

Effects of combined nutrient supply treatments on some physiological parameters of autumn wheat

TÜNDE KAPRINYÁK¹ – RÉKA LÁPOSI¹ – LÁSZLÓ BEKŐ² – SZILÁRD TÓTH³

Eszterházy Károly University

¹Institute of Agricultural Sciences and Environmental Management,
Gyöngyös, Hungary

²Remote Sensing and Rural Development Research Institute,
Gyöngyös, Hungary

³Fleischmann Rudolf Agricultural Research Institute,
Kompolt, Hungary

kaprinyak.tunde@uni-eszterhazy.hu

Summary

The Fleischmann Rudolf Research Institute in Kompolt is not only famous for plant breeding but the institute also surveys the effects of different nutrient supply methods since 1918. In 2017, we joined this research supported by EFOP 3.6.1 project. Our aim was to investigate photochemical processes – which is one of the most determinant in case of yield – of crops by *in vivo* field measurements. We measured the chlorophyll content of leaves using Minolta SPAD 502. We used miniPAM fluorometer to determine actual photochemical efficiency and non-photochemical quenching of PSII during natural light conditions and also to evaluate the pigment (chlorophylls and carotenoids) and water content of leaves we applied field spectrophotometer (ASD FieldSpecPro 3). We utilized these methods by various treatments (1. treatment with soil bacteria + head and base fertilizer; 2. treated by only head fertilizer; 3. treated by only base-fertilizer) in field experiment of autumn wheat (4.1–2.43–1.19 ha) in June, 2017. The difference between treatments was clearly detectable. In the case of the first treatment, physiological processes were more intense and the ripening occurred earlier. The obtained yield was the highest in the case of the area treated by soil bacterial. Based on the results, the first treatment can be recommended in practice.

Keywords: autumn wheat, crop safety, *in vivo* measurements, soil bacteria

Introduction

In the Fleischmann Rudolf Research Institute investigation the effects of various nutrition supply systems has been utmost importance for nearly 100 years. In 2017 we joined these experiments in order to monitor the photochemical processes of plants by *in vivo* field measurements, which can influence the quantity and quality of yield. Among our arable crops, autumn wheat has paramount importance for both food and feed purposes. Due to the excellent ecological conditions of the Carpathian

Basin, Hungary is a major wheat-making country. According to FAO data (Net1), the world's wheat production area exceeds 220 million hectares, from it, Hungary represents approximately 0.5%. All harvested crops have grown since 1990; in 2016 it was 740 million tons. In Hungary, however, it shows a downward trend. In 1990 it still exceeded 6 million t, but in 2016 it did not reach already 4.8 million t. The CSO (Net2) also reported lower values, but in a smaller area (1.044 thousand hectares) a larger quantity (5603 thousand tons) was aggregated. However, the domestic average yield is still increasing. In 2014, 4.7 t ha⁻¹ and in 2016 5.37 t ha⁻¹ wheat were recorded in register of the CSO. This is most likely due to technological development as well as the increasing amount of fertilizer used. The fertilizer per hectare of agricultural land in 2000 was 61 kg; by 2016 it rose to 103 kg. Monoculture cultivation and increasing amounts of fertilizer use are increasingly burdening the environment. However, farmers are forced to increase agricultural production during climate change conditions and demographic explosion of developing countries. The goal is to produce a larger quantity in a smaller area. An important element of sustainable agriculture is the preservation of soil fertility. Crop security is a key issue for achieving successful agriculture in the long term. To achieve this, the use of soil bacteria will help. Several similar products have appeared on the market over the last years.

