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SHORT-TERM IMPACT OF FERTILIZATION ON GROUND VEGETATION OF A DECIDUOUS TREE PLANTATION DEPENDING ON TREE SPECIES AND ENVIRONMENTAL VARIABLES

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Abstract
The study investigates the impact of fertilization with ammonium nitrate and wood ash on ground vegetation in a deciduous tree (*Betula pendula, Alnus glutinosa*, *Cerasus avium*) plantation. The ground vegetation was surveyed two years after fertilization, and the ground cover of each species was estimated visually. Species richness, diversity, differences in composition, and Ellenberg indicator values were compared between fertilized and control (unfertilized) plots. Detrended Correspondence Analysis (DCA) was performed including soil chemical parameters, Shannon diversity index (H') and EIVs as explanatory variables. Results show that several nitrophilous species were observed more frequently and had larger cover in the fertilized parcels. DCA reveals that significant differences in species composition between control and fertilized plots can be observed only in case of *Cerasus avium*. Light is the most influential factor and H' increases along with increasing EIV for light. Also, significant differences in H' values and the number of species between control and fertilized parcels also were found only for sweet cherry. Long-term observations of the plantation are required to determine whether the impact of fertilization observed in this study persists longer.

Keywords: wood ash, ammonium nitrate, ground vegetation, deciduous trees, plantation.

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INTRODUCTION

Carbon sequestration in plant biomass and reduction of greenhouse gas (GHG) emissions by planting trees, provision of renewable energy and restriction of the use of fossil energy are becoming more and more important in order to implement the strategic plans for climate change mitigation (the European Union's Green Deal). The targets are ambitious, while the information on how to achieve them is currently insufficient. The European Union has committed to planting three billion trees by 2030, some of them outside forest land or as a part of afforestation of non-agricultural land (Fetting 2020).

In Latvia, plantation forests are defined as forest stands established through afforestation intended for specific purposes and registered in the State Forest Register (Law on Forests, Chapter I, Section 1, 2000). In the countries of boreal forest zone, the most commonly planted coniferous trees are *Pinus* spp. (e.g., *Pinus sylvestris, P. murayana, P. contorta*), *Picea* spp. (e.g., *Picea abies, P. sithensis*), *Larix* spp. (*Larix decidua, L. sibirica*, *Larix* × *eurolepis*); whereas the most commonly planted deciduous trees are *Betula* spp. (*Betula pendula*, *B. pubescens*), alders (e.g., *Alnus glutinosa*, *A. incana*), aspen and poplar hybrids (e.g. *Populus tremula* × *P. tremuloides*, *Populus* × *canadensis*) and *Salix spp.* (Global Forest Resources Assessment 2000, 2015). On one hand, replacement of vegetation of natural ecosystems by creating artificial environments, e.g., plantations, is considered a threat to biodiversity (Uribe et al. 2021). On the other hand, while species diversity in tree plantations is lower than in natural forests, in several cases it is higher than in agricultural areas and other intensive land uses. Furthermore, tree plantations can significantly rehabilitate degraded areas by improving their flora and fauna, and accelerate regeneration of forests (Carnus et al. 2006, Stephens & Wagner 2007, Weih 2004).

Plants grow by harnessing energy from the sun and capturing CO_2 from the atmosphere through photosynthesis. Although trees are harvested and consumed for energy production, new ones can be cultivated and regrown relatively quickly, therefore woody biomass is a renewable energy source and a sustainable alternative to fossil fuels. Application of fertilizers in forests accelerates tree growth, stand development and carbon sequestration in biomass (Adams et al. 2005, Magnani et al. 2007, Smith et al. 1993). Recently, forest fertilization has been regaining popularity in the Nordic countries as a result of the demand for sustainable energy production. Also, in Latvia a study has been carried out gather knowledge to potentially reintroduce the practice (LVMI "Silava", 2021). The nutrients that usually limit plant growth are nitrogen (N), phosphorus (P) and potassium (K). Nitrogen fertilizers, such as ammonium nitrate (NH_4NO_3), are mostly used in forests on mineral soils. A single dose of N applied with NH_4NO_3 and other N-containing fertilizers to forest soil is 150 kg ha^{-1} and the following growth response of trees is 20–25 m^{3}ha^{-1} (Pukkala 2017). Ash is the solid residue of biomass combustion. Biomass ash, also wood ash is alkaline and contains P, K, calcium (Ca), magnesium (Mg), and a number of trace elements. Due to its alkalinity wood ash can be used a liming material (Bang-Andreasen et al. 2021, Karltun et al. 2008). The recommended dose of K is 40–80 kg ha$^{–1}$ and that of P is 40–50 kg ha$^{–1}$, corresponding to an amount of 2000–5000 kg dry weight of wood ash (Sikström et al. 2010). Wood ash does not contain N, but it can be applied together with NH_4NO to supplement soil N, P and K stocks and to prevent acidification.

