JEE Journal of Ecological Engineering

Journal of Ecological Engineering 2023, 24(2), 247–255
https://doi.org/10.12911/22998993/157019
ISSN 2299–8993, License CC-BY 4.0

Received: 2022.11.12
Accepted: 2022.12.16
Published: 2023.01.01

Total and Protein Nitrogen Content in Potato Tubers under the Influence of Various Care and Nutrition Methods with the Use of Biostimulants

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ABSTRACT

A two-factor field experiment was carried out in 2018–2020 at the Zawady Agricultural Experimental Station (52°03' N and 22°33' E) belonging to the University of Natural Sciences and Humanities in Siedlce in Poland. Tubers of two edible potato cultivars were investigated, i.e. Oberon and Malaga – factor one; in turn, factor two included variants of herbicide application with biostimulators: 1. control object – without preparations, 2. herbicide clomazone + metribuzin (Avatar 293 ZC), 3. herbicide clomazone + metribuzin and biostimulant PlonoStart, 4. herbicide clomazone + metribuzin and biostimulant Aminoplant, 5. herbicide clomazone + metribuzin and biostimulant Agro-Sorb® Folium. The biostimulators used in the research significantly increased the content of total nitrogen and protein nitrogen in tubers, as well as the share of protein nitrogen in total nitrogen in relation to the tubers collected from the control object. The genetic features of the cultivars also determined the concentration of total nitrogen and protein nitrogen in potato tubers.

Keywords: chemical composition; clomazone; growth stimulants; metribuzin; *Solanum tuberosum* L.

INTRODUCTION

The potato (*Solanum tuberosum* L.) is one of the most important food crops after wheat, rice and maize, and is the staple food for the world population. According to the United Nations, potato tubers are a food security crop. The global monthly consumption of potato per capita is 31.3 kg; in Europe it is higher at 87.8 kg/capita (FAO, 2020; Pllana et al., 2018). The advantages potatoes include a rich chemical composition, high nutritional value; moreover, they can grow in any climate and conditions (Silveira et al., 2020). The chemical composition of potato tubers depends on the cultivar, weather conditions, technology of cultivation, fertilization, harvesting and storage conditions (Manjunath et al., 2018; Silveira et al., 2020; Trawczyński, 2018; Zarzecka et al., 2009).

In modern plant cultivation, various plant growth regulators called biostimulants are increasingly used. They are one of the elements of agrotechnics, which, apart from fertilizing and protection plants, can positively affect the size and quality of crops (Yakhin, 2017; Rutkowska, 2016; Shekari et al., 2017). These preparations improve the uptake of nutrients from the soil (Głosek-Sobieraj et al., 2019; Mystkowska and Rogóż-Matyszczak, 2019; Zarzecka et al., 2019) positively affect the intensity of photosynthesis and the course of life processes (Piotrowski and Romanowska-Duda, 2018; Wadas and Dziugieł, 2020**)** increase the resistance of plants to stress factors such as diseases, high temperature, drought (Cwalina-Ambroziak, 2015; Rutkowska, 2016**)** and have a positive effect on the chemical composition as well as yields of plants (Merga and Dechassa, 2019; Trawczyński, 2018; Zarzecka et al., 2009; Zarzecka et al. 2020). Biostimulants include hormones, enzymes, proteins, amino acids, vitamins, microelements, alga extracts and other compounds (Kunicki, 2010; Rutkowska, 2016). The most common biostimulants are: amino acids, proteins, microelements. Plants are able to take up amino acids and peptides both through the leaves and the root system. In addition, plants are capable of self-synthesis of amino acids, but this process requires much energy (Yakhin et al., 2017; Radkowski, 2018). Amino acids are among the most important organic compounds and play an important biological role as building blocks of proteins, enzymes, nucleic acids, antioxidants, hormones and other components. Therefore, the use of these compounds as biostimulants may save energy and improve dynamics of plant development (Shukla et al., 2014). The great advantage of using amino acids as biostimulants is their good mobility and easy transportation in the plant (Kandil et al., 2016). Many authors have shown that biostimulant application in potato cultivation has a positive effect on plant development, yields and chemical composition of tubers (Głosek-Sobieraj et al., 2018; Manjunath et al., 2018; Trawczynski, 2018; Zarzecka et al., 2019; Zarzecka et al., 2020). Therefore, the aim of this study was to determine the effect of biostimulants based on amino acids and microelements and herbicide on the content of total nitrogen and protein nitrogen in potato tubers.

