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The Influence of Different Substrates on the Growth, Yield and Quality of Slovenian Sweetpotato Cultivars under Greenhouse Conditions

Dragan Žnidarčič, Filip Vučanjk, Žarko M. Ilin, Barbara Pipan, Vladimir Meglič and Lovro Sinkovič

Additional information is available at the end of the chapter

Abstract

A greenhouse experiment was conducted to evaluate the genetic relatedness between three Slovenian sweetpotato cultivars; and to assess the effects of different growing substrates on selected agronomic and nutritional traits. Tubers of three cultivars ('Lučka', 'Janja' and 'Martina') with different skin/flesh color were produced in planters under glasshouse conditions in five different growing substrates (perlite, peat, expanded clay, vermiculite and garden soil) from prior raised seedlings. Genetic analysis was performed using a set of eight SSR markers. According to Nei's genetic distance and pairwise population Fst analysis, the most related cultivars are 'Janja' and 'Martina'. The following agronomic traits were evaluated: vine length, thickness of vine-base, number of branches, weight of above ground part, number of leaves plant^{-1}, number of tubers plant^{-1} and tubers weight plant^{-1}. Among nutritional traits, total phenolic content (TPC), antioxidant potential (AOP) and ascorbic acid content (AA) were determined. Significant interactions of growing substrates (factor A) × cultivar (factor B) were observed for thickness of vine-base, weight of above ground part, AOP, TPC and AA. Overall results show different response of cultivars in different growing substrate. Growing substrate provide a discriminant classification of the sweetpotato cultivars according to their agronomic and nutritional traits.

Keywords: growing substrates, genetic analysis, *Ipomoea batatas*, phenolic compounds

1. Introduction

The sweetpotato (*Ipomoea batatas* Lam.) is an herbaceous dicotyledonous perennial plant grown primarily as a root crop. In systematic plant taxonomy, the sweetpotato is assigned

to the family Convolvulaceae Juss., which comprises 55 genera [1]. To distinguish the sweetpotato from the tuberous potato (*Solanum tuberosum* L.), the internationally accepted convention for the common English name is now the one word spelling 'sweetpotato' [2]. Although sweetpotato shoot tips and leaves may be eaten, the swollen root is the main part used for human consumption.

Ranked by current world production, sweetpotato is the 7th major crop, which serves as an energy and phytochemical source of nutrition in more than 100 countries [3]. The origin of sweetpotato is Central America, but at present it is widely cultivated in the tropics and subtropics, and even in some temperate areas at different ecological regions [4]. The main sweetpotato production regions by area include Asia (78.4%), Africa (17.1%), North America (1.8%), South America (1.3%) and Oceania (1.2%). In Europe, where the total production of sweetpotato accounted 45.901 t in 2016, the biggest producers are Portugal (22.591 t), Spain (13.550 t), Italy (6.723 t) and Greece (3.038 t) [5]. In Slovenia, sweetpotato has been quite unknown crop until recently, both for production and human consumption. The environment diversity and specific climatic conditions of this region could enable successful production of that crop in the future [6, 7]. Three new Slovenian sweetpotato cultivars ('Lučka', 'Janja' and 'Martina') were registered in 2015 and are now added to national list of varieties [8]. According of Yamakawa and Yoshimoto [9] of particular importance is the development of novel cultivars with roots contributing to the human diet, both as basic food stuff but with added physiological functions such as antioxidant or specific nutritional traits.

Tubers of sweetpotato are rich in dietary fiber, minerals, vitamins and antioxidants, such as phenolic compounds [10–12]. Besides acting as antioxidants, phenolic compounds and carotenoids also provide sweetpotatoes with their distinctive flesh/skin colors (cream, deep yellow, orange and purple) [13]. Contribution of sweetpotato toward health is acknowledged due to high nutrient content and its anti-carcinogenic and cardiovascular disease preventing properties [4, 14]. In recent years, several reports have indicated that the phytochemicals from sweetpotatoes displayed antioxidative or radical scavenging activity with health-promoting functions [15, 16]. Phenolic acids (i.e., chlorogenic and dicaffeoylquinic acids) contribute to antioxidant activity and other health beneficial properties of color fleshed genotypes [17, 18]. Additionally, cultivars with the same flesh color may differ in total phenolic content, individual phenolic acid profile and antioxidant activity.

Sweetpotato readily produces adventitious roots and has trailing vines, therefore can colonize marginal soils and is not very demanding as regards soil type [19].