Fertilizers are also frequently applied in plantation forests at some point of development. It is relatively easier to apply fertilizers to a plantation forest or short rotation coppice (SRT) plantations, as the movement of forest machines is not hindered by stumps. In Latvia, in plantation forests a lesser number of trees are allowed to be planted than occurring in a natural forest. The number of trees to be planted in SRT plantations per unit area is not regulated by the Cabinet of Ministers of Latvia, therefore the distance between the rows of trees planted can be chosen not to cause damage during the application of fertilizers (Law on Forests, Chapter IV, Section 24. 2000). Fertilization of forest plantations is similar to that of agriculture, however, differences in nutrient cycling need to be taken into account. For trees, the requirement for nutrients is partially met from internal pools of older tissues rather than entirely by uptake from forest soil. A significant amount of nutrients are also obtained from above- and below-ground litter production (Smerthust 2010). These factors lead to lesser amounts of fertilizers used in forestry comparing with agriculture.

Ground vegetation plays a significant role in forest ecosystems, although it frequently receives less attention than trees. Vegetation influences water and nutrient cycling, helps to stabilize soil and maintains biodiversity. Application of fertilizers to soil affects its pH and availability of nutrients, which may cause changes in ground vegetation. However, the magnitude of changes depends on a number of factors, such as site fertility prior to fertilization, light availability to forest floor, the level of moisture, land-use history and forest management activities. Several studies have been carried out in the Nordic countries and Canada, also the Baltic countries, to investigate, how forest fertilization affects the ground vegetation (Hart et al. 2019, Nordin et al. 2005, Ozolinčius et al. 2007). In most cases, changes caused by a single small dose of fertilizers have not been observed, whereas large doses and repeated fertilization may cause significant changes in floristic composition and species diversity in the ground vegetation (Nordin et al. 2005, Van Dobben et al. 1999). Also, the strongest impact has been observed in the least fertile forest sites (Gilliam 2006). Application of fertilizers result in occurrence of species typical of more fertile site types (Kellner 1993). As site productivity increases, rare species and species intolerant to competition are more likely to decrease in abundance and disappear than already abundant species (Davis et al. 1993, Suding et al. 2005).

Fertilization of tree plantations should be as sustainable as possible, therefore studies on the impact on environment, including ground vegetation, are required. The aim of this study was to evaluate a short-term impact on ground vegetation in a young (approximately 10 years old, depending on the parcel) deciduous tree plantation and to determine, which factors have the most significant impact on species richness and diversity. A short-term impact needs to be assessed in order to avoid the negative effect of direct damage of fertilizers to ground vegetation. Long- term observations are expected, taking stabilization of vegetation and establishment of forest ground vegetation species into account.

MATERIALS AND METHODS

Study site

The study was carried out in Ķeipene plantation, where deciduous tree seedlings have been planted in 2012 and 2013 on former agricultural land with mineral soil. The Ķeipene plantation is located in the central part of Latvia, Ogre municipality, Ķeipene parish (56°55’59.3’N 25°08’15.4’E). The parcels with the following tree species were included in this study: silver birch (*Betula pendula* Roth.), black alder (*Alnus glutinosa* (L.) Gaertn.) and sweet cherry (*Cerasus avium* (L.) Moench syn. *Prunus avium* L.). The size of control and fertilized parcels of sweet cherry are 30 x 30 m, whereas the size of parcels for black alder and silver birch

Table 1. Characteristics of the deciduous tree plantation in Keipene and the amount of fertilizers used per parcel.

