, US for cation and DE/S135, Sykam, Germany for anion concentrations) after dilution, as determined by monitoring the electrical conductivity (CM-60S, Toa Electronics Ltd., Japan).

The amount of solution added to the containers (not sprayed outside the trays or leached from the containers) was estimated from the change in tray weight before and about 30 min after irrigation (to allow for leaching). The amount of solution retained in the containers was the basis for the nutrient use efficiency calculation, as described below.

The amount of solution accounted for in the trays, leached from the containers, and sprayed outside of the trays was summed and compared to the amount applied (20 l for each tray cluster for each application). The amount of solution unaccounted for ranged from 1.6 to 3.6% of the amount applied, depending on the species and treatment.

Growth medium sampling and analysis

The growth medium was sampled before and after the experiment. We analyzed one composite pre-treatment sample. At the end of the experiment, we sampled the growth medium from 16 of the containers (one per tray) and composited them by tray cluster to make 4 replicates for each treatment and species combination. Organic matter was measured by dichromate oxidation and titration with ferrous ammonium sulfate (Walkley and Black 1934) and pH was determined from 10 g samples in a 5:1 deionized water: medium mix using a glass electrode (Orion Research Digital Ionanalyser Model 601A). Total N was analyzed in 1 g samples using micro-Kjeldahl methods (Bremner 1965). Extractable P was determined by the Lancaster soil extraction method (Cox 2001). Exchangeable K, Ca, Mg, and Na were extracted from 5 g samples with 50 ml of 1 N NH_4OAc and analyzed by atomic absorption spectroscopy (AA280FS, Varian Inc., USA).

Nutrient flux and data analysis

The amount of each nutrient taken up by plants (U_p) was calculated as the sum of the nutrient contents of shoots and roots. Leached nutrient (N_l) was calculated from the volume and concentration of leachate. The accumulation of nutrients in the containers (N_m) was calculated as the difference between the final and initial nutrient contents. Nutrient use efficiency was calculated as the ratio of uptake to the total nutrient applied; the total nutrient applied was estimated as the sum of U_p , N_l , and N_m . For larch, where the entire plant was used in nutrient analysis, this method agreed with the amount applied as calculated from the concentrations in the fertilization treatments (averaging 2% error), but in the case of yellow poplar, we found 11% more N and 20% more K in the plant, leachate, and medium than was calculated to have been applied, probably because of bias in the sampling of tissues for analysis, as described above.

We used repeated-measures ANOVA to test for differences in height and diameter growth among fertilization treatments, with measurement time as the repeated measure. Analysis of variance procedures with Tukey's honestly significant differences test was used to test the effect of fertilization treatment on biomass growth, properties of the growth medium, and plant nutrient concentrations.

Results and discussion

Seedling growth

Growth rates differed by fertilization treatment over time for yellow poplar (Fig. 1). During the early growth stage (until the second measurement), height and rcd growth were not significantly affected by fertilization treatments, but height growth from third to final measurement was 0.64 cm day^{-1} for the constant treatment, 0.56 cm day^{-1} for the three-stage treatment, and 0.70 cm day^{-1} for the exponential treatment (Fig. 1). As a result, the interaction of fertilization and time was significant for height ($P < 0.01$) and for rcd ($P = 0.03$) for yellow poplar. Height and rcd of yellow poplar were higher in the constant than in the exponential treatment at the third and fourth measurement. At the final measurement, rcd was significantly different among fertilization treatments ($P = 0.03$), but height was not ($P = 0.31$).

