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The Effect of Biostimulants on the Chlorophyll Content and Height of *Solanum tuberosum* L. Plants

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ABSTRACT

The effect of foliar feeding with four biostimulants with active substances (Ecklonia maxima algae extract, titanium, humic substances, plant hormones: auxin and cytokinin) on the chlorophyll content and plant height of three Solanum tuberosum cultivars was investigated. A field experiment was carried out in 2015–2017 in eastern Poland in three growing seasons using the split-plot method. The cultivated varieties significantly differentiated the chlorophyll content (SPAD index) and plant height. The research showed the influence of varieties and many years of research on the height of potato plants. The biostimulants increased the value of the SPAD coefficient and the height of the potato plants.

Keywords: varieties, chlorophyll, potato, variants.

INTRODUCTION

The content of chlorophyll (SPAD index) and plant height (*Solanum tuberosum* L.) are influenced by agrotechnical, genetic and environmental factors [Ricci M. et al. 2019, Salem M.A. et al. 2020]. Contemporary agriculture looks for environmentally friendly farming methods. One of them is the use of biostimulants in plant production [Omidbakhshfard M.A. et.al. 2020]. The use of biostimulants in potato cultivation is justified by increasing the size and quality of the yield and the synthesis of chlorophyll. Their action is to stimulate the development of leaves, stems and roots of plants, to supplement the deficiency of nutrients during the growing season caused, among others, by intensive development of plants, drought, agrotechnical errors. Biostimulators enable a more effective absorption of nutrients from the substrate. An important element in potato agrotechnics is the process of "greening" and the constant reduction of unit cultivation costs. Currently, we are struggling with clearly noticeable climate changes and the associated increasingly extreme weather conditions, which are accompanied by various stress factors for plants, e.g. high and low temperatures, periodic droughts, floods, frosts [Abd EL-Wahab et al. 2016, Fleming T.R. et al. 2019]. In the presence of such unfavourable conditions in plant production, the use of biostimulants is particularly justified. [Kolachevskaya O.O. et al. 2019], increasing the vigour and vitality of plants, which makes it easier for them to survive unfavourable conditions during vegetation [Ahmadi H. et al. 2018, Cassia R. et al. 2018]. The use of biostimulants improving the condition of plants and the quality of the soil environment is in line with the international trend related to the reduction of soil and water chemicalization and the improvement of the quality of agricultural crops. Biostimulants applied during plant vegetation should change their metabolism in such a way that they become stronger and more resistant to pathogen attacks or the effects of unfavourable weather conditions [Trawczyński 2020].

The values of the SPAD index in leaves are strongly related to the content of plant nutrients, especially nitrogen [Su Y. et al. 2007; Udding J. 2007]. The authors found links between the SPAD index in potato leaves, plant height, yield and nitrogen content in this plant [Li R. et al. 2019]. Plant-derived biostimulants increased the SPAD index and height of crops [Caruso R. et al. 2019]. *Solanum tuberosum* L. converts solar energy into human food. The content of chlorophyll measured by the SPAD meter was correlated with the SPAD readings, which ensured a good evaluation of this component in the leaves.

Many researchers Caruso et al. [2019], Dima et al. [2020], Di Mola et al. [2019], Dvořák and Král [2019], report that, with certain readings, the SPAD index indicates that plants are optimally nourished with nitrogen and that yields are at their maximum levels and accumulate nutrients. The aim of the study was to assess the influence of biostimulants on the SPAD index value and the height of edible potato plants.

MATERIAL AND METHODS

The study material included potato tubers obtained from a field experiment carried out in-eastern (51°59' N and 22°47' E) Poland over three growing season, (2015–2017) with different weather conditions. The field experiment was established in a split-plot design with three replications. The examined factors were: factor I – was three edible potato cultivars: medium Jelly and medium early Honorata, and Tajfun; factor II – plant biostimulant: control without biostimulant, Kelpak®SL, Tytanit®, GreenOk®, BrunatneBio Złoto (Table 1). Soil parameters were determined before establishing the experiment (Table 2).