Tree species	Fertilized area, ha	Number of trees per ha	H, m	DBH, cm	NH_4NO_3, kg	Wood ash, kg
Black alder (*Alnus glutinosa*)	0.12	575	5.34	5.60	53	-
Silver birch (*Betula pendula*)	0.12	576	4.12	5.40	53	-
Sweet cherry (*Cerasus avium*)	0.09	1024	3.86	4.11	40	270

are 40 x 80 m). Characteristics of the parcels are shown in Table 1. In 2016, prior to the application of fertilizers, soil and leaf analyses were conducted and P deficiency was diagnosed. Ammonium nitrate was spread manually in 2017 (0.44 t ha^{-1}) and in parcels of sweet cherry, wood ash was spread additionally (3 t ha^{-1}). Wood ash was obtained from SIA Graanul Pellets pellet factory. The element concentration of wood ash was 9.6 g kg^{-1} P, 25.96 g kg $^{-1}$ K, 153.32 g kg^{-1} Ca and 11.58 g kg^{-1} Mg.

Soil analyses

Soil samples at depths of 0–10 cm, 10–20 cm, 20–40 cm and 40–80 cm were collected in 2019. The samples were air-dried, homogenized, and sieved (2 mm pore size) according to the ISO 11465:1993. The samples were further microwave-digested (Mars 6 iWave. CEM) using 65% HNO_3 to measure concentrations of macro-nutrients: Ca (g kg^{-1}), Mg (g kg^{-1}), P (g kg^{-1}) and K (g kg^{-1}). Total nitrogen (g kg^{-1}) content was determined using elementary analysis method by the LVS ISO 13878:1998. Total carbon (g kg^{-1}) content was determined using elementary analysis according to ISO 10694:2006. Soil extracts were analyzed with Flame Atomic Absorption Spectroscopy (FAAS, AAnalyst 200, Perkin Elmer).

Ground vegetation survey

The ground vegetation was assessed two years after the application of fertilizers in double plots (grouped by two) of 1 x 1 m (in total 12 plots each parcel). In each sample plot, the cover of each species in herb layer (vascular plants, shrubs, and tree seedlings up to 0.5 m) in control and fertilized plots was estimated visually in per cent. There was no moss layer in the sample plots.

Shannon diversity index (H’) was selected as the measure to compare species diversity between control and fertilized survey plots. H’ was calculated with Equation 1 using the data on the ground cover of each species instead of the number of individuals (Magurran 1988):

$$H' = -\sum\left(\frac{n_i}{n}\right) log_2\left(\frac{n_i}{n}\right) \qquad (1),$$

where n – the total number of individuals; n_i – the number of species per sample plot.
Ellenberg indicator values (EIVs) (Ellenberg et al. 1992) for light (L), temperature (T), continentality (K), moisture (F), nutrients (N) and soil reaction (R) were calculated for each sample plot and the weighted average for each stand was determined using Equation 2:

$$Weighted\ average = \frac{\sum_{i=1}^{n}(r_{ij} * x)}{\sum_{i=1}^{n} r_{ij}} \qquad (2),$$

where r_{ij} – the response of species i in sample plot j; x_i – the indicator value of species i, I – individual species.

Statistical analysis

The data were processed and analysed with Microsoft Excel and Rstudio softwares. Student’s T-test and Wilcoxon rank sum test with continuity correction were performed (depending on normality of data distribution) to estimate differences of indicators between the control and fertilized plots. Shannon diversity index was calculated, and vegetation ground cover data were processed and the matrices of species and explanatory factors for the Detrended Correspondence Analysis (DCA) were prepared with Microsoft Excel. Student’s T-test and Wilcoxon rank sum test were performed to estimate differences between the control and fertilized areas. The analysis method was chosen depending on the normality of data distribution.

