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Quality of New Potatoes (*Solanum tuberosum* L.) in Response to Plant Biostimulants Application

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Abstract: Background: In sustainable crop production focusing on high-value products, biostimulants have been gaining increasing importance, thus the hypothesis that plant biostimulants could contribute to improving new potatoes quality; Methods: The effects of the seaweed extracts Bio algeen S90 (*Ascophyllum nodosum*) and Kelpak SL (*Ecklonia maxima*), as well as the humic and fulvic acids in HumiPlant (leonardite extract) on the tuber quality of very early potato cultivars ('Denar', 'Lord', 'Miłek') were investigated. Potatoes were harvested 75 days after planting (the end of June); Results: The biostimulants did not affect dry matter, protein, total sugars, monosaccharides and sucrose or L-ascorbic acid content in new potatoes. Bio-algeen S90 increased the starch content in tubers of all potato cultivars tested, on average, by 4.8 g·kg^{-1} compared with control treatment without biostimulant, whereas Kelpak SL and HumiPlant reduced nitrates content only in tubers of 'Denar' cultivar, on average, by 8.50 mg·kg^{-1}, and increased ascorbate-nitrate index (I_{AN}) by 0.29. The biostimulants did not affect potato after-cooking darkening. Both the nutritional value of new potatoes and after-cooking darkening depended on the cultivar and weather conditions during the potato growing period to a great extent; Conclusions: Plant biostimulants slightly affected quality of new potatoes.

Keywords: seaweed extract; humic acids; nutritional value; after-cooking darkening

1. Introduction

Potatoes play an important role in the global food security, nutrition and healthy diet [1]. Potatoes provide a significant amount of high-quality protein, vitamin C (mainly L-ascorbic acid) and group B vitamins, minerals as well as other health-promoting compounds. Consumption of 250–300 g of boiled potatoes provides about 7–8% of the recommended daily intake of protein, 6–11% of carbohydrates, 50% of vitamin C, 30–40% of potassium and about 17% of fibre. Apart from nutrients, potato tubers also contain anti-nutritional substances such as nitrates or glycoalkaloids. The quality of edible potatoes is determined by their nutritional value and low content of anti-nutrients. An important quality characteristic is also sensory properties including after-cooking darkening [2–4]. The chemical composition of potato tubers depends on the cultivar, tuber size and maturity, but may change under environmental (weather and soil conditions) and agronomic factors [5–7].

In sustainable crop production focusing on high-value products, biostimulants have been gaining increasing importance. These natural products enhance nutrition efficiency, abiotic stress tolerance and/or crop quality traits. Plant biostimulant based on seaweed extracts and humic acids have the largest market share [8–10].

Seaweed extracts are complex mixtures with multiple mechanisms of action. Bioactive compounds present in seaweed extracts improve plant growth, enhance nutrient use efficiency, improve plant defences against pathogens and improve crop quality. The bioactivity of seaweed extracts depend on the algae species and the extraction method. Most commercial seaweed products used as plant biostimulants in agriculture and horticulture are manufactured from brown seaweeds (*Phaeophyta*). *Ascophyllum nodosum* (*A. nodosum*) and *Ecklonia maxima* (*E. maxima*) are dominant in this group [11–13]. The *A. nodosum* extract Bio-algeen S90 and the *E. maxima* extract Kelpak SL are used worldwide as a biostimulants for a number of agricultural and horticultural crops. Biostimulants based on *A. nodosum* extracts are used to improwe plant growth and to mitigate abiotic and biotic stresses. The effect of *A. nodosum* extracts on plants are attributed to phytohormone, microelement, and/or alga-specific polysaccharides, betaines, polyamines and phenolic compounds contents [14,15]. *E. maxima* extract Kelpak SL contains auxins, cytokinins, polyamines, abscisic acid, gibberellin, brassinosteriods and a small amounts of macro- and microelements. The active compounds present in Kelpak SL, alone or in combination, bring contribute to enhance plant growth and yield, and improve biotic and abiotic stresses tolerance. Content of some active compounds in Kelpak is higher than in other commercial products based on *E. maxima* extract [16,17]. Commercial seaweed products manufactured from the same seaweed source by different companies, generally marketed as equivalent products, may vary significantly in product composition and in efficacy to induce specific plant responses following appplication, especially under field conditions. Commercial *A. nodosum* extracts Phylgreenmira, Algazone and Ultra-Kelp increased the dry matter and starch content in potato tubers, as well as slightly decreased protein content [18], whereas Bio-algeen S90 did not affect the starch, total nitrogen content or the potato after-cooking darkening [19,20]. Other products based on *A. nodosum* extract Primo increased nitrogen and protein content in potato tubers [21]. Bio-algeen S90 did not affect dry matter content in carrot, but increased L-ascorbic acid and total sugar contents [22]. *E. maxima* extract Kelpak SL did not affect starch or total nitrogen content in potato tubers, but increased vitamin C and nitrate contents [20,23]. Kelpak SL increased nitrate and nitrite content in carrot and protein content in beans, decreased starch content in wheat, but did not affect protein or fat content in winter rape [24–27].

