Jerusalem Artichoke: Quality Response to Potassium Fertilization and Irrigation in Poland

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Received: 27 August 2020; Accepted: 26 September 2020; Published: 6 October 2020

Abstract: The aim of this study was to determine the effects of soil potassium fertilization (150, 250 and 350 kg K₂O ha⁻¹) and irrigation on the tuber quality (content of α-tocopherol, β-carotene, essential and endogenous amino acids) of three Jerusalem artichoke (Helianthus tuberosus L.) cultivars (Topstar, Violette de Rennes, Waldspindel). Jerusalem artichokes were grown during a field experiment in the Agricultural Experiment Station in Tomaszkowo (53°42' N, 20°26' E, north-eastern Poland). The content of α-tocopherol and β-carotene was determined at 1.60–2.65 and 0.75–1.00 mg kg⁻¹ DM, respectively, in all Jerusalem artichoke cultivars produced in north-eastern Poland. High rates of potassium fertilizer (250 and 350 kg K ha⁻¹) increased the content of α-tocopherol in tubers by 47% and 66% on average, respectively. The stimulatory effects of high potassium rates on the content of α-tocopherol (2.5-fold increase) were observed only in response to irrigation. High rates of potassium fertilizer induced a particularly high increase (3.2-fold) in α-tocopherol concentrations in Jerusalem artichokes cv. Waldspindel. Irrigation increased α-tocopherol levels (by 40%) and decreased the concentrations of β-carotene (by 25%) and most essential and endogenous amino acids (isoleucine, leucine, lysine, phenylalanine, valine, alanine, glycine, histidine, serine, threonine). The Topstar cultivar accumulated the highest quantities of essential and endogenous amino acids. Leucine, methionine + cysteine were the limiting amino acids in Jerusalem artichoke tubers. The analyzed tubers were characterized by very high nutritional quality of dietary protein (Essential Amino-Acid Index, 66–78).

Keywords: Helianthus tuberosus L.; fertilization; irrigation; α-tocopherol; β-carotene; amino acids

1. Introduction

Jerusalem artichoke (Helianthus tuberosus L., Asteraceae) originated in North America where it was cultivated by the native population [1,2]. The species is presently grown in Europe and Asia as a valuable source of lignocellulosic biomass for the energy sector [3–9]. Jerusalem artichoke is a crop species that most effectively converts solar energy to biomass in both quantitative and qualitative terms [10–14]. Processed biomass is a valuable resource for the production of biofuels [1,3,4,15,16]. Immature aerial parts of H. tuberosus are used in biogas production, whereas straw is processed into solid fuels (pellets, briquettes), and tubers are a high-yielding resource in the production of bioethanol or substrate/cosubstrate in the methane fermentation process [16–18]. In the temperate climate of Poland, tuber and straw yields can reach 14–30 and 20–50 Mg ha⁻¹ dry matter (DM), respectively [13]. The amount of energy accumulated in the harvested biomass (tubers, straw) ranges from 142–281 [4] to 348 GJ ha⁻¹ [19]. However, high levels of genetic/phenotypic variation in H. tuberosus populations lead to considerable differences in yield, even in relatively similar agricultural production systems in Europe [13,20]. Jerusalem artichoke has low agronomic requirements [1,16], it is resistant to a wide range of biotic and abiotic stressors [1,21–23], and prevents soil erosion [17]. The species
easily adapts to diverse environments [8], therefore it can be effectively grown for energy production on marginal land [4]. Jerusalem artichoke is also a natural source of: (i) inulin, (ii) oligofructose, and (iii) fructose [24–26], compounds with nutritional and functional attributes (food and feed) that are particularly beneficial to individuals with type 2 diabetes, obesity and cardiovascular disorders [25,27,28]. Tubers are abundant in protein, polyphenols and vitamins [26,29]. These qualities make Jerusalem artichoke biomass particularly suitable for biorefining and the development of the bioeconomy [6]. In biorefineries, organic chemical compounds and lignocellulosic materials are recovered, processed and used in the production of biofuels, bioproducts and bioenergy. Jerusalem artichoke biomass is ideally suited for these industrial applications [23].

Jerusalem artichoke tubers can survive (without loss) in frozen soils for several months because they are abundant in inulin. Overwintering success is compromised only in areas where tubers become dehydrated [21]. The species has minimal soil and fertilizer requirements [21,28,30]. It is susceptible to numerous fungal pathogens (Sclerotinia sclerotiorum (Lib.) de Bary, Fusarium acuminatum Ellis & Everh, Botrytis cinerea Pers.) which colonize mainly tubers. Pests of economic importance do not exert pressure on Jerusalem artichoke plantations [23].

The biological value of Jerusalem artichoke tubers is determined by the genetic characteristics of cultivars and the applied production technology [28,30–34]. Fertilization, in particular potassium fertilization, is an agricultural treatment that exerts the greatest impact on the biological value of tubers. Potassium enhances plant growth, development and tuber yields [35], and it plays an important role in the transport of photosynthesis products from leaves and their accumulation in tubers [36,37]. Agricultural crops are increasingly often irrigated due to a higher risk of drought, including in the temperate climate [38–40]. Denorpy [17], Monti et al. [24] and Gao et al. [34] demonstrated that H. tuberosus is sensitive to water stress. Jerusalem artichoke thrives in regions with high levels of precipitation, and irrigation exerts particularly beneficial effects during tuber formation [34]. The aim of this study was to determine the effects of soil potassium fertilization and irrigation on the tuber quality (measured as content of α-tocopherol, β-carotene, amino acids) of three Jerusalem artichoke cultivars representing different maturity groups.