DCA was done with the software PC-ORD 6.08. The planted tree species and the type of fertilizer were specified as categorical factors; soil chemical parameters, H’ and EIVs were specified as quantitative factors. The relative Euclidean distance to the original matrix and Euclidean distance to the ordination matrix were used. Pearson’s correlation coefficients for the three ordination axes with soil chemical parameters, H’ and EIVs were determined.

RESULTS AND DISCUSSION

As the habitat for plant species other than trees, plantations are characterized by restrictions resulted from management. Clearcutting and short rotations favour ruderal species, whereas late-successional species may not be present at all and harvesting disturbance may result in occurrence of invasive species (Allen et al. 1995). The most commonly observed species in the black alder-growing parcels of the studied plantation were *Anthriscus sylvestris*, *Equisetum arvense*, and *Trifolium pratense*. The species mentioned above usually occur in a wide range of habitats such as meadows, pastures, roadside, forests, wasty places. The most commonly observed species in the silver birch- growing parcels besides those mentioned in the alder-growing parcel was *Aogopodium podagraria,* which has a particularly large cover in several plots in both control and fertilized parcels. The most commonly observed species in the sweet cherry-growing parcels with a large cover were *Anthriscus sylvestris*, *Equisetum arvense, Poa angustifolia* and *Artemisia vulgaris.* Graminoids were also commonly observed in all the parcels. Majority of the observed species were heliophiles with the EIV for light (L) ranging from 7 to 9. Full list of species in the sample plots is available upon request of the first author.

Several nitrophilous species either have higher abundance in fertilized parcels than in control or were observed only in the fertilized parcels. In the fertilized parcels of silver birch, *Aegopodium podagraria* was observed more frequently compared to the unfertilized plots. In the sweet cherry-growing area, *Aegopodium podagraria* was observed only in the fertilized parcel. In the cherry-growing parcels, a slightly larger cover was observed for *Artemisia vulgaris*. The nitrophilous *Anthriscus sylvestris* has a slightly larger cover in the fertilized parcels of birch and alder, although it is commonly observed in control parcels as well. *Taraxacum officinale* was observed more frequently compared to the control area. Only in fertilized plots *Lathyrus palustris* and *Heracleum sibiricum* were observed. Only in the fertilized areas of both black alder and wild cherry, *Lolium perenne* was observed. In sweet cherry parcels the nitrophilous *Urtica dioica* was observed, but did it not occur frequently and did not have a large cover. *Tussilago farfara* and *Lolium multiflorum* were observed more frequently in the fertilized cherry parcels comparing with the control parcels. In the fertilized parcels of all the trees, *Phleum pratense* was observed more frequently and had a slightly larger cover. Also, *Potentilla anserina* was observed more frequently in the fertilized parcels of all the trees.

The DCA biplot of vascular plant species composition is shown in Figure 1. The associated eigenvalues and other attributes are shown in Table 2. Only the plots in parcels, where the sweet cherry has been planted, formed separate clusters of control plots and plots fertilized with wood ash + ammonium nitrate. The plots, where the silver birch and the black alder have been planted, are scattered unevenly, partially overlapping and cannot be distinguished as separate clusters of control and fertilized plots in the DCA biplot. This indicates that the species composition and growth conditions between these plots are similar.

Negative correlation ($r < -0.6$) was found between Axis 1 and variables L and H'. Positive correlation ($r > 0.5$) was found between Axis 1 and variables R and N. Correlations between Axis 1 and soil chemical parameters (Corg., pH, Ntot., Ktot., Ca, Mg and P) were relatively weak. The Axis 2 had the strongest correlations with Corg. ($r = 0.567$), Ntot. ($r = 0.541$) and P ($r = 0.470$). The coefficients of correlations between Axis 1, 2 and 3 and soil chemical parameters, H' and EIVs are shown in Table 3. The first axis could be explained by light availability and species diversity (H'). In this case, more light availability indicates a more diverse species community. According to the DCA biplot, the control plots of sweet cherry parcel are characterized by higher species diversity and higher light availability, whereas fertilized plots have higher nutrient availability, which could be the result of fertilization. The second axis could be partially explained by soil nitrogen and organic carbon content.