The biostimulant effects of humic substances are characterized by both structural and physiological changes in plants related to nutrient uptake, assimilation and distribution, and changes in plant primary and secondary metabolism related to abiotic stress tolerance. The biological activity of humic substances depends on their source, chemical structure, and concentrations. Leonardite is the most common commercial source of humic substances [28–30]. Humic substances extracted from leonardite stimulate plant growth, nitrogen metabolism and accumulation of phenolic compounds. The positive effects of humic substances on plant metabolism are attributed to phytohormone-like activity. Biostimulant activity of humic substances extracted from leonardites depends on the origin of the leonardite [31]. Humic acids derived from leonardite applied to soil or introduced into the irrigation system increased dry matter, starch and protein content in potato tuber [32–34]. Soil application of humic acids did not affect dry matter or starch content in potato tubers, whereas the foliar application of fulvic acids did not affect tuber dry matter but increased starch content [35]. Foliar application of humic and fulvic acids in HumiPlant, a commercial extract from leonardite, increased the sugar and carotenoid contents and decreased the nitrate content in carrot [36]. Soil and foliar application of humic acid increased the sugar content in pepper and cucumber [37,38], whereas foliar application of humic and fulvic acids increased the vitamin C content and reduced tomato acidity [39].

To date, few studies have been focused on the effect of plant biostimulants on potato tuber quality. The current study aimed to determine the effect of foliar application of seaweed extracts and humic and fulvic acids on the quality of new potatoes. In the current study, it was hypothesised that seaweed extracts and humic acids could contribute to improving edible potato quality.

2. Materials and Methods

2.1. Plant Material and Experimental Design

The study material included potato tubers obtained from a field experiment carried out in central-eastern Poland (52°03′ N; 22°33′ E) over three growing season, (2012–2014) with different weather conditions (Table 1).

Table 1. Hydrothermal conditions during potato growing period.

Months	Average Daily Temperature (°C)				Rainfall (mm)				Hydrothermal Index		
	Long-Term Average 1981–2010	Deviation from Long-Term Average			Long-Term Average 1981–2010	Deviation from Long-Term Average					
		2012	2013	2014		2012	2013	2014	2012	2013	2014
April	8.3	+0.6	−0.9	+1.5	41.2	−11.3	−5.2	+3.8	1.1	1.6	1.5
May	12.2	+2.4	+3.1	+1.3	53.0	−0.4	+52.9	+39.7	1.2	2.2	2.2
June	16.8	−0.8	+1.2	−1.4	63.8	+12.4	+35.0	−8.4	1.5	1.8	1.2

Hydrothermal index value: up to 0.4 extremely dry; 0.41–0.7 very dry; 0.71–1.0 dry; 1.01–1.3 quite dry; 1.31–1.6 optimal; 1.61–2.0 quite wet; 2.01–2.5 wet; 2.51–3.0 very wet; >3 extremely wet [40].

The field experiment was carried out on Luvisol with pH in KCl from 4.7 to 6.3. The content of total nitrogen in soil ranged from 7 to 11 mg N·kg^{-1}, the content of available phosphorus ranged from 118 to 144 mg P·kg^{-1}, potassium from 124 to 208 mg K·kg^{-1} and magnesium from 22 to 51 mg Mg·kg^{-1} of soil. The soil chemical properties were determined using soil laboratory procedures at the National Chemical and Agricultural Station: pH with potentiometric method in 1 mol·dm^{-3} KCl [41], total nitrogen with the Kjeldahl method [42], available forms of phosphorus with spectrophotometric method [43], potassium with the flame atomic emission spectroscopy (FAES) method [44] and magnesium with flame atomic absorption spectroscopy (FAAS) method [45].

The field experiment was established in a split-plot design with three replications. The examined factors were: factor I–plant biostimulant: control without biostimulant, Bio-algeen S90, Kelpak SL and HumiPlant (Table 2), factor II–potato cultivar: 'Denar', 'Lord' and 'Miłek'(Table 3).

