## RESEARCH



# Effect of innovative peat-free organic growing media and fertilizer on nutrient allocation in pedunculate oak (*Quercus robur* L.) and European beech (*Fagus sylvatica* L.) seedlings after nursery production cycle

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## Abstract

This study evaluates the effects of novel peat-free organic growing media and a novel liquid fertilizer on the biometric features and macronutrient allocation of Quercus robur and Fagus sylvatica seedlings with the view to compare biomass and nutrient allocation of plant organs in seedlings cultivated on peat growing medium against those grown on novel peat-free growing medium and fertilizer. The experimental setup involved four growing medium variants, including peat as the control (R20, R21, R22 and C). The novel growing medium and fertilizer were designed and formulated by the University of Agriculture in Kraków, Poland (UAK). Fertilization used in the state forest nurseries was represented as SR20, SR21, and SR22, while the novel fertilizer of UAK was represented as UR20, UR21, and UR22; meanwhile, SC and UC represented the control growing medium (peat) in both cases, respectively. The experiment was laid in a 2×2×4 experimental design using five seedlings per treatment. Seedlings were assessed for roots, shoots, and leaves biomass after the nursery production cycle. The allocation patterns highlighted the variability of nutrient allocation within the plants, with more nutrients allocated to the root system. Interestingly, treatment UR22 yielded the highest mean root values, root biomass, and virtually all macroelement allocation. The SC solid fertilizer treatment and the UR22 liquid fertilizer treatment consistently showed superior performance across both species and different plant organs. These findings suggest that these treatments are particularly effective in enhancing the nutrient content of oak and beech seedlings, making them suitable choices for optimizing the growth and health of these species.

**Keywords** Peat-free growing media · Liquid fertilizer · Forest tree seedlings · Sustainable cultivation

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# Introduction

The decline of natural resources, including forest, is one of the greatest environmental problems faced in Europe and on other continents. The nutrient cycle plays a vital role in the nourishment and proper development of trees (Cambi et al. 2015). The proper development and high vitality of forest seedlings can be accomplished with an adequate amount and extent of nutrients adapted to the species' requirements (Muschler, 2016). Mineral fertilizers are the main source of nutrients for seedlings growing in forest nurseries, affecting their physiological and morphological properties (Wani et al. 2016). Appropriate mineral fertilization has been reported to enable better nourishment of seedlings, and the aggregated stores of nutrients promote better development, ability to adjust to ecological circumstances, and imperviousness to stress in difficult periods after planting (Loewe-Muñoz, 2024). Moreover, alternative growing medium used in nurseries are prepared based on ingredients such as high peat, bark, sawdust, perlite, vermiculite, or sand, which are poor in mineral compounds and therefore require fertilization (Bosiacki 2009).

In Poland, containers are commonly used for the production of tree seedlings in the nursey (Kormanek and Małek 2023). Over the years, coniferous monocultures have been intensively restructured due to trees' declining health and quality. Fagus sylvatica L. is a temperate species found throughout central and Western Europe (Jaworski 2019). Quercus spp., including Quercus robur L. are a major tree genus in Polish forests. Oak and beech forests play a significant role in the functioning of the biosphere. They produce and accumulate biomass, produce oxygen, regulate the composition of the atmosphere of the planet, and more (Romanov et al. 2022). Meanwhile, the decline and reduction of forests dominated by these species have been observed in Poland and across Europe. Because of their excellent wood quality, beech and oak are becoming more commercially desirable than several conifers because they are the preferred tree genus in any adaptation strategies to climate change for both ecological and economic reasons (Rotowa et al. 2023a). Under the circumstances of global change, it has been established that shifts in tree species and their symbiotic associations impact biogeochemical processes (Crowley et al. 2016; Crowley and Lovett 2017; Yu, 2023). Hence, it is of paramount importance to intensify efforts to raise the health and sustainability of forest stands of these highly sought-after species, especially in this era of transition in Polish forests from pine to other species.

The distribution of dry matter among plant organs is one of the key variables influencing the survival, competitive ability, and productivity of individual plants. It is influenced by several factors, including the availability of minerals in the rhizosphere and the flow of nutrients to the roots (Millner and Kemp 2012; Cortina et al. 2013; Chu et al. 2020; Pająk et al. 2022a). There is no doubt that; the distribution of macronutrients among plant organs plays a critical role in regulating growth rates and is an essential ecological process related to the history of plant life (Tripathi et al. 2014). Earlier studies have predominantly focused on biomass allocation (Vicca et al. 2012; Poorter et al. 2012a, b; Wei et al. 2013; Zhu et al. 2016; Pająk et al. 2022a) or nutrient investment in leaves only (Reed et al. 2012; Richardson et al. 2004; Hu et al. 2014). Aoyagi and Kitayama (2016) took a novel approach by examining how nitrogen and phosphorus investment in plant organs changes in Bornean rain forests to depletion of each element. Notably, Rotowa et al. (2023b) reported the effects of peat-free organic growing medium on the root systems of *F. sylvatica* and *Q. robur*. However, the specific allocation of nutrients between plant organs when grown on novel growing medium has rarely been demonstrated.

Peat is widely recognized as a foundational component within nursery growing medium due to its exceptional good physical, chemical, and biological properties. Its outstanding water-retention capabilities and consistently high quality make it a favored medium for nurturing plants (Gruda 2012). However, the release of carbon from peat soils over time raises concerns about peat's environmental impact. Peat excavation in Europe saw an extraordinary increase from 20,000 cubic meters in 2012 (equivalent to approximately 6,000 tonnes) to 20 million tonnes by 2022 (Gruda 2012; Hirschler and Osterburg 2022). This represents an increase of around 333% over the decade, highlighting a significant expansion in peat excavation activities, which further amplifies environmental degradation (van Beek, 2023). As a result, EU Member States has been on the outlook to reduce peat consumption (FCS, 2012; EPAP 2021; NMCE 2021) and because of upcoming restrictions on the availability of this material (EU, 2018) (GME 2003, Schmilewski, 2015) there is an urgent need to find a material/materials to replace it either partially or completely. To achieve this, it is important to be able to sample the liquid penetrating through the growing medium (both with and without plants growing) during periods of irrigation, fertilization or chemical application. Our technology solution makes this possible.

Therefore, this study aimed to analyze the effect of innovative peat-free organic growing medium and liquid fertilizer developed by the University of Agriculture in Kraków, Poland on the allocation of macroelements in different parts (leaves, shoots, and roots) of Q. *robur* and F. *sylvatica* seedlings toward the end of nursery production, just before planting to the forest site. This research is a preliminary investigation into the effects of innovative peat-free organic growing media and fertilizers on nutrient allocation in *Quercus robur* and *Fagus sylvatica* seedlings. Due to the small sample size, the results should be interpreted as suggestive, laying the groundwork for more extensive studies in the future. The hypotheses put forward assumed that the novel growing medium and fertilizer would effectively support the allocation of nutrients in different parts of individual seedlings, and that the uptake of macronutrients in both species raised on a novel growing medium and liquid fertilizer would enhance the biomass of these seedlings compared to those grown on a standard growing medium (peat plus solid fertilizer).