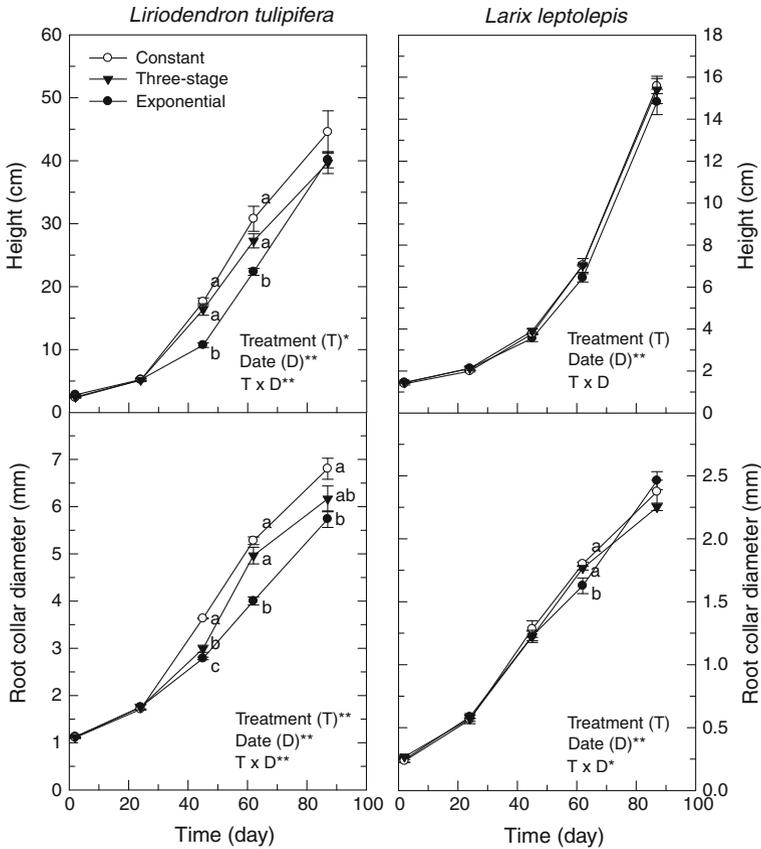


Fig. 1 Height and root collar diameter of yellow poplar (*Liriodendron tulipifera*) and larch (*Larix leptolepis*) in response to three types of solution irrigation for 11 weeks. Vertical bars represent one standard error of the mean ($n = 4$). Different letters show significant differences among fertilization treatments at $\alpha = 0.05$. Repeated-measures ANOVA identified the significance of fertilization treatment, time, and the interaction of fertilization treatment with time: * $P < 0.05$; ** $P < 0.01$

For larch, neither height ($P = 0.42$) nor rcd ($P = 0.58$) differed significantly with fertilization treatment (Fig. 1). Only at the fourth measurement was there a difference in rcd (the exponential treatment being lowest), resulting in a significant interaction of fertilization treatment and time ($P < 0.01$).

Total dry weight of yellow poplar in the constant treatment was higher than that in the three-stage treatment by 9%, but the difference was not statistically significant ($P = 0.53$) (Fig. 2). The total dry weight of larch was the lowest in the three-stage treatment ($P = 0.02$); the exponential treatment did not differ from the constant treatment (Fig. 2). The difference was greatest in aboveground weight, with seedlings in the exponential treatment having 23% more aboveground dry weight than in the three-stage treatment.

Although seedlings in the exponential treatment received half as much fertilizer as in the other treatments, their height, rcd, and dry weight were similar. Other studies have also found plant quality to be similar between conventionally grown and exponentially fertilized seedlings (Close et al. 2005; Qu et al. 2003; Timmer 1996). Outplanting performance was not tested in this study, but other studies have shown exponential fertilization to