2. Materials and Methods

2.1. Field Experiment

Jerusalem artichoke (Helianthus tuberosus L.) was grown in a field experiment in the Agricultural Experiment Station in Tomaszkowo (53°42' N, 20°26' E, NE Poland) in 2018. The experiment had a three-factorial split-split-plot design with three replications. The experimental variables were: (i) cultivar: Topstar (an early edible cultivar with yellow-brown tubers), Violet de Rennes (a mid-late edible cultivar with red tubers), Waldspindel (a mid-late cultivar with red tubers, used in herbal and distilling industries); (ii) soil potassium fertilization-potassium sulfate (kg K₂O ha⁻¹): 150, 250, 350; (iii) irrigation: with and without irrigation.

The studied cultivars had been obtained from an organic farm (Die Topinambur Manufaktur, Heimenkirch, Bavaria, Germany). Tubers were planted in mid-April to a depth of 6–8 cm, with 75 × 30 cm spacing. Before planting, potassium fertilizer was applied in the form of potassium sulfate (50%), 80 kg N ha⁻¹ (urea, 46%), 70 kg P₂O₅ ha⁻¹ (enriched superphosphate, 40%), and 90 CaO kg ha⁻¹ (ground dolomite, 52% CaO, 37% MgCO₃, 48% CaCO₃), according to the experimental design. Organic fertilizer was not applied.

Soil moisture content was controlled from the beginning of tuber formation, during the growth of aerial plant parts, until leaf ageing and the translocation of sugars to tubers (from mid-June to mid-October). The optimal soil moisture content was set at 14.3–16.5%, i.e., 65–75% of field water capacity at a depth of 30 cm. Measurements were conducted twice a week. The crops were irrigated every 5–7 days at 20 dm³ m⁻² when field water capacity decreased below 60% (≤13.2% soil moisture content). Irrigation treatments for Jerusalem artichoke were developed based on the irrigation regime.
for late potato cultivars (*Solanum tuberosum* L.) and field water capacity for different soil types [40]. Soil moisture content was measured with the SM 150-KIT probe (Geomor-Technik sp. z o. o., Szczecin, Poland). Plots were irrigated 11 times during the growing season of *H. tuberosus* (13 and 19 June; 3 and 9 July; 10 and 23 August; 3, 11, 20 and 28 September; 6 October) in the total amount of 220 mm of water.

The experiment was established on Haplic Luvisol loamy sand [41]. Each experimental plot had an area of 2.7 m². Oat (*Avena sativa* L.) was the preceding crop. Composite soil samples were collected from each plot to a depth of 20 cm to determine the chemical properties of soil. Soil pH was determined at 5.4 with a digital pH meter, and soil nutrient levels were determined at 74 mg P kg⁻¹ (Egner-Riehm method), 145 mg K kg⁻¹ (Egner-Riehm method) and 69 mg Mg kg⁻¹ (AAS) [42]. Tubers were hilled once after planting. Jerusalem artichoke was harvested at the beginning of November.

### 2.2. Determination of Tocopherol and β-Carotene Content

The content of β-carotene and tocopherols ([±]-α-tocopherol, β-tocopherol, (+)-γ-tocopherol, (+)-δ-tocopherol) was determined under limited exposure to sunlight. Samples of freeze-dried *H. tuberosus* tubers (Alpha 1-4 LS C Basic laboratory freeze dryer, CHRIST GmbH, Osterode am Harz Germany) were ground in a laboratory mill (Knife Mill GM 300, Retsch GmbH, Haan, Germany). Ground samples of 5 g were combined with 30 cm³ of 20% (v/v) ascorbic acid (aqueous solution) (analytical grade, Sigma-Aldrich Chemie GmbH, Munich, Germany) and extracted in 100 cm³ of naphthyl ether/ethanol mixture (v/v 1:1) (analytical grade, Sigma-Aldrich Chemie GmbH) at room temperature, in dark, for 18 h. The samples were saponified with 50% (v/v) aqueous KOH solution (analytical grade, Sigma-Aldrich Chemie GmbH) at room temperature, in dark, for 6–18 h. The samples were extracted in 4 × 50 cm³ of naphthyl ether (40/60, analytical grade, Sigma-Aldrich Chemie GmbH). The extracts were combined in the separator and rinsed with 10% (v/v) aqueous NaCl solution (analytical grade, Avantor Performance Materials Poland SA, Gliwice, Poland) and deionized water. The eluate was dehydrated with anhydrous sodium sulfate (analytical grade, Sigma-Aldrich Chemie GmbH) and evaporated to dryness in a rotary evaporator at 40 °C (IKA RU 10 digital, Janke & Kunkel IKA–Labortechnik, Baden-Württemberg, Germany). The remainder was dissolved in 5 cm³ of anhydrous n-hexane (analytical grade, Avantor Performance Materials Poland SA), the extract was passed through a PTFE syringe filter with 0.22 µm pore size (30-SF-02, Chromacol Ltd., Hertfordshire, UK) and analyzed in a HPLC system (Shimadzu, Kyoto, Japan).

