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Influence of the storage duration on the health promoting tyrosine, tryptophan, and total phenolics in potato tubers

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Abstract: This research investigated the influence of the storage duration on the health promoting tyrosine, tryptophan and total phenolics in potato tubers. In the course of storage, the total amount of accumulated compounds in the dry mass of organically grown potato tubers increases. This is determined by individual properties of potato variety, storage time, and interaction of these two factors ($P < 0.05$). Organic potato tubers show increased total phenolic compounds in their dry matter during storage. This is due to a variety of characteristics, storage time and the interaction of these two factors ($P < 0.05$). A more pronounced increase in total phenolic compounds during storage was observed in 2022 than in 2021.

Keywords: accumulated compounds; organic farming; *Solanum tuberosum* L.; storage

Potatoes are the most important non-grained crop in the world. According to the Food and Agriculture Organization of the United Nations (FAO), potatoes rank fourth after corn, rice, and wheat in terms of world production volume in tons (FAO 2011). Potatoes deserved such great popularity for their indiscriminateness to growing conditions and high yields and their indisputable nutritional value. Potato tubers are especially useful for human health due to the vitamins and minerals found in them and, of course, the accumulation of significant amounts of phenolic compounds. Chun et al. (2005), after studying the contribution of 34 fruits and vegetables to the intake of phenolic compounds in the American diet, found that potatoes are the third most important source of phenolic compounds in the daily diet after apples and oranges. Since phenolic compounds have strong antioxidant properties, the contribution of potatoes to human health is incredibly significant.

Recently, much attention has been paid to food safety and quality in Lithuania and around the world. Fruits and vegetables grown in a clean and safe environment (without mineral fertilisers and chemical plant protection products, without antibiotics, growth hormones, or genetically modified organisms) are more valued by supporters of healthy food than farms developing intensive agriculture.

The most important indicators are the number of phenolic compounds accumulated and their antioxidant activity. Potatoes' main substances with antioxidant activity are phenolic acids, flavonoids, ascorbic acid, and carotenoids (Vaitkeviciene et al. 2020). The data on the influence of agricultural systems on the number of substances with antioxidant activity accumulated by potatoes are very contradictory. Hajslová et al. (2005) and Hamouz et al. (2005) have the same views. Warman and Havard (1998) and Rembialkowska (1999) argue that there is no difference in vitamin C content in potatoes grown under different conditions. In an article that appeared in 2008, Rosenthal and Jansky (2008) also explained that there is no difference in the antioxidant activity of potato tubers grown under ecological and intensive farming conditions. Thus, there is no unanimous opinion

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about the influence of the agricultural system on the amount of antioxidants stored in potatoes and their antioxidant activity. Still, it is known that the accumulation of phenylpropanoids in potato tubers is stimulated by various abiotic and biotic factors (Torres-Contreras and Jacobo-Velázquez 2021), so it is more likely that phenylpropanoids (chlorogenic acid, ferulic acid, etc.) with antioxidant activity will be more accumulated by organically grown potato tubers since without the use of chemical ones, protective measures are more strongly influenced by such factors that stimulate the synthesis of these compounds as pests and diseases.

It is important not only to know how many and what antioxidants potato tubers accumulate and what their antioxidant activity is but also how these parameters change during the storage of potatoes. Rosenthal and Jansky (2008) also found that stored potato tubers exhibit higher antioxidant activity than fresh ones.

Tyrosine is a substitute amino acid. In other words, the human body can produce it on its own. Tyrosine is synthesised from the essential amino acid phenylalanine with the participation of the enzyme phenylalanine hydroxylase. When the function of this enzyme is impaired, the body can no longer produce tyrosine on its own, phenylketonuria develops (a serious disease manifested by disorders of psychomotor and mental development), and food becomes its only source (van Spronsen et al. 2001).

