Journal of Elementology ISSN 1644-2296

Wierzbowska, J., Cwalina-Ambroziak, B., Waśkiewicz, A. and Bogucka, B. (2023)
'Influence of nitrogen fertilizers on the concentrations of inulin
and micronutrients in Jerusalem artichoke tubers and root chicory',
Journal of Elementology, 28(3), 793-814,
available: https://doi.org/10.5601/jelem.2023.28.2.3024

RECEIVED: 19 May 2023
ACCEPTED: 27 August 2023

ORIGINAL PAPER

Influence of nitrogen fertilizers on the concentrations of inulin and micronutrients in Jerusalem artichoke tubers and root chicory*

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Abstract

The aim of the study was to evaluate the effect of nitrogen fertilization on the content of inulin and microelements in tubers of Jerusalem artichoke (JA) and roots of chicory grown in the climatic and soil conditions of north-eastern Poland. Three cultivars of JA and root chicory were grown in a field experiment. The content of inulin and micronutrients was determined in JA tubers and chicory roots harvested in autumn, JA tubers left in the ground over winter, and stored chicory roots. Jerusalem artichoke tubers fertilized with N (80 and 120 kg ha^{-1}) contained more inulin (803.3 and 737.0 g kg^{-1} DM) than unfertilized plants (635.0 g kg^{-1} DM); the opposite was noted in chicory roots (without N fertilizer – 392.8 and 352.0 and 337.0 g kg^{-1} DM fertilized with 80 and 120 kg N ha^{-1} respectively). Inulin content was lowest in JA tubers and chicory roots harvested in the wet and cold 2017. The highest decrease in inulin content was observed in tubers left in soil over winter and stored roots. The concentrations of Zn, Mn and Fe were higher in JA tubers and chicory roots harvested in the wet seasons of 2016 and 2017, and the content of B and Cu was highest in the warm and dry 2018. The micronutrient content of JA tubers and chicory roots was less affected by cultivar and N dose then years of study. Cultivar exerted a significant influence on the content of Cu and Fe in JA tubers left in soil over winter, and on B content in stored chicory roots. The micronutrient concentrations in JA tubers overwintering in the soil and stored chicory roots were not affected by N fertilization.

Keywords: *Helianthus tuberosus, Cichorium intybus*, N fertilization, inulin, micronutrients

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[*] The results presented in this paper were obtained as part of a comprehensive study financed by the University of Warmia and Mazury in Olsztyn, Faculty of Agriculture and Forestry.

INTRODUCTION

Plants of the family *Asteraceae*, including Jerusalem artichoke – JA (*Helianthus tuberosus* L.) and root chicory (*Cichorium intybus* L. var. *sativum* Bisch.), are a rich source of various phytochemicals. Both JA tubers and chicory roots are rich sources of prebiotics, including inulin, a type of dietary fiber (Jurgoński et al. 2011, Sawicka et al. 2021). Recent years have seen renewed interest in natural plant-derived products which are abundant in biochemical compounds with health-promoting properties, such as phenolic compounds (Amarowicz et al. 2020), inulin (Sawicka et al. 2021), flavonoids and coumarins (Khalaf et al. 2018). JA tubers are renowned for their high nutritional value. They contain (g kg^{-1} FM**)** protein – 30.0, ash – 38.1, carbohydrates – 261.6, total sugar – 144.0, total dietary fiber – 24.0 (Sawicka 2016), mostly inulin (Sawicka et al. 2021), minerals, bioactive compounds, including vitamins: complex B, A, C and E (Sawicka 2016), phenolic acids (Amarowicz et al. 2020), as well as unsaturated fatty acids (Pan et al. 2009). JA tubers exert cholagogic, diuretic, stomachic, and tonic effects, and they can be used as a remedy for diabetes (Kim et al. 2021) and rheumatism (Kays, Nottingham 2007). In addition, proteins have antimicrobial and anticancer properties and protect against Parkinson's and Alzheimer's diseases (Bakku et al. 2022). Owing to their high content of nutrients and minerals, JA tubers are considered a valuable raw material for food processing (Zhang et al. 2021) as well as the cosmetic (Niziol-Lukaszewska et al. 2019) and pharmaceutical industries (Yang et al. 2015); they are also used as animal feed (Pinar et al. 2021). JA tubers are also a raw material for the production of bioethanol (Bhagia et al. 2017).

Chicory is a popular vegetable in the Mediterranean countries, where it is consumed both raw and cooked, and used as a coffee substitute, a source of inulin, and animal feed (Cadalen et al. 2010). Chicory roots contain (g kg^{-1} DM) total carbohydrates (461.5), crude fiber (285.0), fat (65.40), protein (50.60), and crude ash (38.70) – Jangra, Madan (2018). Both chicory roots and leaves contain the following bioactive compounds: inulin, sesquiterpene lactones, coumarins, polyphenols (caffeic acid derivatives) and natural flavonoids (Nwafor et al. 2017, Khalaf et al. 2018**)**, alkaloids, terpenoids, sterols, tannins, various volatile compounds, oils and vitamins (Zarroug et al. 2016). Those found in chicory roots exert anti-inflammatory, antimicrobial and antioxidant (Khalaf et al. 2018), antihyperglycemic (Azay-Milhau et al. 2013), tranquilizing, immunological, hypolipidemic, hepatoprotective (Li et al. 2014), cardioprotective and gastroprotective effects (Al-Snafi 2016). Chicory roots are also an important source of prebiotics – dietary fiber and inulin, and antioxidants – phenolic compounds (Jurgoński et al. 2011). Inulin is hydrolyzed to fructose in the human body. Owing to its health-promoting and prebiotic properties, fructan (inulin) can be consumed by persons with diabetes (Perović et al. 2021). Chicory roots are often used for the industrial-scale production of inulin (Roberfroid 2007).

Plants of the family *Asteraceae* are abundant in trace elements that are essential for human health. These minerals are required for physical and mental well-being (Zhang et al. 2021). Malnutrition is often associated with a micronutrient-deficient diet (FAOSTAT 2018). The adequate intake of micronutrients for adult men and women (per day) is as follows: Fe – 7.0 and 6.0 mg, respectively; Mn – 3 mg; Zn – 7.5 and 9.4 mg, respectively; Cu – 1.3 and 1.6 mg, respectively (EFSA 2019). Unfortunately, the content of trace elements in plant products has decreased significantly in recent decades (Ekholm et al. 2007).

The concentrations of inulin and minerals content in plant material are determined by the varietal traits of crops, soil fertility, weather conditions during the growing season, fertilization, and maturity stage at harvest (De Mastro et al. 2004, Seiler, Campbell 2004, Tartoura et al. 2020).