Tocopherol concentrations were determined using a Nucleosil C₁₈ 250 × 4.6 mm 5 µm column (Sigma-Aldrich Chemie GmbH) under the following conditions: mobile phase–methanol:H₂O (95:5 v/v) (HPLC grade, Sigma-Aldrich Chemie GmbH), flow rate–1 cm³ min⁻¹, detector: RF E₂ 293 and Eₐ 326, sample loop–20 µL. The results were calibrated based on external standards for (±)-α-tocopherol (DL-all-rac α-tocopherol), β-tocopherol, (+)-γ-tocopherol and (+)-δ-tocopherol (Sigma-Aldrich Chemie GmbH). A standard mixture composed of 5 µg of α-T cm⁻³, 2 µg of β-T cm⁻³, 2 µg of γ-T cm⁻³ and 2 µg of δ-T cm⁻³ in anhydrous ethanol (analytical grade, Avantor Performance Materials Poland SA) was prepared. The peak areas of the analyte and the applied standards were compared [43].

The concentration of β-carotene was determined with the use of the Gemini® 5 µm C₁₈ 110 A 250 × 4 mm column (Phenomenex Inc, Torrance, CA, USA) under the following conditions: mobile phase–methanol: tetrahydrofuran (95:5 v/v) (HPLC grade, Sigma-Aldrich Chemie GmbH), flow rate–1 cm³ min⁻¹, detector: UV-vis 450 nm, sample loop–20 µL. Synthetic β-Carotene Type I external standard was applied (Sigma-Aldrich Chemie GmbH) [44,45]. Vitamin A activity was determined in retinol equivalents (RE), where 1 RE = 6 µg of β-carotene [46].

### 2.3. Determination of Amino Acid Content and the Nutritional Value of Protein

The content of amino acids, excluding tryptophan, was determined in dried tuber samples in the AAA-400 amino acid analyzer (INGOS s.r.o, Prague, Czech Republic). The samples were hydrolyzed in 6 M HCl for 24 h at a temperature of 110 °C. The hydrolysate was cooled, filtered, rinsed and evaporated in a water bath, and dry residue was dissolved in a buffer with a pH of 2.2. The obtained specimens
were analyzed in the ninhydrin test. The applied buffers had a pH of 2.6, 3.0, 4.25 and 7.9. Ninhydrin solution was buffered at pH 5.5. The column with a length of 370 mm was packed with ion-exchange resin (Oston ANB, INGOS s.r.o.). Column temperature was 58–74 °C, and reactor temperature was 120 °C. The concentrations of sulfur-containing amino acids (methionine and cysteine) were determined by hydrolysis with a mixture of formic acid and hydrogen peroxide (9:1) at a temperature of 110 °C for 23 h. Cooled samples were handled in accordance with the acid hydrolysis protocol. The applied program (Equation (1)) and the essential amino acid index (EAAI) [48]:

\[
CS = \frac{a_b}{a_w} \times 100
\]

where CS – chemical score, \(a_b\) – content of essential amino acid and \(a_w\) – content of essential amino acid in the reference protein.

2.4. Statistical Analysis

Data were processed statistically by one-way analysis of variance (ANOVA) in the Statistica 13.3 program [49]. Multiple comparison Tukey’s HSD (honestly significant difference) statistical test was applied to assess significant differences (\(p < 0.05\)) between means. The results of the F-test for fixed effects in ANOVA are presented in Table 1.

### Table 1. F-test statistics in ANOVA.

| Parameter                  | Cv. | K | Irrigation (IR) | Cv. × K | Cv. × IR | K × IR | Cv. × K × IR |
|----------------------------|-----|---|-----------------|---------|----------|--------|-------------|
| \(\alpha\)-Tocopherol       | 1.899 ns | 6.254 ** | 2.850 ns | 6.161 ** | 0.053 ns | 7.787 ** | 2.426 ns |
| % \(\alpha\)-Tocopherol in total tocopherols | 1.428 ns | 5.678 ** | 0.004 ns | 2.830 ns | 1.766 ns | 1.098 ns | 1.541 ns |
| \(\beta\)-Carotene          | 1.558 ns | 0.932 ns | 10.066 ns | 0.371 ns | 0.010 ns | 1.370 ns | 1.370 ns |
| Retinol equivalents        | 1.578 ns | 0.928 ns | 10.061 ns | 0.376 ns | 0.010 ns | 1.395 ns | 0.496 ns |
| Alanine                    | 4.001 * | 0.711 ns | 14.589 ** | 0.563 ns | 1.253 ns | 0.879 ns | 1.653 ns |
| Arginine                   | 17.560 ** | 0.199 ns | 10.283 ** | 1.113 ns | 5.001 * | 0.396 ns | 2.395 ns |
| Glycine                    | 5.258 * | 0.717 ns | 23.100 ** | 1.329 ns | 2.885 ns | 0.119 ns | 2.127 ns |
| Histidine                  | 0.482 ns | 0.607 ns | 13.105 ** | 0.519 ns | 1.017 ns | 0.151 ns | 1.236 ns |
| Aspartic acid              | 7.032 ** | 1.674 ns | 0.944 ns | 1.463 ns | 0.729 ns | 0.144 ns | 1.710 ns |
| Glutamic acid              | 20.094 ** | 4.917 * | 8.103 * | 3.383 * | 5.050 * | 1.457 ns | 1.505 ns |
| Proline                    | 2.472 ns | 1.399 ns | 11.632 ** | 2.266 ns | 5.884 * | 1.765 ns | 1.611 ns |
| Serine                     | 2.120 ns | 0.276 ns | 9.025 ** | 0.620 ns | 1.495 ns | 0.235 ns | 1.078 ns |
| Threonine                  | 4.429 * | 0.532 ns | 17.101 ** | 0.712 ns | 1.564 ns | 0.068 ns | 1.669 ns |
| Isoleucine                 | 6.208 ** | 0.196 ns | 17.926 ** | 1.614 ns | 1.617 ns | 0.131 ns | 1.562 ns |
| Leucine                    | 8.587 ** | 0.803 ns | 21.939 ** | 1.109 ns | 3.340 ns | 0.389 ns | 2.297 ns |
| Lysine                     | 1.004 ns | 0.160 ns | 4.445 * | 0.543 ns | 0.677 ns | 0.018 ns | 0.400 ns |
| Methionine                 | 1.369 ns | 0.141 ns | 0.435 ns | 0.265 ns | 0.381 ns | 0.494 ns | 0.690 ns |
| Cystine                    | 1.695 ns | 0.231 ns | 1.326 ns | 0.232 ns | 0.338 ns | 0.980 ns | 0.716 ns |
| Phenylalanine              | 3.666 * | 2.447 ns | 25.536 ** | 3.391 * | 3.268 ns | 0.481 ns | 1.215 ns |
| Threonine                  | 29.488 *** | 0.061 ns | 1.565 ns | 0.817 ns | 0.921 ns | 0.154 ns | 1.911 ns |
| Tryptophan                 | 1.610 ns | 0.093 ns | 0.127 ns | 0.093 ns | 0.129 ns | 0.219 ns | 0.261 ns |
| Valine                     | 7.553 ** | 0.501 ns | 23.428 ** | 1.121 ns | 3.981 * | 0.442 ns | 1.734 ns |