Agrotechnical measures used in the experiment are presented in Table 3. The content of chlorophyll (SPAD indicator) was measured twice at 10-day intervals with the portable SPAD-502 Plus Chlorophyl Meter, which measures the light absorption by the leaves at wavelengths of 650 and 940 nm. Measurements (10 readings) on the BBCH 67–68 scale, for each plot, around 10

Table 1. Description of biostimulants –II Factor

Preparations	Chemical composition
Kelpak®SL	Extract from algae Ecklonia maxima-auxins 11 mg dm^3 and gibberellins 0.031 mg dm^3, dose 0.2 dm^3 ha^{-1}, *
Tytanit®	Titanium, dose 0.2 dm^3 ha^{-1}, *
GreenOk®	Humus substances 20 g·dm^{-1}, dose 0.2 dm^3 ha^{-1}, *
BrunatneBio Złoto®	Plant hormones: auxin – 0.06 mg·dm^{-1} and cytokinin - 12 mg·dm^{-1}, dose 0.2 dm^3 ha^{-1}, *

*Usage: beginning of flowering, fully flowering and after flowering of plants

Table 2. Results of soil analysis in 2014–2016

Year	P available	K available	Mg available	N total	Organic matter
	(mg kg^{-1})				(g kg^{-1})
2014	65.6	145.4	51.0	11.0	15.0
2015	74.4	127.0	52.0	12.6	16.0
2016	114.0	145.0	55.0	12.9	18.7

Table 3. Agrotechnical treatments used in the experiment

Treatments	Specification	Dates
Fertilization	25 t ha^{-1} farmyard P – 44.0 (100 P_2O_5·0.44) kg·ha^{-1} (lubofos for potatoes 7%) and K – 124.5 (150 K_2O·0.83) kg·ha^{-1} (lubofos for potatoes 25%) N 100 kg/ha (nitro-chalk 27%)	Autumn, spring
Planting	Spacing 0.675 x 0.37 m	Second week of April
Insectidides	Actara 25 WG (thiametoksam)w dawce 0,08 kg·ha^{-1} i Calipso 480 S.C. (thiacloprid) w dawce 0.1 dm·ha^{-1}	During vegetation
Fungicides	Ridomil Gold MZ 68 WG (metalaxyl-M+mancozeb) and Copper Max New 50 WP at the rate 2,0 g·kg^{-1} , and Dithane at the rate 2.0 kg·ha^{-1}	During vegetation

o'clock a fully developed leaf, i.e. the fourth or fifth leaf from the top, was taken.

Meteorological conditions

The thermal and humidity conditions during the potato vegetation period were different (Table 4).

The average air temperatures in 2015 and 2016 were higher or close to the long-term average. In 2017, the lowest air temperatures were below the long-term average, the rainfall was similar, and in 2015 and 2017 above the long-term average, although it was unevenly distributed during the potato growth period. The most favorable hydrothermal conditions for harvesting potatoes the cultures were in the warm and moderately wet growing season of 2015. In the following 2016, it was warm and abundant rainfall, while 2017 was cool with abundant rainfall, a period of tuber growth. According to the Sielianinov hydrothermal coefficient, the 2015 growing season can be described as rather dry, 2016 dry and 2017 wet [Skowera et al. 2014].

Statistical analysis

The results of the three year study were analysed statistically with an analysis of variance ANOVA for the two-way split-plot arrangement. The significance of differences between the compared averages was verified using Tukey's test at the significance level P≤0.05. Calculations were performed in Excel using the authors' own algorithm based on the split-plot mathematical model.

$$Yijl = m + ai + gl + e/1/il + b\,j + ab\,ij + e/2/ijl \quad (1)$$

where: Yijl – value of the characteristic researched;
- i – level of A (cultivars),
- j – level of B (cultivars) in the 1_{st} block (replication),
- m – experimental mean,
- ai – effect of i-level of A (cultivars),
- gl – effect of the 1_{st} replication,
- e/1/il – random effect of A (cultivars) with replications,
- bj – effect of j-level of B (biostimulants),
- abij – effect of interaction of A (cultivars) and B (biostimulants),
- e/2/ijl – random effect II.