# **Materials and methods**

#### Composition and formulation of growing medium

The properties of the organic peat-free growing medium, granulometric composition of the growing medium before sowing, and nutrient content of growing media before seed sowing and after seedling production are shown in Tables 1, 2, 3 and 4. The growing medium, based on peat rich in sphagnum used for this study as the control variant (C) was produced at Nursery Farm in Nędza (50.167964 N, 18.3138334 E) Poland. The following granulometric composition, declared by the producer as percentage content of the fraction in a unit of volume: 2.5% of the 10.1–20 mm fraction, 12.5% of the 4.1–10 mm fraction, 12.5% of the 2.1-4. 0 mm, 72.5% of <2.0 mm; maximum degree of decomposition 15%; organic matter content >5%; and elemental content (g/g of 100% dry weight of the growing medium at the

Growing medium	Saw dust (%)	Wood chips (%)	Straw (%)	Wood bark (%)	Perlite (%)	Core wood (%)	Mixed silage (%)
R20	73	10	-	10	4	2	1
R21	20	63	-	10	4	2	1
R22	50	-	10	33	4	2	1

Table 1 Properties of the organic peat free growing medium

Woody materials were sourced from coniferous (mainly Pinus sylvestris L)

Table 2 Mean and standard deviation values of properties of the organic growing medium

Growing medium	Water ca- pacity (%)	Water Out- flow Rate (litre/min)	Co- effi- cient of Vari- ation (%)	Bulk density (g/cm3)	Solid density (g/cm3)	Air capac- ity (%)	Porosity (%)
R20	$53.02 \pm 2.42$	$0.595 \!\pm\! 0.150$	25.2	$0.127 \!\pm\! 0.009$	$1.56 \pm 0.000$	$38.90 \pm 2.90$	$91.85 \pm 0.60$
R21	$45.39 \pm 3.60$	$0.781 \pm 0.114$	14.6	$0.103 \pm 0.013$	$1.61 \pm 0.000$	$48.14 \pm 4.20$	$93.62 \pm 0.83$
R22	$50.71 \pm 2.11$	$0.594 \pm 0.150$	25.3	$0.113 \pm 0.009$	$1.62 \pm 0.000$	$42.35 \pm 2.61$	$93.04 \pm 0.55$
Control	71.44±2.83	$0.417 \pm 0.145$	34.9	$0.091 \pm 0.006$	$1.59 \pm 0.000$	$22.89 \pm 3.15$	$94.25 \pm 0.39$

beginning of the experiment) of  $37.99\pm0.69$  (C),  $0.74\pm0.01$  (N),  $0.02\pm0.01$  (P). On the other hand, the peat-free growing medium (R20, R21, R22) were formulated using a mixture of various components in varying percentage proportions (Table 1). In total, four growing media (R20, R21, R22, and peat) were utilized, each subjected to two fertilization (S and U) variants. The first set received standard solid fertilization (SR20, SR21, and SR22 variants), while the second set was treated with a novel liquid fertilizer also developed by the University of Agriculture in Kraków (UR20, UR21, and UR22). The peat growing medium served as the control in both fertilization scenarios, designated as SC and UC variants.

The properties of the growing medium were calculated according to (Jasik et al. 2023) where the volume of these medium was determined by measuring the top surface of the container using a ruler along with the dimensions of the upper surface of the cell opening and the height of the growing medium, the unoccupied volume (Ve, in cm<sup>3</sup>) within the container was calculated. By subtracting Ve from the total volume of the cell (V, in cm<sup>3</sup>) next, a container was placed beneath the cell to collect both the growing medium and the water that dripped out. The water outflow rate (W (N $\cdot$ s-1) was determined as the ratio of the water weight increase (w) to the time (t) taken for this increase (Bilderback 2022; Kormanek and Małek 2023). The time interval for the differential quotient was set at 60 s. The growing medium was removed from the top using a plastic spoon, and any remaining material that couldn't be spooned out was pushed into the container below. The collected samples from the cell and the container were weighed to determine the wet growing medium weight (mw, in g). After drying the growing medium, its dry mass (ms, in g) was measured. These measurements calculated the dry bulk density (BD, in g/cm3) and the wet bulk density (WBD, in g/cm<sup>3</sup>). BD = ms/Vs and WBD = mw/Vs. In each experimental variant, seedlings of each species underwent cultivation in 75 Marbet V300 polystyrene containers. Each container comprised 53 cells with a volume of 275 cm3.

Table 3 Mean (%) and	d standard deviatio	in values of granulc	metric composition	n of the growing me	dia before sowing			
Growing medium	>10 mm	10<5 mm	5 < 2 mm	2<1 mm	1 < 0.5  mm	0.5 < 0.25  mm	0,25 < 0.1  mm	>0.1 mm
R20	$0.05 \pm 0.10$	$3.77 \pm 1.57$	$14.45\pm 5.90$	$30.53 \pm 9.72$	24.45±2.24	17.72±7.49	$7.71 \pm 4.16$	$1.69 \pm 0.98$
R21	$0.00 \pm 0.00$	$6.40 \pm 1.37$	$25.44 \pm 1.91$	$30.90 \pm 1.11$	$19.11 \pm 0.90$	$12.16\pm0.31$	$5.12 \pm 0.41$	$0.96 \pm 0.11$
R22	$0.08 \pm 0.13$	$3.03 \pm 0.45$	$14.15 \pm 2.36$	$33.36 \pm 2.36$	$25.11 \pm 1.07$	$17.02 \pm 3.21$	7.11±1.72	$1.48 \pm 0.28$
Control	$0.00 \pm 0.00$	$11.27 \pm 0.37$	$25.08 \pm 1.18$	27.77±1.05	$16.20\pm1.05$	$8.42 \pm 0.56$	$3.81 \pm 0.43$	$1.88 \pm 0.21$

#### Seed sowing and germination

The containers were mechanically filled with various growing media, beech and oak seeds were sown manually in the nursery in Suków-Papierna (Daleszyce Forest District). The seeds were sown on April 19 and 20, with the preparation and sowing being carried out by the workers of the container nursery. To improve the germination process, oak seeds were scarified just before sowing, which involved the removal of about one-third of the seed in the cotyledon part. In contrast, beech seeds were stratified without using a stratification medium at a temperature of +3 °C and a humidity of 31%. Regardless of species, the seeds used for all growing medium variants came from the same origin and were accompanied by separate certificates of origin (MR/65848/21/PL for oak and MR/63313/20/PL for beech). After sowing, the containers were placed in the green house for 4 weeks and then transported to an external production field. During the growth of the seedlings, manual weeding was conducted. The seedlings were grown for 5months according to the procedure used in the container nursery (Szabla and Pabian 2009). During the seedling growth period, the total rainfall was only 78 mm, therefore irrigation was applied using an automatic RATHMAK-ERS Gartenbautechnik sprinkler ramp to replenish the water deficit.

Osmocote fertilizer was applied once during preparation of growing medium before sowing at a total dose of 3 kg m-3 of each growing medium, prepared as a mixture of Osmocote 3-4 M (2 kg) and Osmocote 5-6 M (1 kg). The composition of the Osmocote 3-4 M fertilizer was as follows N - 16% including 7.1% N-NO3- and 8.9% N-NH<sub>4</sub>+; P<sub>2</sub>O<sub>5</sub> - 9%, K<sub>2</sub>O -12%; MgO -2.0% and microelements (B, Fe, Cu, Mn, Zn, Mo); 5-6 M: N-15% including 6. 6% N-NO<sub>3</sub>- and 8.4% N-NH<sub>4</sub>+; P<sub>2</sub>O<sub>5</sub> -9.0%; K<sub>2</sub>O -12%; MgO -2.0%; and microelements (B, Fe, Cu, Mn, Zn, Mo). The new liquid fertilizer used was based on two different compositions. The first one consisted of N - 4.78%, P<sub>2</sub>O<sub>5</sub> - 1%, K<sub>2</sub>O - 2.64%, CaO - 2.65%, MgO -1.4%, SO<sup>3</sup> -0.71% and Na<sub>2</sub>O -0.14%. This fertilizer was initially applied with a total volume of 3.14 dm3 (0.048 dm3 -1 m-2). The second fertilizer contained N at 0.798%, P2O5 at 0.166%, K2O at 0.440%, CaO at 0.441%, MgO at 0.234%, SO3 at 0.118% and Na2O at 0.023%. The second fertilizer was applied with a total volume of 15.09 dm3 (0.229 dm3 -1 m-2). In the course of seedling production, the first fertilizer variant was applied eight times at 10-day intervals, while the second variant was applied 15 times at 5-day intervals. It is important to note that the fertilization regimes remained consistent for both beech and oak seedlings.

