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Biology

STUDY OF SOME BIOLOGICAL INDICATORS IN DIFFERENT SYSTEMS OF VEGETATION MANAGEMENT OF OLIVE

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Abstract. The study was carried out over a 9-year period in a 40-year old olive grove of the Frantoio cultivar. In an experimental orchard at Olive Research Institute the use of mulching straw effectively suppressed weed competition, conserved soil moisture for a long period of time and distinctly increased the productivity of olive trees compared with other treatments. Five treatments were experimented as follows: cover crop, untreated control, non-selective herbicide – glyphosate, straw mulch, and ploughing. Statistical analysis has tested the degree of variability and the regressive tendency among the factors. A mulching treatment could replace in the future the use of glyphosate that has also shown good results in controlling weeds in a conventional production system. Research defned the appropriate and harmful levels of nitrogen and water, considered as important during the critical period of endocarp lignifcation. The biological processes were elaborated in strong correlation with the system of soil cultivation, thus highlighting mulching as more favourable for the olive soil. Yield as an expression of the tree metabolism was different and was infuenced by the systems which favoured more water and nitrogen reserves available for the trees.

Keywords: olive grove, ploughing, cultivar, nitrogen, endocarp.

AIMS AND BACKGROUND

The olive tree undergoes recycling processes for the fowering induction, vegetative growth, efforescence, fruiting and fruit growth as well as the synthesis of fatty acids. For these processes the tree needs optimum concentrations of water and minerals, especially nitrogen in the terrestrial solution[1,2]. The olive tree does not distinguish the difference whether nitrogen comes from an organic or mineral source. Nitrogen absorption is carried out through two forms: either nitrate (NO_3^-) or ammoniacal (NH_4^+), whereas organic fertilisers contain their own nitrogen bound with proteins, which through microbial actions are transformed into nitrate and ammoniacal absorbed by the trees[3–5]. On the other hand, the choice of using an organic fertiliser or an intercalary system aims to increase the content of nutrients

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in the terrestrial solution. Organic matter allows a slow and constant absorption, whereas mineral fertilisers are widely used as they can be easily absorbed[4]. Nitrogen (N) is the only common nutritional defciency in the olive tree, whereas other elements, such as: potassium (K), boron (B), phosphorus, calcium, zinc, copper, manganese and magnesium rarely lack in Albanian olive groves[2].

Nitrogen fxation was performed through several bacteria found on the roots of legumes, which proved to have increased the available nitrogen. On the other hand, the terrestrial solution must contain water at optimum levels where the indispensable minerals are dissolved. When water content is lower than the normal values, the trees undergo physiological diffculties, which interrupt or reduce life expectancy. Several scientifc studies/researchers have confrmed the importance of water and nitrogen in the biological and physiological processes of the olive. Under these circumstances this research diagnoses water and nitrogen in the leaves ashes, which is related to several olive cultivations systems. In the meantime it estimates the impact they have on oil quality and tree yield.

EXPERIMENTAL

The study was carried out over a 9-year period in a 40-year old olive grove of the Frantoio cultivar, situated in Shamogjin of the Vlora region. Five treatments were experimented as follows:

T_1 – cover crop: mixed legume and rye for winter growth. The soil was cultivated 100% during October–June with leguminous 350–400 plants/m^2.

T_2 – untreated control: natural meadow wasteland.

T_3 – non-selective herbicide – glyphosate: treatment of the soil twice with glyphosate concentration 1%, 0.1 l/m^2 solution.

T_4 – straw mulch: the soil is dug and covered with cereal straw, up to 10 cm all over the year.

T_5 – ploughing (black fallow): fallow of farmland ploughed and left for a one year period without being sown.

Each treatment plot, containing 25 olive trees, was replicated fve times in a randomised complete block design. An olive tree row served as a buffer between replications. The area of each treatment was 240 m^2. Experimental trees have isolation trees all over the four directions. The trees live in un-irrigated conditions. The entire experimental feld was tilled in the autumn, except the untreated control plot. The cover crop treatment was sown during October with rye and peas at 200 and 150 kg/ha, respectively. Data were collected during the vegetative grown of plants from February to June. On April 20th and May 10th, when most part of the weeds present were at 15–20 cm higher, a treatment with the non-selective herbicide Roundup (glyphosate 36%) at a rate of 5 l/ha using knapsack sprayer calibrated to deliver 400 l/ha, was applied. Straw mulch treatment, using wheat straw before germination of weeds, was applied during January. Examination of weeds for each block is realised during month of January and June. Ploughing treatment was done by tillage, between trees, using a three wheeled tractor and by hand under the olive trees to the depth of 20 cm.