The most innovative technology of plant cultivation in greenhouse conditions is growing in mineral substrates. The origin of substrates is different and they also differ in their physical, chemical and biological properties. According to Kacjan Maršić and Jakše [20] peat, perlite, expanded clay and vermiculite are an efficient growing media in the European market. Peat consists of partially decomposed aquatic, marsh, bog or swamp vegetation. The main advantages of peat lie in its physical properties, which allow an adequate water/air ratio in the root zone, and a high cation exchange capacity able to adequately provide nutrient for plant growth and development [21]. Perlite is a substance made from volcanic rock and often used as a soil additive to increase aeration and draining of the soil. It is also

relatively inexpensive. The biggest drawback to perlite is that it does not retain water very well. Spain is the pioneer among the Mediterranean countries in the commercial use of perlite, mainly for vegetable production [22]. There are several studies on possibilities of using perlite as a substrate and it has been reported that the average yield of fruit-bearing crops cultivated in perlite achieved 2–3 times higher yield than plants grown in soil [23]. It has been also reported that growers in the Mediterranean region prefer perlite to other substrates because it is easily available from local suppliers, it is cheap and can be used for at least three, instead of 2 years, which is the common maximum durability of most other substrates. Expanded clay pellets are made by baking clay in a kiln. Clay pellets are full of tiny air pockets, which give them good drainage. They are best for systems that have frequent watering. Because expanded clay pellets do not have good water-holding capacity, salt accumulation and drying out can be common problems in improperly managed systems. Although the pellets are rather expensive, they are one of the few kinds of medium that can be easily reused [24]. Vermiculite is a micaceous mineral which is expanded when heated in furnaces at temperatures near 109°C. Chemically, it is a hydrated magnesium-aluminum-iron silicate. When expanded, it is very light in weight (96–160 kg/m^3) and neutral in reaction with good buffering properties. It is able to absorb large quantities of water (0.4–0.5 m/cm^3). It has a relatively high cation exchange capacity and thus can hold nutrients in reserve and later release them. It contains some magnesium and potassium which is available to plants [25].

The required physical and chemical characteristics of growth substrates vary notably with crop species and its management, and substrate choice can be influenced by environmental and economic considerations [26]. Afterward, growing substrates are easier to handle and it may provide better growing environment compared to soil culture [27, 28]. To the best of our knowledge, there is no scientific literature regarding cultivation of sweetpotato in different growing substrates.

Application of short sequence repeats (SSR) markers in genetic diversity studies of different agro-economically important species represents informative, effective and reliable marker system [29–34] for distinguishing between different genetic resources. For sweetpotato, which is a hexaploid (2n = 6x = 90) plant species with an out-crossing mating system [35], SSR marker system is highly applicable due its codominant nature [32].

The objective of the study was to analyze the genetic relatedness between three Slovenian sweetpotato cultivars, to examine the effect of different growing substrates on selected agronomic and nutritional traits of these cultivars and to compare responses between cultivars.

2. Materials and methods

2.1. Experimental setup

The experiment was carried out at the Glasshouse experimental station (46°04′N, 14°31′W; altitude 310 m a.s.l.) of the Biotechnical Faculty in Ljubljana, Slovenia. Three new Slovenian

cultivars of sweetpotato (*Ipomoea batatas* L.) were studied: 'Lučka' with orange skin and flesh color, 'Janja' with white skin and flesh color and 'Martina' purple skin and white flesh color (**Figures 1** and **2**). **Table 1** shows the main characteristics of cultivars.

Cuttings and seedlings were grown in styrofoam seed starting trays filled with substrate for seedlings Neuhaus N3 (Humko, Slovenia) and covered with vermiculite.

Polypropylene troughs (Mapal Plastic Agricultural Products Division, Israel) were placed on parallel beds. Each of three troughs—blocks (18 m length, 0.5 m width and 0.2 m height) was divided to plots, separated with polystyrene dams to avoid stirring and filled with growing media. In each plot two seedlings of individual cultivar were planted. The randomized complete block design (RCBD) was split plot with growing media applied to whole plots and cultivars applied to split plots. The experiment was designed to test two factors: different growing substrates (factor A; perlite, peat, expanded clay, vermiculite and garden soil) and different sweetpotato cultivars (factor B).