MATERIAL AND METHODS

The research was carried out in 2018–2020, in the Agricultural Experimental Station Zawady (52°03'N and 22°33'E) which belongs to the University of Natural Sciences and Humanities in Siedlce in Poland. The content of available forms of phosphorus, potassium and magnesium in 2018 was low, and in 2019 and 2020 it was medium. The soil was acidic with a pH of 5.25–5.42 in 1 M KCl (Table 1). Two factors were tested in the split-plot system in three replications: I – two medium-early edible potato cultivars Malaga and Oberon, II – five ways of using herbicide and herbicide with biostimulants: control object – mechanical maintenance, Avatar 293 ZC (clomazone

Table 1. Results of soil analysis in 2018–2020

Year	P available	K available	Mg available	N total	Organic matter
	(mg kg^{-1})				(g kg^{-1})
2018	35.2	102.1	36.6	9.0	20.9
2019	61.0	149.0	61.0	12.6	22.3
2020	60.0	140.0	51.0	11.0	21.1

Table 2. Description of biostimulants and herbicide and their effects

Preparations	Chemical composition and action
PlonoStart	N total – 16.4%, K_2O – 0.75%, CaO – 0.07%, MgO – 0.02%, S – 941 mg·kg^{-1}, actinomycetes, lactic acid bacteria, improves the availability of nutrients, improves plant health, reduces the effects of drought and excessive rainfall, increases the quality and yield of plants
Aminoplant	N total - 9.48%, N organic – 9.2%, $N-NH_4$ - 088%, Corganic - 25%, free amino acids 11.57, organic substance – 87.7, increases the resistance of plants to stress factors, such like drought, frosts, diseases, increases the activity of many enzymes responsible for the uptake of nitrogen and other macronutrients from the substrate, improves the synthesis of sugars and proteins, it accelerates the growth and development of plants in a natural way
Agro-Sorb® Folium	N total – 2.2%, B – 0.02%, Mn – 0.05%, Zn – 0.09%, free amino acids – 10.66%, total amino acids – 13.11%, supports plant regeneration after stressful periods, increases nutrient uptake, has a positive effect on the intensity of photosynthesis,
Avatar 293 ZC	Active ingredients - clomazone (60 g dm^3) + metribuzin (233 g dm^3), it destroys sensitive weeds: *Capsella bursa-pastoris, Chenopodium album, Fumaria officinalis*, *Galeopsis tetrahit, Galinsoga parviflora, Lamium purpureum, Matricaria inodora, Polygonum convolvulus Sinapis arvensis*, *Stellaria media, Thlaspi arvense, Veronica hederifolia, Viola arvensis* and resistant weeds: *Elymus repens*

+ metribuzin) Avatar 293 ZC and Plonostart, Avatar 293 ZC and Aminoplant as well as Avatar 293 ZC and Agro-Sorb Folium (Table 2). The description of the biostimulants and herbicide and their effects are presented in Table 2. The forecrop in all research years was winter triticale. The potatoes were fertilized in autumn with 25 t·ha^{-1} of manure and the 44.0 kg P·ha^{-1} (superphosphate, 17.5% P) and 124.5 kg K·ha^{-1} (potash salt, 50% K) mineral fertilizers were applied. In the spring before planting, 100 kg N·ha^{-1} (ammonium nitrate 34% N) were applied. Potatoes were planted by 18 to 23 April, and were harvested by 4th to 17th September. Pathogens and pests were controlled chemically.

Meteorological conditions

In terms of humidity and thermal conditions, 2018 was the most favorable year for potato development. It was a warm season with evenly distributed rainfall. In 2019, a significant shortage of rainfall was found in relation to the multi-year period. In 2020, the air temperatures were optimal, while the humidity conditions were close to the long-term values, but the rainfall was unevenly distributed during the potato vegetation. Weather data is included in Table 3.