#### Parameter assessment and nutrient analysis

After the nursery's production cycle, a thorough examination of multiple seedlings was conducted. However, due to limitations stemming from the availability of seedling parts for laboratory testing, a specific selection process was employed by Rotowa et al. (2023b). Five seedlings, characterized by standard vigor and biometric parameters, were carefully chosen from each of the eight treatment groups for data collection. This resulted in a total assessment of 80 seedlings for both species in the experiment. The selected containers were distributed diagonally across the experimental field. Data were collected on the biomass of the different parts of the plant organs. These parts (leaves, shoots, and roots) of the sampled seedlings were dried at 65 °C for 48 h. After drying, the samples were ground and mineralized. From each part of the organ, 0.5 g was placed into a flask for mineralization with an

Table 4 Nutrient content (%) of growing media before seed sowing and after seedling production

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Growing medium	С	Ν	Р	K	Ca	Mg	Na
Before sowing							
R 20	48.01	0.297	0.031	0.159	0.452	0.055	0.040
R 21	46.34	0.507	0.068	0.271	0.601	0.072	0.035
R 22	48.90	0.447	0.043	0.404	0.857	0.059	0.042
Control	45.85	0.709	0.015	0.058	1.307	0.585	0.068
After seedling prod	uction						
Fagus sylvatica							
UR20	44.148	0.434	0.030	0.066	0.677	0.055	0.016
UR21	42.518	0.532	0.049	0.072	0.854	0.055	0.015
UR22	42.93	0.578	0.043	0.074	1.179	0.066	0.018
UC	39.784	0.651	0.019	0.071	1.543	0.525	0.072
SR20	42.167	0.596	0.093	0.129	0.721	0.068	0.018
SR21	39.978	0.996	0.134	0.161	0.985	0.087	0.020
SR22	42.167	0.756	0.110	0.156	1.463	0.086	0.023
SC	40.987	0.844	0.096	0.162	1.695	0.476	0.075
Quercus robur							
UR20	44.703	0.383	0.028	0.563	0.594	0.065	0.015
UR21	44.969	0.418	0.032	0.597	0.627	0.042	0.015
UR22	45.422	0.493	0.032	0.650	0.966	0.060	0.015
UC	41.863	0.654	0.016	0.654	1.392	0.472	0.060
SR20	45.455	0.519	0.059	0.991	0.589	0.056	0.014
SR21	43.313	0.942	0.121	1.626	0.879	0.088	0.020
SR22	45.096	0.872	0.114	1.703	1.139	0.081	0.018
SC	41.425	0.805	0.076	1.798	1.424	0.441	0.069

S - State Forest fertilization (solid), U - University novel fertilization (liquid), R - novel growing media, C - controls growing medium (peat-perlite)

added mixture of acids. The mineralized samples were later filtered into a 50 ml flask and the concentration of elements was determined using the ICP-OES apparatus. The samples were analyzed for their nitrogen, sulfur, and carbon contents using a LECO CNS TruMac analyzer and phosphorus, potassium, calcium, and magnesium contents using a Thermo iCAP 6500 DUO ICP-OES spectrometer following mineralization in nitric and hydrochloric acids at a ratio of 3:1. The analyses were performed at the Laboratory of Forest Environment, Geochemistry, and Land Intended for Reclamation in the Department of Ecology and Silviculture and Faculty of Forestry at the University of Agriculture in Kraków, Poland. Before chemical analyses, the root parameters were analyzed using WinRHIZO software in the Laboratory of Forest Biotechnology of the same department.

### Statistical analysis

The experiment was laid in a  $2 \times 2 \times 4$  experimental design, consisting of two species (beech and oak), two fertilizer types (solid and liquid), and four growing medium in each treatment (R20, R21, R22, and control) using five seedlings per treatment. Data analysis was conducted using a multifaceted approach. In order to show the comparative performance between the treatments, the collected data (after verifying that it met the assumptions of ANOVA) were subjected to mean and analysis of variance (ANOVA). Duncan Multiples

Range Test (DMRT) was applied to locate where the significant difference occurs among the treatments at p<0.05. PCA was employed to reduce the dimensionality of the dataset, identifying key variables that collectively explained the variance in the data. Correlation test was further carried out to quantify the strength of the linear relationship between the analysed variables.

## Results

#### Comparative analysis of biomass in Q. robur and F. sylvatica

The preliminary results indicate that growing medium may influence biomass production in both Q. robur and F. sylvatica seedlings, with variations observed across different organs. However, due to the small sample size (n=80), these findings is interpreted as trends rather than conclusive evidence. Solid fertilizer appears to enhance biomass production more consistently than liquid fertilizer in both species and across all seedling organs. The studied species both show enhanced growth under solid fertilizer, but the magnitude of this enhancement varies between the organs (root, shoot, leaf) and types of growing medium. Liquid fertilizer results in consistent leaf and root biomass across growing medium with moderate shoot biomass. While solid fertilizer results in a higher biomass in all organs in oak, While in beech, liquid fertilizer significantly increases root, shoot and leaf biomass.

The observed consistency in leaf biomass among oak seedlings raised with liquid fertilizers suggests a potential stabilizing effect of the treatments (Fig. 1a) Shoot dry mass increases slightly from UR20 to UC, with UR20 being significantly lower (Fig. 1b). Root biomass also shows consistent allocation showing no significant variation (Fig. 1c). In solid fertilizer however, highest biomass was recorded in SC showing slight variations in root, significant increases in shoot and leaves. From the result of beech seedlings raised on liquid fertilizer, leaf biomass is fairly consistent across growing medium. Shoot dry mass is highest in UC and Root dry mass shows a notable increase in UC compared to other growing media. From solid fertilizer treatment, leaf and shoot biomass follows the same trend, with SC having the significantly higher values compared to others, root dry mass is higher in SR22 and SC (Fig. 1). The distinct responses of oak and beech to the treatments stress the importance of fertilization in forestry seedlings and the restoration of forest ecological systems and management.

The result further shows that there exist positive correlations between the below-ground and above-ground organs of the seedlings The relationship between root and shoot biomass indicates balanced growth, which is critical for healthy plant development, Similarly, the relationship between root and leaf biomass indicates efficient nutrient uptake, which is essential for healthy leaf development. In the shoot-leaf biomass correlation among treatments reflects resource allocation strategies influenced by the properties of growing medium and fertilization methods (Fig. 2). Although the result showed large variation, significant differences observed on the SC growing medium but not on others suggested that certain growth characteristics were more sensitive on SC than on others. This means that the novel growing media also affected the biomass of the seedlings toward the end of seedling production just before planting in the forest.