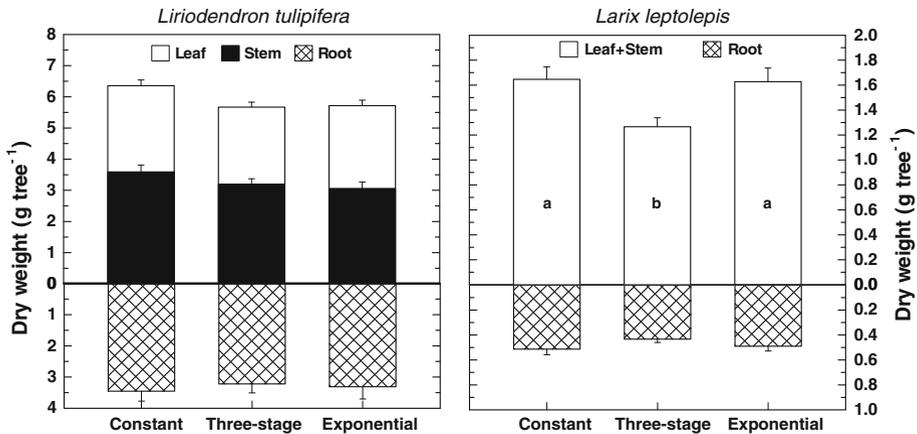


Fig. 2 Biomass of yellow poplar (*Liriodendron tulipifera*) and larch (*Larix leptolepis*) in response to three types of solution irrigation for 11 weeks. Vertical bars represent one standard error of the mean ($n = 4$). Different letters showed significant differences among fertilization treatments at $\alpha = 0.05$

Table 1 Nutrient concentrations in yellow poplar (*Liriodendron tulipifera*) and larch (*Larix leptolepis*) receiving three types of solution irrigation for 11 weeks: a constant rate, a three-stage rate, and exponential fertilization

Species	Tissue	Treatments	N (g kg ⁻¹)	P (g kg ⁻¹)	K (g kg ⁻¹)
<i>Liriodendron tulipifera</i>	Foliage	Constant	30.1 (0.7) ^{ab}	2.6 (0.2)	31.9 (1.3)
		Three-stage	29.4 (1.4) ^b	2.5 (0.1)	33.9 (0.8)
		Exponential	33.3 (0.8) ^a	2.6 (0.1)	35.2 (2.0)
	Stem	Constant	7.1 (0.3)	2.4 (0.1)	19.0 (1.8)
		Three-stage	7.9 (0.4)	2.6 (0.1)	20.8 (1.3)
		Exponential	8.4 (0.3)	2.4 (0.1)	20.6 (0.7)
	Root	Constant	23.9 (2.0)	4.8 (0.2)	33.6 (1.1) ^b
		Three-stage	27.9 (2.1)	5.2 (0.1)	42.5 (2.4) ^a
		Exponential	26.0 (1.8)	4.4 (0.5)	39.1 (1.1) ^a
<i>Larix leptolepis</i>	Foliage + stem	Constant	20.7 (1.2)	5.7 (0.4)	15.5 (0.5) ^b
		Three-stage	20.2 (1.0)	5.4 (0.3)	16.9 (0.8) ^{ab}
		Exponential	20.7 (0.5)	5.9 (0.2)	18.2 (0.6) ^a
	Root	Constant	17.5 (0.3)	6.8 (0.4)	22.0 (4.4)
		Three-stage	16.0 (1.1)	6.2 (0.6)	20.9 (2.6)
		Exponential	18.9 (1.6)	6.7 (0.6)	21.0 (5.5)

Means within a column with the same letter or no letter are not significantly different at $\alpha = 0.05$. Standard errors are in parentheses ($n = 4$)

improve seedling height and rcd after outplanting (Salifu and Timmer 2003; Way et al. 2007) and competitive ability against herbaceous vegetations (Imo and Timmer 1999).

Nutrient loss, uptake, and efficiency

There were few significant differences in foliar, stem, or root nutrient concentrations as a result of fertilization treatments (Table 1). Foliar N concentrations of yellow poplar were

the highest in the exponential treatment and the lowest in the three-stage treatment ($P = 0.05$). There were no significant differences in P concentrations in any tissue type. Root K concentrations of yellow poplar were the lowest in the constant treatment ($P = 0.01$). For larch, aboveground K concentrations of larch were the lowest in the constant treatment and highest in the exponential treatment ($P = 0.05$).