Chemical analysis – determination of nitrogen

During the harvest, 10 potato plants were dug up by hand from each plot and tuber samples (35–60 mm in diameter) were taken for chemical analysis. The tubers taken from the field experiment were washed thoroughly and cut into thin slices. The total nitrogen was determined with Kjeldahl's method on a 2300 Kjeltec Analyser Unit (ISO. ISO 1871, 2009). Protein nitrogen was determined by using the Bernstein method (AOAC 2006; Barbaś and Sawicka, 2017). The determined values was expressed in g·kg^{-1} of dry matter of tubers. The total nitrogen uptake with the tuber yield was calculated as the product of tuber dry matter yield and total nitrogen concentration.

Statistical analysis

Test results were developed statistically with the method of the variance analysis. The significance of the sources of variation was tested with the 'F' Fischler-Snedecor test, and the assessment of the significance of differences with the significance level $P \leq 0.05$ between the compared averages using the multiple Tukey ranges (Trętowski and Wójcik, 1991).

RESULTS AND DISCUSSION

Content of total nitrogen

The nutritional value of a potato is due to its chemical composition. Potato tubers contain approx. 17% of complex carbohydrates (starch), approx. 0.5% of sugars, 2% of proteins, 2.3% of dietary fiber, approx. 0.1% of lipids and a wide

Table 3. Rainfall and average air temperature at the Agricultural Experimental Station – Zawady in 2018–2020

Month	2018	2019	2020	Multi-year mean 1980–2009
	Monthly rainfall (mm)			
4. April	34.5	5.9	6.0	49.6
5. May	27.3	59.8	63.5	48.2
6. June	31.5	35.9	118.5	60.7
7. July	67.1	29.7	67.7	45.7
8. August	54.7	43.9	17.9	53.0
9. Septembr	80.6	17.4	38.8	50.7
Total April–September	295.7	192.6	312.4	307.9
	Average monthly air temperature (°C)			
4. April	13.1	9.8	8.6	7.9
5. May	17.0	13.3	11.7	11.2
6. June	18.3	17.9	19.3	16.7
7. July	20.4	18.5	19.0	19.3
8. August	20.6	19.9	20.2	18.0
9. Septembr	15.9	14.2	15.5	13.0
Mean April–September	17.6	15.6	15.7	14.4

range of vitamins (e.g., A, C, B_1, B_2, B_6) and minerals (e.g., potassium, phosphorus, magnesium, calcium, sodium, iron, zinc, and copper). Moreover, potato tubers contain chemicals important in preventing cancer; they include polyphenols and flavonoids (Baranowska et al., 2018; Ciecholewska-Juśko et al., 2021; Jayanty et al., 2019; Kowalczewski et al., 2019). One of the most important components of potato tubers is protein of high biological value, comparable to chicken egg whites (reference protein). It contains all the essential amino acids in the right amounts: lysine, leucine, isoleucine, phenylalanine, valine, methionine, tryptophan and threonine (Beals, 2019; FAO, 2013; Pęksa et al., 2009).

The authors' own research has shown that the Oberon variety contains more total nitrogen than the Malaga variety (Table 4).

Other authors also showed a different content of this component in tubers depending on the cultivar (Baranowska et al., 2018; Goméz et al., 2017; Gugała et al., 2015; Haider et al., 2012; Wierzbowska et al., 2015). The biostimulants with minerals (PlonoStart) and amino acids (Aminoplant, Agro-Sorb® Folium) and herbicide (Avatar 293 ZC) caused an increase in the content of total protein in comparison with tubers collected from the control object (only mechanical treatments). The highest total nitrogen content was accumulated by tubers collected from the object where the PlonoStart biostimulant was applied twice during the growing season at the overall dose of 2.0 $dm^3 \cdot ha^{-1}$ and from the object sprayed with Agro-Sorb® Folium biostimulant at a dose of 4.0 $dm^3 \cdot ha^{-1}$. Colla et al. (2014) found that the stimulating effect of amino acid biostimulators on plants mainly consists in improving carbon and nitrogen metabolism and increasing nitrogen assimilation. The preparations based on amino acids play an important role in the transport of nitrogen in the plant,

