Nutrition Intensity in Ternary Diagrams Interpretation for Some Ornamental Species Cultivated on Organic Substrate with Increased Biological Activity

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Abstract

Nowadays, many biodegradable organic wastes no longer need to represent an environmental hazard and as a consequence, they could be recycled to obtain horticultural substrates. An experiment was conducted on two deciduous (*Tamarix tetrandra*, *Ligustrum ovalifolium* 'Aureum') and two coniferous species (*Chamaecyparis pisifera* 'Boulevard', *Chamaecyparis lawsoniana* 'Stardust') grown on a soil mixture of forestry compost, leaves compost, peat and grape marc compost. The aim of the research was to investigate the response to fertilization and to obtain valuable information regarding absorption rate of nutritive elements during vegetation. Nitrogen data show a lowering of its nutritive equilibrium point towards autumn in the leaves of deciduous shrubs species. Resorption of nutrients before leaves fall occurs due to conservation process in woody plants with deciduous leaves. In the case of coniferous species the lowering of nitrogen content is less intense. The monthly fertilization with Coïc solution indicates no influence on nitrogen metabolism of this species. The exception was on *Ligustrum* with differences between nitrogen content in fertilized and unfertilized plants. The phosphorus nutritive equilibrium point reveals a decrease during the summer months (July - August), the species presenting the lowest values in this period and the cause of this behaviour was the plants adaptation to high temperature and low humidity. Potassium nutritive equilibrium data indicates small differences in the unfertilized plants in comparison with those fertilized in all species. The novelty of the research is represented by the ternary diagrams N-P-K that were constructed, interpreted and reported for dendrologic species.

Keywords: dendrologic species, N-P-K diagrams, substrate, grape marc compost

Introduction

In European Union countries, where recycling and reintroduction of numerous waste vegetal circuits is a widespread activity, compost marketing is regulated by several laws. Composts for horticultural market are labelled mandatory wearing explanation of the origin, the components of the substrate, as any natural or synthetic materials, organic or mineral elements that may alter the mechanical, physical, chemical or biological character of the substrate. Feasibility of mixtures of substrates is virtually numerous, but substrates characterized by phytotoxicity, instability, high cost or limited availability are constantly rejected.

For intensive horticulture is mandatory to assure maximum quality and to use environmental friendly methods. A literature survey concerning different substrates used in horticulture revealed that until at a certain moment, the cultures on growing media have been carried out with a limited number of substrates (peat, rockwool, glasswool, perlite, vermiculite, charcoal chips) (Olle *et al.*, 2012; Restrepo *et al.*, 2013; Santiago *et al.*, 1989).

Lately, people are keen on the research and development of ecologically friendly substrates, which recently have been proven as a viable alternative to other traditional methods.

Nowadays, there is a growing interest in the exploitation of the residues generated by wine industry. Grape marc (waste skins, seeds and stalks), a valuable waste product in wine producing regions, is generated from the crushing, draining and pressing stages of the wine production, and contains approximately 8% seeds, 10% stems, 25% skins, 57% pulp (Laurenson *et al.*, 2012). There are researches that

present the effects of composted grape marc as substrate for ornamental plants (Chen *et al.*, 1988), vegetables (Baran *et al.*, 2001; Carmona *et al.*, 2012; Leoni *et al.*, 1992). Therefore, identifying new materials suitable to obtain substrates for horticultural purposes is an important requirement. Grape marc composts possess chemical particularities that are responsible for fertility improvement (high contents of potassium and organic matter and significant contents of nitrogen and phosphorus) (Bustamante *et al.*, 2005, 2007, 2010), but there are some environmental problems related with its acidic character, high content of phenolic compounds and with phytotoxic and antimicrobial effects (Carmona *et al.*, 2012). Sometimes, substrates used for different ornamental species are insufficiently rich in nutrients. Providing ornamental plant nutrients can be handled by applying the fertilizer in the form of liquid or solid substrate.

Having in view that many biodegradable organic wastes can be recycled to ensure sustainability, this being an inexpensive alternative to obtain horticulture substrates, an experiment was designed and focused on four dendrologic species grown on a substrate composed of forestry compost, leaves compost, peat and grape marc compost, with the aim to investigate the plants' response to fertilization and to obtain ternary diagrams (N-P-K) that will provide information suitable to interpret species particularities, the absorption rate of nutritive elements during vegetation period.