Table 4. Rainfall and average air temperature in 2015–2017

Month	2015	2016	2017	Multi-year mean 1980–2009
	Monthly rainfall (mm)			
4. April	30.0	28.7	59.6	33.6
5. May	100.2	54.8	49.5	58.3
6. June	43.3	36.9	57.9	59.6
7. July	62.6	35.2	23.6	57.5
8. August	11.9	31.7	54.7	59.9
9. Septembr	47.1	13.6	80.1	42.3
Total April-September	295.1	200.9	325.4	335.4
Average monthly air temperature (°C)				
4. April	8.2	19.1	6.9	8.0
5. May	12.3	15.1	13.9	13.5
6. June	16.5	18.4	17.8	17.0
7. July	18.7	19.1	16.9	19.7
8. August	21.0	18.0	18.4	18.5
9. Septembr	14.5	14.9	13.9	13.5
Mean April-September	15.2	15.8	14.6	15.0

Hydrothermal Index							
Year	4. April	5. May	6. June	7. July	8. August	9. September	Mean
2015	1.35	2.91	0.84	1.20	0.20	1.20	1.30
2016	1.08	1.47	0.72	0.64	0.62	0.28	0.80
2017	3.82	1.52	1.07	0.47	1.01	1.92	1.63

Hydrothermal index value: up to 0.4 extremely dry; 0.41–0.7 very dry; 0.71–1.0 dry; 1.01–1.3 rather dry; 1.31–1.6, optimal; 1.61–2 rather humid; 2.01–2.5 humid; 2.51–3 very humid; >3 extremely humid [Skowera et al. 2014]

RESULTS AND DISCUSSION

The content of chlorophyll - index SPAD compiled in the first term was on average 40.47 units, and depending on the cultivars, variants of using biostimulants and the hydrothermal conditions in sthe the study, it ranged from 39.17 to 41.87 (Tables 5, 6). In the second term of determinations, the SPAD indicator was higher – it amounted to an average of 40.84, depending on the variants and cultivars, it ranged from 40.21 to 41.87 units. Similar values of the SPAD index were shown by Trawczyński (2019).

The cultivars differed significantly in terms of the SPAD index. The highest significant value of chlorophyll content was recorded for the variety Honorata, significantly lower for Jelly, and the lowest for the Tajfun cultivar (Table 5). In the study by Zarzyńska and Pietraszko (2017), potato varieties differed significantly in terms of the Spad index In the authors' own research, no interactions of cultivated varieties with variants of biostimulants used in both study dates were found.

In the conducted studies, the values SPAD index depended on the variants of the biostimulants used. These preparations increased SPAD parameters compared to the control variant, and the highest readings were recorded after the application of the BrunatneBio Złoto® biostimulant (Table 5, 6). Similar research results were obtained by Dvořák et al. (2016), Trawczyński (2019) noticed the influence of safe preparations for the environment, producers and consumers, such as biostimulants, on the value of the SPAD index in potato leaves, while Wadas and Dziugieł (2020) did not note the effect of biostimulants on the SPAD values. The own research found the influence of years of research on the leaf greenness index. The highest SPAD values were recorded in 2015, the season in which the average temperatures were about 15.2^{o}C and the rainfall was 295.1mm, and the lowest SPAD values in 2017, which turned out to be humid, but with the lowest average air temperature. A significant influence of study years on SPAD was found during the first term of the study, while in the second term, an insignificant effect of study years on this trait was demonstrated, and no interactions of years with variants of biostimulant application on SPAD determined on both dates were found (Table 6). Wadas and Dziugieł (2020) in their research confirmed the influence of years of research on the values of the SPAD index, which turned out to be the highest in the warm and wet season.

The height of potato plants was significantly determined by the cultivated varieties, variants of biostimulant application and weather conditions (Table 7, 8). The potato plants of the Honorata cultivar achieved the highest height, and Tajfun cultivars the lowest. The applied biostimulants

Table 5. Index SPAD depending on cultivar

Variants of biostimulants (II)	Cultivars (I)			Mean
	Jelly	Honorata	Tajfun	
SPAD I termin				
1. Control variant	40.17A	40.10A	39.89A	40.05c
2. Kelpak SL®	40.52A	41.21A	40.03A	40.58b
3. Tytanit®	40.15A	40.78A	40.12A	40.35b
4. GreenOK®	40.13A	40.89A	40.17A	40.39b
5. BrunatneBio Złoto®	41.13A	41.54A	40.32A	40.99a
Mean	40.42b	40.90a	40.10b	40.47
SPAD II termin				
6. Control variant	40.67A	40.35A	40.21A	40.41b
7. Kelpak SL®	40.76A	41.35A	40.34A	40.81b
8. Tytanit®	40.88A	41.23A	40.45A	40.85b
9. GreenOK®	40.92A	41.43A	40.55A	40.96b
10. BrunatneBio Złoto®	40.98A	41.87A	40.65A	41.16a
Mean	40.84b	41.24a	40.44b	40.84