Under the influence of tryptophan hydroxylase, it is translated into 5-hydroxytryptophan, which, in the course of further reactions, turns into serotonin (a hormone that regulates neurotransmitters, appetite, mood, etc.) and melatonin (a hormone that regulates circadian rhythm) (Wang et al. 2002). During the next metabolic pathway, tryptophan is converted to nicotinic acid (vitamin B3) (Heijnen et al. 2013). From it, nicotinamide adenine dinucleotide (NAD) is synthesised during further enzymatic reactions and is involved as a coenzyme in oxidation-reduction reactions (Revollo et al. 2004). Tryptophan is essential for normal protein synthesis, growth, mood, behaviour, and immune response (Klaessens et al. 2022).

Tyrosine and tryptophan are also important by themselves as antioxidants. Although they are not specific binders of radicals, their high concentrations in the body significantly contribute to the fight against free radicals (Brazinskiene et al. 2017).

The aim of the study is to evaluate the effect of storage time on the levels of tyrosine and tryptophan and total phenolic compounds accumulated in organically grown potato tubers.

MATERIAL AND METHODS

Plant material. Potato (*Solanum tuberosum* L.) tubers of several varieties with different times of maturity: very early – 'VB Venta', 'Fresco', 'Acapella'; early – 'Sante', 'Goda', 'VB Liepa'; medium – 'Lady Rosetta', 'Red Lady', 'Courage'; and late – 'Saturna', 'VB Rasa', 'VB Aista' (Table 1).

Table 1. Pedigree information of potato varieties

Potato varieties	Characters				
	maturity	country	national list	tuber: the colour of skin	tuber: the colour of flesh
VB Venta	very early	LT	2009	yellow	light yellow
Fresco		NL	1982	yellow	light yellow
Acapella		D	2000	yellow	light yellow
Sante	early	NL	1981	yellow	light yellow
Goda		LT	2004	yellow	light yellow
VB Liepa		LT	2016	yellow	light yellow
Lady Rosetta	medium	NL	1988	red	light yellow
Red Lady		D	2004	red	yellow
Courage		NL	1998	red	light yellow
Saturna	late	NL	1964	yellow	light yellow
VB Rasa		LT	2006	red	yellow
VB Aista		LT	2006	yellow	white

LT – Lithuania; NL – Netherlands; D – Germany

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Organic farming conditions. According to the FAO and United Nations Educational, Scientific and Cultural Organization (UNESCO) classification of Haplic Luvisol (LVh), potatoes were grown in 2021–2022 in Lithuania at the Voke Branch of the Lithuanian Research Centre for Agriculture and Forestry in breeding plots with sandy loam soil on carbonated fluvioglacial eluvified gravel (JDp): The soil's pH potassium chloride (KCl) value was 5.9, the amount of accessible phosphorus (P_2O_5) was 230 mg·kg^{-1}, and the amount of available potassium (K_2O) was 310 mg·kg^{-1}. In 2021, the potatoes were planted on May 17 and harvested on September 19; in 2022, they were planted on May 11 and harvested on September 10.

The potatoes are organically grown. In the potato variety trials, the plot length was 10.5 m, width 1.4 m and area 14.7 m^2. Test variations in repetitions are arranged in a randomised way in 3 repetitions. 60 tubers are planted in each plot. The soil was deeply tilled in the autumn. The field was worked twice in the spring before being cultivated with a rotary cultivator to a depth of 0.25 m. Before sowing the potatoes, the field was furrowed. The potatoes were planted by hand. After planting, the potato rows are loosened twice every 7 days with rotary hoes. In the case of organic farming, 60 kg·ha^{-1} of nitrogen (Provita, Germany), 60 kg·ha^{-1} of phosphorus (phosphorite powder), and 90 kg·ha^{-1} of potassium (Patentkali – potassium and magnesium) were inserted at planting time (Burakova et al. 2020). Potatoes were twice harrowed after being earthed up. Following planting, a rotary hiller was used to climb the interrows twice (every 7 days). Mechanical means were applied to combat weeds and Colorado beetle larvae (*Leptinotarsa decemlineata*) in the organic potato crop.