Materials and methods

Substrate composition and plant material

The experiments were set up at the greenhouse from University of Agronomic Sciences and Veterinary Medicine Bucharest. Four dendrologic species were planted in vegetation pots (8 L capacity) that contained substrate formulated according to the recipe described below.

The organic materials used for the substrate composition were forestry compost (FC), leaves compost (LC), peat (P) and grape marc compost (GM) according to volumetric ratio FC: LC: P: GM = 1:1:1:0.5. The disinfection of the substrate components for pathogen destruction was made at 55 °C.

There were analyzed two deciduous species (*Tamarix tetrandra*, *Ligustrum ovalifolium* 'Aureum') and two coniferous species (*Chamaecyparis pisifera* 'Boulevard', *Chamaecyparis lawsoniana* 'Stardust'). During vegetation period it were performed the cutting of the branches to uniform the plants (in the middle of May), watering, mechanic weed control and application of phytosanitary treatments. In the experiments, it was used Coïc acidophile solution (Davidescu *et al.*, 2001) as source of macro- and microelements.

In June, the substrate was characterized from agrochemical point of view and it has been applied as the first fertilization treatment with Coïc acidophile solution, for a part from potted plants. At 30 days (July) the substrate was analyzed after fertilizer has been applied, the operation being repeated another 2 times (August, September).

Substrate agrochemical characterization and plant analysis

The substrate analyses were performed at the beginning of the experiment. The agrochemical characterization of the substrate was made by assessment of pH, EC (electrical conductivity), soluble forms (1:10, v/v ratio) as nitrate, ammonium, phosphorus, potassium and sodium (Tab. 1). Also, mobile phosphorus and exchangeable forms of main cations (K^+, Na^+, Ca^{+2}, Mg^{+2}) were determined in CH_3COONH_4 0.5M, pH=4.65, 1:3 v/v ratio (Tab. 2).

During the experiment at four moments (M1 – June, M2 – July, M3 – August, M4 – September) in order to establish the absorption rate of nutritive ions, for each plant species it were achieved total forms of nitrogen, phosphorus and potassium, using acknowledged methods (see Analytical methods and techniques section) (Tab. 3).

The nutritive equilibrium is given by the ratio of each element (N, P, K) expressed as percent against the sum of their concentrations, as it is presented below:

$$\text{En}_N = \frac{\%N}{\sum N,P,K} \cdot 100 \, ; \quad \text{En}_P = \frac{\%P}{\sum N,P,K} \cdot 100 \, ;$$

$$\text{En}_K = \frac{\%K}{\sum N,P,K} \cdot 100 \, .$$

Biotic and enzymatic characterization of substrates

The biotic tests carried out on substrates have investigated the respiration and cellulolytic activity (Dashtaban *et al.*, 2010). Enzymatic tests were based on sugar hydrolysis (saccharase activity) (Balezentiene *et al.*, 2009; Shi *et al.*, 2008), urea hydrolysis (urease activity) (Shi *et al.*, 2008) and phosphorus hydrolysis from organic compounds (phosphatase activity) (Balezentiene *et al.*, 2009; Shi *et al.*, 2008). In addition, catalase activity (Shi *et al.*, 2008) was determined in order to obtain information concerning oxidation processes from substrates.

Analytical methods and techniques

Analytical grade chemicals were used throughout the study without any further purification. The pH values were carried out through potentiometric method, meanwhile EC (electrical conductivity) was determined using conductivity method. Soluble and total phosphorus contents were evaluated by spectrophotometric molybdenum blue method. The levels of nitrate and ammonium species were assessed by spectrophotometric means, also using phenoldisulfonic acid and Nessler reagent, respectively. Total form of nitrogen was determined using Kjeldahl method, meanwhile potassium and sodium contents were assessed using atomic emission spectrometry.

Calcium and magnesium concentrations were determined through flame atomic absorption spectrometry (FAAS). Measurements were carried out using a Thermo Scientific AA Spectrometer. Selected wavelengths were 422.7 nm for calcium and 285.2 nm for magnesium. The calibration curves for calcium and magnesium are linear for the studied ranges and were plotted by running different concentrations of standard solutions prepared from stock solutions of 1000 mg/L calcium and magnesium, respectively.

Results and discussions

Substrate characterization

The main agrochemical characteristics of the substrates are presented in Tabs. 1 and 2. Also, biotic and enzymatic tests were carried out for substrates and the results are presented in Tab. 3.