Fig. 1 Alphabets 'a' and 'b' denote homogeneous groups under liquid fertilization and 'e, f' and 'g' denote homogeneous groups under solid fertilization. A-C biomass allocation across different growing medium in oak and beech species

# Analysis of the allocation of nutrients in *Q. robur* and *F. sylvatica* seedlings as affected by treatments

The results in Tables 5 and 6 show the allocation of macroelement contents in different parts (root, shoot, leaf) of *Q. robur* and *F. sylvatica* seedlings, respectively. In the roots of oak seedlings, the UR22 treatment under liquid fertilizer demonstrated the highest accumulation of crucial macro-elements, including NPK. This suggests that UR22 was the most effective liquid fertilizer treatment for promoting root nutrient content in oak seedlings. Among solid fertilizers, the SC treatment outperformed others, particularly in nitrogen and calcium content, indicating its superior efficacy for root nutrient uptake in this species. For the shoots of oak seedlings, UR22 again emerged as the best-performing liquid fertilizer treatment, yielding the highest levels of C, P, K and Ca. This indicates that UR22 not only supports root development but also enhances nutrient accumulation in the above-ground biomass. Among solid fertilizers, the SC treatment was particularly effective, producing the highest concentrations of C, N, P, K, and Ca in the shoots, making it the most robust solid fertilizer treatment for shoot growth. In the leaves, the UR20 liquid fertilizer treatment led to the highest accumulation of all tested macro-elements. This suggests that UR20 is especially beneficial for leaf nutrient content in oak seedlings. Similarly, the SC solid fertilizer treatment



Fig.2 A-C correlation among assessed biomass of seedling organs. S - State Forest solid fertilization, U -University novel liquid fertilization, R - novel growing media, C - controls growing medium (peat-perlite)

ment outperformed others, achieving the highest levels of all macro-elements in the leaves, which underscores its overall superiority among the solid fertilizer options. The result of analysis of variance revealed significant differences in macro-element content among the treatments. For oak seedlings, significant differences were observed in the nutrient content of shoots and leaves under liquid fertilizers, particularly for elements such as P, K, Ca and Mg. Under solid fertilizers, significant differences were also noted in the nutrient content of both shoots and leaves, indicating that the type of growing medium and fertilizer significantly influenced nutrient allocation in these organs (Table 5).

In the roots of beech seedlings, the UR22 treatment under liquid fertilizer was most effective, resulting in the highest levels of crucial nutrient including NPK. This indicates that UR22 is the most suitable liquid fertilizer for enhancing root nutrient content in beech seedlings. Among solid fertilizers, SR21 was the most effective, particularly in increasing N, P, Ca and Mg. For the shoots of beech seedlings, the UR22 liquid fertilizer treatment once again proved superior, showing the highest accumulation of phosphorus, potassium, and calcium. This highlights its consistent performance across different organs of the plant. In contrast, the SC solid fertilizer treatment led to the highest concentrations of C, N, P, K and Mg in the shoots, confirming its strong overall performance for shoot nutrient accumu-

Table 5 Mean	and SD and	alysis showing the al	llocation of macro	elements content in	n different parts of	Q. Robur seedling	s for each treatmen	ıt	
Treatment	Part	Fertilizer type	C (g/kg)	Ν	Р	K	s	Ca	Mg
			(mg/kg)						
	Roots								
UR20		Liquid	$4.09 \pm 0.69^{a}$	$36.8 \pm 7.10^{a}$	$6.00\pm1.10^{ab}$	$32.6\pm 5.40^{a}$	$4.10 \pm 0.60^{b}$	$33.10 \pm 6.00^{bc}$	$10.80 \pm 1.90^{a}$
UR21			$5.48 \pm 0.69^{a}$	$48.3 \pm 6.71^{a}$	$6.00\pm0.50^{\rm ab}$	$36.4 \pm 4.20^{a}$	$5.10 \pm 0.50^{ab}$	$37.80 \pm 5.20^{ab}$	$10.90 \pm 1.40^{a}$
UR22			$5.94 \pm 0.60^{a}$	$50.7\pm 5.20^{a}$	$7.00 \pm 0.80^{a}$	$42.6 \pm 4.30^{a}$	$6.20 \pm 0.60^{a}$	$49.80 \pm 4.50^{a}$	$13.50\pm1.50^{a}$
UC			$4.05 \pm 0.44^{a}$	$42.8 \pm 5.40^{a}$	$2.90 \pm 0.30^{\rm b}$	$26.2\pm2.50^{a}$	$4.20 \pm 0.60^{b}$	$22.00\pm 2.90^{\circ}$	$13.00\pm1.30^{a}$
Total			$4.86 \pm 0.34$	$44.27 \pm 6.17$	$5.64 \pm 0.86$	$34.3 \pm 3.41$	$4.90 \pm 0.61$	$35.32 \pm 4.25$	$11.77 \pm 1.63$
p-value			0.069 <sup>ns</sup>	$0.408^{ns}$	0.046*	0.078 <sup>ns</sup>	0.050*	$0.005^{**}$	0.673 <sup>ns</sup>
SR20		Solid	$3.16 \pm 0.36^{\circ}$	$36.0\pm10.2^{\circ}$	$5.10 \pm 1.30^{\circ}$	$22.1\pm6.10^{\circ}$	$4.20 \pm 1.40^{\circ}$	$29.80 \pm 4.20^{\circ}$	$8.40 \pm 2.00^{\circ}$
SR21			$4.15 \pm 0.74^{e}$	$49.8 \pm 14.9^{\circ}$	$8.20 \pm 1.40^{\circ}$	$34.0\pm 5.50^{\circ}$	$4.00\pm0.70^{e}$	$31.30 \pm 4.80^{\circ}$	$9.70 \pm 1.60^{\circ}$
SR22			3.46±0.23°	44.6±16.2°	$7.20\pm1.80^{\circ}$	$27.6 \pm 4.20^{\circ}$	$4.50 \pm 0.50^{\circ}$	$25.70 \pm 4.10^{\circ}$	$9.40 \pm 1.70^{\circ}$
SC			2.76±0.38°	$51.7 \pm 9.20^{\circ}$	$7.70 \pm 0.70^{\circ}$	$25.8 \pm 3.60^{\circ}$	$4.70 \pm 0.80^{\circ}$	$20.80 \pm 3.00^{\circ}$	$11.10\pm1.20^{e}$
Total			$3.37 \pm 0.24$	$45.5\pm15.8$	$7.06 \pm 1.40$	27.4±4.47	$4.36\pm0.72$	$26.87 \pm 3.08$	$9.64 \pm 1.50$
p-value			$0.238^{ns}$	0.429 <sup>ns</sup>	0.323 <sup>ns</sup>	0.263 <sup>ns</sup>	0.893 <sup>ns</sup>	0.325 <sup>ns</sup>	0.612 <sup>ns</sup>
	Shoots								
UR20		Liquid	$0.85 \pm 0.07^{ab}$	$10.9 \pm 0.80^{a}$	$1.20\pm0.12^a$	$4.90 \pm 0.60^{a}$	$1.00 \pm 0.10^{a}$	$17.70 \pm 1.90^{b}$	$3.00\pm0.20^{b}$
UR21			$0.77 \pm 0.03^{ab}$	$9.30 \pm 0.80^{a}$	$1.00 \pm 0.10^{ab}$	$4.30 \pm 0.20^{ab}$	$0.90 \pm 0.10^{a}$	$15.20 \pm 1.50^{b}$	$2.10 \pm 0.10^{\circ}$
UR22			$0.93 \pm 0.07^{a}$	$10.6 {\pm} 0.80^{a}$	$1.20 \pm 0.20^{a}$	$5.20 \pm 0.50^{a}$	$1.00 \pm 0.60^{a}$	$18.40\pm0.90^{\rm b}$	$2.60 \pm 0.20^{ab}$
UC			$0.76 \pm 0.02^{b}$	$10.3 \pm 0.40^{a}$	$0.61\pm0.03^{ m d}$	$3.40 \pm 0.20^{\rm b}$	$0.90 \pm 0.10^{a}$	$10.10 \pm 1.00^{a}$	$3.30 \pm 0.30^{a}$
Total			$0.83 \pm 0.04$	$10.29 \pm 0.35$	$0.99\pm0.08$	$4.45 \pm 0.35$	$0.97 \pm 0.04$	$15.34 \pm 0.98$	$2.75 \pm 0.20$
p-value			0.118 <sup>ns</sup>	0.423 <sup>ns</sup>	$0.015^{*}$	0.030*	0.629 <sup>ns</sup>	0.003 **	$0.002^{**}$
SR20		Solid	$0.94 {\pm} 0.09^{ m f}$	$12.4 \pm 1.10^{f}$	$1.40 \pm 0.20^{\rm f}$	$4.90 \pm 0.50^{\rm f}$	$1.30 \pm 0.20^{\rm f}$	$14.50 \pm 1.80^{\rm f}$	$2.50 \pm 0.30^{\mathrm{f}}$
SR21			$0.86 {\pm} 0.12^{\rm f}$	$12.6 \pm 1.70^{f}$	$1.30\pm0.20^{\mathrm{f}}$	$5.00 \pm 0.70^{f}$	$1.20 \pm 0.20^{\rm f}$	$15.50 \pm 1.80^{\rm f}$	$2.70 \pm 0.40^{f}$
SR22			$0.96 \pm 0.04^{\rm f}$	$17.8 \pm 3.40^{f}$	$1.60 \pm 0.20^{\rm f}$	$5.70 \pm 1.05^{f}$	$1.60 \pm 0.20^{f}$	$13.68 \pm 1.30^{f}$	$2.20 \pm 0.20^{f}$
SC			$1.96 \pm 0.26^{\circ}$	$34.1\pm 6.30^{\circ}$	$3.20\pm0.40^{\circ}$	$11.9 \pm 1.30^{e}$	$3.60 \pm 0.60^{\circ}$	$22.21 \pm 3.70^{e}$	$7.40 \pm 1.10^{\circ}$
Total			$1.29 \pm 0.07$	$14.8 \pm 1.50$	$1.40 \pm 0.24$	$5.70 \pm 0.94$	$1.40\pm0.32$	$15.90\pm 2.80$	$3.20 \pm 0.83$
p-value			0.003 **	0.002**	$0.000^{**}$	0.000**	0.000 **	$0.007^{**}$	$0.000^{**}$
	leave								