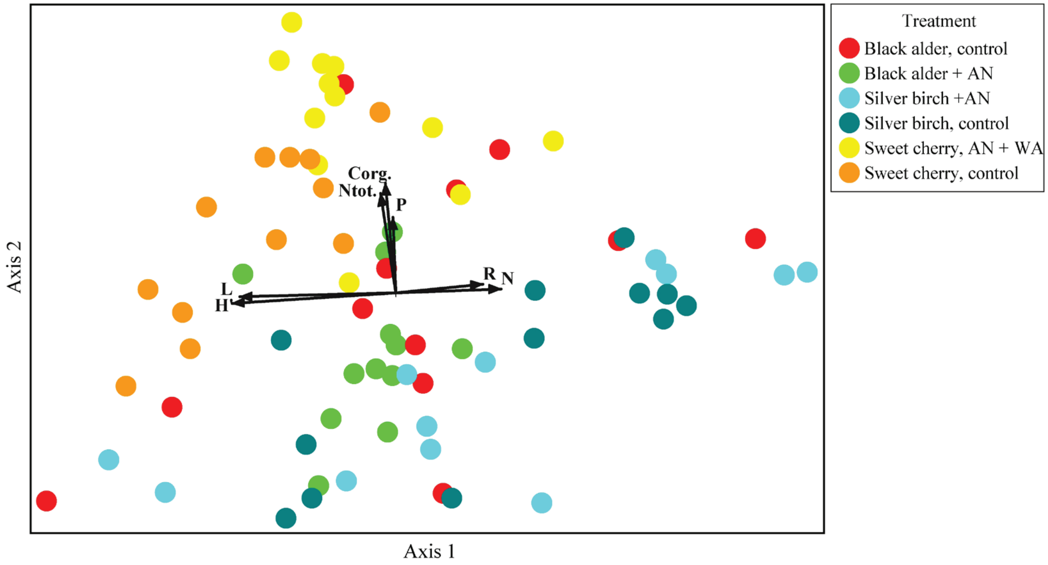

Figure 1. A DCA diagram showing the relationship between ground vegetation species composition in sample plots, soil chemical parameters, Ellenberg indicator values and Shannon diversity index in control and fertilized plots in a deciduous tree plantation. Abbreviations: AN – ammonium nitrate, WA – wood ash, Corg. – organic carbon, Ntot. – total nitrogen content, Ca – calcium, pH, L – light, N – soil fertility, R – reaction, F – humidity, T – temperature, K – continentality, H' – Shannon diversity index.

Although water and nutrient availability are essential for plant growth, light has a crucial role in utilization of nutrients and photosynthesis, therefore it is most likely the limiting resource for the understory layer and also seems to be the factor that determines species diversity in forests (Strengbom & Nordin 2008; Alvarez-Clare et al. 2013, Record et al. 2016). A study focusing on temperate forests suggest that species diversity of ground vegetation does not increase with light availability, whereas species richness increases (Dormann et al. 2020). Another study indicates that heterogeneity of light, not the average availability results in higher species richness (Bartels & Chen 2010). We also compared the total number of species, the average number of species in 1 x 1 m plots and H' in control and fertilized areas. The average number of species in the sweet cherry parcel is higher in the control plots (18 species) compared to the fertilized plots (14 species), and the difference is statistically significant ($p < 0.05$). Statistically significant differences were not found in case of parcels of other trees

Table 2. The eigenvalues, gradient length, increment (R^2) and the cumulative percentage of explained variance of Axis 1, Axis 2, and Axis 3 of DCA ordination.

Axis	Eigenvalue	Gradient length	Increment (R^2)	Cumulative percentage of explained variance (R^2)
1	0.49218	3.440	0.237	0.237
2	0.24624	2.245	0.177	0.414
3	0.18019	2.453	0.046	0.460

Table 3. Pearson correlation coefficients (r) and r^2 between the three DCA axes and soil chemical parameters, H' and EIVs.