Respiration activity indicates the global biological activity of a substrate and the obtained value for this biotic test was 93.36 mg CO_2/100 g substrate (d.w.). Cellulolytic potential of the substrate is correlated with organic matter, the latest one representing the substrate for cellulolytic microflora. Catalase and saccharase activities are related with high organic matter content of the substrates. The urease potential of the analyzed substrate is high (662 mg NH_4^+/100 g substrate), this indicating the possibility of ammonia loses through volatilisation.

Phosphatase activity of the substrate is quantified through phosphorus that is released enzymatic and the obtained value is 5.72 mg P/100 g substrate (d.w.). The increased biological activity is due to the high C/N ratio (20.8-26.85) in comparison with humus level (10-12%), this indicating that organic matter is still decomposing.

Nutritive equilibrium

The establishment of nutritive equilibrium was done using the method based on the global nutrition calculation, according to experimental analysis performed during the vegetation period. The global nutrition is the sum of N, P, K concentrations (as total forms) expressed as percents from dry matter. The values obtained after plant analysis during vegetation period at certain moments are summarized in Tab. 4.

The interpretation was expressed through ternary diagram N-P-K using a triangular system. For each of the dendrologic specie that were subjects of the experiment, it were constructed ternary diagrams (N-P-K) that allowed to interpret the species particularities, the absorption rate of nutritive elements for fertilized and unfertilized variant.

The ternary diagrams with highlighted theoretic nutritive equilibrium obtained for *Tamarix tetrandra*, *Ligustrum ovalifolium* 'Aureum', *Chamaecyparis pisifera* 'Boulevard', *Chamaecyparis lawsoniana* 'Stardust' represent a new edge concerning nutritive equilibrium of N, P, K for ornamental species. Also, the values for nutritive equilibrium of the same elements after plant analysis were evidenced.

Nutritive equilibrium for Tamarix tetrandra

The points for nutritive equilibrium resulted for unfertilized variant *Tamarix tetrandra* describe the equilibrium area with trilinear coordinates for N between 48.36% and 61.67%, for phosphorus between 3.91% and 14.02% and for potassium between 24.32% and 40.46%.

The analysis revealed that at the beginning of the vegetation period the nitrogen absorption was higher and decreased as dry matter accumulation increased. The phosphorus absorption is intense at the beginning, and decrease to a third, during vegetation period, so that at the end to increase at values comparable with those recorded at vegetation start stage. Potassium absorption increase slightly during vegetation period as it is shown by the equilibrium points resulted from En_K coordinates (Fig. 1, a).

Equilibrium points resulted from the variant fertilized with acidophile Coïc solution marks the nutrient equilibrium for species *Tamarix tetrandra* by a shift of equilibrium in behalf of N absorption and less intense for P and K compared to the unfertilized (control) variant (Fig. 1, a).

Nutritive equilibrium for Ligustrum ovalifolium

Ligustrum ovalifolium equilibrium points obtained from the unfertilized variant describe the nutritive equilibrium area with trilinear coordinates for En_N between 41.43% and 57.13%, for En_P between 3.62% and 14.38% and for En_K between 32.87% and 50.50%. Nitrogen uptake is more intense at the early vegetation stage, then has an equilibrium plateau and finally decreases slightly.

Phosphorus uptake is intense in June and then decreases in July and August about three times; in early September it was observed the increase of the absorption intensity of phosphorus, which exceeds the values recorded at the beginning of the vegetation period.

During the vegetation, potassium absorption is intense and constantly increases, with highest values in July, until it records a slight decline marked by a stable balance at the end of the vegetation stage (Fig. 1, b).

Equilibrium area delimited by equilibrium points in fertilized variants exhibits a constant absorption of phosphorus, significant variation of nitrogen absorption intensity that is intense at the beginning of vegetation stage, but decreases as dry matter accumulate and grows again in the following stages. Potassium absorption intensity shows large increases to a maximum and decreases at the end of the vegetation period.

Nutritive equilibrium for Chamaecyparis pisifera 'Boulevard'

The equilibrium areas for *Chamaecyparis pisifera* 'Boulevard' bounded by the equilibrium points resulted after data calculation, presents differences between unfertilized and fertilized variants. It was observed that the uptake of N was more intense at the early vegetation period for unfertilized variant in comparison with the fertilized one. The absorption intensity values for P are undifferentiated by fertilization and constant for K at fertilized variant, meanwhile the unfertilized variant it registered an increasing trend. The coordinates for ternary diagram take values between 46.86% and 57.75% for En_N, 4.40% and 12.49% for En_P and 37.38% and 46.06% for En_K (Fig. 1, c).

