

IMPACT OF BIOFERTILIZERS ON VEGETATIVE GROWTH AND SOIL RESPIRATION IN *Triticum spelta* L. GROWN IN ORGANIC FARMING

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Abstract

*Intensive agricultural production alters the natural cycle of nutrients in the soil, very often leading to a reduction in organic matter and soil degradation. The use of biofertilizers would solve these problems and make the ecosystem healthier. The biofertilizers used in the study have a positive effect on *Triticum spelta* L. growth. The results of the treated variants with Litovit, Tryven, Baikal EM and Amalgerol, show a higher height of the plants in the stem elongation phase compared to the control, by 20%, 15%, 12% and 10.8%, respectively. At full ripeness, the general stimulating effect of the applied foliar biofertilizers on the height, better nutrition and growth of the plants is taken into account. Greater soil microbial activity in the studied biofertilizers was observed on the 14th day after treatment. Higher values are reported for all variants of combined fertilization (foliar fertilization on basic fertilization with Agriorgan pellet) compared to unfertilized control and self-fertilization with Agriorgan pellet, which is statistically proven.*

Key words: biofertilizers, organic agriculture, *Triticum spelta* L., soil respiration, vegetative growth.

INTRODUCTION

Agriculture alters the natural cycling of nutrients in soil (Mishra & Dash, 2014) especially when there is an intensification in agriculture which has a negative impact to agriculture ecosystem that is including decrease of organic matter (Altuhaish & Tjahjoleksono, 2014), export of nutrients (Minev et al., 2019) and soil degradation. Organic farming methods (such as the use of biofertilizers) would solve these issues and make the ecosystem healthier (Mishra & Dash, 2014). Biofertilizers has been identified as an alternative to chemical fertilizers to increase soil fertility and crop production in sustainable farming (Mazid et al., 2011b; Mishra et al., 2015; Javoreková et al., 2015). The biofertilization was beneficial in stimulating the growth for plants (Rivera-Cruz et al., 2008; Abd El- Gleel Mosa et al., 2018). Many authors point out that energy renewables in the agro-ecosystem, food security and the sustainability of agriculture depends on healthy and fertile soil (Singh et al., 2017; Sabbagh et al., 2017). This is achieved by applying environmental friendly source of plant nutrient, such as biofertilizers (Bhattacharjee & Dey, 2014). Javoreková et al. (2015) and Mazid et al. (2011a) points out that according to Vessey,

(2003) biofertilizer as a substance which contains living microorganisms which, when applied to seed, plant surfaces or soil, colonizes the rhizosphere or the interior of the plant and promotes growth by increasing the supply or availability of primary nutrients to the host plant. Biofertilizers play an important role in improving nutrient supplies for crops (Pal et al., 2015) and hold a great promise to improve crop yields through environmentally better nutrient supplies (Wu et al., 2005). Mazid & Khan (2014) points out that according to Ghosh (2004) liquid biofertilizers are special liquid formulations containing not only the desired microorganisms and their nutrients but also special cell protectants or chemicals that promote formation of resting spores or cysts for longer shelf life and tolerance to adverse conditions. Agricultural practices that improve soil quality and agricultural sustainability have received much attention by researchers and farmers. The role of organic manures in plant nutrition is now attracting the attention of agriculturists and soil scientists throughout the world (Jat et al., 2015). Organic manures releasing nutrients rather slowly and steadily over a longer period (Makinde et al., 2007) and can be used to promote the healthy population of beneficial organisms in the soil (Rather et

al., 2018). Soil organisms are very important for the management of soil fertility in agriculture (Bing-Ru et al., 2006) and microorganisms that perform important functions in the soil (Rizvanov et al., 1988; Araújo & Monteiro, 2006). Soil microbial respiration is the most characteristic indicator for determining soil microbial activity (Tu et al., 2006). Soils that maintain a high content of microbial biomass have an accumulation of nutrients that are involved in the cycle of the soil system (Gregorich, 1994).