Table 4. Content of total nitrogen, protein nitrogen in potato tubers (g kg^{-1} dry matter) and uptake of total nitrogen with the yield of tubers (kg ha^{-1})

Application of herbicide and biostimulants	Cultivars		Mean
	Oberon	Malaga	
Total nitrogen			
1. Control - only mechanical treatments	23.40A	18.18A	20.79c
2. Avatar 293 ZC 1.5 $dm^3 ha^{-1}$ (BBCH 00–08)	24.08A	19.59A	21.83b
3. Avatar 293 ZC + PlonoStart 1.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 1.0 $dm^3 ha^{-1}$ (BBCH 31–35)	26.82A	20.32A	23.57a
4. Avatar 293 ZC + Aminoplant 1.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 0.5 $dm^3 ha^{-1}$ (BBCH 31–35)	25.37A	20.01A	22.69ab
5. Avatar 293 ZC + Agro-Sorb® Folium 2.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 2.0 $dm^3 ha^{-1}$ (BBCH 31–35)	26.25A	20.06A	23.16a
Mean	25.19a	19.63b	22.41
Protein nitrogen			
1. Control - only mechanical treatments	16.20A	12.45A	14.33c
2. Avatar 293 ZC 1.5 $dm^3 ha^{-1}$ (BBCH 00–08)	16.93A	13.39A	15.16b
3. Avatar 293 ZC + PlonoStart 1.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 1.0 $dm^3 ha^{-1}$ (BBCH 31–35)	19.71A	14.91A	17.31a
4. Avatar 293 ZC + Aminoplant 1.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 0.5 $dm^3 ha^{-1}$ (BBCH 31–35)	18.42A	14.40A	16.41b
5. Avatar 293 ZC + Agro-Sorb® Folium 2.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 2.0 $dm^3 ha^{-1}$ (BBCH 31–35)	19.19A	14.65A	16.92a
Mean	18.09a	13.96b	16.03
Uptake of total nitrogen			
1. Control - only mechanical treatments	171.7C	91.6C	131.6d
2. Avatar 293 ZC 1.5 $dm^3 ha^{-1}$ (BBCH 00–08)	188.5C	117.2B	152.9c
3. Avatar 293 ZC + PlonoStart 1.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 1.0 $dm^3 ha^{-1}$ (BBCH 31–35)	236.4A	129.8A	183.1ab
4. Avatar 293 ZC + Aminoplant 1.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 0.5 $dm^3 ha^{-1}$ (BBCH 31–35)	213.3B	123.2AB	168.3bc
5. Avatar 293 ZC + Agro-Sorb® Folium 2.0 $dm^3 ha^{-1}$ (BBCH 13–19) and 2.0 $dm^3 ha^{-1}$ (BBCH 31–35)	249.1A	141.1A	195.1a
Mean	211.8a	120.5b	166.2

Note: means followed by the same letters do not differ significantly at $P \leq 0.05$. Means in columns marked with capital letters refer to interactions between the factors. Means in the last column and means in the last row (followed by lowercase) are for treatments and, cultivars.

which is stored in greater amounts under stressful conditions (Van Oostenet al., 2017; Ahmad et al., 2013; Dos Reis et al., 2012). The herbicide Avatar 293 ZC (metribuzin, clomazone) used in the research probably also influenced the plant's metabolism and contributed to an increase in total nitrogen accumulation. Gugała et al. (2015) and Wichrowska et al. (2009) found that plant protection products increase the total nitrogen content in potato tubers.

The conducted research also showed that the weather conditions during the experiment differentiated the content of total nitrogen. The highest total nitrogen content in potato was accumulated by tubers during the growing season of 2018, when in July and August (the months of crop formation) rainfall was similar to the long-term values (67.1 and 54.7 mm) and a stable air temperature for growth and development of potato plants (20.4 and 20.6 °C) (Table 3, 5).