Materials and methods

Properties of the experimental area

The experiment was carried out at the Fleischmann Rudolf Research Institute in Kompolt, which is located in the southern part of the southern mountain range of Mátra between Eger and Gyöngyös, an average of 125 meters above sea level. Its weather is extreme, especially during the summer period, with few precipitation (Holló et al., 2009ab). In 2017, the annual precipitation amounted to 529.8 mm, from which rainfall reached 418.3 mm in summer. The average number of sunny hours is 2108, and 1528 hours in the vegetation period. The average annual temperature was 10 °C, which was 10.67 °C in 2017. Over the vegetation period, in multiannual average of 17.3 °C (Herczeg, 2013), while in 2017 the average temperature was 17.05 °C according to local meteorological station data. In the area of the research institute, chernozem brown forest soil is the main soil type, the thickness of the humus layer is between 0.5 and 0.8 m (Holló and Kádár, 2003). Based on the latest soil analysis (*Table 1*) the area can be characterized by poor phosphorus content, with sufficient potassium content and low humus content.

Table 1. Soil parameters of the experimental area (Károly Róbert College, Fleischmann Rudolf Research Institute, 2015)

Sample depth (cm)	pH (KCl)	Gold's binding	Soluble salt in water % (m/m)	Humus content % (m/m)	Soluble potassium (AL) in K ₂ O mg/kg	Soluble phosphorus (AL) in P ₂ O ₅ mg/kg	Soluble (NO ₂ +NO ₃)-N (KCl) mg/kg
0-30	4.47	43	0.07	2.1	227	123	18

Experiment features

Preparation of experiment

In the year 2016, the forecrop was an autumn cabbage rape. The basic fertilizer NPK=8:24:24; a total of 174.96 kg ha⁻¹ was on the total area. The soil bacteria treatment on 4.1 ha was 1 l ha⁻¹ with a spraying machine (Gambetti in 1000 liters of water). Sowing time was on 23rd of November in 2016. Ortegus variety of autumn wheat was selected, of which 250 kg ha⁻¹ seed was needed. On 2nd of February in 2017 96.15 kg ha⁻¹ head fertilizer and 27% Péti salt were applied. Dispensing of the head fertilizer was carried out in case of soil bacteria treated 4.1 ha and in case of 2.43 ha without soil bacteria (control with head and basic fertilizer). The remained 1.19 ha only received basic fertilizer. The 1.19 ha without head fertilizer only received basic fertilizer (control with only basic fertilizer). The experiment was set up in 4 repetitions in the following ways:

- With soil bacteria: 10 250 m² per repeat;
- With head and basic fertilizer: 5350 m² per repeat;
- With basic fertilizer: 2975 m² per repeat.

The applied soil microbe product contains free-living, nitrogen-binding and phosphor-mobilization soil-dwelling bacteria (50% *Pseudomonas*, 25% *Azotobacter* és 25% *Azospirillum* strains) and microelements (Dilci, 2008; Szabados, 2008; Net3). The preparation does not contain any chemicals or hormones, its soil improver effect can reduce the amount of fertilizer applied (Net3). The seemingly costly treatment of bacterial preparations is suitable to reduce the adverse effects of climate change, thus achieving better quality and more crops. (Káta, 2011; Net4).

Their use helps to create and maintain healthy, equilibrium soil life. They have already been supported by Agri-environment schemes (AES) (Net5). The beneficial effect can be solved only by living soil bacteria; therefore it is indispensable to stay alive until they are incorporated into the soil. The product has to be protected from light and high temperature until use (Net3; Net6).