Preparation of samples for analysis. Each potato variety's storage was emptied of its tubers, and five were randomly picked. The potato tuber was washed and air-dried before being sliced into 4 mm thick slices and quickly frozen at –35 °C in a freezer with air circulation. The lyophilisation process took 3.0–3.5 days. In a dryer (FD8512S lyophilyser; Ilshin Freze, Netherlands), the frozen potato slices were lyophilised at 5 mbar pressure (condenser temperature –85 °C). The lyophilised potato was ground into powder using a grinder (Grindomix GM 200; Retsch, Germany). 1 g of the acquired power was put into an analytical flask and covered with acetic acid, methanol, and water (2 : 39 : 59; *v*/*v*/*v*) to make 10 mL of the analytical sample. After that, the mixture was placed for 20 min in an ultrasonic cleaner (Biosonic UC100; Coltene/Whaledent, USA). The resulting potato extract was filtered through paper (Albet 400; Albet LabSience, Germany) and a syringe filter with a nylon membrane with a 0.22 m pore size (Rotilabo; Carl Roth GmbH, Germany) before passing through. This kind. of the extract was examined. For each potato, three extracts were made.

High-performance liquid chromatography ultraviolet-visible (HPLC UV-VIS) analysis. A chromatograph (Waters 2695, Waters, USA) was used for the analysis. Advanced Chromatographic Technologies, Scotland, provided the 4.6 × 250 mm, 5 µm ACE C18 250 mm column to separate active chemicals. Throughout the study, the column was kept at an external Waters temperature control module, which maintained a temperature of 25 °C. Ten micrometres of the test solution were injected during the analysis. The flow rate for the mobile phase was 1 mL per 1 min. The gradient system employed was as follows: solvent A was 0.5% acetic acid in water and solvent B was methanol. The time intervals between each solvent were as follows: 0 min (95% A and 5% B), 40 min (40% A and 60% B), 41 min (10% A and 90% B), 55 min (10% A and 90% B), and 56 min (95% A and 5% B). Using a photodiode array detector (Waters 996 PDA; Waters, USA) tyrosine and tryptophan at wavelengths ensured their maximal absorption. The Waters Millennium 2000 chromatographic manager system (version 2000) was used to gather and analyse the data.

Statistical analysis. The analysis of variance (ANOVA) method was used to conduct the statistical analysis. Multiple comparisons should be used if averages show statistically significant differences. It allowed for a certain variety's statistically significant distinct averages and attributes to be determined at the 0.05 significance level.

RESULTS AND DISCUSSION

Previous studies on the phytochemical analysis of tubers of *S. tuberosum* show that potatoes accumulate a wide range of biologically active compounds during storage. Still, one of the more important ones is the substances belonging to the two classes of phenolic compounds, phenolic acids and flavonoids. As shown in Table 2, climatic conditions in Lithuania varied between 2021 and 2022, especially in June. June 2021 was much warmer (average temperature 2.6 °C above the long-term average), with almost twice the normal rainfall. By contrast, June 2022 was almost a degree cooler and had one-third more rainfall than the long-term average. May 2022 was drier and warmer

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Table 2. Average monthly daily temperature and amount of precipitation

Month	Year		Multiannual average
	2021	2022	
temperature (°C)			
May	12.9	13.8	12.5
June	18.3	14.8	15.7
July	19.6	19.5	16.9
August	17.3	16.5	16.3
precipitation (mm)			
May	79	38	60
June	41	99	77
July	155	81	78
August	101	83	68

than the long-term average and 2021, while 2021 was wetter and slightly warmer than the long-term average. July was about 2.5 °C warmer than the long-term average in both years of the study. Precipitation was also higher in both cases, but in 2021, it was almost twice as high as the long-term average and in 2022. Autumn 2021 was warmer and wetter than in 2012 and the long-term average. Thus, 2021 was warmer overall. June was notable for its low rainfall and July for its high rainfall compared to 2022 the long-term average.