Table 2. Characteristics of plant biostimulants; according to manufacturers.

Plant Biostimulant	Active Compounds
Bio-algeen S90 *Ascophyllum nodosum* extract	amino acids, vitamins, alginic acids and other active components of seaweeds, macronutrients (N, P, K, Ca, Mg) and micronutrients (B, Fe, Cu, Mn, Zn, Se, Co)
Kelpak SL *Ecklonia maxima* extract	auksin (11 mg·dm^{-3}), cytokinin (0.031 mg·dm^{-3})
HumiPlant leonardite extract	humic acid (12%), fulvic acid (6%), macronutrients (K, Ca, Mg, S) and micronutrients (Fe, Mn, B, Mo, Zn, Cu)

Table 3. Characteristics of potato cultivars [46].

Cultivar	Country of Origin	Cooking Type	Plants' Soil Requirements	Plants' Water Requirements
Denar	Poland	AB	medium–large	medium–large
Lord	Poland	AB	medium	tolerant
Miłek	Poland	BC	medium	medium–large

Cooking type: AB–multi-purpose type to salad type, BC–multi-purpose type to floury type.

6-weeks pre-sprouted seed potatoes were planted on 12 April 2012, 18 April 2013 and 7 April 2014, with a row spacing of 0.250 m and 0.675 m between rows. The plots were six rows wide and 4 m long (96 plants per plot). Potato cultivation was carried out according to common agronomical practice.

The biostimulants were applied twice, in one or two-weeks intervals, according to the manufacturers' recommendations: Bio-algeen S90 at the beginning of leaf development stage (according to a uniform coding of phenologically similar growth stages of plant species of Biologische Bundesanstalt, Bundessortenamt and Chemical Industry the BBCH 10–11 stage) and two weeks after the first treatment, Kelpak SL at the leaf development stage (BBCH 14–16) and two weeks after the first treatment, HumiPlant at the leaf of development stage (BBCH 14–16) and one week after the first treatment [47]. In each treatment, the biostimulants were applied at the dose of 2 $dm^3 \cdot ha^{-1}$. Potato plants sprayed with water were used as a control without a biostimulant. Due to different active compounds, the application of the tested biostimulants at different time points in the early stages of potato growth ensure better root system development and better the plants growth at later time. Potatoes were harvested 75 days after planting (the end of June).

2.2. Laboratory Analysis

Laboratory studies were conducted on samples of 50 different-sized tubers taken from each plot. Fresh potatoes were analysed immediately after sampling.

Potato tubers were analysed for:

- dry matter–with the gravimetric method by drying to the constant weight at 105 °C [48],
- starch–with the polarimetric method according to Ewers after hydrolyze with 0.1 N (0.1 M) hydrochloric acid (HCl); the optical rotation was measured at a wavelength of 589 nm [49],
- total sugars (glucose, fructose and sucrose), monosaccharides (glucose and fructose) and sucrose–with the Luff-Schoorl method after inversion to reducing sugars and reduction, under alkaline medium, cupric (Cu^{2+}) ions of copper sulphate ($CuSO_4$) to cuprous (Cu^+) oxide (Cu_2O); sucrose content was calculated from the difference of the total sugars after inversion and the reducing sugars, using the conversion factor 0.95 [50],
- total protein–with the Kjeldahl method using the conversion factor of total nitrogen to total protein 6.25 [51],
- L-ascorbic acid–with the titration method with 2,6-dichlorophenolindophenol (DCPIP) according to Tillmans after extraction with 2% oxalic acid ($C_2H_2O_4 \cdot H_2O$) [52],
- nitrates–with the spectrophotometric method based on the Griess reaction after reduction of the nitrates to nitrites with cadmium dust; the absorbance was measured at a wavelength of 538 nm [53].

The results were expressed as grams or milligrams per kilogram of fresh weight (FW) of potatoes. The ascorbate-nitrate index (I_{AN}) was calculated as the ratio of L-ascorbic acid amount-to-nitrate amount in potato tubers [54].

Potato after-cooking darkening was also determined. The assessment of potato after-cooking darkening was performed after 10 min and 2 h following boiling in water, using the 9-point Danish scale on which 9 means no darkening and 1 denotes the strongest darkening. The assessment of potato after-cooking darkening was conducted on ten tubers per plot [55].