Methods of measurements

We investigated some photochemical parameters using in vivo chlorophyll fluorescence induction method with miniPAM fluorometer (WALZ GmbH, Germany) under unclouded natural light conditions between 11–13 a.m. (1200–1600 $\mu\text{mol m}^{-2} \text{s}^{-1}$ light intensity). We determined the actual photochemical efficiency of the PSII system ($\Delta F/F_m' = F_m' - F/F_m'$; Genty et al., 1989) and the rate of electron transport ($\text{ETR} = (\Delta F/F_m' \times \text{PAR} \times 0.5 \times 0.84)$) and the ratio of photochemical ($qP = F_m' - F/F_m' - F_0$) and non-photochemical processes ($qN = F_m - F_m'/F_m - F_0$, where F_m – maximum fluorescence, F_0 – basic fluorescence) (Net7; Schreiber et al., 1998). The relative chlorophyll content was measured by the Minolta SPAD 502 automatic meter. The device measures at the red light, since absorption of this is not affected by the contents of the leaf carotene (Markwell, 1995; Hawkins et al., 2009). The second leaves of the plants were measured in the upper third. For each plant 2 measurements were made to reduce measurement errors, it means 80 measurements per plot. The measurements were executed on 19th of June in 2017, at the advanced maturation stage. We carried out spectroscopic reflectance measurements by ASD FieldSpec3 portable spectroradiometer (USA) and determined vegetation indices (NDVI, RDVI, EVI, TCARI/OSAVI, SIPI, PRI, ARI, CRI, PWI, SRWI) from which chlorophyll, carotenoid and water content of the leaves can be calculated. The method is used to isolate the reflection properties of plants (Spinetti et al., 2009).

During the vegetation period (2016-06) the following parameters were recorded with remote sensing methods:

– Structural indices

Normalized Difference Vegetation Index (mean spectra):

$$\text{NDVI} = (R_{\text{NIR}} - R_{\text{red}}) / (R_{\text{NIR}} + R_{\text{red}}) \text{ (Rouse et al., 1974),}$$

Renormalized Difference Vegetation:

$$\text{RDVI} = (R_{800} - R_{670}) / \sqrt{(R_{800} + R_{670})} \text{ (Rougean and Breon, 1995);}$$

– Leaf indices

Structure Insensitive Pigment Index:

$$\text{SIPI} = (R_{800} - R_{450}) / (R_{800} + R_{650}) \text{ (Peñuelas et al., 1995),}$$

Carotenoid Reflectance Index:

$$\text{CRI} = 1 / B_{550} - 1 / B_{700} \text{ (Gitelson et al., 2002),}$$

Anthocyanin Reflectance Index:

$$\text{ARI} = B_{840} \times (1 / B_{550} - 1 / B_{700}) \text{ (Gitelson et al., 2001),}$$

Optimized Soil-Adjusted Vegetation Index:

$$\text{OSAVI} = (1 + 0.16) * (R_{800} - R_{670}) / (R_{800} + R_{670} + 0.16) \text{ (Rondeaux et al., 1996),}$$

Enhanced Vegetation Index

$$\text{(EVI)} = 2.5 \times (B_{840} - B_{670}) / (B_{840} + (6 \times B_{670}) - (7.5 \times B_{450}) + 1) \text{ (Huete et al., 2002);}$$

– Light utilization

Photochemical Reflectance Index:

$$PRI_1 = (R_{528} - R_{567}) / (R_{528} + R_{567}),$$

$$PRI_2 = (R_{531} - R_{570}) / (R_{531} + R_{570}),$$

$$PRI_3 = (R_{570} - R_{539}) / (R_{570} + R_{539}) \text{ (Gamon et al., 1992);}$$

– Water content

Plant Water Index:

$$PWI = R_{970} / R_{900} \text{ (Peñuelas et al., 1997),}$$

SRWI (Simple Ratio Water Index): R_{858} / R_{1240} (Zarco-Tejada et al., 2003).

We recorded at harvesting (22. 07. 2017) the yields ($t \text{ ha}^{-1}$) and the grain moisture (%) values.

NDVI, RDVI vegetation indices were also determined by aerial photographs, which on a rigid, unmanned aerial vehicle (UAV) fixed 4-channel multispectral camera system were made. After photogrammetric processing was the mosaic recording geometric resolution is 16 cm.

Channel distribution of the multispectral camera system: green: 530–570 nm; red: 640–680 nm; red edge: 730–740 nm; nearly infrared: 770–810 nm.

During the vegetation period, soil samples were taken, soil samples at depths 0–20 and 20–40 cm. Soil parameters were determined in the laboratory and the contents of the macro-, meso- and micro element of the soil, plant. The latter were also analyzed from stored crops.

Evaluation of differences between measured parameters in case of different treatments was performed using SPSS 20 statistical program, One-Way ANOVA method.