Nutritive equilibrium for Chamaecyparis lawsoniana 'Stardust'

The equilibrium areas delimited by the equilibrium points shows that between unfertilized and fertilized variants there are no significant differences for *Chamaecyparis lawsoniana* 'Stardust'; the areas are almost overlapped, this behavior indicating that fertilization does not influence the absorption of nutritive elements N-P-K. The coordinates for nutritive equilibrium for unfertilized variant (as control), vary between 56.42% and 72.85% for En_N, 5.10% and 15.32% for En_P and between 13.05% and 38.20% for En_K (Fig. 1, d).

Tab. 1. The agrochemical characterization (soluble forms) of the substrates with potted plants

Substrate with potted plants	pH	EC dS·m⁻¹	PO₄³⁻ ppm	NO₃⁻ ppm	NH₄⁺ ppm	Ca²⁺ ppm	Mg²⁺ ppm	K⁺ ppm	Na⁺ ppm
Tamarix tetrandra	7.83	0.1359	22.00	0.50	25.00	112.23	13.693	140	20
Ligustrum ovalifolium 'Aureum'	8.14	0.2687	38.40	trace	23.00	163.24	24.604	290	17
Chamaecyparis pisifera 'Boulevard'	8.03	0.1468	42.40	4.00	23.00	193.85	24.192	145	11
Chamaecyparis lawsoniana 'Stardust'	7.80	0.1037	29.80	3.00	9.00	102.03	14.434	62	20

After chemical plant analysis at moments M1-M4 (June - September), there were obtained results that were compared with limits from literature data (Davidescu *et al.*, 2001). An overview based on the obtained results is detailed in Tab. 5. The studies of nutrition intensity of dendrologic species cultivated in substrate made of leaves compost, forestry compost, peat and grape marc compost (ratio 1: 1: 1: 0.5) reveals significant differences between deciduous (*Tamarix tetrandra, Ligustrum ovalifolium* 'Aureum') and coniferous species (*Chamaecyparis pisifera* 'Boulevard', *Chamaecyparis lawsoniana* 'Stardust'), but not significant differences in applied fertilizers as Coïc solution during vegetation period (June-September) in comparison with unfertilized plants for all studied species.

Discussion

Ternary diagrams are used in various areas (chemistry, geology, industry, biology, agriculture) and represent graphic tools that permit to evaluate the interaction between systems and the interactions between systems and environment (Almeida *et al.*, 2007; Giannetti *et al.*, 2006, 2013), allow the estimation of nutrients balance in soil (Parent *et al.*, 2012), visualization of relation among the three humus fractions (Watanabe *et al.*, 1991) or variation of sugar composition in nectars (Herrera *et al.*, 2006). In literature, there is lack of information concerning ternary diagrams correlated with nutritive equilibrium in the case of dendrology species. Notwithstanding, some similar researches have been presented for potato (Thomas, 1938).

The data obtained in this experiment conducted with the aim to evaluate the absorption rate of nutritive elements during vegetation period for different dendrologic species, allowed us to construct ternary diagrams N-P-K. The four studied species belonging to the two different groups of deciduous and coniferous trees, with distinct metabolism and diverse biological particularities, have responded different to the application of fertilizers. Their behaviour after nutritive

Tab. 2. Mobile phosphorus and exchangeable forms of main cations in substrate with potted plants

Substrate with potted plants	P (ppm)	Ca (mE/100g)	Mg (mE/100g)	K (mE/100g)	Na (mE/100g)
Tamarix tetrandra	262.00	59.05	2.566	0.910	0.1413
Ligustrum ovalifolium 'Aureum'	278.00	52.94	2.553	1.028	0.1217
Chamaecyparis pisifera 'Boulevard'	162.75	42.76	2.027	0.910	0.1000
Chamaecyparis lawsoniana 'Stardust'	290.00	54.47	2.537	0.769	0.1300

Tab. 3. Biotic and enzymatic tests developed on substrate

| Biotic tests		Enzymatic tests			
Respiration activity (mg CO₂/100 g)	Cellulolytic activity (g cellulose/ 100 g cotton)	Catalase activity (cm³ O₂/ 100 g)	Saccharase activity (mg saccharose/ 100 g)	Urease activity (mg NH₄⁺/ 100 g)	Phosphatase activity (mg P/ 100 g)
93.36	76.20	953	2104	662	5.72