* Significant at \(p < 0.05\), ** significant at \(p < 0.01\), *** significant at \(p < 0.001\), ns—not significant.

2.5. Weather Conditions

The growing season of 2018 lasted for 201 days. Jerusalem artichokes were harvested in the first week of November. Weather conditions during the experimental period are presented in Table 2. During the growing season, mean monthly air temperatures did not differ considerably from the
long-term average of 1981–2010. Rapid growth of aerial plant parts and tuber setting began in mid-June. Total precipitation during the growing season reached 418.8 mm, and it was 7% lower than the long-term average (450.1 mm). Rainfall was unevenly distributed, with dry spells in May and September. Precipitation levels were lowest in May and September, 57% and 64% lower than the long-term average, respectively, and irrigation was required (3, 11, 20 and 28 September). As described in the Methods section, the optimal moisture content of soil was determined based on the irrigation system for late-maturing potato varieties. A comparison of the water requirements of plants with precipitation levels shows that rainfall was 27% lower than the optimal water supply also in August. Only in July precipitation exceeded the long-term average by 90% and the water requirements of plants by 45%. Despite the above, measurements of the soil moisture status revealed the need for irrigation since the field water capacity dropped below 60%, i.e., to ≤13.2%.

Table 2. Meteorological data for the growing season of 2018 and long-term average for 1981–2010.

| Specification         | April | May | June | July | August | September | October | November |
|-----------------------|-------|-----|------|------|--------|-----------|---------|----------|
| Mean air temperature (°C) | 10.8  | 15.7| 17.2 | 19.7 | 19.2   | 14.5      | 8.7     | 3.3      |
| 1981–2010             | 7.7   | 13.5| 16.1 | 18.7 | 17.9   | 12.8      | 8.0     | 2.9      |
| Rainfall total (mm)   | 33.5  | 25.0| 53.7 | 141.0| 44.6   | 20.3      | 84.7    | 16.0     |
| 1981–2010             | 33.3  | 58.5| 80.4 | 74.2 | 59.4   | 56.9      | 42.6    | 44.8     |

3. Results and Discussion

3.1. Content of α-Tocopherol and β-Carotene

α-, β-, γ- and δ-Tocopherols (lipid-soluble compounds with vitamin E activity) act as antioxidants that exert anti-sclerotic effects and improve vascular function. According to Sram et al. [50], vitamin E is essential for healthy brain function and a 12-month course of vitamin E improves short-term memory, psychomotor functions and mood. The optimal daily dose of supplemental vitamin E is 200–400 mg of tocopheryl acetate [51]. α-Tocopherol has the highest biological activity (100%), and the activity of the remaining tocopherols (β, γ, and δ) ranges from 2 to 35% [52]. Tocopherols are thermally stable even at a temperature of 200 °C (only 10% of tocopherol is lost during cooking) [52]. In the present study, the content of α-tocopherol in Jerusalem artichoke tubers ranged from 1.60 to 2.65 mg kg⁻¹ DM (Table 3).