Spelled wheat (*Triticum spelta* L.) belongs to the group of hexaploid wheat, and until the beginning of the XXth century it was the predominant cereal and fodder crop in many regions of Southwest Germany (Blatter et al., 2004), as well as in parts of Switzerland and Austria (Poltoretskyi et al., 2018). Today it is a relic culture in Central Europe and northern Spain. As it has a number of valuable properties - it improves blood circulation and digestion, facilitates kidney function, finds its prestigious place as a healthy food (Kohajdová & Karovičová, 2008; Arzani & Ashraf, 2017) and although low yield is very suitable for preparation of pasta because it contains a balanced amount of gluten, which further enhances the interest in this crop grown especially in the system of organic production (Ugrenović et al., 2018). The aim is to study the influence of the applied biofertilizers on the vegetative growth and soil respiration of *Triticum spelta* L., in the conditions of a plant-growing organic farm.

MATERIALS AND METHODS

The study was conducted during 2014-2017 at the Agroecological Center at the Agricultural University, Bulgaria. The following factors were studied: Vegetation year (2014/2015; 2015/2016; 2016/2017); Wheat species - *Triticum spelta* L.; Fertilizers - Agriorgan pellet biofertilizer in dose 100 kg/da is used as a basic fertilization of the whole experimental area and the Amalgerol, Lithovit, Baikal EM and Tryven as foliar application in the different variants. All foliar fertilizers are applied twice in the tillering and the stem elongation phases in the following concentrations, respectively: Amalgerol- 200 ml/da in tillering phase and

500 ml/da in the stem elongation phase; Lithovit- in a dose of 150 g/da; Baikal EM - with 0.1% solution; Tryven- 400 ml/da. The soil of the Agroecological Center is alluvial-meadow (Mollic Fluvisols in the FAO) classification, with a slightly alkaline reaction, and the profile is represented by a humus horizon with a thickness of 10 to 40 cm, below which follow alluvial deposits unaffected by soil formation. Alluvial-meadow soils are loose, ventilated and warm. Most of these soils are characterized by high fertility on the soil type (Popova & Sevov, 2010).

Characteristics of used biofertilizers: **Amalgerol**- is a liquid emulsion concentrate rich in hydrocarbons and natural plant growth hormones. Contains seaweed extracts, distilled paraffin oil, vegetable oils, distilled herbal extracts. **Lithovit**- is a high quality nanotechnology product created by tribodynamic activation and micronization. Contains (CaCO₃) - 79.19%; (MgCO₃) - 4.62%; (Fe) - 1.31% and others. **Baikal EM** - is a probiotic product containing beneficial microorganisms (lactic acid bacteria, yeast, bifidobacteria, enzymes and spore bacteria) which are antagonists of pathogenic and opportunistic microflora. **Tryven** - is a complex mixture of NPK, contains (N)- 24.4%; Organic nitrogen- 17.3%; (P₂O₅)- 17.2%; (K₂O)- 7.42%. It has a good systemic effect, especially nitrogen. **Agriorgan pellet** - is an organic fertilizer from sheep manure, enriched with microorganisms and a supplement of trace elements. Contains: Organic nitrogen (N) - 2.5%; (P₂O₅) - 3.0%; (K₂O)- 1.0%; Organic carbon (C) - 28.5%; Humic acids - 6.0%; pH - 6, fulvic acids.

Agrotechnics of experience. Agriorgan pellet fertilizer in a dose of 100 kg/da is imported in the soil at depth of 10-15 cm together with crushing of the pepper predecessor plant biomass and mixing with the soil. Plant residues enrich the soil with organic matter necessary for soil organisms and maintaining soil fertility. Against late autumn and early spring weeds all the experimental area was harrowed with harrow, in the early spring in March when the crop is in tillering phenophase and the weeds are in their early development. This method is extremely successful for

controlling weeds in cereals for organic production.

Study Parameters: Plant height - measure the height of 10 plants of each variant and each repetition in the phases tillering, stem elongation, full ripenes.

'Soil respiration' - Total microbiological activity is determined by the amount of CO₂ released (determination of the intensity of CO₂ release). The method used is a modification of the method of Stotzky (1965) (Sapundzhieva et al., 2010).

The statistical processing of the experimental data was performed using SPSS V. 9.4 for Microsoft Windows (SAS Institute Inc., 1999), by Duncan, Anova. A Duncan multiple-range test was also performed to identify the homogeneous type of the data sets among the different treatments at P<0.05 level (Duncan, 1955).