Tab. 4. Total forms of nutritive elements after plant analysis

Var	June			July			August			September		
	Total forms (%)											
	N	P	K	N	P	K	N	P	K	N	P	K
V1	4.1850	0.952	1.65	3.2517	0.218	2.10	2.5542	0.174	1.65	2.8686	0.663	2.40
V2	4.2734	0.872	1.50	3.2223	0.244	1.95	2.6132	0.168	1.50	2.7900	0.625	2.00
V3	3.3893	0.593	1.95	3.1339	0.247	3.45	2.9472	0.214	2.60	2.4265	0.844	2.60
V4	3.1339	0.614	1.70	2.2497	0.236	3.40	2.7212	0.235	2.65	2.6819	0.718	2.40
V5	2.2399	0.189	1.45	2.1514	0.191	2.00	2.3086	0.189	1.50	2.1907	0.582	1.90
V6	2.1908	0.171	1.60	2.2006	0.190	1.90	2.2890	0.156	1.90	2.3784	0.560	1.95
V7	2.7900	0.540	0.50	2.1416	0.204	1.45	2.4953	0.204	1.30	2.3086	0.653	1.30
V8	3.0651	0.558	0.65	2.1907	0.200	1.50	2.5542	0.191	1.25	2.3577	0.561	1.35

V1 - *Tamarix tetrandra* (unfertilized); V2 - *Tamarix tetrandra* (fertilized); V3 - *Ligustrum ovalifolium* (unfertilized); V4 - *Ligustrum ovalifolium* (fertilized); V5 - *Chamaecyparis pisifera* 'Boulevard' (unfertilized); V6 - *Chamaecyparis pisifera* 'Boulevard' (fertilized); V7 - *Chamaecyparis lawsoniana* 'Stardust' (unfertilized); V8 - *Chamaecyparis lawsoniana* 'Stardust' (fertilized)

Tab. 5. General overview on nutritive equilibrium for ornamental species

| Var.	Specie	June E_n (%)			July E_n (%)			August E_n (%)			September E_n (%)		
		N	P	K	N	P	K	N	P	K	N	P	K
V1	*Tamarix tetrandra*												
V2	*Tamarix tetrandra* (fertilized)												
V3	*Ligustrum ovalifolium*												
V4	*Ligustrum ovalifolium* (fertilized)												
V5	*Chamaecyparis* 'Boulevard'												
V6	*Chamaecyparis* 'Boulevard' (fertilized)												
V7	*Chamaecyparis* 'Stardust'												
V8	*Chamaecyparis* 'Stardust' (fertilized)												