The aim of the study was to evaluate the effect of nitrogen fertilization on the content of inulin and microelements in tubers of three JA cultivars and roots of three chicory cultivars grown in the climatic and soil conditions of north-eastern Poland.

MATERIALS AND METHODS

Location of the experiment, methodological assumptions

In 2016-2018, a plot experiment was conducted at the Agricultural Experiment Station in Tomaszkowo near Olsztyn, Poland. The station is owned by the University of Warmia and Mazury in Olsztyn (53°41′N, 20°24′E). The experiment was established on a Eutric cambisol with the granulometric composition of medium loamy sand and loamy sand (agricultural suitability class 5, soil quality class IVb) – WRB (2015). Each year, before establishing the experiment, topsoil samples (0-20 cm) were collected for chemical analyses. Soil pH and the content of available macronutrients and micronutrients, C-org. and N-total are presented in Table 1**.**

Three JA cultivars: Albik (club-shaped, white; Poland), Rubik (irregular to oval shaped tubers, purple; Poland) and Gute Gelbe (oval and round shaped, white; Germany), and three root chicory cultivars: Polanowicka (Poland), Chrysolite (France) and Orches (France) were grown. A separate field experiment was established for each plant species; in the treatment without organic fertilization, the preceding crop was oats. Agronomic treatments were identical in all plots, and weeds were removed mechanically.

The factors of the experiment were: N fertilizer doses and JA/root chicory cultivars. The experiment had a randomized subblock design (blocks – N fertilizer doses, subblocks – cultivars), with three replicates. Three levels of topsoil N fertilization were applied (urea – 46% N, single application) before sowing: 0, 80 and 120 kg ha^{-1}. The fertilization regime included also 32.5 kg P (granular triple superphosphate – 20.1% P) and 95.8 kg K (potash

Table 1

Soil characteristics before establishing the experiment

Characteristics		Units	Content	Determination method
pH		–	5.04-5.54	PN-ISO 10390:1997
C-org.		(g kg^{-1})	9.40-9.85	Vario Max Cube CN elemental analyzer
N-total			0.71-0.76	
Available forms of	P	(mg kg^{-1})	35.6-48.8	(PN-R-04023:1996)
	K		94.2-124.0	(PN-R-04022: 1996+Az1:2002)
	Mg		38.0-42.0	(PN-R-04020:1994+Az1:2004)
	B		3.33-4.25	in a 1 mol HCl dm^{-3} extract (Ostrowska et al. 1991)
	Cu		0.90-1.05	
	Zn		4.50-5.59	
	Mn		110.2-139.0	
	Fe		1250-1700	

salt – 50% K) per ha. Jerusalem artichoke tubers were planted in mid-April, in heated soil, at a depth of 8 cm, 40 cm apart; inter-row spacing was 62.5 cm, and plant density was six plants per m^2. Tubers were harvested in late autumn (until mid-November) and early spring (until mid-March). Chicory seeds were sown in the last ten days of April, in three rows, 15 cm apart; inter-row spacing was 40 cm; single plot area was 3.6 m^2 (3 x 1.2 m). Roots were harvested between 10 and 20 October. They were weighed and stored for five months under controlled conditions (temp. 4°C).

Chemical analyses

Five randomly selected JA tubers (from autumn harvest and after wintering in the ground) and chicory roots (weighing around 0.5 kg in total) were harvested from each plot and subjected to chemical analyses. Chemical analyses were also performed on chicory roots after 5-month storage. After rinsing under running water, they were diced into 1 cm × 1 cm × 1 cm cubes, freeze dried (Alpha 1-4LD laboratory freeze-dryer, Doncerv®-Martin Christ Gefriertrocknungsanlagen GmbH,), and ground in a laboratory mill (A11 basic, IKA®-Werke GmbH & CO. KG Germany).

Inulin

Inulin was extracted from plant samples (5 g) using 20 mL of water. Following each extraction, tubes containing the extracts were centrifuged at 5000 x g for 15 min. Before the chromatographic analysis, the supernatants were passed through a 0.20 μm filter (Chromafil Pet 20/15 MS, Macherey-Nagel, Steinheim, Germany). Inulin was identified and quantified in the plant extracts by liquid chromatography using the 2695 Waters high-performance liquid chromatograph (HPLC) system with a 2414 refractive index (RI) detector (Waters, Milford, MA, USA) and a Bio-Rad Aminex HPX-87H column (Bio-Rad, Woodinville, WA, USA); column temp. – 40°C, software – Empower™ 1 (Waters, Milford MA, USA); mobile phase – 0.5 mM H_2SO_4, flow rate – 0.5 cm^3 min^{-1}. In order to quantify inulin, peak areas were measured at specified retention time, based on calibration curves.

Micronutrients

The collected plant material was wet mineralized (Büchi Speed Digester K-439) in a mixture of nitric acid (HNO_3) and chloric acid ($HClO_4$) (4:1 ratio) with the addition of hydrochloric acid (HCl). The content of Cu, Fe, Mn and Zn was determined by atomic absorption spectrophotometry (AAS) on a Shimadzu AA-6800 spectrophotometer (Ostrowska et al. 1991). For B content determination, plant samples were dry mineralized (520°C) in the presence of calcium oxide (CaO), the resulting ash was dissolved in HCl (0.5 mol dm^{-3}), and the B content was determined by the azomethine-H colorimetric method on a Shimadzu UV – 1201 V spectrophotometer (Benedycka, Rusek 1994). The content of all chemical elements was expressed on a dry matter basis (drying temp. of 105°C).

Statistical analysis

The results were analyzed statistically by split-plot ANOVA (three-year series). Calculations were performed in the Statistica® program (1984-2017 TIBCO Software Inc.). Differences between means were determined by the Tukey's HSD test, and they were regarded as significant at p=0.05.

Weather conditions

In 2016, mean monthly temperatures from April to November were comparable to the long-term average of 1981–2010 (Table 2). In 2017, mean monthly temperatures were lower than the long-term average (excluding September and October). In 2018, mean monthly temperatures were higher than the long-term average. Precipitation levels were high in the first two years of the study, exceeding the long-term average by 25.7% and 45.5%, respectively. Abundant precipitation was noted in July, September and October 2017. In September 2016 and May 2017, precipitation levels were below the long-term average. In 2018, the amount of precipitation recorded during the growing season was 7% lower than the long-term average; May, June and September were very dry, whereas rainfall in July was almost twice as high as the long-term average.