RESULTS AND DISCUSSIONS

1. Agroclimatic characteristics. The vegetation year 2014-2015 is characterized as relatively warm, and precipitation is above normal, except January and April, which defines the year as relatively humid. After sowing in abundant rainfall conditions in the third ten days (116 mm/m²), combined with temperatures above the norm (12.8°C) allowed timely emergence of plants. The winter period is characterized with high rainfall values and temperatures, which is good for all the processes, related to the plant cold resistance good enter the tillering phenophase, and successfully overwinter. The spring vegetation is in heavy rainfall conditions (138 mm/m²), 94 mm/m² above the norm, which is with positive effect on crop development in the tillering and stem elongation phenophases. During May and June, when wheat is heading, temperatures and precipitations are found in values above the norm (Table 1).

The vegetation year 2015-2016 is characterized as relatively warm, with temperatures above the norm and relatively humid. The heavy rainfall in October (70.3 mm/m²) contributes to a good supply of soil with moisture for normal crop

germination. Temperatures in the period December-February are above the norm, which allows successful overwintering of the plants. From March, the temperature values are slightly above the norm, precipitation in May (64.7 mm/m²) twice over the long-term period, which gives its positive attitude to the plants development (Table 2). 2016-2017 is characterized as relatively warm with uneven precipitation. The beginning of crop vegetation in the autumn takes place under favorable conditions, characterized by temperature values close to the long-term period. The renewed vegetation in March is characterized by temperatures above the norm (by 3.7°C) and precipitation above the norm of 47.9 mm/m², favorable for the crop development.

2. Plant height, cm. The influence of the foliar biofertilizers on *Triticum spelta* plants height is presented on Table 3. In 2015, during tillering phase, higher values were reported in the variants treated with Lithovit and Amalgerol, respectively 12.87 cm and 12.80 cm. The tendency for higher plant is maintained in the stem elongation phase with the same treatment options, with values of 60.67 cm and 58.33 cm, and in full ripenes when treated with biofertilizer Baikal EM- 84.35 cm, followed by the variant treated with Lithovit and Agriorgan pellets.

In 2016, during the tillering phase, the values differ, but close to the control. In the stem elongation phase, is observed positive effect on the plant height at combined foliar and soil fertilization in all variants, compared to the control plants. It is announced with unfavorable conditions for nutrients absorption in February and March - high air temperatures and low precipitation values. The highest plants were reported at Lithovit treated variants (21%), Tryven and Baikal EM (11% above the control). This confirms the effectiveness of foliar fertilization as a complementary element in plant nutrition. At full ripenes, higher values of the indicator were reported in the variants, treated with Amalgerol - 87.52 cm (5% above control) and Tryven - 87.15 cm (4% above control).

Table 1. Average daily air temperatures by ten days (°C), 2014-2017

Months	IX			X			XI			XII			I			II			III			IV			V			VI			
	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	
Ten days	20.7	18.6	14.8	14.1	15.2	9.2	8.8	10.2	4.8	4.8	6.7	4.3	4.4	0.5	2.4	6.5	3.3	0.8	6.9	4.7	5.5	9.9	7.9	14.8	14.6	19.3	19.3	19.4	20.9	22.7	19.6
Av. monthly t°C	18.1			12.8			7.9			5.1			3.1			3.7			6.7			12.4			19.3			21.1			
2015/16	22.9	20.3	18.4	14.9	14.5	9.2	9.6	13.2	11.2	7.1	3.4	4.8	0.1	1.5	-2.0	5.1	9.3	9.7	10.0	7.2	10.6	15.4	17.4	13.7	14.1	16.8	20.1	19.8	24.3	25.6	
Av. monthly t°C	21			12.8			11.3			5.1			-0.1			8.0			9.3			15.5			17.0			23.3			
2016/17	21.8	21.7	15.2	14.9	11.2	12.1	10.3	5.6	3.9	4.1	0.4	2.0	-4.4	-2.0	-5.4	-0.8	1.4	9.0	8.8	9.3	11.1	11.5	13.2	13.4	16.4	18.3	18.0	21.1	22.1	27.4	
Av. monthly t°C	19.6			12.7			6.6			2.2			-3.9			3.2			9.7			12.7			17.6			22.8			
Av. for the period 1965/95	18.3			12.6			7.4			2.2			-0.4			2.2			6			12.2			17.2			20.9			