Results

As shown in *Figure 1*, at all parameters were high the deviation, this has significantly influenced the result. The control had the highest relative chlorophyll content. However, its photosynthesis activity was also outstanding. However, for the three parcels, the light intensity was not the same. When evaluating the sunscreen mechanism of the plant, higher photochemical efficiency ($\Delta F / F_m^2$) and lower non-photochemical photochemical extinction (qN) were measured at the control. The electron transport rate (ETR), which is calculated by the instrument based on $\Delta F / F_m$ 'and the actual light intensity significantly was different for the three parcels, the control was higher.

It is apparent from the data in *Table 2* that, like the SPAD value, the second treatment had lower the NDVI index, while the TCARI/OSAVI index was the lowest for treatment I.

Figure 1. Relative chlorophyll content (SPAD value), photochemical activity ($\Delta F/F_m'$), electron transport rate (ETR), non-photochemical quenching of PSII (q_N) of autumn wheat (Kompolt, 19/06/2017)

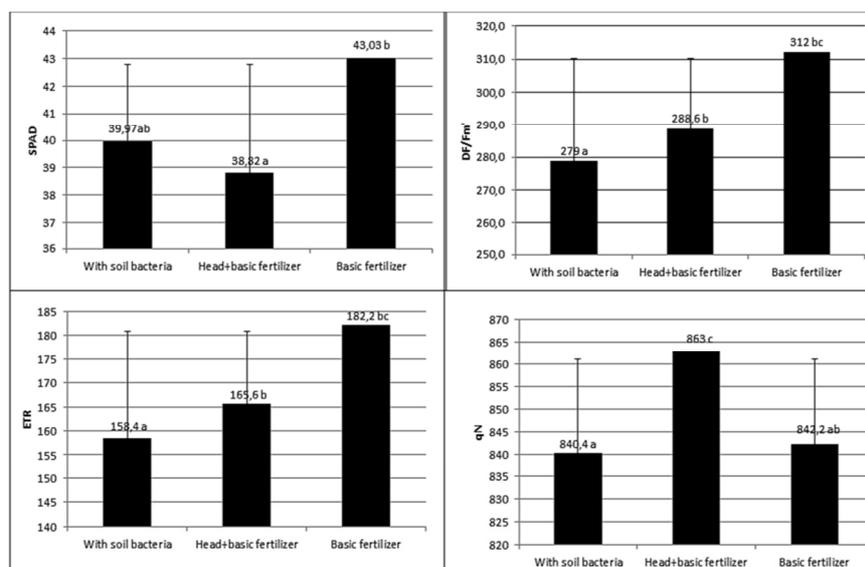


Table 2. Vegetation indices determined by field spectrophotometer (Kompolt, 2017)

Parameter	Significance	I. treatment	II. treatment	Control
NDVI	*** 231-aab	0.824±0.01	0.777±0.037	0.790±0.012
TCARI/OSAVI	*** 132-aab	0.176±0.023	0.203±0.005	0.177±0.05
RDVI	*** 231-aab	0.552±0.011	0.513±0.028	0.521±0.014
PRI3	*** 321-abb	-0.081±0.013	-0.07±0.018	-0.052±0.004
EVI	*** 231-aab	0.646±0.02	0.587±0.039	0.592±0.03
ARI	*** 321-aab	-2.067±0.703	-3.182±0.658	-3.167±0.571
CRI	*** 321-abb	15.412±1.31	15.304±1.742	14.631±3.77
SIPI	*** 231-aab	0.866±0.002	0.845±0.018	0.847±0.014
PWI	*** 132-abb	0.843±0.015	0.852±0.006	0.850±0.01
SRWI	*** 231-aab	1.439±0.001	1.381±0.057	1.382±0.055