The influence of weather conditions on the accumulation of the discussed component is confirmed by the studies of other authors (Gugała et al., 2015; Wierzbowska et al., 2015; Zarzecka et al., 2009). The demonstrated interaction of the cultivars with the years of research indicates that the total nitrogen content in the cultivated cultivars was shaped by the humidity and thermal in the years of the study.

Content of protein nitrogen

Chemical analyses have shown, and statistical calculations have confirmed, a significant influence of cultivars, application of herbicide and biostimulants, as well as weather conditions in the years of research on the content of protein nitrogen in potato tubers. Under the conditions of the experiment, the average protein nitrogen content in potato tubers ranged from 12.45 to 19.71 $g \cdot kg^{-1}$ dry matter (Tables 4, 5). The Oberon cultivar had a statistically higher concentration of protein nitrogen than the Malaga cultivar. According to Mystkowska (2018) and Gugala et al. (2017) the content of true protein and thus the content of protein nitrogen in potato tubers depended on the genetic characteristics of cultivars. In the conducted experiment, the herbicide (object 2) and biostimulants (objects 3–5) significantly increased the content of protein nitrogen in potato tubers with the control combination. The largest significant increase in the content of this macronutrient in tubers was found after the application of the Avatar 293 ZC herbicide as well as the biostimulant and herbicide Plonostart and Agro-Sorb® Folium biostimulant. Similar results were obtained in the authors' previous work (Zarzecka et al., 2020) using a single herbicide Sencor 70 WG (metribuzin) and a mixture of the Sencor 70 WG herbicide (metribuzin) with the Asahi SL biostimulant. Grabowska et al. (2012) and Kunicki

Table 5. Content of total nitrogen, protein nitrogen in potato tubers (g kg^{-1} dry matter) and uptake of total nitrogen with the yield of tubers (kg ha^{-1}) in study years

Years	Cultivars		Mean
	Oberon	Malaga	
Total nitrogen			
2018	26.96A	19.37A	23.17a
2019	22.66B	19.47A	21.07b
2020	25.95A	20.05A	23.00a
Mean	25.19a	19.63b	22.41
Protein nitrogen			
2018	19.52A	13.82A	16.67a
2019	16.12B	13.87A	15.00b
2020	18.63A	14.20A	16.42a
Mean	18.09a	13.96a	16.03
Uptake of total nitrogen			
2018	264.3A	109.4A	186.9a
2019	185.9B	129.8A	157.9b
2020	185.2B	122.4A	153.8b
Mean	211.8a	120.5b	166.2

Note: means followed by the same letters do not differ significantly at $P \leq 0.05$. Means in columns marked with capital letters refer to interactions between the factors. Means in the last row (followed by lowercase) are for cultivars and years.

Table 6. Share of protein nitrogen in total nitrogen depending on application of herbicide and biostimulants and cultivars

Application of herbicide and biostimulants	Cultivars		Mean
	Oberon	Malaga	
Total nitrogen			
1. Control - only mechanical treatments	69.23	68.48	68.86
2. Avatar 293 ZC 1.5 dm^3 ha^{-1} (BBCH 00–08)	70.31	68.35	69.33
3. Avatar 293 ZC + PlonoStart 1.0 dm^3 ha^{-1} (BBCH 13–19) and 1.0 dm^3 ha^{-1} (BBCH 31–35)	73.48	73.38	73.43
4. Avatar 293 ZC + Aminoplant 1.0 dm^3 ha^{-1} (BBCH 13–19) and 0.5 dm^3 ha^{-1} (BBCH 31–35)	72.61	71.96	72.29
5. Avatar 293 ZC + Agro-Sorb® Folium 2.0 dm^3 ha^{-1} (BBCH 13–19) and 2.0 dm^3 ha^{-1} (BBCH 31–35)	73.10	73.03	73.07
Mean	71.75	71.04	71.40