Table 3. The effect of experimental factors on the vitamin content of Jerusalem artichoke tubers.

| Parameter                  | α-Tocopherol (mg kg⁻¹ Lyophilisate) | α-Tocopherol (% Total Tocopherols) | β-Carotene (mg kg⁻¹ Lyophilisate) | Retinol Equivalents |
|----------------------------|-------------------------------------|-----------------------------------|----------------------------------|---------------------|
| Cultivar                   |                                     |                                   |                                  |                     |
| Violette de Rennes         | 2.04                                | 76.1                              | 0.82                             | 137                 |
| Waldspindel                | 2.55                                | 70.2                              | 0.83                             | 138                 |
| Topstar                    | 2.02                                | 71.2                              | 0.97                             | 162                 |
| Potassium fertilization (kg K₂O ha⁻¹) |                                    |                                   |                                  |                     |
| 150                        | 1.60 b                              | 68.0 b                            | 0.81                             | 135                 |
| 250                        | 2.35 ab                             | 69.8 b                            | 0.94                             | 157                 |
| 350                        | 2.65 a                              | 79.6 a                            | 0.88                             | 147                 |
| Irrigation                 |                                     |                                   |                                  |                     |
| Irrigated                  | 2.57 a                              | 72.4                              | 0.75 b                           | 125 b               |
| Not irrigated              | 1.83 b                              | 72.6                              | 1.00 a                           | 167 a               |

Means with the same letters do not differ significantly at p ≤ 0.05 in Tukey’s test. The absence of letters denotes non-significant main effects.
Wheat germ oil is the richest source of α-tocopherol (2500 mg kg\(^{-1}\)) [52]. In edible vegetable oils, the mean concentration of α-tocopherol ranges from 50–100 (soybean oil) to 300–400 mg kg\(^{-1}\) (canola and sunflower oil) [53,54]. The concentration of α-tocopherol was determined at 1–9 mg kg\(^{-1}\) in the seeds of oilseed crops (linseed, mustard, poppy, pumpkin), 1–15 mg kg\(^{-1}\) in cereal grain (buckwheat, barley, maize, rye, spelt, millet), and 16–104 mg kg\(^{-1}\) in legume seeds (chick peas, lentils, peas) [55]. Potato tubers contain 0.5–2.8 [56] to even 20.8 mg kg\(^{-1}\) DM of α-tocopherol (native Andean cultivars) [57].

In the current study, an increase in the potassium fertilizer rate led to a significant increase in the α-tocopherol content of Jerusalem artichoke tubers (Table 3). This correlation was particularly pronounced in cv. Waldspindel where α-tocopherol levels increased 3.2-fold when the potassium rate was increased from 150 to 350 kg K\(_2\)O ha\(^{-1}\) (Figure 1). Irrigation increased α-tocopherol concentrations by 41% on average in the studied Jerusalem artichoke cultivars (Table 3). Potassium fertilizer enhanced α-tocopherol levels only in irrigated plots. In non-irrigated plots, potassium fertilization was not correlated with α-tocopherol concentrations in \(H.\ tuberosus\) tubers (Figure 2). α-Tocopherol is the most biologically active form of tocopherol, and its proportion in total tocopherols is an important consideration [52]. Potassium rate was the only experimental variable which significantly differentiated the proportion of α-tocopherol in total tocopherols (Table 1). The highest percentage of α-tocopherol in total tocopherols (79.6% on average) was noted in tubers supplied with potassium at 350 kg ha\(^{-1}\), regardless of cultivar or irrigation (Table 3).

β-Carotene is a vitamin A precursor which is essential for the maintenance of normal vision, healthy skin and immunity [58]. The recommended daily allowance of vitamin A is 4.8 mg of β-carotene [59]. Foods of animal origin and carotene-containing plants are the main sources of vitamin A in the human diet [52,60,61]. In this study, the β-carotene content of \(H.\ tuberosus\) tubers ranged from 750 to 1000 μg kg\(^{-1}\) DM (Table 3), and it was 1.8-to 2.5-fold higher than in potato tubers (400 μg kg\(^{-1}\) DM), and 3.7-to 5-fold higher than in sugar beetroots (200 μg kg\(^{-1}\) DM) [62]. In the present study cultivar, potassium rate and irrigation did not affect β-carotene synthesis in Jerusalem artichoke tubers (Table 1). In a study by Wierzbicka and Hallmann [61], agronomic factors also exerted a weak influence on β-carotene synthesis in potato tubers.

**Figure 1.** The effect of potassium fertilization on the α-tocopherol content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at \(p \leq 0.05\) in Tukey’s test. The absence of letters denotes non-significant main effects.
Figure 2. The effects of irrigation and potassium fertilization on the α-tocopherol content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.

Vitamin A activity is determined in retinol equivalents (RE) [46,62]. Raju et al. [46] analyzed the activity of provitamin A in 30 species of leafy vegetables and reported the highest value in broccoli (641 RE) and the lowest value in sorrel (12 RE). In the present experiment, vitamin A activity in Jerusalem artichokes was determined in the range of 125–167 RE, and it was higher in non-irrigated tubers (Table 3).

3.2. Amino acid Content and Nutritional Value of Protein

Jerusalem artichoke contains highly nutritional protein with a balanced amino acid profile [63]. In this study, amino acid concentrations differed across the examined cultivars (Table 1).

The highest content of most essential (isoleucine, leucine, phenylalanine, thyrosine and valine) and endogenous amino acids (alanine, glycine, threonine) was noted in cv. Topstar. Jerusalem artichoke cv. Waldspindel was most abundant in thyrosine and arginine, whereas the tubers of cv. Violette de Rennes were characterized by the highest concentrations of aspartic acid and glutamic acid (Tables 4 and 5).