Table 5 (contin	(pən								
Treatment	Part Fertilize	er type	C (g/kg)	Ν	Ρ	K	S	Ca	Mg
			(mg/kg)						
UR20	Liquid		$1.36\pm0.05^{a}$	$21.20\pm3.60^{a}$	$1.70 \pm 0.30^{a}$	$9.2 \pm 1.40^{a}$	$2.06\pm0.39^{a}$	$58.8 \pm 4.58^{a}$	$8.40\pm0.18^{\rm a}$
UR21			$0.84 \pm 0.10^{\rm b}$	$11.30 \pm 1.30^{b}$	$0.70 \pm 0.10^{\rm b}$	$7.2 \pm 1.020^{ab}$	$1.16\pm0.17^{c}$	$33.8 \pm 4.55^{\rm b}$	$3.6\pm0.40^{b}$
UR22			$0.78 \pm 0.09^{b}$	$19.40\pm 2.80^{\rm b}$	$0.80\pm0.10^{\mathrm{b}}$	$8.3\pm1.30^{ab}$	$0.92\pm0.13^{\circ}$	$33.8\pm3.73^{\rm b}$	$4.0 \pm 0.54^{b}$
UC			$0.82 \pm 0.13^{b}$	$15.20\pm1.60^{b}$	$0.50 \pm 0.20^{\rm b}$	$6.0 \pm 0.95^{b}$	$1.54\pm0.87^{ m b}$	$28.1 \pm 3.10^{b}$	$8.2 \pm 0.90^{a}$
Total			$0.94 \pm 0.09$	$14.24\pm 2.37$	$0.95 \pm 0.13$	$7.69 \pm 1.98$	$1.42 \pm 0.11$	$38.62 \pm 4.05$	$6.05 \pm 0.57$
p-value			$0.000^{**}$	$0.003^{**}$	$0.000^{**}$	$0.050^{**}$	$0.000^{**}$	$0.000^{**}$	$0.000^{**}$
SR20	Solid		$1.49 \pm 0.11^{\circ}$	$32.80\pm 2.90^{\circ}$	$4.90 \pm 0.40^{\circ}$	$19.1 \pm 1.70^{\circ}$	$3.80 \pm 0.30^{\circ}$	$64.9\pm6.4^{\circ}$	7.5±0.61 <sup>f</sup>
SR21			$1.10 \pm 0.09^{\rm f}$	$26.80\pm2.80^{\circ}$	$3.70 \pm 0.60^{\circ}$	$15.8\pm2.30^{\circ}$	$3.20 \pm 0.40^{\circ}$	$40.1 \pm 5.8^{\circ}$	$5.8 \pm 0.90^{\mathrm{f}}$
SR22			$1.26 \pm 0.03^{\circ}$	$38.80 \pm 3.70^{\circ}$	$4.70 \pm 1.00^{\circ}$	$19.8 \pm 3.80^{\circ}$	$3.50 \pm 0.70^{\circ}$	$35.0\pm 3.7^{\rm e}$	$5.2 \pm 0.20^{f}$
SC			$1.728 \pm 0.33^{\circ}$	74.10±4.90°	$6.90 \pm 2.30^{\circ}$	$26.3 \pm 8.50^{\circ}$	$6.30 \pm 1.80^{\circ}$	$49.3 \pm 7.5^{\circ}$	$13.6\pm 2.20^{\circ}$
Total			$2.10 \pm 0.16$	$29.50\pm 2.10$	$3.60 \pm 0.30$	$16.8\pm 2.20$	$3.00 \pm 0.20$	$30.0 \pm 15$	$7.00 \pm 0.40$
p-value			0.114 <sup>ns</sup>	$0.107^{ns}$	$0.390^{\mathrm{ns}}$	$0.497^{ns}$	$0.150^{ns}$	0.150 <sup>ns</sup>	$0.001^{*}$
S - State Forest	fertilization, U - Un	uversity fe	stilization, R - nc	vel growing media	a, C - controls gro	wing medium (pea	tt-perlite)		
alphabets 'a' an	d 'b' denote homoge	sneous gro	ups under liquid	fertilization, 'e, f'	and 'g' denote ho	mogeneous groups	under solid fertili	ization. $p=0.05$	