In winter, when JA tubers were left in the ground, above-zero temp. (max. 4°C) were recorded in December in all years of the study, in March 2016, and in February and March 2018. Below-zero temperatures were in the range of -0.4 to -4.6°C. Precipitation (rain and snow) levels in winter approximated (December 2017) or exceeded the long-term average, and they were lower than normal in January 2016, and in February and March 2017.

Table 2

Meteorological data during the experiment according to the Meteorological Station in Tomaszkowo

Month	2016/2017		2017/2018		2018/2019		1981-2010	
	x °C	∑ mm	x °C	∑ mm	x °C	∑ mm	x °C	∑ mm
April	7.4	28.8	5.7	59.1	10.8	33.5	7.7	33.3
May	13.7	56.9	12.1	25.1	15.7	25.0	13.5	58.5
June	17.1	69.3	15.7	74.5	17.2	53.7	16.1	80.4
July	18.1	130.4	16.8	107.6	19.7	141.0	18.7	74.2
August	17.1	70.4	17.4	63.1	19.2	44.6	17.9	59.4
September	13.6	21.1	12.8	168.1	14.5	20.3	12.8	56.9
October	6.1	104.3	8.7	114.9	8.7	84.7	8.0	42.6
November	2.4	84,5	3,9	42,4	3.3	16.0	2,9	44,8
December	0.8	41.1	1.8	35.2	0.9	58.8	-0.9	38.2
January	-3.4	20.2	-0.4	41.5	-2.5	43.5	-2.4	36.4
February	-1.4	47.6	-4.6	3.1	1.8	31.5	-1.7	24.2
March	4.0	45.3	-1.3	10.4	3.9	47.2	1.8	32.9
x monthly/ ∑	8.1	721.2	7.4	745.0	9.4	599.8	7.9	581.8

RESULTS AND DISSCUSION

Inulin content

The inulin content of JA tubers was significantly affected by weather conditions during the study, cultivars, and N dose (Table 3). Inulin content was highest in JA tubers harvested in the first year of the study (2016), and lowest in those harvested in the second year (2017), which was wet and relatively cold (means for years of the study). Polish JA cultivars (Rubik and Albik) had similar inulin content, which was over 26% higher in the German cultivar Gute Gelbe (significant differences at $p<0.05$). The inulin content of JA tubers was significantly highest in cv. Gute Gelbe in the first year of the study (836.7 g kg^{-1} DM), and significantly lowest (533.3 g kg^{-1} DM, a difference of approx. 36%) in cv. Albik in the second year of the study. The N rate of 80 kg ha^{-1} induced the highest (significant) increase in the inulin content of JA tubers, whereas JA tubers grown without N fertilization had the lowest inulin content.

In previous studies, the inulin content of Polish JA cvs. Albik and Rubik ranged from approximately 410 to 504 g kg^{-1} DM (Florkiewicz et al. 2007). Greater amounts of inulin in JA tubers (from approx. 460 to 609 g kg^{-1} DM) were reported by Michalska-Ciechanowska et al. (2019). Inulin accumulation was significantly higher in the early-maturing cv. Topstar than in the mid-

Table 3

Inulin content of JA tubers harvested in autumn (g kg^{-1} DM; mean±SE)

Variable		Cultivar			Mean for year of study
		Rubik	Albik	Gute Gelbe	
Year of study	2016	817.8±16.41 *a*	722.4±32.18 *ab*	836.7±5.46 *a*	792.3±15.26 *A*
	2017	628.9±26.86 *b-d*	533.3±61.23 *d*	815.6±10.84 *a*	659.3±31.60 *B*
	2018	615.1±50.92 *cd*	785.1±32.67 *a*	722.2±14.03 *a-c*	724.1±25.00 *AB*
N rate (kg ha^{-1})	0	570.7±51.95 *c*	549.1±43.55 *c*	785.1±16.87 *ab*	635.0±30.61 *C*
	80	763.3±19.66 *ab*	823.8±18.08 *a*	824.2±9.99 *a*	803.3±10.72 *A*
	120	727.8±32.81 *ab*	668.0±56.39 *bc*	815.1±11.17 *ab*	737.0±24.28 *B*
Means for cultivars		687.3±26.39 *B*	680.3±32.26 *B*	808.1±7.93 *A*	–

Values followed by the different letters are significantly different according to the Tukey's HSD test ($p<0.05$).

late cultivars. In experiments conducted by Singh et al. (2019), the concentrations of inulin and fructooligosaccharides (FOS) in JA tubers were determined at 450 to 750 g kg^{-1} DM. In turn, Redondo-Cuenca et al. (2021) found that JA tubers had a high inulin content of 811 g kg^{-1} DM. In the work of Brkljača et al. (2014), the inulin content of JA tubers ranged from 82 to 135 g kg^{-1} FM.

In the present study, the inulin content of JA tubers (Table 3) was significantly modified by weather conditions and cultivar. According to Chekroun et al. (1994), weather conditions exert a considerable influence on the accumulation of oligofructans in JA tubers. Sawicka et al. (2021) evaluated the inulin content of JA tubers grown in an organic system in Poland and Lithuania, and found that it was higher in a wet growing season than in a season with lower precipitation (172 and 163 g kg^{-1} FM, respectively). More inulin was in tubers cv. Albik than in cv. Rubik (185 and 158 g kg^{-1} FM, respectively). Puttha et al. (2012) demonstrated that the inulin content of JA tubers (553 to 740 g kg^{-1} DM) was determined by plant genotype. According to Puangbut et al. (2017), a genotype exerts a greater effect on inulin accumulation in JA tubers than the interaction between a genotype and environmental conditions.

In the current study, inulin concentration increased significantly in the tubers of JA plants fertilized with N, and it peaked when N was applied at 80 kg N ha^{-1} (Table 3). Matias et al. (2013) observed no differences in the inulin content of JA tubers in response to higher doses of NPK fertilizer. Michalska-Ciechanowska et al. (2019) analyzed the inulin content of JA tubers fertilized with K at three different doses (150, 250 and 350 kg K_2O ha^{-1}) and found that it peaked in response to the highest K dose. Cultivars responded differently to increasing K doses. Pinmongkhonkul et al. (2021) demonstrated that in organic cultivation areas, the inulin content of JA tubers was positively correlated with the soil levels of N, P and K.

The inulin content of JA tubers left in the ground over winter decreased in all years of the study due to biochemical processes, and the decrease ranged from around 37% in 2016 to 62% in 2017 – significant differences at $p<0.05$ (Figure 1) The decrease in the inulin content of JA tubers left in the ground over winter varied across cultivars, but their effect was not significant. The decrease in inulin content was greatest in cv. Rubik (approx. 53%) and smallest in cv. Gute Gelbe (approx. 44%). The inulin content of JA tubers grown without N fertilization decreased after winter by around 44.5%, and this decrease was significantly smaller relative to JA tubers fertilized with 80 and 120 kg N ha^{-1} (48% and 53%, respectively).