Table 2. Sum of rainfall (mm/m²) by ten days in the study period 2014-2017

Months	IX			X			XI			XII			I			II			III			IV			V			VI		
	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III
Ten days	78.2	66.5	51	5	0.1	116	0.5	48.7	0.3	78.3	6.2	8.5	0.1	0.0	17.3	72.4	0.6	3.6	102.0	28.7	7.3	12.6	0.2	1.2	28.6	36.2	4.7	3.0	49.1	24.6
Monthly amounts	195.7			121.1			49.5			93.0			17.4			76.6			138.0			14.0			69.5			76.7		
2015/16	3.0	71.8	25.8	12.4	18.3	39.6	0	0	39.6	0	2.2	1.4	15.3	54.3	0	15.1	5.3	4.0	7.8	23.0	3.1	0.1	15.4	15.2	24.8	32.2	7.7	37.2	9.4	13.1
Monthly amounts	100.6			70.3			39.6			3.6			69.6			24.4			33.9			30.7			64.7			59.7		
2016/17	0.2	0.1	2.0	2.2	17.1	0.4	5.9	3.6	23.4	0.0	0.0	2.4	22.2	46.8	1.1	5.4	3.2	2.5	33.3	10.3	4.3	1.9	20.9	3.3	27.2	0.7	24.8	2.8	3.3	9.3
Monthly amounts	2.3			19.7			32.9			2.4			70.1			11.1			47.9			26.1			52.7			15.4		
Av. for the period 1965/95	65			47			35			36			40			48			44			39			32			36		

In 2017, during the tillering phase, higher values for plant height were reported in the

variant with self-fertilization with Agriorgan pellets - 14.80 cm (14% above the control).

Table 3. Height of plants by phenological phases and organic fertilization variants, *Triticum spelta* L. (2015-2017), cm

Year Variants	Phenological phases														
	Tillering					Stem elongation					Full ripeness				
	2015	2016	2017	Average	%	2015	2016	2017	Average	%	2015	2016	2017	Average	%
Control	-	17.47	12.97	15.22	100	-	45.97	48.87	47.42	100	-	83.48	92.97	88.22	100
Agriorgan pellets	11.93	16.03	14.80	14.25	93.6	56.10	43.57	48.30	49.32	104.0	81.87	83.30	96.20	87.12	98.7
Amalgerol	12.80	15.88	14.43	14.37	94.4	58.33	49.10	50.20	52.54	110.8	75.57	87.52	101.4	88.16	99.9
Lithovit	12.87	15.67	12.47	13.67	89.8	60.67	55.60	54.86	57.04	120.3	83.27	81.65	97.77	87.56	99.2
Baikal EM	11.63	15.37	14.30	13.77	90.5	54.93	51.07	53.23	53.08	111.9	84.35	68.03	99.60	83.99	95.2
Tryven	12.30	16.87	13.37	14.18	93.2	55.97	51.13	56.93	54.68	115.3	76.93	87.15	96.97	87.02	98.6

Consecutive release of nutrients from the organic fertilizer and their absorption by the plants in the soil coenosis promotes their fuller assimilation and good vegetative growth. Higher plants were also reported in the variants, treated with Amalgerol (11% above control) and Baikal EM (10% above control). In the stem elongation phase there is an increased effect on growth and higher plant height than already absorbed foliar fertilizers, and this effect is best expressed in the variants treated with Tryven- 16%, Lithovit- 12% and Baikal- 9% above control. The tendency for higher plants is maintained in full ripeness phase at the variants, treated with Amalgerol (9% above control) and Baikal (7% above control). It is very clearly stated that in all variants with applied foliar fertilizers there are higher values for plant height compared, to the control values. This proves that foliar vegetation nutrition has a positive and timely effect on plant nutrition. Average values for the study period show, that in the tillering phase, the heights of the plants from the different fertilization variants are close. This can be explained by the still weak reaction of plants to feeding with foliar fertilizers in the phenological phase itself. A more noticeable

effect is observed at variants, treated with Amalgerol and Tryven. In the stem elongation phase, higher values of the indicator are reported when treated with Lithovit, Tryven, Baikal EM and Amalgerol, respectively 20%, 15%, 12% and 10.8% above the control variant. There is a noticeable increase in plant height at phenological phase in the treated variants and it shows that the combined application of biofertilizers in the growing technology leads to better nutrition and better vegetative growth of the plants. In the phase of full ripeness it is noticed that the application of biofertilizers have a stimulating effect on the plants height at *Triticum spelta* and their usage is a reliable measure to support better nutrition and vegetative growth of plants to achieve higher grain yields.