2.3. Statistical Analysis

The results of the three-year study were analysed statistically using a two-factor analysis of variance (ANOVA) for the split-plot design (plant biostimulant × potato cultivar × years). The significance of the sources of variability was tested using the Fisher-Snedecor test, and the significance of differences between the compared averages was verified using Tukey's test at the significance level $p \leq 0.05$. Statistical calculations were performed in Excel software using the authors' own algorithm based on the split-plot mathematical model.

3. Results

3.1. Dry Matter and Carbohydrates Content

The biostimulants used in the experiment had no effect on dry matter content in tubers (Table 4). The dry matter content in potato tuber depended on the cultivar and weather conditions to a greater extent.

Table 4. Dry matter content in potato tubers; g·kg^{-1} FW.

Plant Biostimulant	Years			Cultivar			Mean
	2012	2013	2014	Denar	Lord	Miłek	
Without biostimulant	168.8 a	194.4 a	184.3 a	179.8 a	175.0 a	192.8 a	182.5 A
Bio-algeen S90	166.7 a	192.0 a	184.0 a	177.6 a	173.7 a	191.4 a	180.9 A
Kelpak SL	165.7 a	188.0 a	188.7 a	174.9 a	178.0 a	189.6 a	180.8 A
HumiPlant	168.7 a	197.9 a	184.4 a	174.2 a	179.6 a	197.2 a	183.7 A
Mean	167.5 B	193.1 A	185.4 A	176.6 B	176.6 B	192.7 A	182.0

Means within columns/rows followed by the same lowercase/uppercase letters do not differ significantly at $p \leq 0.05$. Means in columns marked with lowercase refer to interactions: plant biostimulant × years, plant biostimulant × cultivar. Means in the last column and means in the last row marked with uppercase are for plant biostimulants, years and cultivars.

Regardless of the treatment (with or without biostimulant), tubers of 'Miłek' contained more dry matter than 'Denar' and 'Lord'. The most dry matter was accumulated by potato tubers in the warm and moist growing season of 2013. The plant biostimulant and potato cultivar interaction effect on the dry matter content in tubers was not found.

The plant biostimulant had a significant effect on starch content but did not affect the content of total sugars (glucose, fructose and sucrose), monosaccharides (glucose and fructose) or sucrose (Table 5).

Table 5. Carbohydrates content in potato tubers.

Plant Biostimulant	Years			Cultivar			Mean
	2012	2013	2014	Denar	Lord	Miłek	
Starch (g·kg^{-1} FW)							
Without biostimulant	119.0 a	130.4 a	98.1 b	108.3 a	115.7 a	123.5 a	115.9 B
Bio-algeen S90	119.1 a	133.2 a	109.8 a	113.6 a	116.1 a	132.4 a	120.7 A
Kelpak SL	119.3 a	134.3 a	97.0 b	112.8 a	113.0 a	124.9 a	116.9 B
HumiPlant	120.8 a	133.8 a	97.2 b	113.3 a	114.6 a	123.9 a	117.3 AB
Mean	119.6 B	132.9 A	100.5 B	112.0 C	114.8 B	126.2 A	117.7
Total sugars (g·kg^{-1} FW)							
Without biostimulant	7.92 a	8.13 a	6.30 a	7.32 a	7.28 a	7.76 a	7.45 A
Bio-algeen S90	8.44 a	7.68 a	6.18 a	7.38 a	7.40 a	7.52 a	7.43 A
Kelpak SL	7.58 a	8.21 a	6.40 a	7.24 a	7.37 a	7.58 a	7.40 A
HumiPlant	7.41 a	7.99 a	6.18 a	7.14 a	7.30 a	7.13 a	7.19 A
Mean	7.84 A	8.00 A	6.26 B	7.27 A	7.34 A	7.50 A	7.37
Monosaccharides (g·kg^{-1} FW)							
Without biostimulant	2.39 a	2.36 a	2.40 a	2.17 a	2.38 a	2.60 a	2.38 A
Bio-algeen S90	2.47 a	2.27 a	2.17 a	2.31 a	2.32 a	2.27 a	2.30 A
Kelpak SL	2.29 a	2.63 a	2.23 a	2.37 a	2.36 a	2.43 a	2.39 A
HumiPlant	2.24 a	2.22 a	2.38 a	2.33 a	2.09 a	2.32 a	2.25 A
Mean	2.35 A	2.37 A	2.27 A	2.29 A	2.29 A	2.41 A	2.33