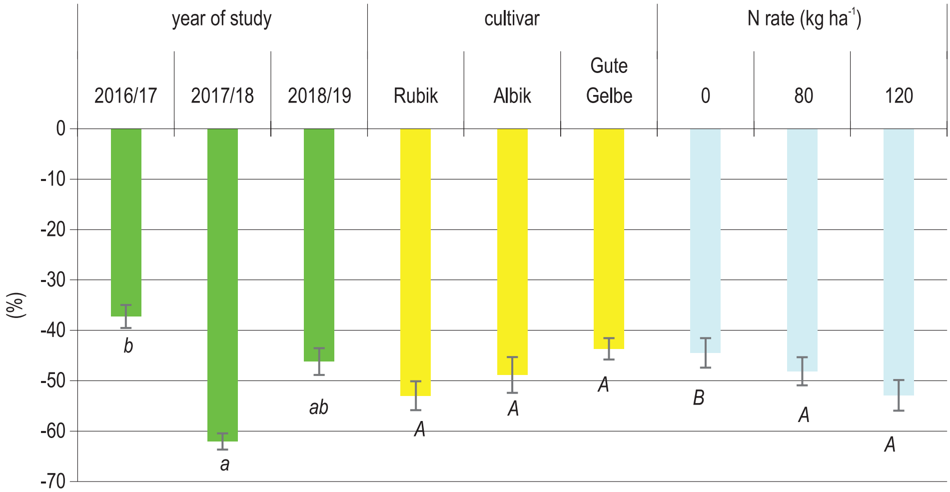

Fig. 1. Changes in the inulin content of JA tubers left in the ground over winter (mean±SE; lower case letters for year of study, upper case letters for cultivars, and italicized upper-case letters for N dose; bars followed by the same letters do not differ significantly according to the Tukey's HSD test at p<0.05)

Černiauskiene et al. (2018) demonstrated that the largest amounts of inulin were obtained in spring from JA cvs. Albik and Rubik (from 330.7 to 423.7 g kg^{-1} DM in March and April), and in autumn from cv. Sauliai (452.3 g kg^{-1} DM in October). According to Michalska-Ciechanowska et al. (2019), JA tubers are most abundant in inulin between mid-October and December, whereas the concentration of polyfructose decreases in successive months. In the cited study, inulin content was significantly lower in the tubers of medium-late maturing cultivars (Violette de Rennes, Waldspindel) than in those of an early-maturing cultivar (Topstar). The inulin content of JA tubers stored in a refrigerator for one to five months ranged from 35.9% DM (after two months of storage) to 43.7% DM (after four months of storage), compared with 47.8% DM in freshly-harvested tubers (Sennoi, Puttha 2021). Inulin concentration varied significantly across cultivars regardless of storage duration.

Similarly to JA tubers, the inulin content of chicory roots was significantly affected by the weather conditions during the growing seasons, genotype, and N dose (Table 4). The inulin content of chicory roots was the highest in the humid and relatively warm 2016 (404.5 g kg^{-1} DM), and the lowest in the wet and relatively cold 2017 (difference of approx. 24%). The roots of cv. Orchies were most abundant in inulin, and the roots of cv. Polanowicka were least abundant in this polysaccharide. The inulin content was significantly the highest in the roots of cv. Orchies grown in the first year of the study (464.4 g kg^{-1} DM), and the lowest in the roots of cv. Polanowicka grown in the second year (267.8 g kg^{-1} DM). Increasing N doses had a negative influence on the inulin content of chicory roots. The roots of chicory plants grown without N fertilization had the significantly highest inulin content (392.8 g kg^{-1} DM). The N dose of 120 kg ha^{-1} induced a significant decrease (over 14%) in the inulin content of chicory roots.

Table 4

Inulin content of chicory roots at harvest (g kg^{-1} DM, mean±SE)

Variable		Cultivar			Mean for year of study
		Polanowicka	Chrysolite	Orchies	
Year of study	2016	323.3±10.91 *ef*	428.8±7.23 *ab*	464.4±14.21 *a*	404.5±13.28 *A*
	2017	267.8±9.59 *f*	301.1±9.63 *ef*	357.8±10.50 *d*	308.9±9.13 *C*
	2018	362.3±9.58 *d*	333.7±10.13 *de*	406.2 ±9.82 *cd*	367.4±8.00 *B*
N dose (kg ha^{-1})	0	353.8±14.82 *b-d*	382.1±18.00 *a-c*	442.5±18.92 *a*	392.8±12.04 *A*
	80	305.3±12.15 *cd*	335.7±17.27 *cd*	414.9±15.67 *ab*	352.0±12.37 *AB*
	120	294.3±15.79 *d*	345.9±24.02 *b-d*	371.1±13.90 *bc*	337.1±12.00 *B*
Means for cultivars		317.8±9.43 *C*	354.5±11.76 *B*	409.5±10.72 *A*	–

Values followed by the different letters are significantly different according to the Tukey's HSD test ($p<0.05$).

Shoaib et al. (2016) reported that the inulin concentration in chicory roots varied between 420 and 760 g kg^{-1} DM, and Redondo-Cuenca et al. (2021) demonstrated that the content of inulin and FOS in chicory roots reached 705 g kg^{-1} DM. Gałązka and Czarnecka (2002) found that inulin yields varied depending on the size of chicory roots and harvest date. The highest inulin yield was obtained from the largest roots harvested until mid-October. Jurgoński et al. (2011) found that the ethanol extract of dried chicory roots had the highest inulin content (601 inulin g kg^{-1} FM), and the extract of dried seeds had the highest mineral content.