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Table 6 Mear	and SD and	alysis showing the al	llocation of macro	elements content in	n different parts of a	F. Sylvatica seedling	gs for each treatme	ent	
Treatment	Part	Fertilizer type	C (g/kg)	Ν	Р	K	S	Ca	Mg
			(mg/kg)						
	Roots								
UR20		Liquid	$3.31 \pm 0.60^{a}$	$34.40\pm 2.50^{a}$	$3.50 \pm 0.21^{\circ}$	$21.80 \pm 3.50^{\rm b}$	$3.30 \pm 0.30^{a}$	$30.80\pm 2.19^{a}$	$7.20 \pm 0.98^{a}$
UR21			$3.09\pm0.51^{a}$	$34.20\pm 2.90^{a}$	$6.10 \pm 0.90^{b}$	$23.50 \pm 4.00^{ab}$	$3.50\pm0.50^{a}$	$35.00\pm 6.30^{a}$	$8.10 \pm 1.60^{a}$
UR22			$3.49\pm0.56^{a}$	$40.60 \pm 3.40^{a}$	$9.30 \pm 1.00^{a}$	$25.50\pm 5.70^{a}$	$4.20 \pm 0.40^{a}$	$34.10\pm 5.60^{a}$	$9.30 \pm 1.70^{a}$
UC			$2.66\pm0.61^{\rm b}$	$42.10 \pm 3.90^{a}$	$2.10\pm0.30^{\circ}$	$16.30\pm3.01^{\circ}$	$3.10 \pm 0.30^{a}$	$13.30\pm6.00^{\rm b}$	$7.40 \pm 1.90^{a}$
Total			$3.12 \pm 0.62$	$36.92 \pm 3.31$	$2.89 \pm 0.74$	$20.56 \pm 3.89$	$3.52 \pm 0.19$	$28.92 \pm 9.38$	$7.95 \pm 1.69$
p-value			0.125 <sup>ns</sup>	0.470 <sup>ns</sup>	$0.000^{**}$	$0.000^{**}$	$0.194^{ns}$	$0.000^{**}$	$0.135^{ns}$
SR20		Solid	$3.25\pm0.74^{e}$	$38.80 \pm 4.20^{\circ}$	$8.20\pm0.80^{\circ}$	$22.10\pm 5.30^{e}$	$3.80 \pm 0.60^{\circ}$	$35.80 \pm 4.50^{\circ}$	$8.40 \pm 1.90^{\circ}$
SR21			$3.20 \pm 0.94^{\circ}$	$38.50\pm 5.50^{\circ}$	$9.80 \pm 1.60^{\circ}$	$21.10\pm7.20^{\circ}$	$3.50 \pm 0.90^{\circ}$	$39.50 \pm 7.50^{\circ}$	$9.60 \pm 3.70^{\circ}$
SR22			$2.30 \pm 0.86^{ef}$	$40.80\pm8.10^{\circ}$	$6.60\pm0.80^{\circ}$	$20.30 \pm 4.80^{\circ}$	$3.10 \pm 0.90^{\circ}$	$30.10 \pm 1.90^{\circ}$	$8.40\pm 2.20^{\circ}$
SC			$1.99\pm0.86^{\mathrm{f}}$	$38.00 \pm 4.70^{\circ}$	$7.00\pm0.80^{\circ}$	$18.40\pm7.10^{\circ}$	$3.40\pm0.80^{\circ}$	$12.00\pm 2.10^{f}$	$7.30 \pm 1.90^{\circ}$
Total			$2.68 \pm 0.93$	$38.40 \pm 5.60$	$6.60 \pm 0.85$	$22.10\pm 5.10$	$3.50 \pm 0.80$	$28.80 \pm 3.90$	$8.20{\pm}2.30^{a}$
p-value			0.049*	0.988 <sup>ns</sup>	0.186 <sup>ns</sup>	0.797 <sup>ns</sup>	0.715 <sup>ns</sup>	$0.003^{**}$	0.620 <sup>ns</sup>
	Shoots								
UR20		Liquid	$0.65 \pm 0.05^{a}$	$11.91 \pm 0.79^{a}$	$1.31 \pm 0.04^{b}$	$4.55 \pm 0.61^{a}$	$0.85 \pm 0.01^{a}$	$11.08 \pm 1.08^{a}$	$1.83 \pm 0.05^{b}$
UR21			$0.70 {\pm} 0.05^{a}$	$12.28 \pm 1.04^{a}$	$1.88 \pm 0.23^{ab}$	$5.61 \pm 0.84^{a}$	$0.89 \pm 0.05^{a}$	$11.86 \pm 0.36^{a}$	$1.97 \pm 0.05^{b}$
UR22			$0.73 \pm 0.10^{a}$	$12.64 \pm 3.46^{a}$	$2.38 \pm 0.34^{a}$	$7.21 \pm 1.74^{a}$	$0.95 \pm 0.12^{a}$	$12.56\pm 3.56^{a}$	$2.02\pm0.55^{b}$
UC			$0.91\pm0.15^{a}$	$13.43 \pm 6.47^{a}$	$0.92\pm0.13^{\circ}$	$5.09 \pm 1.26^{a}$	$1.08 \pm 0.19^{a}$	$9.93 \pm 2.14^{a}$	$2.84 \pm 0.96^{a}$
Total			$0.75 \pm 0.04$	$13.56 \pm 4.14$	$1.63 \pm 0.16$	$5.62 \pm 0.98$	$0.95 \pm 0.06$	$11.35\pm 2.21$	$2.16 \pm 0.25$
p-value			0.242 <sup>ns</sup>	0.105 <sup>ns</sup>	$0.001^{**}$	0.166 <sup>ns</sup>	0.518 <sup>ns</sup>	0.287 <sup>ns</sup>	0.047*
SR20		Solid	$1.31 \pm 0.11^{\rm f}$	$14.80 \pm 11.3^{b}$	$2.50{\pm}0.30^{\rm f}$	$5.80 \pm 0.60^{f}$	$1.00 \pm 0.80^{\rm f}$	$15.70\pm 2.40^{\circ}$	$2.90 \pm 0.80^{\rm f}$
SR21			$0.82 \pm 0.07^{\rm f}$	$16.50 \pm 1.20^{\rm b}$	$2.40 \pm 0.10^{\rm f}$	$6.00 \pm 0.50^{\rm f}$	$0.90 \pm 0.10^{\rm f}$	$16.20\pm 2.00^{\circ}$	$2.10 \pm 0.77^{f}$
SR22			$0.88 {\pm} 0.09^{\rm f}$	$22.50 \pm 1.60^{ab}$	$4.70 \pm 1.60^{\rm ef}$	$9.80 \pm 0.80^{\circ}$	$2.00 \pm 0.60^{\circ}$	$22.80 \pm 3.00^{\circ}$	$3.20 \pm 0.93^{ef}$
SC			2.79±0.85°	$24.40 \pm 0.70^{\circ}$	$6.50 \pm 0.10^{\circ}$	$9.30 \pm 0.80^{\circ}$	$3.70 \pm 0.12^{e}$	$24.70 \pm 3.40^{\circ}$	$5.10 \pm 0.50^{\circ}$
Total			$1.10 {\pm} 0.15$	$17.80 \pm 1.10$	$3.80 \pm 0.30$	$7.80 \pm 0.67$	$1.30 \pm 0.17$	$19.50 \pm 2.20$	$2.85 \pm 0.30$
p-value			0.018*	0.013*	0.007**	0.007**	$0.028^{*}$	0.280 <sup>ns</sup>	$0.006^{**}$
	leave								