Tryptophan is an essential amino acid and can only be obtained from food. A daily intake of 500–700 mg of tryptophan is recommended. Tryptophan is essential for normal protein synthesis, growth, mood, behaviour and immune response (Wang et al. 2002). The content of free tryptophan in potato tubers was not significantly affected by climatic conditions during the entire storage of the tubers ($P > 0.05$). Figure 1 shows the average tryptophan content for all the measurements performed. Figure 1 shows that during the six months of storage, the tryptophan content in tubers increased by 2–122%, depending on the variety. During the storage period, tryptophan levels increased the most in 'Courage' tubers (on average 23.5 μg^{-1}), and the lowest in 'Goda' tubers (on average –7.1 μg^{-1}). The vegetation time of the potatoes did not significantly affect the increase in tyrosine levels ($P > 0.05$).

Thus, it can be concluded that the content of this amino acid in potato tubers depends on the variety ($P < 0.5$) and increases during storage ($P < 0.05$).

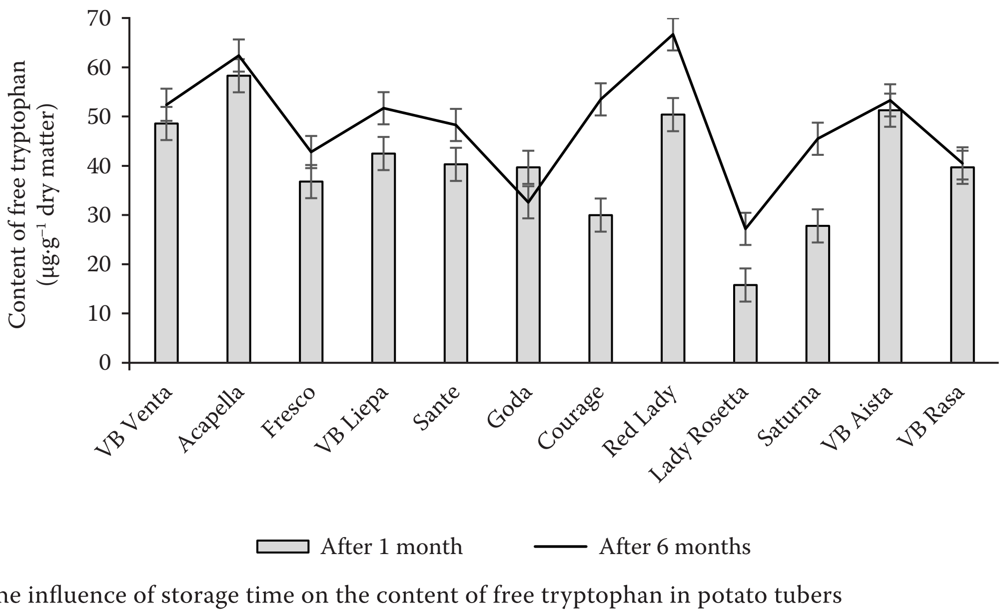

Figure 1. The influence of storage time on the content of free tryptophan in potato tubers

LSD – Least Significant Difference; $LSD_{0.05(storage)} = 2.52$, $LSD_{0.05(variety)} = 6.16$, $LSD_{0.05(storage \times variety)} = 8.72$

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The enzyme phenylalanine hydroxylase synthesises Tyrosine from the essential amino acid phenylalanine. When this enzyme becomes dysfunctional, the body can no longer make tyrosine on its own, and phenylketonuria (a severe disorder of psychomotor and mental development) develops. The body is left with the sole source of tyrosine in its food (van Spronsen et al. 2001). The amount of free tyrosine in potato tubers increased significantly over 6 months of storage (Figure 2) (P < 0.05). However, as in the case of tryptophan, this was not influenced by the climatic conditions in which the potatoes grew (P > 0.05), so Figure 2 gives the mean values for all measurements.

During the storage period, tyrosine levels increased the most in 'Red Lady' tubers (on average by 145.3 μg^{-1}), and the lowest in 'Goda' tubers (on average by 35.4 μg^{-1}). The vegetation time of the potatoes did not significantly affect the increase in tyrosine levels (P > 0.05).