Table 5. *Cont.*

Plant Biostimulant	Years			Cultivar			Mean
	2012	2013	2014	Denar	Lord	Miłek	
	Sucrose (g·kg^{-1} FW)						
Without biostimulant	5.26 a	5.29 a	3.70 a	4.80 a	4.66 a	4.79 a	4.75 A
Bio-algeen S90	5.68 a	5.14 a	3.81 a	4.81 a	4.82 a	4.99 a	4.88 A
Kelpak SL	5.02 a	5.30 a	3.96 a	4.63 a	4.76 a	4.89 a	4.76 A
HumiPlant	4.91 a	5.48 a	3.72 a	4.58 a	4.95 a	4.57 a	4.70 A
Mean	5.22 A	5.30 A	3.80 B	4.71 A	4.80 A	4.81 A	4.77

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Following the application of Bio-algeen S90, the starch content was higher, on average, by 4.8 g·kg^{-1} FW, compared with the control treatment without biostimulant. The greatest difference was found in the year with the lowest air temperature and a drought in the tuber growth period. In that year (2014), following the Bio-algeen S90 application, the starch content was higher by 11.7 g·kg^{-1} FW, compared with the control treatment without biostimulant. The plant biostimulant and potato cultivar interaction effect on the starch and sugars content in tubers were not found.

The starch and sugars content in potato tuber depended on the cultivar and weather conditions to a greater extent (Table 5). Regardless of the treatment (with or without biostimulant), tubers of ‘Miłek’ contained more starch than ‘Denar’ and ‘Lord’ whereas the content of total sugars, monosaccharides and sucrose in tubers of the potato cultivars tested were similar. The ratio of starch-to-sugars amounted, on average, 15.5/1 for ‘Denar’ and ‘Lord’, and 16.8/1 for ‘Miłek’, whereas the ratio of sucrose-to-monosaccharides for the potato cultivars tested amounted 2.1/1, on average. The most starch were accumulated by potato tubers in the warm and quite wet growing season of 2013, whereas the least total sugars were accumulated in tubers in 2014, with the lowest air temperature and with a drought periods during tuber growth.

3.2. Protein Content

The biostimulants used in the experiment had no effect on protein content in tubers (Table 6). The protein content in potato tuber depended on the cultivar and weather conditions during the potato growing period to a greater extent.

Table 6. Protein content in potato tubers; g·kg^{-1} FW.

Plant Biostimulant	Years			Cultivar			Mean
	2012	2013	2014	Denar	Lord	Miłek	
Without biostimulant	13.70 a	16.04 a	14.33 a	14.36 a	14.24 a	15.48 a	14.69 A
Bio-algeen S90	13.31 a	15.42 a	14.09 a	13.57 a	13.81 a	15.44 a	14.27 A
Kelpak SL	13.28 a	14.87 a	14.29 a	13.64 a	14.41 a	14.38 a	14.14 A
HumiPlant	14.20 a	15.84 a	14.13 a	13.98 a	14.47 a	15.74 a	14.73 A
Mean	13.62 B	15.54 A	14.21 B	13.88 B	14.23 B	15.26 A	14.46

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Regardless of the treatment (with or without biostimulant), tubers of ‘Miłek’ contained more protein than ‘Denar’ and ‘Lord’. The most protein was accumulated by potato tubers in the warm and moist growing season of 2013 (Table 6). The plant biostimulant and potato cultivar interaction effect on the protein content in tubers was not found.

3.3. L-ascorbic Acid and Nitrates Content

The biostimulants used in the experiment had no effect on L-ascorbic acid or nitrate content in tubers (Table 7). Following the biostimulant application, there was only a slight increase in the L-ascorbic acid content and a slight decrease in the nitrate content. The differences were not statistically confirmed.

Table 7. L-ascorbic acid and nitrates content in potato tubers.