3. 'Soil respiration' in *Triticum spelta* L. The results for 'soil respiration' after the first vegetative feeding in 2016 at *Triticum spelta* L., show that Amalgerol application, leads to highest soil activity on the 7th day and confirmed on the 14th day, which is preserved during the second reporting of the heading phase (Table 4).

Table 4. Data on 'Soil respiration' - ($\mu\text{g CO}_2/\text{h/g}$) in *Triticum spelta* L. (2016)

Variants	7 th day	14 th day	21 st day
	Average \pm St. dev.	Average \pm St. dev.	Average \pm St. dev.
	First reporting (phase stem elongation)		
Control	12.23 \pm 0.208 ^a	12.00 \pm 1.000 ^{ab}	7.13 \pm 0.115 ^b
Agriorgan pellets	9.80 \pm 0.173 ^c	10.67 \pm 0.577 ^b	9.03 \pm 1.266 ^a
Amalgerol	12.60 \pm 0.100 ^a	12.33 \pm 0.577 ^a	9.37 \pm 0.321 ^a
Lithovit	10.90 \pm 1.217 ^{bc}	11.67 \pm 0.577 ^{ab}	9.13 \pm 0.503 ^a
Baikal EM	10.37 \pm 0.208 ^c	11.00 \pm 1.000 ^{ab}	9.53 \pm 0.058 ^a
Tryven	12.00 \pm 0.985 ^{ab}	11.67 \pm 0.577 ^{ab}	9.70 \pm 0.265 ^a
	Second reporting (phase heading)		
Control	8.3 \pm 0.723 ^b	8.4 \pm 0.115 ^d	7.6 \pm 0.404 ^b
Agriorgan pellets	8.0 \pm 0.208 ^b	8.1 \pm 0.173 ^d	8.1 \pm 0.265 ^b
Amalgerol	10.2 \pm 1.852 ^a	9.9 \pm 0.115 ^a	9.3 \pm 0.493 ^a
Lithovit	8.3 \pm 0.115 ^b	9.4 \pm 0.208 ^b	7.4 \pm 1.159 ^b
Baikal EM	8.7 \pm 0.416 ^{ab}	9.8 \pm 0.289 ^a	9.2 \pm 0.265 ^a
Tryven	8.9 \pm 0.265 ^{ab}	8.97 \pm 0.208 ^c	9.6 \pm 0.404 ^a

*Means followed by the same letter are not statistically different ($P < 0.05$) by Duncan's multiple range test

This gives a reason to attribute this effect to the composition of the fertilizer and the increased microbial activity in the rhizosphere zone, which has an impact on the acceleration of assimilation processes, expressed in maximum height in plants during full ripeness phase - 87.52 cm.

It can be pointed out that at the first reporting (stem elongation) on the 14th day is observed an increase in soil activity at the variants, treated with Lithovit and Tryven, which has its impact on vegetative growth in the stem elongation, as plants treated with Lithovit having the largest height- 55.60 cm, followed by Tryven- 51.13 cm.

The activation of the rhizosphere microflora under the influence of Lithovit biofertilizer is associated with the rich in micronutrients composition of the fertilizer.

It is necessary to point out that in the first reporting (stem elongation) on the 21st day, in

addition to activity in Tryven- 9.70 $\mu\text{g CO}_2/\text{h/g}$, there is a significant activity in the plants of the variant treated with Baikal EM- 9.53 $\mu\text{g CO}_2/\text{h/g}$, which also affects the greater height of the plants in the stem elongation phase- 51.13 cm at Tryven treated variant, and 51.07 cm when Baikal EM is used.