After the initial watering of the substrate and seedlings, T-tape tubes (T-Tape® TSX 500 Model) were placed over the growing substrate. Basic fertilization was performed with water soluble NPK fertilizer Entec Perfect (14-7-17, EuroChem Agro, Germany; 350 kg ha^{-1}) during planting of seedlings in growing substrate in the beginning of June. Two weeks after transplantation and throughout the growing period, the plants were fertilized three times per week with

Figure 1. Cv. 'Janja' (left) and cv. 'Lučka' (right) (photo: D. Žnidarčič).

Figure 2. Cv. 'Martina' (photo: D. Žnidarčič).

Characteristics	Cultivar					
	'Lučka'		'Janja'		'Martina'	
	State of expression	Note	State of expression	Note	State of expression	Note
Plant growth habit	Spreading	5	Spreading	5	Spreading	5
Length of primary shoots	Medium	5	Short	3	Short	3
Length of internode	Medium	5	Medium	5	Medium	5
Diameter of internode	Medium	5	Medium	5	Very large	9
Anthocyanin coloration of internode	Absent or week	1	Absent or week	1	Absent or week	1
Anthocyanin coloration of tip	Medium	2	Absent or week	1	Absent or week	1
Anthocyanin coloration of node	Medium	2	Absent or week	1	Absent or week	1
Pubescens of tip	Absent or sparse	1	Absent or sparse	1	Dense	3
Leaf blade: lobes	Absent	1	Three lobes	2	Absent	1
Leaf blade: shape	Triangular	2	Triangular	2	Triangular	2
Leaf blade: depth of lobing	—	—	Very shallow	1	—	—
Leaf blade: color	Green	2	Green	2	Green	2
Leaf blade: anthocyanin coloration of upper side	Absent or week	1	Absent or week	1	Absent or week	1
Leaf blade: extent of anthocyanin coloration on abaxial veins	Small	3	Absent or very small	1	Absent or very small	1
Leaf blade: intensity of anthocyanin coloration on abaxial veins	Weak	3	Very weak	1	Very weak	1
Young leaf blade: main color on upper side	Purplish brown	7	Medium green	3	Medium green	3
Petiole: anthocyanin coloration	Absent or very week	1	Absent or very week	1	Absent or very week	1
Petiole: length	Short	3	Short	3	Short	3
Storage root: shape	Ovate	1	Ovate	1	Ovate	1
Storage root: ratio length/width	Medium	5	Moderately elongated	7	Medium	5
Storage root: thickness of cortex relative to overall diameter	Thick	7	Thick	7	Medium	5
Storage root: main color of skin	Brownish orange	5	Light beige	2	Light purple	9
Storage root: secondary color of skin	Brown	8	Pink	5	Pink	5
Storage root: main color of flesh	Orange	4	Beige	2	Beige	2

Characteristics	Cultivar					
	'Lučka'		'Janja'		'Martina'	
	State of expression	Note	State of expression	Note	State of expression	Note
Storage root: intensity main color of flesh	Medium	2	Light	1	Light	1
Storage root: secondary color of flesh	Yellow	3	Yellow	3	White	1
Storage root: depth of eyes	Shallow	1	Shallow	1	Shallow	1

Table 1. Characteristics of sweetpotato cultivars (included in the UPOV test guidelines, CPVO technical protocol or reporting authority's test guidelines).

nutrient solution prepared with tap water-containing water soluble NPK fertilizer Polifid (16-8-32, Haifa, Israel; 1 g L^{-1}). During the growth period the following measures were implemented: removing weeds, monitoring the functioning of the irrigation system, cleaning dead plant parts and monitoring the presence of pests and diseases.

At harvest, after 128 days growing period, the following agronomic traits (growth and yield parameters) were evaluated for individual cultivar and growing substrate: vine length (cm), thickness of vine-base (mm), number of branches, weight of above ground part (g), number of leaves (plant^{-1}), number of tubers (plant^{-1}) and tubers weight (kg plant^{-1}). The proximate analysis of the tubers was also assessed. For the analysis of total phenolic content (TPC), antioxidant potential (AOP) and ascorbic acid content (AA), random tubers of each cultivar and growing substrate were used. For the sample extraction, 8 g of fresh tuber slices (flesh and skin) were mixed with 10 g of 2% metaphosphoric acid dissolved in distilled water. The tissue was homogenized using an Ultraturax T 25 (20,500 rpm). Homogenized samples were centrifuged and filtered through a 0.45 μm filters (17 mm syringe filter CA). The extracts were stored at −80°C until analyzed.