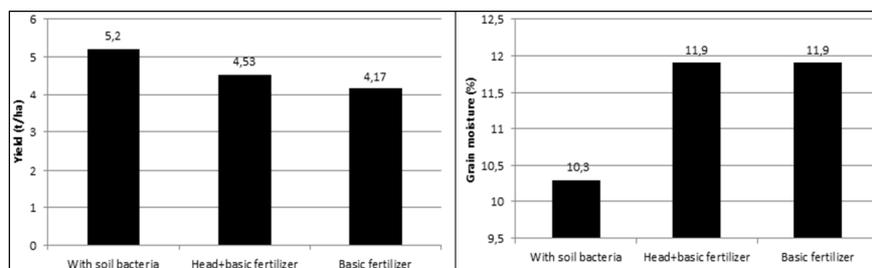
Note: a, b, c index – Tukey-Test significance groups ($p < 0.05$); *** ANOVA significance $p < 0.001$; 1 – I. with soil bacteria treatment, 2 – II. head+basic fertilizer; 3 – basic fertilizer.

The photochemical reflectance index (PRI) was the highest in I. treatment. The anthocyanin (ARI) in the treatment II., while the carotenoid (CRI) reflectance index was the highest in the soil bacterial parcel. The leaf structure independent pigment index (SIPI) indicating carotenoid/chlorophyll ratio was of outstanding value in I. treatment, which also indicates the state of the plants' stress (Peñuelas et al., 1995). The soil bacterial parcel had also lower water content based on the PWI index, which explains the more stressful situation than the other two

treatments. The vegetation indices used for autumn wheat similarly to physiological measurements show that the two treatments improve the stress tolerance of the plants.

On the basis of measurements, with soil bacteria treatment is beneficial in yield, while the head fertilizer is slightly lower. The moisture content measured after harvest also confirms the soil bacteria treatment, as 1.6% less than in the other two treatments (*Figure 2*).

Figure 2. Autumn wheat harvest characteristics (Kompolt, 2017)



The aerial photographs show the heterogeneity of plots (*Figure 3*). On the parcels are scattered spots visible. The soil bacterium treated seems homogeneous unlike the other two parcels.

The soil bacteria treatment presumably provided more balanced nutrient and water supply to crops, increased the parcel homogeneity, which is important for crop safety.

In table K7 soil samples were taken at 20 and 40 cm deep from the three parcels. According to our results (*Table 3*) In the 20 cm soil depth the soil moisture content was the lowest in the head fertilizer treatment and the dry matter content the highest. The same could be detected at a depth of 40 cm, too. The pH was the lowest in the soil bacteria treatment parcel. Based on Gold's binding a sample taken from the ground 20 cm deep showed the low value of the head fertilized treatment, but in the 40 cm depth sample it was already the same as the.

According to the results of *Table 4*. 20 cm in soil depth nitrate at head fertilizer treatment, nitrite, phosphorus at the control, potassium, calcium, magnesium, selenium, iron and lead in the soil bacteria treatment was the highest. At 40 cm depth nitrate, phosphorus, potassium at control, nitrite, calcium, magnesium, iron and lead at the head fertilizer treatment, selenium at soil bacteria treatment was high.

The nutrient values from stored grain yields showed high value at the control phosphorus, calcium, magnesium, iron, selenium, while in case of soil bacteria treatment they exhibited high sodium values. The head fertilizer treatment showed the values between the two treatments, alone,

the potassium content was outstanding. It can be concluded that control grain extracted most nutrients from the soil (Table 5).

Figure 3. The NDVI index with the multispectral camera on the three parcels (Kompolt, 2017)