The inulin content of chicory roots decreased after five months of cold storage (Figure 2). The decrease in the inulin content was significantly the highest in chicory roots harvested in 2017 (approx. 70%) and significantly the smallest (approx. 65%) in those harvested in 2018. The decrease in the inulin content of chicory roots varied significantly across cultivars. The inulin

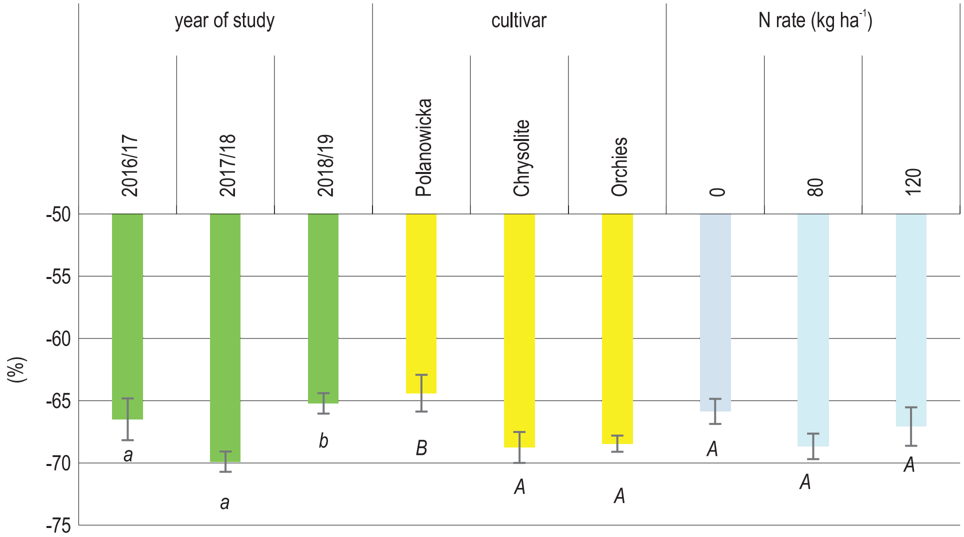

Fig. 2. Changes in the inulin content of chicory roots during storage (mean±SE; lower case letters for year of study, upper case letters for cultivars, and italicized upper-case letters for N dose; bars followed by the same letters do not differ significantly according to the Tukey's HSD test at $p<0.05$)

content decreased by around 64% in the roots of cv. Polanowicka, whereas a significantly greater decrease (by approx. 68%) was noted in the roots of cvs. Chrysolite and Orchies. The decrease in the inulin content was greater in chicory roots fertilized with 80 and 120 kg N ha^{-1} than in the unfertilized treatment (approx. 69% and 67%, respectively vs. approx. 66%; non-significant differences). Also Cabezas et al. (2002) reported that the content of inulin, sucrose, fructose and glucose decreased in chicory roots and JA tubers stored in plastic bags in darkness at different temperatures. In the work of Haggag et al. (2017), the inulin content of artichoke (*Cynara scolymus* L.) heads decreased to a greater extent during storage at 5°C than at 0°C.

Micronutrient content

The micronutrient content of JA tubers is presented in Table 5. The weather conditions had the greatest influence on micronutrient concentrations in JA tubers, and significant differences were found between the growing seasons. The wet seasons of 2016 and 2017 were conducive to the accumulation of Zn, Mn and Fe (excluding 2016) in JA tubers. In the warm and relatively dry 2018, JA tubers were most abundant in B and Cu, and least abundant in Zn and Mn. The cultivar exerted a minor effect on the micronutrient content of JA tubers. Gute Gelbe had the highest concentrations of Cu, Zn and Mn, but a significant difference relative to the other cultivars was noted only for the Cu content. In comparison with the control (unfertilized) treatment, N doses of 80 and 120 kg ha^{-1} increased the concentrations of the analyzed

Table 5

Micronutrient content of JA tubers harvested in autumn (mg kg^{-1} DM; mean±SE)

Variable	Cu	Zn	Mn	Fe	B
Year of study					
2016	6.43±0.77 *b*	23.67±5.35 *a*	24.63±7.43 *a*	174.68±35.07 *b*	3.28±0.45 *b*
2017	7.01±1.20 *ab*	16.44±2.69 *b*	24.32±4.71 *a*	212.32±49.46 *a*	2.48±0.24 *c*
2018	7.32±1.19 *a*	13.50±2.60 *c*	11.31±2.13 *b*	181.19±69.12 *b*	3.56±0.60 *a*
Cultivar					
Rubik	6.57±1.02 *b*	16.43±5.42 *a*	19.10±8.00 *a*	198.00±40.82 *a*	3.09±0.58 *a*
Albik	6.80±0.92 *ab*	17.65±6.00 *a*	19.52±7.81 *a*	185.35±54.48 *a*	3.26±0.72 *a*
Gute Gelbe	7.38±1.27 *a*	19.53±5.35 *a*	21.63±9.48 *a*	184.35±49.32 *a*	2.96±0.60 *a*
N dose (kg ha^{-1})					
0	7.15±1.02 *a*	16.75±5.08 *a*	18.87±8.51 *a*	175.16±57.92 *a*	3.01±0.61 *a*
80	6.91±0.92 *a*	18.64±6.77 *a*	20.35±7.52 *a*	189.78±55.77 *a*	3.17±0.75 *a*
120	6.70±1.37 *a*	18.23±5.04 *a*	21.09±9.43 *a*	203.26±50.73 *b*	3.14±0.56 *a*
Harvest date					
Autumn	6.87±1.07 *a*	15.62±3.71 *b*	20.29 ±9.16 *a*	193.32±50.56 *a*	3.11 0.61 *a*
Spring	6.97±1.19 *a*	20.12±6.43 *a*	19.88±7.90 *a*	185.48±53.78 *a*	3.11±0.68 *a*

Values followed by the different letters are significantly different according to the Tukey's HSD test ($p<0.05$).

micronutrients, except for Cu. However, significant differences were observed only in the Fe content, which was higher in JA tubers fertilized with 120 kg N ha^{-1}. In comparison with JA tubers harvested in autumn, JA tubers left in the ground over winter had a significantly higher Zn content. The concentrations of the remaining micronutrients in JA tubers were not affected by the harvest date.

The concentrations of micronutrients (excluding B) increased in the tubers of cvs. Gute Gelbe and Albik (excluding B) wintering in the ground (Figure 3). The greatest increase in the content of Cu, Zn and Mn was noted in the tubers of cv. Albik. Only the Zn content increased by around 25%-32% in JA tubers of all cultivars which were left in the ground over winter. The Fe content of JA tubers varied across cultivars. Over winter, the Fe content decreased by around 15% in the tubers of cv. Rubik, and increased by around 12% in the tubers of cv. Gute Gelbe. Only changes in the concentrations of Cu and Fe in JA tubers were significant.