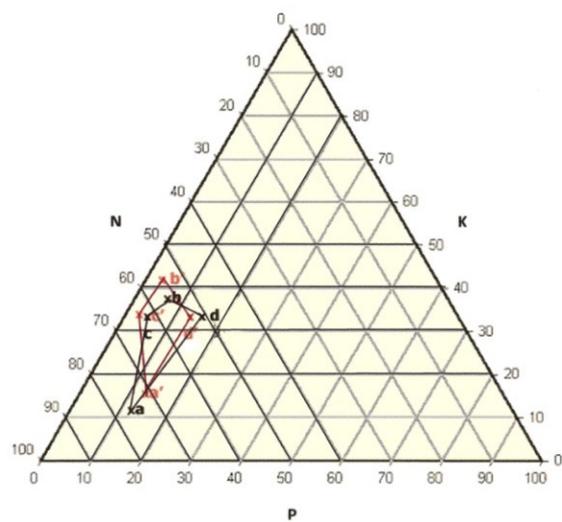Legend: very low ; low ; normal ; high ; very high .
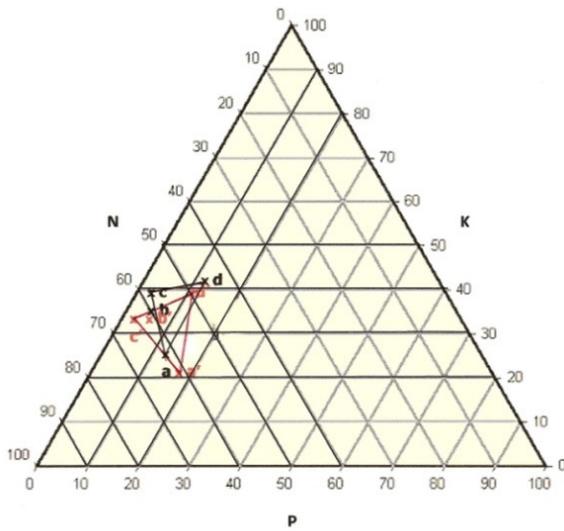
a) Ternary diagram N-P-K for *Tamarix tetrandra*
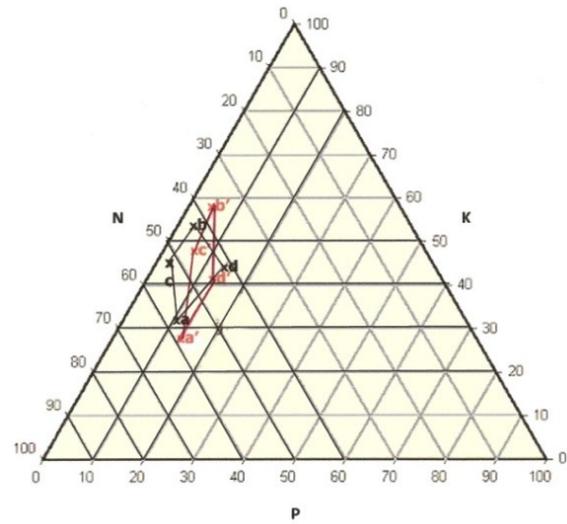
b) Ternary diagram N-P-K for *Ligustrum ovalifolium* 'Aureum'
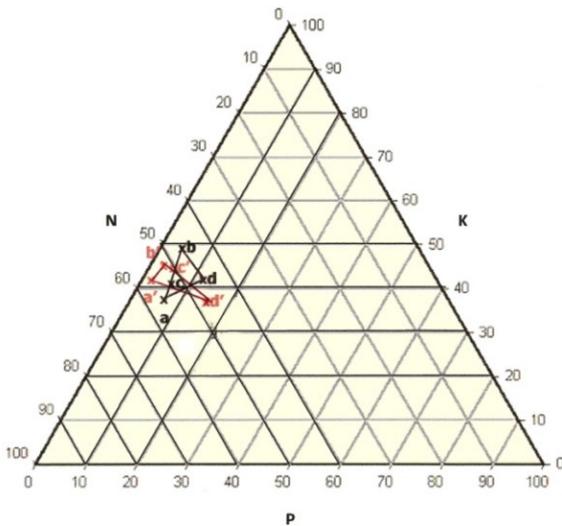
c) Ternary diagram N-P-K for *Chamaecyparis pisifera* 'Boulevard'

d) Ternary diagram N-P-K for *Chamaecyparis lawsoniana* 'Stardust'

Fig. 1. Ternary diagram for studied dendrologic species (black line - unfertilized; red line - fertilized)

solution application on the same substrate is different concerning nutrient absorption capacity, nutrient balance during the vegetation period and the relationship between species, enzyme substrate and biotic indicators of used substrate.

Equilibrium points represented in the ternary diagrams N-P-K resulted from fertilized variant with acidophile Coïc solution, marks the nutrient equilibrium area for specie *Tamarix tetrandra* by a shift of nutrient balance for the absorption of N and less intense for P and K compared to the control variant.

The equilibrium area delimited by the points of equilibrium for *Ligustrum ovalifolium* 'Aureum' in the case of fertilized variants present constant absorption of P, large variations of the intensity of N absorption that is intense at the beginning of the vegetation period and decrease as the dry matter accumulate, then increase again during the rest of the following period. The intensity of K absorption shows large increases up to a maxima and then decreases at the end of vegetation period.

The equilibrium areas for *Chamaecyparis pisifera* 'Boulevard' bounded by the equilibrium points resulted after calculation shows differences between unfertilized and fertilized variants with more intense consumption at the beginning of the vegetation period for N at unfertilized variant in comparison with the fertilized one. The values that describe the intensity of the P absorption are undifferentiated from fertilization and are constant for K at fertilized variant, with increasing values for unfertilized variant.

For *Chamaecyparis lawsoniana* 'Stardust' there is no differences between fertilized and unfertilized plants related with the nutrients absorption, the fertilization leading to an undetectable influence. On the other hand, there is a higher difference of nutritive elements absorption intensity in comparison with *Chamaecyparis pisifera* 'Boulevard'.

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