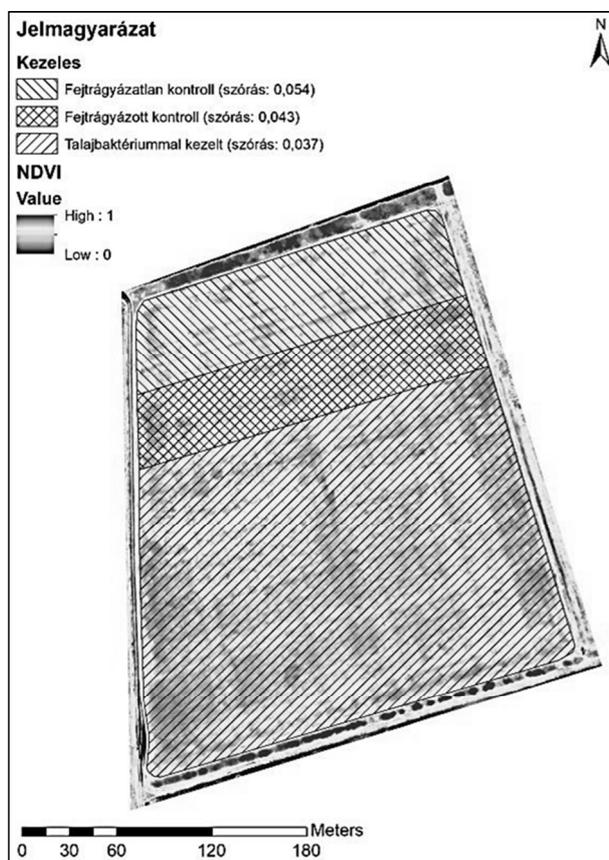


Table 3. Characteristics of soil properties (Kompolt, 2017)

Depth (cm)	Treatment	Moisture content %	Dry matter content %	pH (KCl)	Conductivity S m ⁻¹	Soluble salt in water % (m/m)	Gold's binding
20	Control	22.10	77.90	4.71	335	0.04	45
40	Control	20.10	79.90	4.68	464	0.05	47
20	I. treatment	19.45	80.55	4.70	304	0.03	42
40	I. treatment	18.20	81.80	4.70	351	0.04	47
20	II. treatment	21.63	78.37	4.57	372	0.04	45
40	II. treatment	20.81	79.19	4.52	325	0.03	44

Table 4. The nutrients content values of soil samples of different depths (mg kg^{-1}) (Kompolt, 2017)

Depth (cm)	Treatment	P	K	Ca	NO ₃ -N	NO ₂ -N	Mg	Se	Pb	Fe
20	Control	49118	9687	4606	24.2	0.04	5469	24.8	19.1	16733
40	Control	48935	9655	3053	9.9	0.02	5270	23.6	17.0	16066
20	I. treatment	47290	8960	4629	26.6	0.02	5285	24.6	19.0	16457
40	I. treatment	41642	8641	5500	0.9	0.04	6578	37.3	21.7	29196
20	II. treatment	46436	10115	5116	7.2	0.03	5612	25.7	19.9	28857
40	II. treatment	44184	9134	5223	2.5	0.03	5996	40.4	19.7	29112

Table 5. Values of nutrient content of stored grain yield (Kompolt, 2017)

Treatment	P	K	Na	Ca	Mg	Fe	Se	Pb
Control	2596	4438	258	1077	1502	51	6.2	0.9
I. treatment	2341	4440	282	903	1288	45	4.3	1.1
II. treatment	2175	4110	306	930	1233	41	3.5	1.2

Conclusions

The leaves of healthy, nearly the same size and color plants were measured, therefore measurements are not subjective. Presumably, under optimum conditions, the yield-enhancing effect is expected in the proportions of the experiment results. On aerial photographs, it is clear that parcels are heterogeneous, the most the control. Based on the results, the soil bacteria treatment can be recommended in public production, for which physiological processes were more intense, the ripening occurred sooner, and the moisture content at harvest was 1.6% lower on the soil bacteria than on the other two treatments. When evaluating soil samples, the head fertilization resulted the soil to have a lower moisture content but higher dry matter content. At depths of 20 cm on the soil bacteria treatment was measured low nitrate, phosphorus, but high potassium values. In the depths of 40 cm, the result was high selenium and iron content. In the case of the grain yield, were also lower phosphorus, potassium, calcium, magnesium, iron and selenium value than on the other two treatments. The soil bacteria treatment has a beneficial effect on soil pH reduction, deeper soil level in moisture conservation and nutrient utilization.

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