The zinc content, regardless of fertilization, was around 30% higher in JA tubers left in the ground over winter than in those harvested in autumn (Figure 4). In comparison with JA tubers harvested in autumn, the Mn content of tubers harvested in spring from the control treatment and the treatment fertilized with 80 kg N ha^{-1} was approximately 10% higher, and it decreased by around 8.5% in response to the application of 120 kg N ha^{-1}. Overwin-

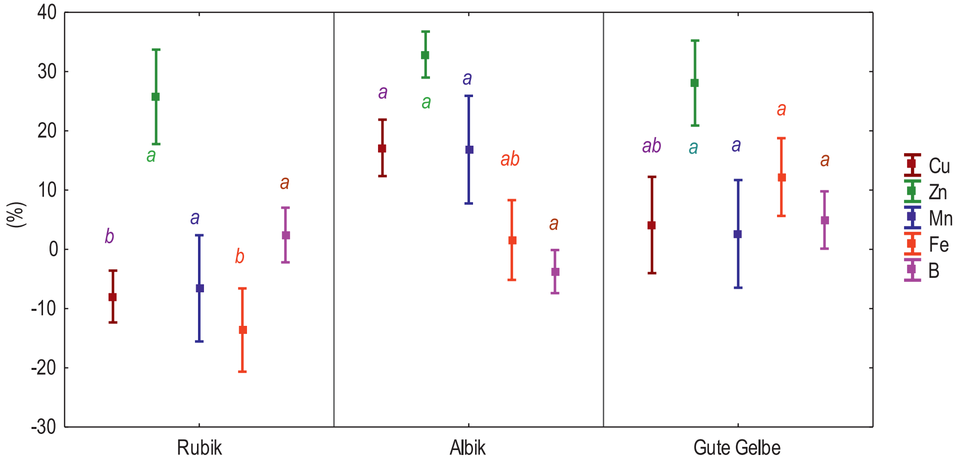

Fig. 3. The effect of a cultivar on changes in the micronutrient content of JA tubers left in the ground over winter (mean±SE, each element is marked with a different color; values for a single element followed by the same letters do not differ significantly according to the Tukey's HSD test at $p<0.05$).

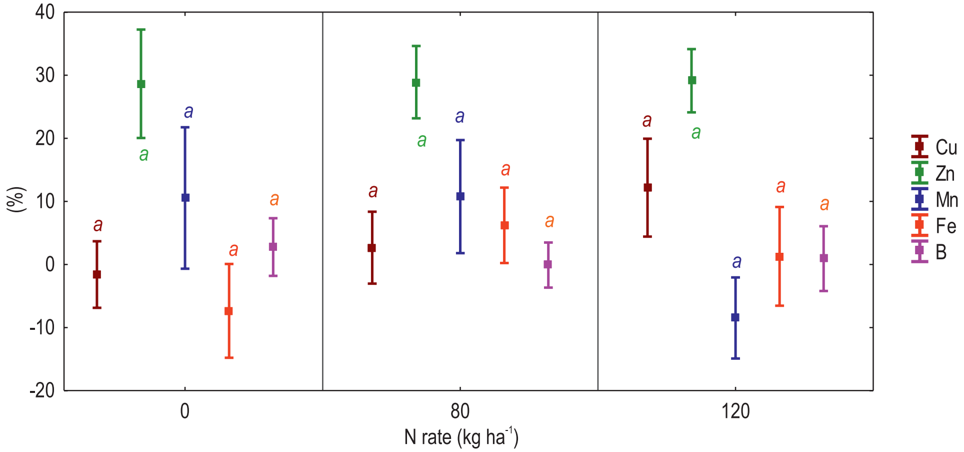

Fig. 4. The effect of N dose on changes in the micronutrient content of JA tubers left in the ground over winter (mean±SE; each element is marked with a different color; values for a single element followed by the same letters do not differ significantly according to the Tukey's HSD test at $p<0.05$).

tering affected also the Cu content of JA tubers; changes in the Cu content varied from -1.6% (unfertilized treatment) to 12.2% (120 kg N ha^{-1}). Changes in the Fe content of JA tubers varied from -7.4% (unfertilized treatment) to 6.2% (80 kg N ha^{-1}). Changes in the B content of JA tubers over winter were small and varied from -0.1% (80 kg N ha^{-1}) to 2.8% (unfertilized treatment). The statistical analysis revealed that N fertilization had no significant influence on changes in the micronutrient content of JA tubers left in the ground over winter.

The chemical composition and mineral content of JA tubers are determined by the environmental and soil conditions, cultivar, agronomic factors, and harvest date (Černiauskiene et al. 2018, Bogucka, Jankowski 2020, Sawicka et al. 2021, Wierzbowska et al. 2021). Research shows that micronutrient concentrations in JA tubers are as follows: Fe – 18 to 150, Cu – 2 to 10, Mn – 2.0 to 100, Zn – 2 to 35.0, Se – 0.02, B – 9 to 13 mg kg^{-1} DM **(**Danilčenko et al. 2011, Ma et al. 2011). In this study, the micronutrient content of JA tubers was modified by the weather conditions, cultivar and N dose (Table 5). The levels of Cu, Zn and Mn were within the ranges reported by the cited authors, whereas the B content was considerably lower, and the Fe content was higher. The weather conditions exerted the strongest effect on the micronutrient content of JA tubers. Sawicka (1996) and Cieślik (1998) also found that environmental conditions were the most important factor affecting the accumulation of Fe and Cu in JA tubers, which was promoted by abundant rainfall and moderate temperatures. According to Sawicka (1996), high Cu levels in JA tubers are noted in favorable weather conditions, whereas during dry spells, JA tubers accumulate more Zn and less Cu. Seiler (1990) observed significant differences in the content of Mn, Zn, Cu, and Fe between JA genotypes grown under identical conditions. In comparison with wild genotypes, the tubers of cultivated genotypes had higher concentrations of Mn, Zn and Fe, and a lower Cu content. The iron concentration was higher in JA tubers than in other root crops (rutabaga, turnip, potato). According to Danilčenko et al. (2011), high genetic variation in nutrient concentrations in various plant parts may be a key to identifying the origin of JA cultivars and clones. Terzić et al. (2012) analyzed the chemical composition of JA tubers in wild genotypes collected in Montenegro and the USA, and reported the following concentrations of essential elements (mg kg^{-1} DM): Mn – 0.31 to 10.80, Fe – 25.91 to 389.10, Zn – 4.82 to 94.38, Cu – 3.35 to 17.12. The levels of Mn, Fe and Cu were higher in American genotypes, whereas the Zn content was higher in Montenegrin genotypes. Sawicka and Kalembasa (2011) demonstrated that JA cv. Rubik was more abundant in minerals than cv. Albik, but the concentrations of the analyzed micronutrients were more stable in the latter cultivar.

Catană et al. (2018) observed differences in the Fe content of JA tubers between red and white varieties (135.2 mg kg^{-1} DM and 126.1 mg kg^{-1} DM on average, respectively). In the current study, the tubers of cv. Rubik with red skin also had a higher Fe content (by approx. 7.5%) than the tubers of cvs. Albik and Gute Gelbe, both with white skin, but the noted differences were not significant at $p<0.05$ (Table 5).