Differences in the chemical properties of the growth medium after 11 weeks of treatment were also few (Table 2). The pH was low in the constant treatment for larch ($P < 0.01$). Exchangeable K^+ ($P < 0.01$) and Mg^{2+} ($P = 0.02$) were low in the constant treatment for poplar.

The concentrations of N (nitrate plus ammonium), P, and K in leached solutions were very different among fertilization treatments and they fluctuated during the experiment (Fig. 3). Generally, nutrient concentrations were 2–3 times higher in the constant and the three-stage treatment than the exponential treatment. It is not surprising that losses were lowest in the treatment that applied the least nutrients. These nutrient concentrations were multiplied by the volumes of solution leached to calculate nutrient losses from leaching during the experiment (Fig. 4).

Nitrogen accumulation in plants was similar among fertilization treatments in both species and N accumulation in the growth media was negligible (Fig. 4). However, the amounts of N leached from the containers were 2.4 times higher in the constant and the three-stage treatments than in the exponential treatment. As a result, N use efficiency was highest in the exponential treatment. Nitrogen use efficiency of yellow poplar was 63, 61, and 85% for the constant, three-stage, and exponential treatments, respectively. Nitrogen use efficiency of larch was 35, 30, and 53% for the constant, three-stage, and exponential treatments. Our results for N use efficiency are comparable to those of Dumroese et al. (2005), in which N use efficiency was 50% in three-stage treated *P. monticola* seedlings compared to 75% in exponentially treated seedlings.

Phosphorus use efficiency was also higher in the exponential treatment than other treatments (Fig. 4). As for N, the P contents of plants of both species were very similar among fertilization treatments. The amount of P leached was 2.5 times greater in the constant and three-stage treatments than in the exponential treatment. Phosphorus accumulation in the growth medium averaged $7.3 \text{ mg container}^{-1}$ and did not differ by treatment (Fig. 4). Phosphorus use efficiency of yellow poplar was 15, 14, and 28% for the constant, three-stage, and exponential treatments, respectively. Phosphorus use efficiency of larch varied from 8 to 24% by treatment. Most of the applied P contents were leached: from 70 to 85% for yellow poplar and from 73 to 91% for larch.

Most of the applied K was taken up by yellow poplar (Fig. 4); leaching losses were greater for larch. As for the other nutrients, more K was leached (3.0 times more) in the constant and three-stage treatments than in the exponential treatment. The use efficiency of K was high compared to N and especially P (Fig. 4). Potassium use efficiency of yellow poplar was 84, 80, and 93% for the constant, three-stage, and exponential treatments. Potassium use efficiency of larch was lower than that for yellow poplar: 54, 50, and 77%. Of the three nutrients measured, K had the lowest losses to leaching and to the growth medium.

Most studies of nutrient use efficiency have focused on N, because of its importance to plant production and because of concerns about NO_3 in drinking water. The fertilizer application rates in our study were based on N, so it is not surprising that P use efficiency was low. The optimal rate of P addition is probably quite a bit less than that of N addition, while our fertilizer applied equal amounts of N, P (as P_2O_5), and K (as K_2O). The N:P ratio on a mass basis was 2.3, which is very high. Selecting an optimal fertilizer grade could further reduce losses of non-limiting nutrients. Similarly, the study by Dumroese et al.

Table 2 Chemical properties of the growth medium receiving three types of solution irrigation for 11 weeks: a constant rate, a three-stage rate, and exponential fertilization