The amount of tyrosine, like tryptophan, increases during storage (P > 0.05) and depends on the individual characteristics of the variety (P > 0.05).

The total phenolic compounds accumulated by the organic potato varieties in 2021 and 2022 after one month of storage were significantly related to the varietal traits (P > 0.05) and after 5 months. Among the organic potatoes, in 2021 after one month of storage, the highest total phenolic compounds were found in 'VB Rasa' and the lowest in 'Fresco' tubers. After 5 months of storage, the most phenolic compounds were found in the organic 'Saturna' and the lowest in 'Sante' tubers. There was a significant effect of storage time on the phenolic content of potato tubers (P < 0.05). During 5 months of storage in 2021, the phenolic content of organically grown potatoes increased from 4% to 91%, depending on the variety, in the tubers of all (except 'Goda' and 'VB Aista') varieties tested (Figure 3). Various environmental stresses, such as pathogens, promote the synthesis of phenylpropanoids in plants. Phenolic compounds have a protective function during drought. Phenolic compounds are very important for plants under stress because of their ability to directly neutralise reactive oxygen species and to stop the chain reaction of lipid peroxidation. With the further storage of potatoes, climatic conditions, potato genotypes, and interactions affected the content of phenolic compounds (P > 0.05). Of the organic potatoes grown in 2022 after one month of storage, the highest total phenolic compounds were found in 'VB Rasa' and the lowest in 'Lady Rosetta' tubers. After 5 months of storage, the highest amount of phenolic compounds was found in the organic 'VB Rasa' and the lowest in the 'Fresco' tubers. Storage time had a significant effect on the phenolic content of potato tubers (P > 0.05). In 2022, during 5 months of storage, the phenolic content of organic potatoes increased from 23% to 170%, depending on the variety. The total phenolic content of organic potato tubers (dry matter) increases during storage. This is due to cultivar characteristics, storage time and the interaction between the two (P > 0.05). A more pronounced increase in total phenolic compounds during storage was observed in 2022 than in 2021.

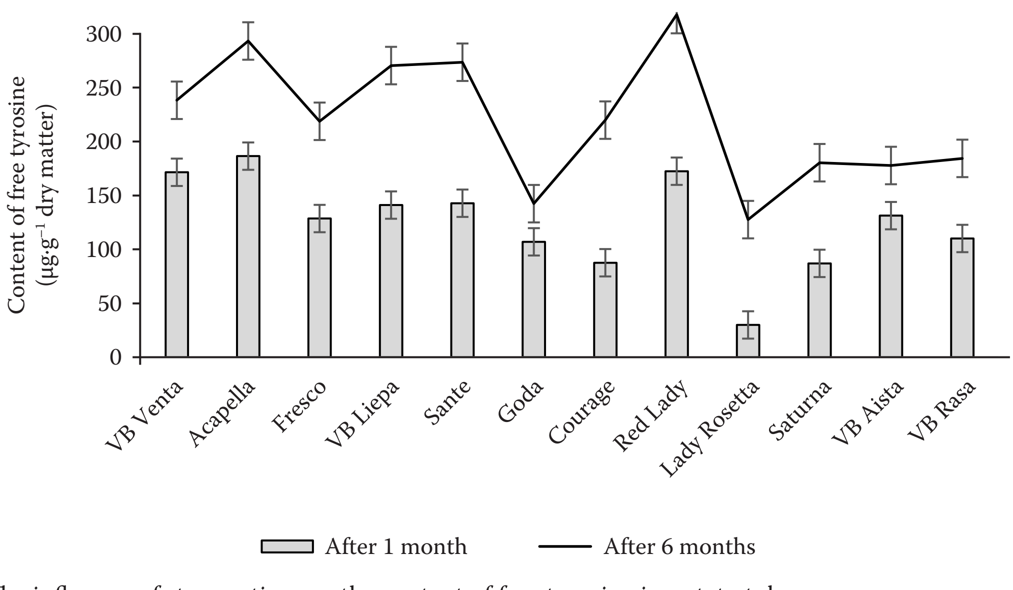

Figure 2. The influence of storage time on the content of free tyrosine in potato tubers