The content of lysine, methionine, cystine, tryptophan, histidine, proline and serine was not differentiated by the genetic factor (Table 1). An increase in the potassium fertilizer rate to 250 kg K$_2$O ha$^{-1}$ decreased the content of glutamic acid (by 1.22 g 100 g$^{-1}$ of protein) in Jerusalem artichoke tubers (Table 5). This correlation was particularly visible in cvs. Violette de Rennes and Topstar where glutamic acid levels decreased by 2.57 g 100 g$^{-1}$ of protein on average in response to higher potassium rates (Figure 3). The phenylalanine content of the examined cultivars was also influenced by potassium fertilization (Cv. $\times$ K interaction) (Table 1). Phenylalanine levels were highest in cv. Topstar supplied with the lowest potassium rate (150 kg K$_2$O ha$^{-1}$). In the tubers of cv. Waldspindel, similar concentrations of phenylalanine were noted only in response to the highest potassium rate (350 kg K$_2$O ha$^{-1}$) (Figure 4).
Table 4. The effect of experimental factors on the content of essential amino acids (g 100 g$^{-1}$ of protein) in Jerusalem artichoke tubers.

| Parameter                        | Isoleucine | Leucine | Lysine | Methionine | Cysteine | Phenylalanine | Thyrosine | Tryptophan | Valine |
|----------------------------------|------------|---------|--------|------------|----------|---------------|-----------|------------|--------|
| Cultivar                         |            |         |        |            |          |               |           |            |        |
| Violette de Rennes               | 2.12 b     | 2.99 b  | 3.72   | 0.59       | 0.433    | 4.79 b        | 1.41 b    | 0.67       | 2.59 b |
| Waldspindel                      | 2.09 b     | 2.97 b  | 3.94   | 0.67       | 0.532    | 5.01 ab       | 2.44 a    | 0.82       | 2.56 b |
| Topstar                          | 2.49 a     | 3.48 a  | 4.23   | 0.76       | 0.570    | 5.36 a        | 2.82 a    | 0.68       | 2.98 a |
| Potassium fertilization (kg ha$^{-1}$) |          |         |        |            |          |               |           |            |        |
| 150                              | 2.20       | 3.08    | 3.99   | 0.69       | 0.523    | 4.98          | 2.22      | 0.71       | 2.70   |
| 250                              | 2.22       | 3.11    | 3.86   | 0.64       | 0.482    | 4.87          | 2.20      | 0.72       | 2.65   |
| 350                              | 2.28       | 3.25    | 4.06   | 0.69       | 0.531    | 5.32          | 2.26      | 0.75       | 2.77   |
| Irrigation                       |            |         |        |            |          |               |           |            |        |
| Irrigated                        | 2.02 b     | 2.88 b  | 3.66 b | 0.65       | 0.476    | 4.62 b        | 2.32      | 0.71       | 2.47 b |
| Not irrigated                    | 2.45 a     | 3.41 a  | 4.28 a | 0.70       | 0.548    | 5.49 a        | 2.13      | 0.74       | 2.95 a |

Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.

Table 5. The effect of experimental factors on the content of endogenous amino acids (g 100 g$^{-1}$ of protein) in Jerusalem artichoke tubers.

| Parameter                        | Alanine | Arginine | Glycine | Histidine | Aspartic acid | Glutamic acid | Proline | Serine | Threonine |
|----------------------------------|---------|----------|---------|-----------|---------------|---------------|---------|--------|-----------|
| Cultivar                         |         |          |         |           |               |               |         |        |           |
| Violette de Rennes               | 3.04 b  | 16.86 b  | 2.69 b  | 2.10      | 8.92 a        | 9.84 a        | 2.99    | 2.18   | 2.63 b   |
| Waldspindel                      | 3.23 ab | 22.07 a  | 2.75 ab | 2.14      | 7.34 b        | 7.31 b        | 2.28    | 2.24   | 2.73 ab  |
| Topstar                          | 3.50 a  | 14.68 b  | 3.11 a  | 2.22      | 7.43 b        | 8.08 b        | 2.55    | 2.50   | 3.11 a   |
| Potassium fertilization (kg K$_2$O ha$^{-1}$) |        |          |         |           |               |               |         |        |           |
| 150                              | 3.23     | 17.83    | 2.81    | 2.11      | 8.40          | 8.90 a        | 2.52    | 2.26   | 2.76     |
| 250                              | 3.18     | 17.48    | 2.79    | 2.13      | 7.67          | 7.68 b        | 2.91    | 2.29   | 2.78     |
| 350                              | 3.37     | 18.29    | 2.95    | 2.23      | 7.62          | 8.64 ab       | 2.39    | 2.38   | 2.93     |
| Irrigation                       |         |          |         |           |               |               |         |        |           |
| Irrigated                        | 3.00 b  | 19.55 a  | 2.58 b  | 1.98 b    | 7.71          | 8.88 a        | 3.05 a  | 2.11 b | 2.52 b   |
| Not irrigated                    | 3.51 a  | 16.19 b  | 3.13 a  | 2.33 a    | 8.09          | 7.93 b        | 2.16 b  | 2.51 a | 3.12 a   |

Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.
Figure 3. The effect of potassium fertilization on the glutamic acid content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.