Vegetative growth, phenology and yield indices were analysed using 200 shoots collected around olive tree canopies. Flowering and fruit set measurements were studied analysing 7000–8000 fowers from 10 trees/treatment distributed uniformly. The number of inforescences and numbers of fowers per inforescence were recorded. The number of fruit sets were observed and performed in July and September. Indices as total number of fruits and yield for each treatment, biometry of fruits, fruit weight, and % of olive oil content in fruits, dry matter and the acidity of olive oil were analysed during the harvest period. Oil percentage in the fruit was analysed using the Soxhlet method. *Defnition of acidity* (%) was realised using 5–10 ml oil, dissolved in 50–150 ml (petroleum ether + ethyl alcohol), adding some drops of phenolphthalein with alcoholic dip of 0.1 N KOH until the pink colour remains for 30 s. Diagnosis of N traces in the ashes of 500 burnt leaves was realised in three phases: during plant dormancy, in phase of efforescence and phase of endocarp ligneous. *Water content of leaves* was measured by the method of determining the humidity using a Denver Instrument; grinding 100 g of leaves and the paste obtained in 8–10 g, 5–10 min to 130°C, during dormancy, efforescence and endocarp ligneous[6]. All statistical analyses of the observed and measurement indices were realised using JMP software[7,8].

RESULTS AND DISCUSSION

GENERAL REVIEWS

The results related to indices measured are given in Table 1. The trees were grown and fed within the same soil, which resembles a reservoir that is flled up via precipitation, while emptied by the trees with their evaporation. The trees have consumed differently water and mineral solution of the soil. The different state of H_2O and N has created vegetative growth, blooming, fruiting and different yield per tree. The soil and trees were intimately related to carbon dioxide content CO_2, methane CH_4, and nitrogen in diatomic molecular form N_2 found in the atmosphere. Recycling of the three elements through the physiological processes of respiration and photosynthesis was different and under the infuence of treatment. The trees have absorbed ammonia NH_3 and nitrate ions NO_3^-. In T_2, T_3,T_4 and T_5, conversion of the atmospheric nitrogen N_2 was carried out via the bacteria dwelling in the soil, whereas in T_1, the process was favoured by specifc bacteria that live in symbiosis with leguminous plants, as of chemical reaction:

$$2N_2 + 3CH_2O + 3H_2O \rightarrow 4NH_3 + 3CO_2.$$

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Table 1. Descriptive statistical analysis of quantitative traits evaluated for 5 treatments of végétation management of olive

Treatment	Level							
	Veget. Growth	Flower Cluster	Mean (% fruiting)	Fruit weight (g)	Field (kg/tree)	Acidity (%)	Oil (%)	Oil (hl/ha)
Cover crop	9.4±0.50 c	12.7±025 c	3.6±0.16d	2.3±0.09 c	22.3±0.57 c	0.83±0.03 b	20.5±0.20 a	11.4±0.13c
Control	8.4±0.50 d	10.8±0.20 d	2.8±0.13d	2.1±0.12d	19.0±1.00d	1.13±0.04a	20.9±0.15a	9.9±0.10e
Gliphosate	9.0±0.15cd	12.4±0.15c	4.4±0.15c	2.3±0.15c	20.0±1.00e	0.78±0.03 b	21.3±0.20a	10.6±0.23 d
Straw mulch	14.9±0.17a	14.4±0.28 a	7.6±0.26 a	2.6±0.18a	37.6±1.52a	0.5±0.08 d	21.1±0.60a	19.8±0.20 a
Black fallow	10.8±0.20b	13.2±0.14b	5.0±0.12b	2.4±0.08b	29.0±1.00b	0.63±0.02 c	20.7±0.30 a	15.0±0.20b