Species	Treatments	Organic matter (%)	pH	Total N (g kg ⁻¹)	Extractable phosphorus (mg kg ⁻¹)	Exchangeable K ⁺ (cmol _c kg ⁻¹)	Exchangeable Ca ²⁺ (cmol _c kg ⁻¹)	Exchangeable Mg ²⁺ (cmol _c kg ⁻¹)	Exchangeable Na ⁺ (cmol _c kg ⁻¹)
<i>Liriodendron tulipifera</i>	Pretreatment	46.4	5.3	3.7	13.6	0.20	1.52	0.85	3.2
	Constant	33.5 (1.3)	4.1 (0.0)	3.1 (0.3)	56.1 (29.2)	0.28 (0.01) ^a	1.70 (0.08)	0.65 (0.01) ^a	0.12 (0.02)
	Three-stage	31.6 (0.8)	4.1 (0.1)	2.6 (0.1)	73.2 (7.2)	0.19 (0.01) ^b	1.69 (0.05)	0.69 (0.04) ^b	0.14 (0.02)
<i>Larix leptolepis</i>	Exponential	28.5 (0.9)	4.1 (0.0)	2.7 (0.1)	79.1 (15.8)	0.36 (0.02) ^b	1.84 (0.07)	0.84 (0.01) ^b	0.12 (0.00)
	Constant	32.6 (0.1)	4.5 (0.1) ^a	3.1 (0.2)	90.1 (17.2)	0.29 (0.02)	2.42 (0.14)	0.84 (0.03)	0.20 (0.03)
	Three-stage	33.2 (1.5)	4.3 (0.0) ^b	3.2 (0.0)	67.3 (8.4)	0.28 (0.02)	1.96 (0.05)	0.86 (0.04)	0.21 (0.02)
	Exponential	34.4 (0.3)	5.1 (0.1) ^b	3.2 (0.1)	80.0 (20.8)	0.37 (0.03)	2.21 (0.12)	0.84 (0.01)	0.54 (0.20)

Means with the same column with the same letter or no letter are not significantly different at $\alpha = 0.05$. Standard errors are in parentheses ($n = 2$)

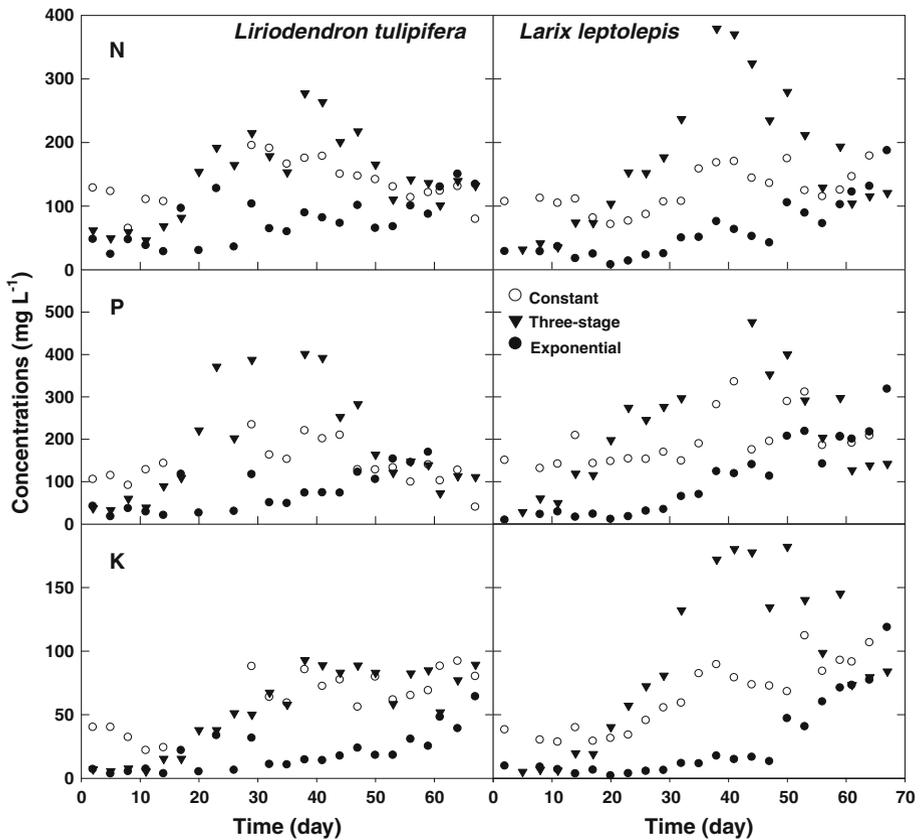


Fig. 3 Nutrient concentrations in solutions leached from trays receiving three types of solution irrigation for 11 weeks

(2005) reported low P use efficiency in container-grown *P. monticola* seedlings (12% for three-stage and 19% for exponentially treated seedlings).