In the present experiment (Table 5), as in the studies of Gruca-Królikowska and Wacławek (2006), increasing N doses had no significant effect on the content of Cu, Zn, Mn, and B in JA tubers, only the Fe content increased in response to higher N doses. However, a rational and balanced mineral fertilization regime is required to achieve and maintain high nutritional value of JA tubers. In a study by Sawicka and Kalembasa (2011), the highest content of Cu, Mn and Zn was noted in the tubers of JA plants fertilized with 150 kg N ha^{-1}, whereas the content of Mo and Fe was the highest in treatments without NPK fertilization. The concentrations of Fe, Mo and Mn were significantly affected by PK fertilization.

Izsaki and Kadi (2013) reported that the Fe content of JA tubers of the early-maturing cv. Tápiói Korai (determined on day 155 of the growing season and at harvest, on day 195) increased to 836 mg kg^{-1} DM, whereas the concentrations of other micronutrients decreased as follows: Cu – 11, Zn – 17, and Mn – 26 mg kg^{-1} DM. In the late-maturing cv. Tápiói Sima, the concentrations of the analyzed micronutrients decreased at harvest, on day 225 of the growing season (relative to earlier stages of tuber formation, i.e. days 155 and 195): Cu – 10, Zn – 20, Mn – 7, and Fe – 110 mg kg^{-1} DM.

Similarly to JA tubers, the accumulation of micronutrients in chicory roots was significantly modified by the weather conditions. (Table 6). Humid and warm weather (2016) was conducive to the accumulation of Zn and Mn, whereas a relatively dry and warm growing season (2018) contributed to an increase in the Cu and B content of chicory roots. High soil moisture content and relatively low temperatures were favorable for the accumulation of Fe in chicory roots. The concentrations of B, Zn and Fe were the highest in the roots of cv. Polanowicka, and the lowest in the roots of cv. Orchies (additionally Mn). The roots of cv. Orchies had the highest Cu content, and the roots of cv. Chrysolite had the highest Mn content. However, significant differences between cultivars were observed only in the Fe and B content. Nitrogen fertilization had no significant influence on the concentrations of Cu, Zn, Fe and B in chicory roots. In comparison with unfertilized plants, the content

Table 6

Micronutrient content of chicory roots at harvest (mg kg^{-1} DM; mean±SE)

Variable	Cu	Zn	Mn	Fe	B
Year of the study					
2016	5.57±0.92 *b*	25.80±2.35 *a*	51.55±11.62 *a*	164.01±31.72 *b*	1.36 ±0.31 *b*
2017	5.47±1.32 *b*	22.03±3.75 *b*	22.99±4.14 *c*	215.02±74.52 *a*	1.64 ±0.49 *ab*
2018	9.71±2.11 *a*	21.41±2.35 *b*	29.09±3.94 *b*	173.08±45.96 *b*	1.74 ±0.47 *b*
Cultivar					
Polanowicka	7.29±2.72 *a*	23.65±3.24 *a*	33.28±9.97 *a*	204.17±71.03 *a*	1.80 ±0.60 *a*
Chrysolite	7.35±2.23 *a*	22.68±2.97 *a*	35.28±14.77 *a*	170.93±41.27 *b*	1.52 ±0.34 *ab*
Orchies	8.19±3.10 *a*	21.65±3.46 *a*	30.99±13.43 *a*	168.79±36.66 *b*	1.43 ±0.29 *b*
N dose					
0	8.42±2.79 *a*	22.70±3.02 *a*	31.80±11.65 *b*	169.66±37.48 *a*	1.53 ±0.36 *a*
80	7.47±2.78 *a*	23.21±3.36 *a*	30.89±9.14 *b*	175.00±43.89 *a*	1.64 ±0.41 *a*
120	6.95±2.40 *a*	22.07±3.51 *a*	36.85±13.33 *a*	199.20±73.77 *a*	1.58 ±0.58 *a*

Values followed by the different letters are significantly different according to the Tukey's HSD test ($p<0.05$).

of Mn and Fe in chicory roots increased significantly in response to the application of 120 kg N ha^{-1}. The Cu content of chicory roots tended to decrease with increasing N doses. The concentrations of B and Zn were the highest in chicory roots fertilized with 80 kg N ha^{-1}.

Massoud et al. (2009) determined the ash content of chicory (*C. intybus* L.) roots at 42.5 g kg^{-1} DM, whereas micronutrient concentrations were arranged in the following descending order (mg kg^{-1} DM): Fe – 17.7, Zn – 3.90, Cu – 3.62, and Mn – 3.12. Chicory is a rich source of minerals, in particular Fe. Jurgoński et al. (2011) reported that the ash content of chicory root ethanol extract was 38 mg kg^{-1} FM. In other studies, the crude ash content of chicory roots ranged from 38.7 (Jangra, Madan 2018) to 56.0 g kg^{-1} (El Zeny et al. 2019). In another experiment, the ash content of chicory roots was 26.7 g kg^{-1} DM, and micronutrient concentrations (mg kg^{-1} DM) were as follows: Fe – 40.10, Zn – 1.90, and Cu 0.35 (Zheng et al. 2003). Nwafor et al. (2017) determined the mineral content (mg kg^{-1} DM) of chicory roots at Cu – 3.60, Mn – 3.10, and Zn – 3.90. In different parts of wild *C. intybus* plants grown in Romania (Stanciu et al. 2019), micronutrient levels were ranked as follows: Cu > Zn > Fe >Mn (highest content in leaves, except for Fe whose content was highest in flowers).

Root vegetables undergo physiological, chemical and biological changes during storage, which lead to quantitative and qualitative yield losses. Changes in the chemical composition of stored vegetables are affected by the cultivar, weather conditions during the growing season, and storage conditions (Wierzbowska et al. 2021). Changes in micronutrient concentrations in chicory roots during storage varied across cultivars (Figure 5). In the roots of cv. Polanowicka, the content of Zn and Fe decreased by around 14%,