In the work of Eppendorfer [64], potassium deficiency induced the greatest decrease in the concentration of proline, followed by glutamic acid, but it increased histidine and aspartic acid levels. These findings were not corroborated by the present study where the synthesis of endogenous amino acids was not significantly affected by higher potassium rates (Table 5). The content of aspartic acid in Jerusalem artichoke tubers was a cultivar-dependent trait (Table 1). The highest accumulation of aspartic acid was noted in cv. Violette de Rennes (Table 5). According to Eppendorfer [64], as cited by Peksa et al. [65], the content of essential amino acids is inversely correlated with fertilizer rates. In this experiment, the applied rates of potassium fertilizer were not correlated with the synthesis of essential amino acids (isoleucine, leucine, lysine, methionine, cystine, phenylalanine, thyrosine, tryptophan, valine).
Irrigation decreased the content of most essential (isoleucine, leucine, lysine, phenylalanine, valine) and endogenous amino acids (alanine, glycine, histidine, serine, threonine) (Tables 4 and 5). A particularly high decline (25%) in valine concentration was noted in irrigated tubers cv. Topstar (Figure 5). Irrigation enhanced the concentrations of only selected endogenous amino acids (arginine, glutamic acid, proline) (Tables 4 and 5). In irrigated plots, an above-average increase was noted in arginine levels in tubers cv. Waldspindel and in glutamic acid and proline concentrations in cv. Violette de Rennes (Figures 6–8).

**Figure 5.** The effect of irrigation on the valine content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.

**Figure 6.** The effect of irrigation on the arginine content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.
Figure 7. The effect of irrigation on the glutamic acid content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.

Figure 8. The effect of irrigation on the proline content of tubers in the analyzed Jerusalem artichoke cultivars. Means with the same letters do not differ significantly at $p \leq 0.05$ in Tukey’s test. The absence of letters denotes non-significant main effects.

The essential amino acid index and the limiting amino acid index support evaluations of the nutritional quality of protein relative to chicken egg protein as the reference. New reference proteins which contain smaller amounts of amino acids (even twice smaller for methionine + cysteine, threonine, tryptophan, phenylalanine + thyrosine) and correspond to human nutritional requirements have been introduced in 2007 [66]. The nutritional value of the studied protein preparations was evaluated based on their chemical scores (SC). The chemical scores of cvs. Topstar and Waldspindel exceeded the reference value only for phenylalanine + thyrosine (by 29.8% and 18.2%, respectively), and leucine was the limiting amino acid (52.7% and 45.0%, respectively). In a study by Mitrus et al. [67], leucine was also the first limiting amino acid in *S. tuberosum* (36.5%). In Jerusalem artichokes cv. Violette de
Rennes, the content of all amino acids was below the reference value, from 1.6% (phenylalanine + tyrosine) to 21.8–59.2% (the remaining amino acids), and methionine + cysteine were the limiting amino acids (40.8%) (Table 6). Methionine + cysteine were also identified as the limiting amino acids in Jerusalem artichoke tubers analyzed by Danilcenko et al. [68] and Pęksa et al. [69]. In the work of Ciborowska and Rudnicka [70], the nutritional value of potatoes was limited by methionine + cysteine. According to Zhu et al. [71] and Pęksa et al. [65,69], the limiting amino acids in potato and Jerusalem artichoke tubers are determined mainly by cultivar and geographic location. Jerusalem artichokes are abundant in protein of high biological value. The species contains all essential amino acids in optimal proportions. Cieślak and Filipiak-Florkiewicz [63] reported higher levels of methionine in Jerusalem artichoke than in potato tubers. Contrary results were noted in this study, in particular in cv. Violette de Rennes. The nutritional value of protein evaluated based on the EAAI ranged from 66% (cv. Violette de Rennes) to 71–78% (cvs. Topstar and Waldspindel) (Table 6). Oser [48] and Sawicka [23] determined the EAAI of potatoes at 56-68%. The EAAI of other tuber crops of economic importance ranges from 54% (cassava, Manihot esculenta Crantz) to 82% (sweet potatoes, Ipomoea batatas L./Poir.) [48]. The EAAI of Jerusalem artichoke tubers exceeds the mean values for S. tuberosum and M. esculenta.

Table 6. Nutritional value of protein in tubers of the analyzed Jerusalem artichoke cultivars.

| Amino Acids (g 100 g⁻¹ of Protein) | cv. Topstar | cv. Waldspindel | cv. Violette de Rennes |
|-----------------------------------|-------------|-----------------|-----------------------|
|                                   | Mean CS EAAI| Mean CS EAAI    | Mean CS EAAI          |
| Isoleucine                        | 2.49 88.90  | 2.09 74.60      | 2.12 78.20            |
| Leucine                           | 3.48 52.70  | 2.97 45.00      | 2.99 45.30            |
| Lysine                            | 4.23 72.90  | 3.94 67.90      | 3.72 64.30            |
| Methionine + Cysteine             | 1.33 53.20  | 1.20 48.00      | 1.02 40.80 66.00     |
| Phenylalanine + Tyrosine          | 8.18 129.80 | 7.45 118.20     | 6.20 98.40            |
| Tryptophan                        | 0.68 61.80  | 0.82 74.50      | 0.67 60.90            |
| Valine                            | 2.98 85.10  | 2.56 73.10      | 2.59 74.00            |

Chicken egg albumin as the reference standard (WHO/FAO/UNU 2007) (g 100 g⁻¹ of protein): isoleucine-2.80; leucine-6.60; lysine-5.80; methionine + cysteine-2.50; phenylalanine + tyrosine-6.30; tryptophan-1.10; valine-3.50.