Finally, it is important to note that nutrients can be lost in the application process; we did not include solution sprayed outside of the trays in our calculation of nutrient use efficiency. Our loss rates (87–88%) were probably higher, because of the small area of trays, than in commercial production systems. In a Finnish nursery production system, 33–85% of applied N was lost during production of forest seedlings in containers (Juntunen et al. 2002). For this reason, improving nutrient delivery systems, such as by supplying nutrient solutions to the bottom of the containers (Landis and Wilkinson 2004; Bumgarner et al. 2008), using taller containers (Landis et al. 1990), or irrigating based on monitoring soil moisture (Heiskanen 1995; Stowe et al. 2010), has the potential to further reduce nutrient losses beyond those achieved by optimizing the fertilizer concentrations applied to containers.

Conclusions

Exponential fertilization can reduce nutrient losses in tree nursery production systems. Our experiments with yellow poplar and larch showed improved nutrient use efficiency of N, P,

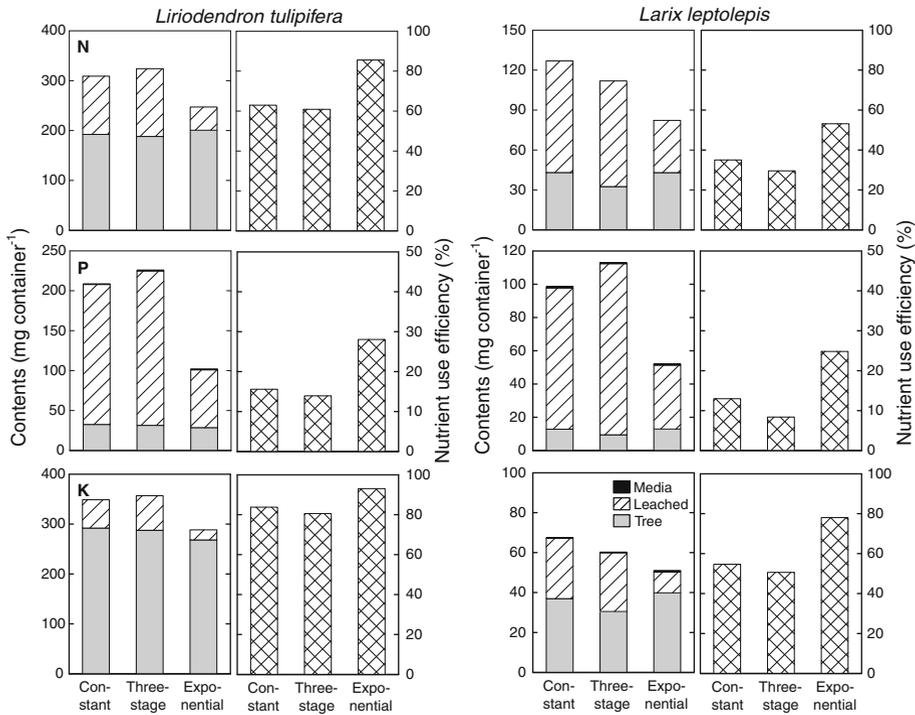


Fig. 4 Nitrogen, P, and K contents in biomass, leached solutions, and soil media (left panels) in response to three types of solution irrigation for 11 weeks. Nutrient use efficiency (right panels) was calculated as the ratio of nutrient contents of the plant to the sum of plant, leached nutrients, and accumulated nutrients in the containers

and K under exponential fertilization relative to constant and three-stage treatment regimes. Adjusting fertilization rates can reduce soil and water contamination around the nursery without compromising growth performance, which reduces both production costs and environmental impacts.

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