Diagnosing of nitrogen content. The trees absorbed nitrogen to produce proteins and nucleic acids, for the vegetative and reproductive processes. Nitrifcation of the organic matter transformed the products NH_4^+ in nitrate and nitrite, oxidation reaction through enzyme catalysis in the agrobacterium of leguminous plants, as well as those of terrestrial solution as of chain reaction: $NH_4^+ \leftrightarrow NO_2^- \leftrightarrow NO_3^-$. Treatments T_4 and T_1, stimulated more denitrifcation processes, conversion of air nitrogen into molecular form N_2, in direct correlation with CO_2 and nitrogen dioxide N_2O. These processes were performed through the bacteria which transformed organic matter as of the following reaction:

$$4NO_3^- + 5CH_2O + 4H^+ \rightarrow 2N_2 + 5CO_2 + 7H_2O.$$

Each cultivation system created different amounts of nitrogen compounds in the soil (NH_4^+, NH_3) and nitrate (NO_3^-). During the endocarp ligneous phase, the content of nitrogen in the leaf was highest at the T_4 treatment (1.67%). On the contrary, the T_2 treatment represented a low content of nitrogen in the leaf (1.21%). In the control (T_2) treatment considerable amounts of fruits fell down as the trees supported the more critical condition[9]. In this study the treatments were the source of variability on nitrogen content. Comparison of nitrogen content for each treatment related to control range treatments as follow: $T_4 > T_1 > T_5 > T_3 > T_2$. The low content of nitrogen in the T_2 could be explained by unfavourable system for nitrogen recycling because of the competitive spontaneous weeds and lack of agro-technics (Table 2). In addition, the highest nitrogen content at T_4 treatment (1.67% N), followed by cover crop, T_1 (1.51% N) were dedicated to favourable system created by usage of mixed legumes and rye, where the nitrogen accumulation process was favoured by the specifc bacteria able to fxe molecular nitrogen and to transform organic matter, whereas T_5 – plowing (1.37% N), and T_3 – gliphosate, were not proved as favourable systems for this important cycling for the trees[10,11] (Table 2, Fig. 1).

Table 2. Content of nitrogen (%) and water (%) in the leaves in relation to the level of treatment vegetation management of olive

Level / Treatment	N in leaf (%)	H_2O in leaf (%)	Mean-mean^0 /Std^0	Score sum	Score mean	P-Value
Cover crop/30/8	1.51±0.07de	46.0±2.0 **b**	-0.883	-1.764	-0.3528	0.0001*
Cover crop/30/I	1.77±0.12bc	56.0±1.0 **a**	1.073	2.142	0.4284	0.0001*
Control/30/8	1.21±0.04 f	39.0±1.0 **c**	-3.998	-1.5967	-7.983	0.0001*
Control/30/I	1.72±0.10bc	56.0±1.0 **a**	0.509	0.2034	1.017	0.0001*
Gliphosat/30/8	1.36 ±0.09 ef	49.0±2.0 **b**	-2.228	-0.8896	-4.448	0.0001*
Gliphosat/30/I	1.79±0.06 bc	56.0±1.0 **a**	1.116	0.4456	2.228	0.0001*
Straw mulch/30/8	1.67±0.13cd	55.0±1.0 **a**	0.158	0.0630	0.315	0.0001*
Black fallow/30/8	1.37±0.07ef	47.0±1.0 **b**	-2.025	-0.8086	-4.043	0.0001*
Black fallow/30/1	1.90±0.07ef	56.0±1.0 **a**	2.339	0.9341	4.670	0.0001*

Levels not connected by same letter are significantly different

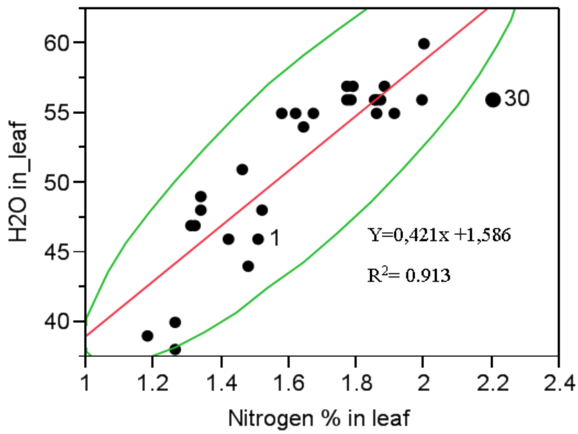

Fig. 1. Plot illustrating the multivariate corrélations estimated by REML method, between nitrogen and ELO % in the leaf analysis of fve Systems of végétation management of olive