Plant Biostimulant	Years			Cultivar			Mean
	2012	2013	2014	Denar	Lord	Miłek	
L-ascorbic acid (mg·kg^{-1} FW)							
Without biostimulant	126.3 a	126.7 a	117.2 a	123.2 a	125.7 a	121.3 a	123.4 A
Bio-algeen S90	130.3 a	129.8 a	114.6 a	124.7 a	123.9 a	126.1 a	124.9 A
Kelpak SL	130.9 a	127.6 a	119.0 a	129.7 a	125.4 a	122.3 a	125.8 A
HumiPlant	125.8 a	128.3 a	124.6 a	128.8 a	127.0 a	122.9 a	126.2 A
Mean	128.3 A	128.1 A	118.8 B	126.6 A	125.5 A	123.2 A	125.1
Nitrates (mg·kg^{-1} FW)							
Without biostimulant	69.78 a	71.44 a	69.67	73.22 a	65.56 a	72.11 a	70.30 A
Bio-algeen S90	66.56 a	68.44 a	66.44	68.56 ab	58.33 a	74.56 a	67.15 A
Kelpak SL	67.89 a	66.78 a	66.78	64.78 b	65.33a	71.33 a	67.15 A
HumiPlant	70.44 a	68.56 a	66.22	64.67 b	65.00 a	75.56 a	68.41 A
Mean	68.67 A	68.81 A	67.28 A	67.81 B	63.56 B	73.39 A	68.25
Ascorbate-nitrate index (I_{AN})							
Without biostimulant	1.82 a	1.79 a	1.74 a	1.71 b	1.95 a	1.70 a	1.78 A
Bio-algeen S90	1.99 a	1.93 a	1.78 a	1.85 ab	2.14 a	1.71 a	1.90 A
Kelpak SL	1.95 a	1.92 a	1.79 a	2.01 a	1.93 a	1.72 a	1.89 A
HumiPlant	1.82 a	1.88 a	1.90 a	2.00 a	1.96 a	1.63 a	1.86 A
Mean	1.89 A	1.88 A	1.80 B	1.89 AB	2.00 A	1.71 B	1.86

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The plant biostimulant and potato cultivar interaction effect on the L-ascorbic acid content in tubers was not found (Table 7). The plant biostimulant applied had a greater effect on the nitrate content of 'Denar' than 'Lord' and 'Miłek'. Following the application of Kelpak SL and HumiPlant, the nitrate contents in the tubers of 'Denar' were lower, on average, by 8.50 mg·kg^{-1} FW, compared with the control treatment without biostimulant, while the ratio of L-ascorbic acid amount-to-nitrate amount in tubers (I_{AN}) was higher by 0.29.

Regardless of the plant biostimulant applied, tubers of the tested potato cultivars had similar L-ascorbic acid, but 'Miłek' accumulated the most nitrates (Table 7). The I_{AN} value for 'Miłek' tubers was 0.29 lower than for 'Lord', on average, and was 0.18 lower compared to 'Denar'. The lowest L-ascorbic acid was accumulated by potato tubers in the year with the lowest air temperature and a moisture shortage in the tuber growth period (2014). The weather conditions during the potato growing period had no effect on the nitrate accumulation in potato tubers.

3.4. Potato After-Cooking Darkening

Biostimulants did not affect the potato after-cooking darkening (Table 8). This quality characteristic of edible potatoes depended to a greater extent on the cultivar and weather conditions during the potato growing season.

Table 8. Potato after-cooking darkening.

Plant Biostimulant	Years			Cultivar			Mean
	2012	2013	2014	Denar	Lord	Miłek	
10 min after cooking (9-point Danish scale)							
Without biostimulant	8.72 a	8.94 a	8.94 a	8.94 a	9.00 a	8.67 a	8.87 A
Bio-algeen S90	8.72 a	8.89 a	9.00 a	8.94 a	8.94 a	8.72 a	8.87 A
Kelpak SL	8.89 a	8.83 a	9.00 a	8.94 a	8.94 a	8.83 a	8.91 A
HumiPlant	9.00 a	8.83 a	9.00 a	8.94 a	9.00 a	8.89 a	8.94 A
Mean	8.83 B	8.88 AB	8.99 A	8.94 A	8.97 A	8.78 B	8.90
2 h after cooking (9-point Danish scale)							
Without biostimulant	8.61 a	8.89 a	8.83 a	8.89 a	8.89 a	8.56 a	8.78 A
Bio-algeen S90	8.61 a	8.61 a	8.83 a	8.83 a	8.78 a	8.44 a	8.68 A
Kelpak SL	8.78 a	8.61 a	8.78 a	8.83 a	8.72 a	8.61 a	8.72 A
HumiPlant	8.83 a	8.72 a	8.89 a	8.89 a	8.89 a	8.67 a	8.82 A
Mean	8.71 A	8.71 A	8.83 A	8.86 A	8.82 A	8.57 B	8.75

Means within columns/rows followed by the same lowercase/uppercase letters do not differ significantly at $p \leq 0.05$. Means in columns marked with lowercase refer to interactions: plant biostimulant × years, plant biostimulant × cultivar. Means in the last column and means in the last row marked with uppercase are for plant biostimulants, years and cultivars.

'Miłek' tubers showed greater susceptibility to darkening, both directly after cooking and two hours after cooking. The greatest potato susceptibility to after-cooking darkening, especially directly after cooking, was observed in the warm and moderately wet growing season of 2012. The plant biostimulant and cultivar interaction effect on the potato after-cooking darkening was not found.