In the days before sampling, the environmental conditions were reported as favorable, improving the microbial activity, namely - precipitation 14.3 mm, soil temperature (at 10 cm depth) - 17.7 °C, air temperature - 15.9°C and relative humidity - 60% (Table 1).

The results of soil microbial activity in 2017 after the first feeding show that on the 7th day in all variants with foliar biofertilizers there is a proven higher soil activity in plants, and it is best expressed in the variant with biofertilizer Amalgerol (Table 5).

Table 5. Data on 'Soil respiration' - ($\mu\text{g CO}_2/\text{h/g}$) in *Triticum spelta* L. (2017)

Variants	7 th day	14 th day	21 st day
	Average \pm St. dev.	Average \pm St. dev.	Average \pm St. dev.
	First reporting (phase stem elongation)		
Control	9.10 \pm 0.376 ^b	9.00 \pm 0.608 ^c	8.1 \pm 0.153 ^d
Agriorgan pellets	9.30 \pm 0.095 ^b	9.03 \pm 0.569 ^c	8.2 \pm 0.100 ^d
Amalgerol	11.9 \pm 0.648 ^a	11.89 \pm 0.081 ^a	11.4 \pm 0.100 ^a
Lithovit	11.30 \pm 0.458 ^a	11.50 \pm 0.520 ^{ab}	9.6 \pm 0.100 ^c
Baikal EM	11.7 \pm 0.060 ^a	11.70 \pm 0.105 ^a	10 \pm 0.265 ^b
Tryven	11.5 \pm 0.202 ^a	10.80 \pm 0.173 ^b	9.6 \pm 0.100 ^c
	Second reporting (phase heading)		
Control	9.0 \pm 0.361 ^d	9.1 \pm 0.058 ^c	8.0 \pm 0.153 ^d
Agriorgan pellets	9.2 \pm 0.115 ^d	9.2 \pm 0.208 ^c	8.2 \pm 0.252 ^{cd}
Amalgerol	11.1 \pm 0.252 ^a	10.9 \pm 0.252 ^a	10.6 \pm 0.265 ^a
Lithovit	9.8 \pm 0.513 ^c	10.1 \pm 0.208 ^b	8.5 \pm 0.100 ^c
Baikal EM	10.4 \pm 0.115 ^b	10.7 \pm 0.058 ^a	9.6 \pm 0.100 ^b
Tryven	10.7 \pm 0.153 ^{ab}	10.83 \pm 0.058 ^a	10.9 \pm 0.265 ^a

*Means followed by the same letter are not statistically different ($P < 0.05$) by Duncan's multiple range test

This tendency for higher activity when treated with Amalgerol is maintained on the 14th and 21st day after treatment, which leads to greater vegetative growth by *Tr. spelta* in the phase of full ripeness, where the plants reach the highest height of 101.4 cm (Table 3). A similar consistent effect is observed when Baikal EM is applied, which leads to values for the greater height in the phase of full ripeness - 99.60 cm, compared to the control. In the second reporting at heading phase, the same trend is observed for overall positive impact of feeding with the studied biofertilizers, as again the variant treated with Amalgerol comes to the fore, in which proven higher values of soil respiration from 7th to 21st are reported and the day after treatment of the plants. A similar trend is observed at the variant treated with Tryven, followed by the variant treated with Baikal EM.

CONCLUSIONS

Higher plant height as a result of the applied biofertilizers was reported at tillering phase 14 days after treatment with Lithovit, Tryven, Baikal EM and Amalgerol, where the increase of the indicator compared to the control is by

20%, 15%, 12% and 10.8%. The combined application of biofertilizers in the growing technology leads to better nutrition and better vegetative growth of plants.

Over the years proven higher values of soil respiration 7 days after treatment were reported in the variants treated with Amalgerol, Baikal EM and Tryven. Greater soil microbial activity in the studied biofertilizers was observed on the 14th day after treatment. Higher values are reported for all variants of combined fertilization (foliar fertilization on basic fertilization with Agriorgan pellet) compared to unfertilized control and self-fertilization with Agriorgan pellet, which is statistically proven. On the 21st day after biological fertilizers application, a slight decrease in the values of the soil microbial activity is observed in all treated variants and it starts to approach the value of the control.

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