During dormancy nitrogen within the leaves was less used by the trees, but had high values. During endocarp ligneous phase the trees consumed considérable amounts of nitrogen and had a better performance when this élément had a concentration of 1.51 up to 1.7% N. It was categorically harmful for the trees when nitrogen was lower than 1.4%. Nitrogen content on the leaf 2% occurred only during relative dormancy, whereas during the active phase it was reduced in corrélation with the stage of development and nature of treatment[12] (Fig. 1).

Water content. In Table 2 and Fig. 1 is shown that the dynamics of the H_20 percentage in the leaf for the period 15 December - 28 February (dormancy period) was 54-56%, whereas leaf weight 0.29-0.33 g, followed by coeffcient of variation (CV) = 1.04-3.8%. Lateron when the température exceeded the biological zéro, the need for H_20 increased and became serious during efforescence, fruiting and endocarp ligneous phase[13]. During thèse stages H_20 traces in the leaf were différent, because they were like this even in the terrestrial solution. During the active phase only T_4 - straw mulch and T_5 treatment had a normal turgor because their leaves had 55 and 47% H_20. The values of this vital élément in % ranked the treatments as follows: T, > T, > T, > T. > T, (% FLO). The macro-microelements which are indispensable for the tree are dissolved in the soil water of the roots, thus comprising the feld capacity aquifer. This solution is strongly absorbed by the soil and roots (P_F) which increases proportionally with humidity decrease. When H_20 in the leaf decreases to 44% the tree and soil absorb water with the same rate, thus enabling the appearance of the shrivelling phenomenon[13]. The shrivelling phenomenon occurred in T_2 and T_3 treatments, but not in T_4 and T_5 treatments, where during the endocarp ligneous phase the humidity content on leaf from 48 to 56% were optimum. On the contrary, the leaf humidity content under 44% was categorically inappropriate.

Analysis of ail the biological processes, during the annual cycle, proved strong corrélations between the content in % of water and nitrogen in the leaf $r^2 = 0.91$ (Fig. 1). Water content in the leaf above 50% occurred during two months of the winter cycle (dormancy phase), while during the other phases the content of water decreased under this limit in relation to the cultivation systems[14,15] (Fig. 1, Table 2).

Yield as an expression of the tree metabolism (Table 1) was different and fuctuated from 16 kg/tree T_3 – gliphosate up to 37 kg/tree T_4 – straw mulch. It was infuenced by the systems which favoured more water and nitrogen reserves available for the trees. Yield was related to fruit quantity and weight[16,17]. The trees with a system of straw mulch, (T_4) provided superior yield as they had 7.6% fruit and 0.36 g fruit weight more than the following treatment and 4.8% fruit and 0.50 g more than control. The most favourable yield received in T_4 – straw mulch, became gradually unfavourable ranking treatments as follows: $T_4 > T_5 > T_3 > T_1 > T_2$ (yield, kg) (Table 1), displays the yield with a strong correlation of water content $r^2 = 0.94$, and nitrogen content in the leaves $r^2 = 0.95$. The systems had different effects on the soil structure, the process of nitrogen recycling and water conservation available for the trees. As a result fruit weight is a product of this function which was reduced respectively from T_4 – straw mulch 2.6 g up to T_2 – control, 2.1 g ($T_4 > T_5 > T_3 – T_1 > T_2$ (weight fruit). Variation of the yield indices was 8.9% CV as opposed to T_4, T_3 21.6, T2 18.6 and T_1 15.3, etc. Fruit dimensions were lower when the trees had high yield and a high number of fruits[18].

The results presented in Table 1 show that the olive trees in mulching and ploughing treatment produced 28 and 15%, respectively, higher yield than that on untreated control. Similar results were evident also for the mean weight of olive fruits. In mulching and ploughing treatments compared to the control treatment, the weight was 30 and 25% higher, respectively. Regarding the biometry of fruits differences on *D* and *d* were evident in all treatments respect to the untreated control. Signifcantly higher those parameters were observed on mulching and ploughing treatments.