Means followed by the same letters do not differ significantly at P≤0.05. Means in columns marked with capital letters refer to interactions between the factors. Means in the last row (followed by lowercase) are for variants and cultivars.

increased the plant height in relation to the control object. The highest potato height was recorded after the application of the BrunatneBio Złoto® biostimulant (Table 7). The positive effect of biostimulants on the height of potato plants was shown by Trawczyński (2020).

Table 6. Index SPAD depending on weather conditions during the years 2015–2017

Variants of biostimulants (II)	Years			Mean
	2015	2016	2017	
SPAD I termin				
1. Control Variant	40.60A	39.17A	40.39A	40.05c
2. Kelpak SL®	41.31A	40.22A	40.23A	40.58b
3. Tytanit®	40.58A	40.25A	40.22A	40.35b
4. GreenOK®	40.60A	40.32A	40.27A	40.39b
5. BrunatneBio Złoto®	41.45A	41.17A	40.37A	40.99a
Mean	40.90a	40.22b	40.30b	40.47
SPAD II termin				
1. Control Variant	40.42A	40.40A	40.41A	40.41b
2. Kelpak SL®	40.56A	41.45A	40.44A	40.81b
3. Tytanit®	41.68A	40.43A	40.45A	40.85b
4. GreenOK®	40.72A	41.33A	40.85A	40.96ab
5. BrunatneBio Złoto®	41.78A	40.87A	40.85A	41.16a
Mean	41.03a	40.9a	40.6a	40.84

Means followed by the same letters do not differ significantly at P≤0.05. Means in columns marked with capital letters refer to interactions between the factors. Means in the last row (followed by lowercase) are for variants and years.

Table 7. Plant height in potato depending on cultivar

Variants of biostimulants	Cultivars			Mean
	Jelly	Honorata	Tajfun	
Potato plant height (cm)				
1. Control Variant	61.10A	61.50A	60.02A	60.87d
2. Kelpak SL®	61.53A	61.65A	60.67A	61.28c
3. Tytanit®	61.67A	61.69A	60.73A	61.36c
4. GreenOK®	61.58A	62.30A	60.83A	61.57b
5. BrunatneBioZłoto®	61.89A	62.60A	60.90A	61.80a
Mean	61.55b	61.95a	60.63c	61.37

Means followed by the same letters do not differ significantly at P≤0.05. Means in columns marked with capital letters refer to interactions between the factors. Means in the last row (followed by lowercase) are for variants and cultivars.

Table 8. Plant height in potato depending on weather conditions during the study years

Variants of biostimulants	Years			Mean
	2015	2016	2017	
Potato plant height (cm)				
1. Control Variant	59.02A	60.10A	63.50A	60.87c
2. Kelpak SL®	60.37A	60.53A	62.95A	61.28b
3. Tytanit®	60.63A	60.47A	62.99A	61.36b
4. GreenOK®	60.53A	60.38A	63.80A	61.57ab
5. BrunatneBioZłoto®	59.50A	60.59A	65.30A	61.80a
Mean	60.01b	60.41b	63.70a	61.37

Means followed by the same letters do not differ significantly at P≤0.05. Means in columns marked with capital letters refer to interactions between the factors. Means in the last row (followed by lowercase) are for variants and years.

Weather conditions during vegetation significantly differentiated plant height. The largest plants were in 2017 and the smallest in 2015. The studies proved that the leaf greenness index SPAD at both determination dates was strongly related to height in all cultivars (Table 8).

CONCLUSION

The biostimulants used in the experiment increased the leaf greenness index SPAD and the plant height of three potato cultivars compared to the control object. The own research revealed the effect of varieties and years of research on the height of potato plants and the SPAD index determined with the Konica Minolta SPAD-502Plus measuring apparatus.

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