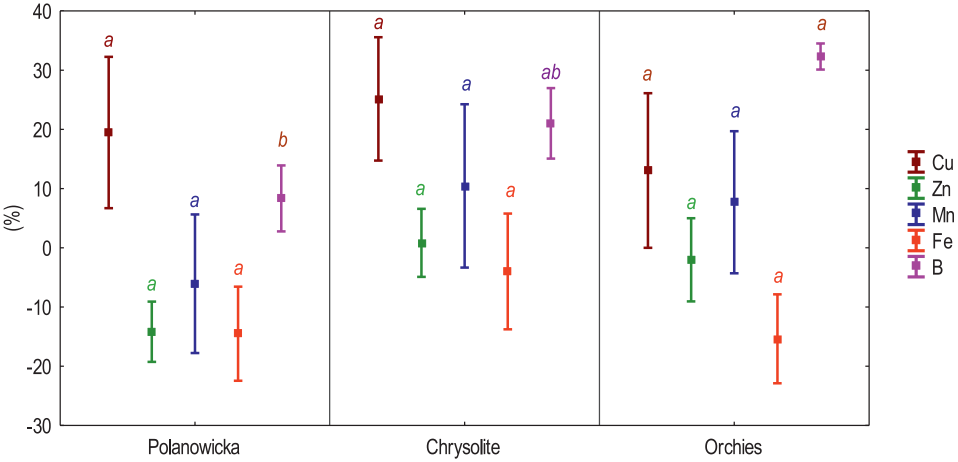

Fig. 5. The effect of cultivar on changes in the micronutrient content of chicory roots during storage (mean±SE; each element is marked with a different color; values for a single element followed by the same letters do not differ significantly according to the Tukey's HSD test at $p<0.05$)

the Mn content decreased by 6%, whereas the content of Cu and B increased by >19% and >8%, respectively. The roots of cv. Chrysolite were characterized by the highest increase in the concentrations of Cu (approx. 25%) and B (20%), followed by Mn (approx. 10%). After storage, the content of B, Cu and Mn in the roots of cv. Orchies increased by 33%, 13% and approximately 8%, respectively. The iron content decreased by around 15% in the roots of cv. Orchies, and by 4% in the roots of cv. Chrysolite. The cultivar had a significant ($p<0.05$) influence on changes in the B content of chicory roots during storage.

The effect of N doses on changes in the micronutrient content of chicory roots during storage is presented in Figure 6. The content of Cu and B in chicory roots increased with increasing N doses (by 11%-29% and 14.5-26%, respectively). A decrease in the concentrations of Zn (1.6% to 9.3%) and Fe (6.3% to 16.0%) in chicory roots was observed in all treatments (0, 80 and 120 kg N ha^{-1}). Changes in the Mn content of chicory roots were ambiguous. Different levels of N fertilization had no significant influence on changes in the micronutrient content of chicory roots during storage.

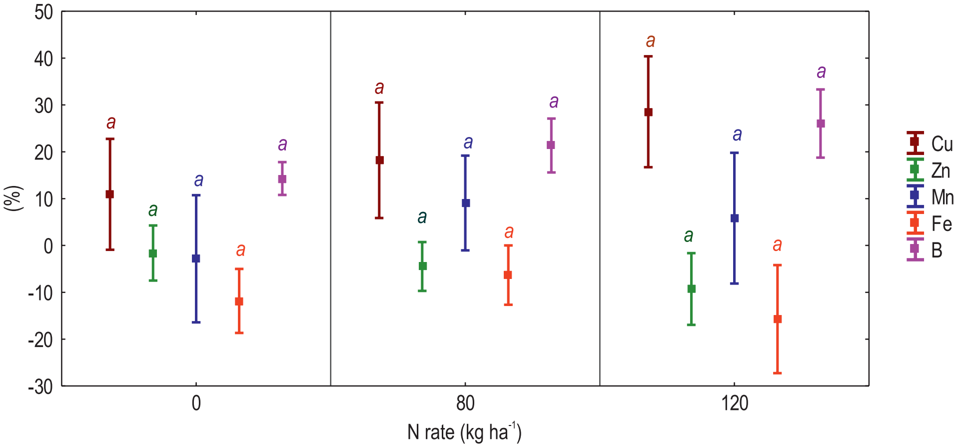

Fig. 6. The effect of N dose on changes in the micronutrient content of chicory roots during storage (mean±SE; each element is marked with a different color; values for a single element followed by the same letters do not differ significantly according to the Tukey's HSD test at $p<0.05$)

CONCLUSIONS

Jerusalem artichoke tubers were a richer source of inulin than chicory roots. The inulin content of JA tubers and chicory roots varied significantly across cultivars. Unlike chicory roots, JA tubers fertilized with N had higher inulin content than unfertilized plants. Weather conditions exerted the greatest influence on the inulin content of JA tubers and chicory roots. Inulin content was lowest in JA tubers and chicory roots harvested in the wet and cold 2017. The highest decrease in inulin content was noted in JA tubers left in the ground over winter and in stored chicory roots, harvested in 2017. The decrease in the inulin content of JA tubers and chicory roots was greater in unfertilized treatments (different N doses induced significant differences in inulin accumulation in JA tubers).

In comparison with chicory roots, JA tubers accumulated more B, less Cu, Zn and Mn, and comparable quantities of Fe. The content of Zn, Mn and Fe was higher in JA tubers and chicory roots harvested in wet growing seasons (2016 and 2017), and the content of B and Cu was highest in the warm season with average precipitation (2018). Jerusalem artichoke and chicory cultivars had a minor effect on micronutrient concentrations, only Cu content was significantly higher in the tubers of cv. Gute Gelbe, and the content of Fe and B was significantly higher in the roots of cv. Polanowicka. Increasing N doses induced an increase in the Fe content of JA tubers, and a decrease in the content of Fe and B in chicory roots. The Zn content in tubers of all JA cultivars was higher after overwintering than harvested in the fall. The N dose had no effect on changes in the content of micronutrients in JA tubers overwintering in the soil. The concentrations of the analyzed micronutrients increased in cvs. Albik (excluding B) and Gute Gelbe. The concentrations of Cu, B and Mn increased in stored chicory roots (except for Mn content in the 0 kg N ha^{-1} treatment). The roots of cvs. Chrysolite and Orchies were characterized by the greatest increase in micronutrient content (except for Fe content in both cultivars, and Zn content in cv. Orchies).

Author contributions

J.W., B.C-A. – conceptualization; J.W., A.W. – formal analysis; J.W., B.C-A., B.B. – funding acquisition; J.W., B.C-A., B.B. A.W – methodology; J.W. – visualization; J.W., B.C-A. – writing-original draft preparation; J.W. – writing-review and editing. All authors have read and agreed to the published version of the manuscript.

ACKNOWLEDGMENTS

The publication was written as a result of the author's internship in Slovak University of Agriculture in Nitra Faculty of Engineering, Institute of Agricultural Engineering, Transport and Bioenergetics, co-financed by the European Union under the European Social Fund (Operational Program Knowledge Education Development), carried out in the project Development Program at the University of Warmia and Mazury in Olsztyn (POWR.03.05.00-00-Z310/17).

Conflicts of interest

The authors declare no conflict of interest.

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