et al. (2010) observed a beneficial effect in the cultivation of carrots and spinach using Aminoplant (amino acid extract). The Agro-Sorb® Folium growth stimulator (amino acid extract) used in the studies by Wojdyła and Sobolewski (2016) reduced the symptoms of white mold in the cultivation of beans. The years in which the research was conducted also differed the discussed feature. In 2018, a optimally moist and warm year, tubers accumulated the most protein nitrogen. According to Mazurczyk and Lis (2001) and Baranowska et al. (2018) the content of total protein, especially specific protein, is stimulated by a warm and sunny vegetation period, and in the period of tuber setting the temperature around 20 °C is the most favorable, because assimilation occurs most vigorously then. In turn, Lis et al. (2002) demonstrated that excessive precipitation in the period of yield accumulation (June and July) may lead to nitrogen leaching from soil, which results in limited uptake of this nutrient by the tuber crop. The effect of weather conditions on protein content is confirmed by other works (Gugała et al., 2017; Piotrowski and Romanowska-Duda, 2018; Pęksa et al., 2009; Zarzecka et al., 2009). In the conducted experiment, an interaction of potato cultivars and weather conditions in the years of research on this trait was found.

In the human diet, potato is one of the main sources of nutrients (Flis et al., 2012). Hence, an important feature, from the consumer's point of view, is the share of proper protein in total protein, and thus protein nitrogen in total nitrogen, which in the experiment was quite high and ranged from 68.35% to 73.4% (Table 6).

In the conducted research, both the genetic characteristics of the cultivars and the application of herbicide and biostimulants significantly influenced the share of protein nitrogen in total nitrogen (Table 6). The cultivar Oberon had a greater share than the cultivar Malaga. The applied biostimulants increased the share of protein nitrogen in total nitrogen compared to the control object and to the variant where only the herbicide was applied. In the studies by Barbaś and Sawicka (2017), this share was within the range of 68.03%–80.48% depending on the cultivar, and 59.20–89.65% in dry matter, depending on the year of the study. In Bárta and Bártová (2008), this share was in the range of 44.5–62.1%. Many authors emphasize that potato protein is unique, free from allergens, gluten and lactose, is rich in exogenous amino acids and has a high functional value (Beals, 2019; Merga and Dechassa, 2019; Pęksa et al., 2009; Pęksa and Miedzianka, 2021; Pllana et al., 2018) Hence, analyzing the content of protein nitrogen under the conditions of biostimulant application provides valuable information.

Uptake of total nitrogen with the yield of tubers

The uptake of nutrients with the yield is determined mainly by the harvested tuber yield and the content of macroelements in tubers, but may also be determined by agrotechnical and environmental factors (Mystkowska and Rogóż-Matyszczak, 2019; Zarzecka et al., 2019). The conducted analysis and calculations showed that the uptake of total nitrogen with the yield of tubers depended significantly on cultivated cultivars, application of herbicide and biostimulants and the weather conditions during plant growth (Table 4, 5). Cultivar Oberon (211.8 kg ha^{-1}) was characterized by higher mean total nitrogen uptake (120.5 kg ha^{-1}) than cultivar Malaga. The highest total nitrogen uptake capacity was recorded in the object where the Avatar 293 ZC herbicide and the Agro-Sorb® Folium biostimulant were used (195.1 kg·ha^{-1}). In the other objects with herbicide and biostimulants (objects 2–4), the intake of this component was significantly higher compared to the control object. The highest total nitrogen uptake was found in 2018, when it was warm, and the sum of rainfall was similar to the values from the long-term period. It was a favorable year for potato yielding and total nitrogen accumulation. Similar results were obtained by Wierzbowska et al. (2015).

CONCLUSIONS

New cultivation technologies with the use of biostimulants reduce the negative environmental impact and at the same time improve the quality of the harvested crops. The present research shows that the application of herbicide and biostimulants on potato plantation increased the content of total nitrogen and protein nitrogen in tubers. The best effects were obtained with the use of the Avatar 293 ZC herbicide as well as PlonoStart and Agro-Sorb® Folium biostimulants, compared to the control object. At the same time, it was found that the share of protein nitrogen in total nitrogen was the highest after spraying potato plants with biostimulants.

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