Factor	Axis1		Axis2		Axis3	
	r	r^2	r	r^2	r	r^2
$C_{org.}$	-0.183	0.033	0.567	0.322	0.053	0.003
pH	0.167	0.028	0.146	0.021	-0.443	0.197
$N_{tot.}$	-0.214	0.046	0.541	0.293	0.093	0.009
$K_{tot.}$	0.098	0.010	0.167	0.028	-0.382	0.146
Ca	0.167	0.028	0.226	0.051	-0.421	0.177
Mg	0.256	0.066	0.001	0.000	-0.406	0.165
P	-0.101	0.010	0.470	0.221	0.008	0.000
L	-0.676	0.458	-0.111	0.012	-0.345	0.119
T	-0.271	0.074	0.170	0.029	-0.161	0.026
K	-0.181	0.033	0.221	0.049	-0.134	0.018
F	0.122	0.015	0.032	0.001	0.553	0.306
R	0.507	0.257	0.162	0.026	0.067	0.005
N	0.557	0.310	0.113	0.013	0.116	0.013
H'	-0.694	0.482	-0.179	0.032	-0.131	0.017

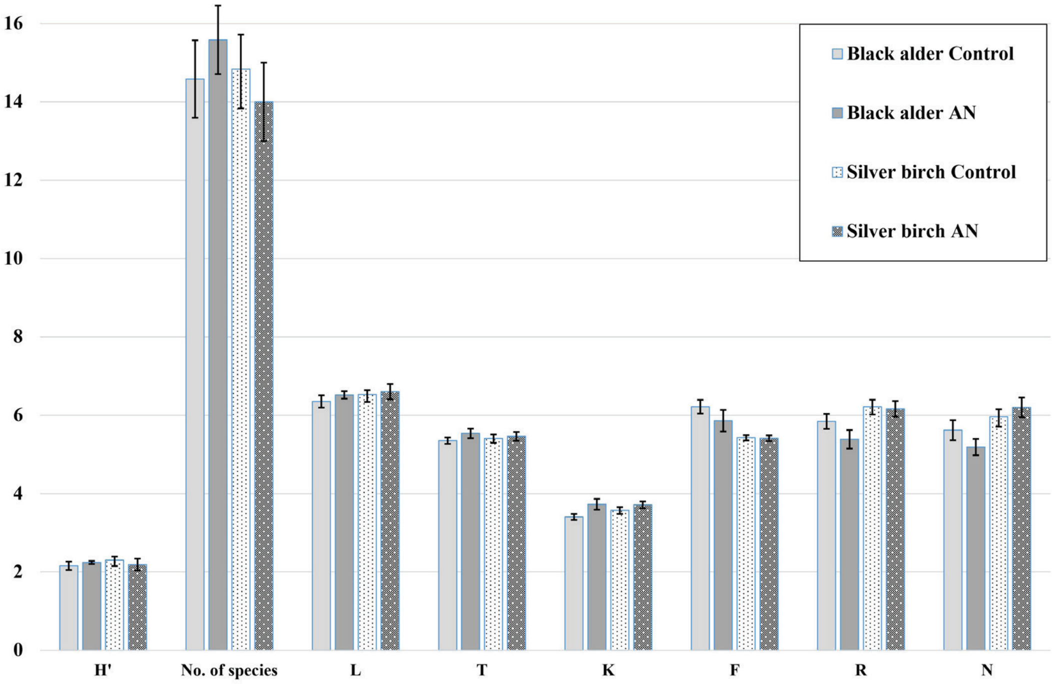

Figure 2. Species diversity, richness (the number of species) and Ellenberg indicator values for light (L), temperature (T), continentality (K), reaction (R) and nitrogen/nutrient level (N) in control and fertilized vegetation survey plots in the black alder and silver birch growing parcels in Ķeipene plantation.