Vegetative growth. In Table 1, when the trees were kept fallow (T_5 – plowing) they had vegetative growth 10.8 cm, but when they were straw mulch (T_4 – straw mulch) they had a more active one 14.9 cm (CV: 6.6%). Leguminous cultures were in a race with the olive trees for water and mineral matter during the active phase and did not favour vegetative growth of the olive. The linear growth of shoots were higher in the straw mulch and ploughing treatments. (T_1 – cover crop limited (13%) vegetative growth compared to the fallow system (T_5 – plowing and 37% as compared to (T_4 – straw mulch). From this point of view Index test defned the differences with control (T_2), gliphosate (T_3) and cover crop (T_1), respectively; (6.4, 5.8, 5.4) and fallow T_5 (4.1) (see Table 1). Vegetative growth resulted from recycling the carbon hydrates and was infuenced by water and nitrogen concentration[17,18]. Vegetation was favoured in T_4 and unfavoured in descending order up to T_2 ($T_4 > T_5 > T_1 > T_3 > T_2$ (vegetative growth).

Intercalary culture (T_1 – cover crop, displayed a strong correlation when it is co-accompanied with the olive ($r^2 = 0.828$), whereas the olive did not favour this co-habitation and displayed a poor correlation $r^2 = 0.324$.

Efforescence. The fruit begins with the ovum fertility and growth in the embryonic sac. Formation and development of the fruit is improved continuously until the fruit becomes an 'extra' organ for its own branch. Fruiting had a strong correlation with nitrogen and water content. In Table 1, when these correlations were strong efforescence fall was low and the opposite. Dynamic of fruiting was 2.8–7% referring to the fowering biological basis. Water and nitrogen content in the plant tissues caused proportionally defects on pollination, fruiting, and endocarp ligneous up to ripeness. Trees of water and nitrogen defcits eliminated the fruits to preserve tissue turgor. Under these circumstances can the highest percentage of fruiting in T_4 be explained (7%), index which is reduced up to the minimum limits in T_2. Fruiting as per treatment is ranked in percentage: ($T_4 > T_5 > T_1 > T_3 > T_2$ (% fruiting). The highest percentage of fruiting is related to the positive impact of straw mulch (T_4) to preserve oil humidity as a basic element for fruiting[13,15].

Percentage of oil and acidity. Oil percentage in the fruit is a synthetic product of metabolism, which pointed out the genetic character with some slight differences, whereas quantity of the oil was different depending on fruit yield. When the fruit was red to purple oil percentage varied from 03 to 0.8% from treatment to treatment and resulted higher in T_3 – glyphosate 21.3% and lower in T_1 20.5%. The maximum quantity of oil was when the trees were cultivated with the system T_4 – straw mulch 19, 8 hl/ha, whereas T_2 – control 9.9 hl/ha corresponded to CV = 47%. Coeffcient of variation as opposed to T_4 – Straw mulch, increases up to Treatment control ($T_4 > T_5 > T_1 > T_3 > T_2$ (% CV). The lowest values of acidity were in T_4 – straw mulch and T_5 – plowing; 0.5 and 0.63%, because of the normal processes of the lipid synthesis in the fruit, whereas T_2 – control is characterised by higher acidity values 1.13% as a result of the negative effects of the respective cultivation systems (Table 1).

CONCLUSIONS

Cycling of the main elements through the physiological processes of respiration and photosynthesis was different as well as the interdependence of the systems of tree cultivation. The real values of nitrogen matter and water were tested well in the tissues of the leaves and were in correlation with the biophysiological processes.

Research defned the appropriate and harmful levels of nitrogen and water percentage, considered as important during the critical period of endocarp lignifcation.

The biological processes were elaborated in strong correlation with the system of soil cultivation, thus highlighting mulching as more favourable for the olive soil. Cycling of the biological and chemical processes was competitive and served the amount of recycling organic and nitrogen matter in the soil.

The highest waterpercentage enabled fruiting, fruit weight and yield as more favourable. Natural preservation of trees negatively infuenced the development and yield and was estimated as an inappropriate system for the olive.

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