The chemical score calculated for different potassium fertilization rates exceeded the reference value for phenylalanine + tyrosine (by 12% at 250 kg K₂O ha⁻¹, by 14% at 150 kg K₂O ha⁻¹, and by 20% for 350 kg K₂O ha⁻¹) (Table 7). The limiting amino acid was leucine at 150 kg K₂O ha⁻¹ (47%), and methionine + cysteine at 250 and 350 kg K₂O ha⁻¹ (45% and 49%, respectively). The nutritional value of protein expressed by the EAAI relative to the reference protein ranged from 70 to 74% for the applied rates of potassium fertilizer (Table 7).

Table 7. The effect of potassium fertilization on the nutritional value of protein in Jerusalem artichoke tubers.

| Amino Acids (g 100 g⁻¹ of Protein) | 150 kg K₂O ha⁻¹ | 250 kg K₂O ha⁻¹ | 350 kg K₂O ha⁻¹ |
|-----------------------------------|-----------------|-----------------|-----------------|
|                                   | Mean CS EAAI    | Mean CS EAAI    | Mean CS EAAI    |
| Isoleucine                        | 2.20 78.57      | 2.22 79.28      | 2.28 81.43      |
| Leucine                           | 3.08 46.67      | 3.11 47.12      | 3.25 49.24      |
| Lysine                            | 3.99 68.79      | 3.86 66.55      | 4.06 70.00      |
| Methionine + Cysteine             | 1.21 48.40      | 1.12 48.80      | 1.22 48.80 74.00|
| Phenylalanine + Tyrosine          | 7.20 114.28     | 7.07 112.22     | 7.58 120.32     |
| Tryptophan                        | 0.71 64.54      | 0.72 65.45      | 0.75 68.18      |
| Valine                            | 2.70 77.14      | 2.65 75.71      | 2.77 79.14      |

Chicken egg albumin as the reference standard (WHO/FAO/UNU 2007) (g 100 g⁻¹ of protein): isoleucine-2.80; leucine-6.60; lysine-5.80; methionine + cysteine-2.50; phenylalanine + tyrosine-6.30; tryptophan-1.10; valine-3.50.
The CS of phenylalanine + thyrosine exceeded the reference value by 10% in irrigated plots and by 20% in the absence of irrigation (Table 8). The limiting amino acid was leucine regardless of the irrigation regime (44–52%). The nutritional value of protein expressed by the EAAI relative to the reference protein ranged from 67 to 76% in irrigated and non-irrigated tubers (Table 8).

Table 8. The effect of irrigation on the nutritional value of protein in Jerusalem artichoke tubers.

| Amino Acids (g 100 g⁻¹ of protein) | Irrigation | No Irrigation |
|----------------------------------|------------|--------------|
|                                  | Mean       | CS | EAAI | Mean | CS | EAAI |
| Isoleucine                       | 2.02       | 72.14 |       | 2.45 | 87.50 |       |
| Leucine                          | 2.88       | 43.64 |       | 3.41 | 51.67 |       |
| Lysine                           | 3.66       | 63.10 |       | 4.28 | 73.79 |       |
| Methionine + Cysteine            | 1.13       | 45.20 | 67.00 | 1.25 | 50.00 | 76.00 |
| Phenylalanine + Thyrosine        | 6.94       | 110.16 |       | 7.62 | 120.95 |       |
| Tryptophan                       | 0.71       | 64.54 |       | 0.74 | 67.27 |       |
| Valine                           | 2.47       | 70.57 |       | 2.95 | 84.28 |       |

Chicken egg albumin as the reference standard (WHO/FAO/UNU 2007) (g 100 g⁻¹ of protein): isoleucine-2.80; leucine-6.60; lysine-5.80; methionine + cysteine-2.50; phenylalanine + tyrosine-6.30; tryptophan-1.10; valine-3.50.

4. Conclusions

The present study confirmed that the genetic factor (cultivar) strongly affects the biological value of Jerusalem artichoke tubers. Topstar cultivar was most abundant in essential (isoleucine, leucine, phenylalanine, thyrosine, valine) and endogenous amino acids (alanine, glycine, threonine). High rates of potassium fertilizer (250 and 350 kg K₂O ha⁻¹) increased the absolute content of α-tocopherol and its proportion in total tocopherols, and decreased the content of glutamic acid in tubers. Irrigation increased α-tocopherol levels and decreased the concentrations of β-carotene and most essential and endogenous amino acids. The limiting amino acids in Jerusalem artichoke tubers were leucine (cv. Topstar and Waldspindel) and methionine + cysteine (cv. Violette de Rennes). Leucine was the limiting amino acid in tubers supplied with low potassium rates (150 kg K₂O ha⁻¹), whereas in plots with moderate and high potassium rates (250 and 350 kg K₂O ha⁻¹, respectively), the limiting amino acids were methionine + cysteine. Jerusalem artichoke tubers were characterized by very high nutritional value of protein, expressed by the EAAI.

Author Contributions: Conceptualization, methodology, B.B., formal analysis, writing—original draft preparation, B.B. and K.J. All authors have read and agreed to the published version of the manuscript.

Funding: Project financially co-supported by Minister of Science and Higher Education in the range of the program entitled “Regional Initiative of Excellence” for the years 2019–2022, Project No. 010/RID/2018/19, amount of funding 12.000.000 PLN.”

Acknowledgments: The results presented in this paper were obtained as part of a comprehensive study financed by the University of Warmia and Mazury in Olsztyn (grant No. 20.610.020-110).

Conflicts of Interest: The authors declare no conflict of interest.

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