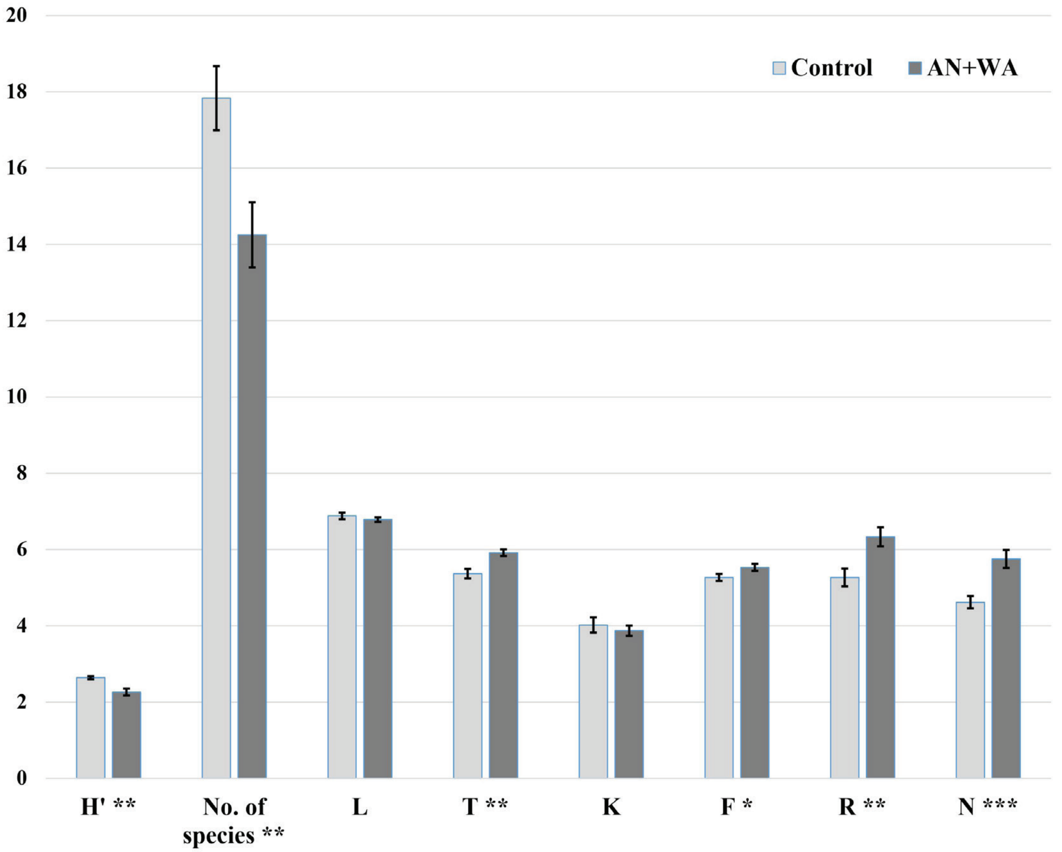

Figure 3. Species diversity, richness (the number of species) and Ellenberg indicator values for light (L), temperature (T), continentality (K), reaction (R) and nitrogen/nutrient level (N) in control and fertilized vegetation survey plots in the sweet cherry growing parcel in Ķeipene plantation. Significance levels are indicated with *, where * – p ≤0.05, ** – p≤0.01, *** – p≤0.001.

planted. The total number of species in control area is 39 in sweet cherry-growing parcels, 36 – in silver birch-growing parcels and 42 in black alder-growing parcels. In the fertilized areas, the total number of species in sweet cherry-growing parcels is 33, in birch-growing parcel – 42, and in alder-growing parcel – 41. The total number of species in fertilized parcels is slightly lower those of sweet cherry and black alder, comparing with the control plots, whereas in fertilized birch-growing parcels the number is slightly higher. The average H' value in cherry-growing parcel control plots is 2.64 ± 0.05, but in fertilized plots it is 2.26 ± 0.09, and the difference between control and fertilized plots is statistically significant ($p<0.05$). The comparison of species diversity, species richness and Ellenberg indicator values is shown in Figures 2 and 3.

CONCLUSION

Lower species richness and differences in species composition (presence or increased occurrence of nitrophilous species) of the ground vegetation in the fertilized sweet cherry-growing parcel comparing with the control parcel in Ķeipene plantation might be the result of wood ash and ammonium nitrate application. The slightly lower species diversity and species richness might result from reduced light availability due to increase of tree canopies. However, two years is a relatively short time to assess ground vegetation development and the impact of fertilization in a young deciduous tree plantation. Species composition will change as the plantation grows older becoming suitable for shade-tolerant species. Longer-term observations of vegetation in the Ķeipene plantation are required to draw in-depth conclusions.

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