LSD – Least Significant Difference; $LSD_{0.05(storage)}$ = 6.99, $LSD_{0.05(variety)}$ = 16.32, $LSD_{0.05(storage \times variety)}$ = 23.06

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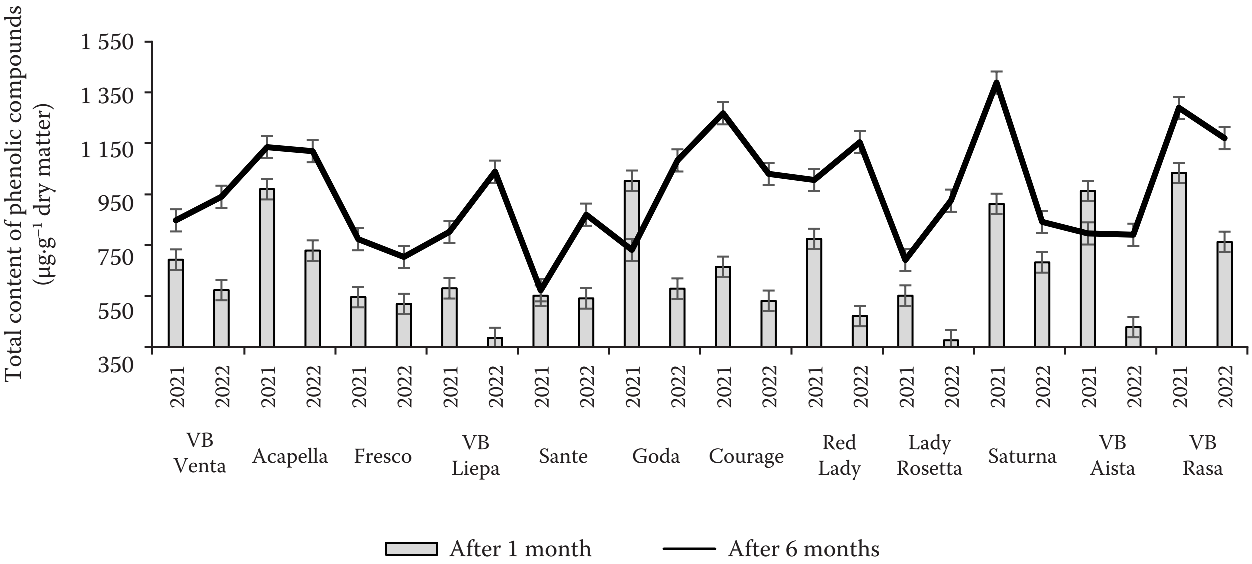

Figure 3. Influence of storage time on the total content of phenolic compounds in potato tubers

LSD – Least Significant Difference; 2021: $LSD_{0.05(storage)}$ = 41.63, $LSD_{0.05(variety)}$ = 101.97, $LSD_{0.05(storage \times variety)}$ = 128.99; 2022: $LSD_{0.05(storage)}$ = 41.90, $LSD_{0.05(variety)}$ = 102.64, $LSD_{0.05(storage \times variety)}$ = 145.154

During storage, the content of phenolic compounds in potato tubers increases ($P < 0.05$). The same conclusions that the number of phenolic compounds increases during storage were drawn by Lewis et al. (1999) and Blessington et al. (2010).

CONCLUSION

According to the present results, the overall concentration of tryptophan and tyrosine chemicals found in potato tubers during storage rises in the dry bulk of organically cultivated potato tubers. The specific characteristics of the potato type, the length of storage, and the interaction between these two parameters all play a role in this ($P < 0.05$). During storage, organic potato tubers have increased total phenolic compounds in their dry matter. This is due to cultivar characteristics, storage duration and the interaction of these two factors ($P < 0.05$). A more pronounced increase in total phenolic compounds during storage was observed in 2022 than in 2021. The study's results verified that the chemical changes that occurred at tubers in the long storage depend on potato cultivars.

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