Table 6 (continued)								
Treatment Part	Fertilizer type	C (g/kg)	Ν	Р	K	s	Ca	Mg
		(mg/kg)						
UR20	Liquid	$0.50 \pm 0.01^{\rm b}$	$7.50 \pm 1.80^{\rm b}$	$0.55\pm0.05^{\rm b}$	$4.90\pm0.15^{a}$	$0.74 \pm 0.08^{b}$	$24.68\pm3.06^{a}$	$3.07\pm0.42^{b}$
UR21		$0.49\pm0.04^{\mathrm{b}}$	$7.60 \pm 1.80^{\rm b}$	$0.79 \pm 0.14^{\rm b}$	$4.20\pm0.13^{a}$	$0.76 \pm 0.09^{b}$	$24.86\pm6.07^{a}$	$2.93 \pm 0.74^{b}$
UR22		$0.44\pm0.07^{ m b}$	$6.10\pm1.50^{b}$	$1.01\pm0.13^{b}$	$5.50\pm0.30^{a}$	$0.67 \pm 0.09^{b}$	$22.75\pm7.42^{a}$	$2.48 \pm 0.96^{\rm b}$
UC		$0.71\pm0.07^{a}$	$15.10\pm 2.33^{a}$	$0.59 \pm 0.03^{a}$	$4.60\pm0.28^{a}$	$1.35\pm0.12^{a}$	$23.08\pm4.49^{a}$	$4.30 \pm 1.03^{a}$
Total		$0.54 \pm 0.03$	$9.21 \pm 1.75$	$0.74 \pm 0.06$	$4.68 \pm 0.23$	$0.87 \pm 0.08$	$23.84 \pm 5.15$	$3.19 \pm 1.03$
p-value		$0.011^{*}$	$0.000^{**}$	$0.019^{**}$	$0.227^{ns}$	$0.001^{**}$	0.901 <sup>ns</sup>	0.019*
SR20	Solid	$0.51\pm0.07^{\rm f}$	$17.40\pm 2.50^{g}$	$2.20 \pm 0.30^{\rm f}$	$7.80 \pm 0.85^{\rm f}$	$3.30 \pm 0.55^{f}$	$19.60\pm 3.50^{\rm f}$	$3.00 \pm 0.30^{\rm f}$
SR21		$0.72\pm0.11^{\mathrm{f}}$	$20.50 \pm 9.80^{g}$	$2.50 \pm 0.49^{f}$	$9.20 \pm 1.00^{\rm f}$	$3.70 \pm 0.70^{f}$	$33.70 \pm 3.80^{\mathrm{f}}$	$3.40 \pm 0.55^{\rm f}$
SR 22		$1.03\pm0.18^{\rm ef}$	$29.50\pm 2.20^{f}$	$4.50 \pm 0.80^{\circ}$	$11.00 \pm 1.90^{ef}$	$3.93\pm0.82^{\rm ef}$	$42.80 \pm 4.00^{\circ}$	$4.20\pm0.84^{\mathrm{ef}}$
sc		$1.20\pm0.22^{\rm e}$	$34.20\pm 2.90^{\circ}$	$4.10 \pm 0.78^{\circ}$	$15.50 \pm 1.22^{\circ}$	$4.50\pm0.88^{\rm e}$	43.00±4.39°	$5.10 \pm 0.90^{\circ}$
Total		$0.70 \pm 0.06$	$19.80 \pm 2.80$	$2.10 \pm 0.63$	$7.80 \pm 1.07$	$3.50 \pm 0.72$	$27.40 \pm 3.30$	$3.80 \pm 0.40$
p-value		0.028*	$0.002^{**}$	0.053 *	0.019*	0.006*	$0.030^{*}$	$0.002^{**}$
S - State Forest solid homogeneous group	fertilization, U - Univer s under liquid fertilizatio	sity novel liquid fe on, 'e, f' and 'g' de	ertilization, R - nov enote homogeneou	el growing media, s groups under sol	C - controls growin id fertilization. $p = 0$	ng medium (peat-J 0.05	perlite). alphabets '	a' and 'b' denote

lation in beech seedlings. In the leaves, the UC liquid fertilizer treatment performed best. For solid fertilizers, the SC treatment demonstrated the highest levels of all tested macroelements. ANOVA result showed significant differences in the nutrient content of roots and leaves, especially under solid fertilizers (Table 6).

The comparative analysis of the average allocation of various elements in photosynthetic (leaves) and non-photosynthetic (shoots and roots) organs of oak and beech seedlings established that the allocation of nutrients varied significantly within and between both species. These results suggest that oak seedlings have a higher overall demand for or efficiency in absorbing nutrient, and that they allocate more resources to growth and development compared to beech seedlings, especially in roots, which play a key role in the adaptive process of crops after planting. The response to different treatments also highlights the species-specific adaptations and needs for nutrients. Oak generally showed a higher allocation of nutrient in its roots compared to beech, suggesting that the oak roots possessed a higher nutrient uptake and storage capacity than beech. Overall, the SC solid fertilizer treatment and the UR22 liquid fertilizer treatment consistently showed superior performance across both species and different plant organs.

#### **Correlation analysis of nutrient components**

The correlation analysis of nutrient components revealed a linear relationship among the assessed variables. The correlation coefficient between carbon and biomass shows a perfect positive relationship, meaning that biomass was highly related to the carbon and other element content of the samples, indicating that higher allocation of these elements was associated with higher biomass. Significant positive relationships were shown to also exist between nitrogen and other elements. This could imply that different treatments resulted in different patterns of nutrient allocation in seedlings of oak and beech. This trend suggests a strong positive relationship between the treatment applied and the allocation metric may continue beyond the nursery growth (Table 7).

### Result of principal component analysis of Q. robur, the F. sylvatica

The results of the PCA showed the distribution of macroelement allocation in the root system of oak and beech seedlings under different fertilizer treatments. The influence of growing medium was less significant, as illustrated in Figs. 3 and 4. Here, the macroelement composition in the roots of both oak and beech seedlings appeared consistent across different growing medium, with points clustered closely together. This implies that variation in growing medium had minimal impact on the macroelement profiles in the roots (Figs. 3 and 4). In contrast, the impact of UR and SR fertilizers was particularly noticeable, as shown by the diverging points in Fig. 5. This indicated a significant effect of fertilization on nutrient content for both species, with the points being spatially separated. The variation observed in these graphs suggests that the primary factor influencing growth is the type of fertilizer treatment rather than the growing medium treatment.

	С	N	P	K	S	Ca	Mg
Oak							
Ν	$0.649^{**}$						
Р	$0.727^{**}$	$0.890^{**}$					
Κ	$0.899^{**}$	$0.857^{**}$	0.910**				
S	$0.754^{**}$	$0.959^{**}$	$0.895^{**}$	$0.905^{**}$			
Ca	0.339**	$0.447^{**}$	0.436**	$0.495^{**}$	$0.518^{**}$		
Mg	$0.791^{**}$	$0.797^{**}$	0.735**	$0.847^{**}$	0.843**	$0.590^{**}$	
Biomass	$0.914^{**}$	$0.607^{**}$	$0.700^{**}$	0.839**	$0.687^{**}$	$0.288^{**}$	$0.729^{**}$
Beech							
Ν	$0.770^{**}$						
Р	$0.829^{**}$	0.753**					
K	$0.925^{**}$	0.841**	0.873**				
S	$0.862^{**}$	0.938**	0.831**	0.933**			
Ca	0.491**	$0.608^{**}$	$0.590^{**}$	$0.579^{**}$	0.624**		
Mg	$0.860^{**}$	0.913**	$0.818^{**}$	$0.908^{**}$	$0.956^{**}$	$0.669^{**}$	
Biomass	$0.997^{**}$	$0.783^{**}$	0.833**	$0.932^{**}$	$0.872^{**}$	$0.489^{**}$	$0.876^{**}$