4. Discussion

The biostimulants used in the experiment slightly affected potato tuber quality. The dry matter content is one of the most important characteristics of new potatoes. When potatoes are harvested early, low dry matter content can result in a soggy texture and decrease the quality of new potatoes. The biostimulants used in the experiment had no effect on dry matter content in immature tubers of very early potato cultivars 'Denar', 'Lord' and 'Miłek'. In a study carried out by other authors, the biostimulants based on *A. nodosum* extracts (Phylgreenmira, Algazone, Ultra-Kelp) increased dry matter content in mature tubers of early ('Arizona', 'Riviera') and medium-early ('Agria') potato cultivars [18]. In a previous research, foliar application of fulvic acids had no effect on tuber dry matter of medium-early cultivar 'Atlantic' [35]. The dry matter content in potato tuber is determined by leaf assimilation area and chlorophyll content in leaves. Reducing the assimilation area along with increasing the chlorophyll *a* content and simultaneously decreasing the chlorophyll *b* content in leaves increased the dry matter content in tubers [56]. The biostimulants used in the experiment enlarged the assimilation area of very early potato cultivars, but did not affect the chlorophyll content in leaves [57].

The quality of edible potatoes is determined by their starch and sugar contents [4]. Among the biostimulants used in the experiment, only Bio-algeen S90 (*A. nodosum* extract) increased starch content in tubers of very early potato cultivars 75 days after planting, especially in a year with a low air temperature and a drought in the tuber growth period. In the three years of the study, following application of this biostimulant, the average starch content was higher by 4.8 g·kg^{-1} FW compared to the average for the untreated control tubers. Bio-algeen S90 has more bioactive compounds that may promote starch synthesis through the induction of carbon metabolism and activities of starch synthesis enzymes. In a study carried out by other authors, Bio-algeen S90 did not affect the starch content in tubers of the medium-early cultivar 'Muza' [19]. Other biostimulants based on *A. nodosum* extracts (Phylgreenmira, Algazone, Ultra-Kelp) increased starch content in tubers of early ('Arizona', 'Riviera') and medium-early ('Agria') potato cultivars [18]. The bioactivity of seaweed extracts depends on the extraction method [11,12] and on the date and dose of application [13,21]. In the present study, humic and fulvic acids from leonardite in HumiPlant (fulvic acid 6% and humic acid 12%) did not affect starch content in immature tubers of very early potato cultivars. In a study carried out by other authors, humic acid from leonardite in Huma K (humic acid 56% and fulvic acid 30%) introduced into the irrigation system increased starch content in tubers of medium-early cultivar 'Hermes' [33], and soil application of crude humic acids from leonardite in Agro-Lig (total humic acid 85%) increased starch content in tubers of late maturity cultivar 'Caspar' [34]. Foliar application of Natural Canadian fulvic acids (80% liquid) increased starch content in tubers of the late cultivar 'Atlantic' [35]. Humic substances affect the activity of the main enzymes involved in carbohydrate metabolism and ADP-glucose pyrophosphorylase (AGPase) has a major role in starch synthesis. The effect of humic substances on enzyme activities depends on humic molecular size, molecular characteristics and concentration [28,31]. The biostimulants used in the experiment did not affect the sugar content in immature tubers of very early potato cultivars. Starch content in potato tubers is associated with assimilation leaf area, the chlorophyll *a* content in leaves and efficiency of the photosystem in the dark, whereas the sugars content depend on chlorophyll content in leaves and fluorescence yield [56]. The biostimulants used in the experiment did not affect starch and sugar content in immature tubers of very early potato cultivars, except for Bio-algeen S90, although it enlarged the assimilation leaf area [57].

Protein and vitamin C are very important nutritional compounds in potatoes. The protein present in potato tubers has a higher biological value than other crops due to the content of all exogenous amino acids, and especially a high content of lysine [4]. The biostimulants used in the experiment did not affect protein or L-ascorbic acid content in immature tubers of very early potato cultivars. Protein accumulation in potato tuber is determined by leaf assimilation area and the chlorophyll *a* content in leaves, while vitamin C content is only determined by chlorophyll *a* content [56]. In the present study, enlargement of the leaf assimilation area as a result of biostimulant application [57] had no effect on the protein content in tubers. In a study carried out by other authors, Bio-algeen S90 and Kelpak SL also did not affect total nitrogen content in tubers of very early ('Volumia') or medium-early ('Irga', 'Satina', 'Silvana') cultivars [20], but Kelpak SL increased the vitamin C content in tubers of a medium-late cultivar ('Bryza') [23].

Although potato tubers accumulate small amounts of nitrates [4], due to high potato consumption they can be a source of substantial quantities of these compounds in the human diet. Nitrates are accumulated in potato tubers when their uptake is greater than the possibility of the plant to utilise them in organic nitrogenous compounds. In general, immaturity in potato tubers has been connected with high nitrate levels. The later the potatoes are harvested, the lower the nitrate contents are in tubers, but the relationship between nitrate content and tuber maturity differs between genotypes [58]. Bio-algeen S90 did not affect nitrate content in immature tubers of very early potato cultivars tested, whereas Kelpak SL and HumiPlant reduced the nitrate content only in tubers of the 'Denar' cultivar. Following the application of those biostimulants, the nitrate content in tubers of 'Denar' was lower, on average, by 8.50 $mg \cdot kg^{-1}$ FW. In a study carried out by other authors, Bio-algeen S90 had no effect on nitrate content in tubers of very early ('Volumia') and medium-early ('Irga', 'Satina', 'Silvana') cultivars, whereas Kelpak SL increased the content of these compounds [20]. Seaweed extracts and humic substances may promote nitrogen metabolism. A positive dose-dependent effect of seaweed extracts and humic acids on the activities of the main enzymes involved in the reduction and assimilation of inorganic nitrogen (nitrate reductase, glutamate dehydrogenase and glutamine synthetase) was found [12,29]. Results of the present study suggest, that after the application of biostimulants, the amount of transcripts of regulatory of enzymes related to the nitrogen metabolism of Denar cultivar increased more than of Lord and Miłek cultivars.

There is a significant correlation between the vitamin C content and nitrate level in potato tubers. A higher content of vitamin C is accompanied by a lower nitrate content [59]. The relative levels of ascorbic acid content and nitrate content in potato tubers may be expressed using the ascorbate-nitrate index (I_{AN}), which is one of the indicators of food safety. A higher index value reflected higher food safety [54]. The biostimulants used in the experiment had no effect on the ratio of L-ascorbic acid amount-to-nitrate amount in tubers of 'Lord' and 'Miłek' cultivars, whereas Kelpak SL and HumiPlant increased this ratio in tubers of the 'Denar' cultivar, on average, by 0.29. Regardless of the treatment (with or without biostimulant), the ratio of L-ascorbic acid amount-to-nitrate amount (I_{AN}) in immature tubers of potato cultivars tested was about 2/1, which indicates that the new potatoes were safe for human health regarding the nitrate content [54].

After-cooking darkening is an important quality characteristic of edible potatoes. It is caused by non-enzymatic oxidation of the chlorogenic acid-iron compound after cooking. The severity of the darkening depends on the ratio of chlorogenic acid-to-citric acid concentration in the potato tubers [60]. The biostimulants used in the experiment did not affect the susceptibility of new potatoes to after-cooking darkening. This characteristics of table potato quality depends on the cultivar [60], which was confirmed in the present study. In a study carried by other authors, Bio-algeen S90 did not affect after-cooking darkening of the 'Muza' medium-early cultivar [19].

The nutrient content in potatoes depends on several factors, with cultivar being among the most important [3]. The nutrient content in potatoes depends on the cultivar and weather conditions during potato growth, to a greater extent than on the biostimulants applied, which was confirmed in a study carried out by other authors [20].

The results of our study showed, that the application of plant biostimulants Bio-algeen S90, Kelpak SL or HumiPlant improved the plant growth and early crop potato yield [57,61] without any negative effect on the nutritional value of new potatoes.

5. Conclusions

The foliar application of seaweed extracts *A. nodosum* (Bio-algeen S90) and *E. maxima* (Kielpak SL), as well as humic and fulvic acids from leonardite (HumiPlant), did not affect dry matter, protein, total sugars, monosaccharides and sucrose or L-ascorbic acid content in tubers of very early potato cultivars 75 days after planting. Bio-algeen S90 increased starch content in tubers of all tested potato cultivars, on average, by 4.8 $g \cdot kg^{-1}$ FW compared with control treatment without biostimulant, whereas Kelpak SL and HumiPlant reduced the nitrate content only in tubers of the 'Denar' cultivar, on average, by 8.50 $mg \cdot kg^{-1}$ FW, and increased I_{AN} by 0.29. The biostimulants did not affect potato after-cooking darkening. The nutritional value of new potatoes and after-cooking darkening depends to a greater extent on the cultivar and weather conditions.

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