 Table 7 Correlation analysis between chemical elements and biomass

\*\* Correlation is significant at the 0.01 level





# Discussion

The results of this study provide information on how different growing medium treatments affect the growth (biomass) and nutrient allocation (macroelements) in and within oak and beech seedlings after production in the nursery (just before establishing them as a crop). This pilot study provides some preliminary evidence that innovative peat-free growing media and liquid fertilizers can result in an improved nutrient status of *Q. robur* and *F. sylvatica* seedlings, with the UR22 treatment being particularly promising in this case. Although the findings provide some evidence of potential benefits for nursery management in relation to reduced peat use. In essence, the aboveground characteristics of the seedlings cultivated on the peat-free growing medium, coupled with the liquid fertilizer developed by the University of Agriculture in Kraków, were in every respect close to those of the seedlings grown



Fig. 5 Nutrient allocation in oak and beech root grown on different fertilization methods

on the state forest growing medium comprising peat and solid fertilizer and these supports the earlier stated hypothesis. The novelty of this research is the examination of sustainable peat substitutes for use in forestry nurseries.

The distribution of plant biomass has been identified as a crucial factor that aids allocation of nutrients in plant organs (Rumpf et al. 2011; Meiwes et al. 2012; Freschet et al. 2015; Husmann et al. 2018; Klimešová et al. 2018; Yue et al. 2021). The importance of nitrogen and phosphorus partitioning between plant organs as a critical factor in regulating growth rate has been highlighted in previous studies (Laliberté et al. 2012; Minden and Kleyer 2014; Tang et al. 2018; Malhotra et al. 2018; Zhang et al. 2018; Yang et al. 2021) The results of this study show contrasting trends in nutrient allocation between nitrogen and phosphorus as both were influenced from negative side of distribution to positive as a result of the effect of fertilization. A similar divergence in nutrient availability was observed by Aoyagi and Kitayama (2016) in their investigation of nutrient allocation between plant organs in Bornean rainforests. In contrast to the findings of Zhao et al. (2020) that nutrient allocation among organs of tree species is intimately related to it demand, the application of this novel fertilizer, as depicted in Fig. 5, appears to be markedly different. The storage of carbon and nutrients in long-lived organs, such as shoots and roots, is considered essential to compensate for biomass losses due to fallen leaves and branches (Aoyagi and Kitayama 2016), or to natural enemies (Fricke et al. 2014; Comita and stump 2020) It is important to note that although the role of shoot phosphorus is less well established in oak, the shoot could potentially act as a storage organ, as previously suggested by Sardans and Peñuelas (2015).

Although the allocation of C, N and P in the studied species recorded higher values than those reported by Aoyagi and Kitayama (2016) However, assessment of nutrient allocation in the studied organs are similar with the values reported by Wei et al. 2013; Pająk et al. 2022a, 2022b; Marušić et al. 2023. The lower concentrations of elements especially, N,P, K and Mg observed in leaves agrees with the study of Hidaka and Kitayama (2011) indicating a withdrawal of nutrient into stable organs, while the calcium content was high because it is immobile in leaves (Peuke and Rennenberg 2011; Loewe-Muñoz et al. 2024). As observed in this study, the seedlings performed well on R22 growing medium, this could be as a result of the addition of straw in the formulation which has been reported to be rich in nutrient content (Xie et al. 2012; Viera et al. 2017; Liu et al. 2018; Guan et al. 2020) Beech seedlings have been reported to grow better in media rich in calcium, magnesium, and potassium (Pająk et al. 2022b). Moreover, Balcar et al. (2011) used dolomite (containing calcium and magnesium) to fertilize beech trees in plantations, and reported a positive effect on their survival and growth.

Variety in nutrient allocation was not only found to exist between the tree species, but also within different organs of each species. This concurs with various investigations on European beech and pedunculate oak (Poorter et al. 2012a, b; André 2010, Pretzsch 2014, Husmann et al. 2018). Unlike beech, oak displayed essentially higher nutritional accumulation in the roots, shoots, and leaves. This study also re-established that nutrient allocation is generally higher in below ground organ than those that are above ground and this applied to the two species (Haase & Jacobs 2013, Liu et al. 2016). Prominently, nutrient response efficiencies varied significantly among the studied species, with treatment on R22 of novel growing medium and UAK fertilizer formulation accumulating more nutrients in the root. This corroborates previous studies (Freschet et al. 2015; Husmann et al. 2018; Klimešová et al. 2018; Rumpf et al. 2011; Meiwes et al. 2012; Pająk et al. 2022b; Rotowa et al. 2023b) that focused solely on the nutrient content of aboveground and belowground biomass.

The different growing media exhibited significant effects on the biomass characteristics of seedlings, especially in oak. This agrees with other studies that have highlighted the influence of peat growing medium on nutrient contents. For example, Pająk et al. (2022a) reported higher macronutrient allocation in root of European beech seedlings grown on differently compacted peat growing medium in a container nurseryRotowa et al. (2023b) reported the influence of various growing medium and fertilizers on the root systems of oak and beech. In addition, Banach et al. (2013) experiment conducted on *F. sylvatica* and *Abies alba* seedlings in a sawdust–peat growing medium, finding that well-aerated growing medium are essential for good growth in beech seedlings. Additionally, Freschet et al. (2015) reported that nutrient allocation additionally relied on the quality of the medium on which they were grown. Rotowa et al. (2023b) in a study carried out on the root systems of the same seedlings, reported treatment UR20 to be best in root morphology and characterization. In this study, however, the treatment UR22 showed the best performance in nutrient allocation in the roots of both species' seedlings. This therefore implies that peat-free growing medium with the recommended dosage of fertilizer (Pająk et al. 2022a; Jasik et al. 2023;

Rotowa et al. 2023b) could be a viable alternative to traditional peat-based medium, offering a sustainable approach to seedling cultivation in forest nurseries.

# Conclusions

The confirmation of the earlier-stated hypotheses highlights the efficacy of the peat-free organic growing medium for supporting the growth of Q. robur and F. sylvatica seedlings in the nursery. The first hypothesis, predicting an improvement in nutrient allocation across different parts (leaves, shoots, and roots) of the seedlings with the novel growing medium and fertilizer, was confirmed. According to the PCA results, the utilization of the novel growing medium and liquid fertilizer indeed resulted in an efficient distribution of nutrients in a way that was not worse than the traditional practice within individual crops. Moreover, the study validated that the uptake of macroelements in both species under the improved root system was comparable and in some cases better than those grown on a standard growing medium (peat plus solid fertilizer). This affirmation emphasizes the potential of the novel growing medium and fertilizer.

Based on the trends observed in this pilot study, the use of peat-free organic growing media and fertilizers developed by the University of Agriculture in Kraków shows potential to support nutrient allocation and growth in pedunculate oak and European beech seedlings at the end of the growing season in the nursery. Significant variations were evident across the different novel growing media treatments especially UR22. Moreover, the use of liquid fertilizer also formulated by the University of Agriculture in Kraków, for the needs of the seedlings, further supports nutrient allocation in various organs. Our concept of growing media and novel liquid fertilizers has potential to provide a new approach of growing tree seedlings for establishing forest plantations in Europe. Furthermore, it may offer a sustainable alternative to peat growing media in the long run.

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Data availability No datasets were generated or analysed during the current study.

# Declarations

Competing interests The authors declare no competing interests.

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