2.2. Genetic analysis

Genomic DNA was extracted from frozen leaves of six different plants collected individually from each of three cultivars grown in garden soil. BioSprint 15 DNA Plant Kit (Qiagen, Germany) and MagMax (Applied Biosystems, USA) nucleic acids isolation robot, following the modified method from manufacturer's instructions, were used. Dilutions of 1 ng μL^{-1} of DNA were used for PCR amplification. Eight primer pairs: Ib-316, Ib-318, Ib-242, Ib-248, Ib-255F1, Ib-255, Ib-286 and Ib-297 [35, 36] were applied for SSR assessment. PCR reactions were performed in a final volume of 11 μL, containing 1 ng of genomic DNA and following reagents with starting concentrations of: 10× PCR buffer (Biotools, Spain), 10 mM of each dNTP's, 50 mM MgCl$_2$ (Biotools, Spain), 10 μM of each primer, 10 μM 5' fluorescently labeled universal primer (6-FAM, NED and HEX) and 0.5 U of Taq DNA polymerase (Biotools, Spain). The forward primer of each SSR was appended with 18 bp tail sequence 5'-TGTAAAACGACGGCCAGT-3' (M13(−21)) as described by Schuelke [37]. PCR analyses were performed on ABI 9700 (Applied Biosystems, USA) under the following 'touch-down' conditions: 94°C for 4 min, 30 cycles at

94°C for 1 min, auto increment temperature from 49.5°C for 0.5°C per cycle for 30 s, 72°C for 1 min, followed by 30 cycles at 94°C for 30 s, auto increment temperature from 49.5°C for 0.5°C per cycle for 30 s, 72°C for 1 min and final extension for 5 min at 72°C. Fragment analysis was performed on 3130XL Genetic Analyzer (Applied Biosystems, USA), the allele lengths were determined by comparison with size standard GeneScan-350 ROX (Applied Biosystems, USA) using GeneMapper 4.0 (Applied Biosystems, USA). Parameters of genetic diversity among loci and varieties, including number of migrants (Nm), inbreeding coefficients (Fst), % of polymorphic loci, numbers of effective alleles, total expected heterozygosities (Ht), Shannon's information index, pairwise Nei's genetic correlations, pairwise population Fst analysis, analysis of molecular variance via R-statistics under 999 permutations (AMOVA) and principal coordinate analysis (PCoA) were conducted applying GenAlEx v.6.4 [38].

2.3. Analysis of nutritional traits

Analyses of bioactive compounds included evaluation of TPC, AOP and AA in tubers of sweetpotato. The TPC was determined using the Folin-Ciocalteu method, as described by Singleton and Rossi, and slightly modified [39]. Gallic acid (Merck, Germany) was used for six point calibration curve, which ranged from 3 to 150 mg L^{-1} (R^2 = 0.9998). The results were expressed as gallic acid equivalents (mg GAE 100 g^{-1} FW; fresh weight). The AOP was evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl; Sigma-Aldrich, Saint Louis, MO, USA) free radical scavenging method [40]. Trolox (220 mg L^{-1}; Sigma-Aldrich, Saint Louis, MO, USA) was used for six point calibration curve, which ranged from 40 to 220 mg L^{-1} (R^2 = 0.9900). The results were expressed as Trolox equivalents (mg TE g^{-1} FW). AA analysis was performed on an HPLC system (Agilent 1260; Agilent Technologies) using a diode array detector, with the wavelength set at 254 nm. The determination of AA was carried out on a 100 × 2 mm i.d., 3 μm Scherzo SM-C18 column (Imtakt, Japan), at a flow rate of 0.3 mL min^{-1}. The mobile phase consisted of water (A) and acetonitrile (B), both of which contained 0.3% formic acid. The following elution gradient was used for solvent B: 0–3 min, 0–10%; 3–4 min, 10–100% and 4–6 min, 100%. The temperature of the column was maintained at 30°C, while the temperature of the automatic sample feeder was set at 4°C. AA was calculated using an external standard method and expressed as mg 100 g^{-1} FW.

2.4. Statistical analyses

Statistical analyses were performed using the Centurion Statgraphics XVI statistical analysis program. Prior statistical analyses data was tested for normal distribution using Shapiro-Wilk test. If the data were not normally distributed, log transformation was used prior further analysis. For easier interpretation the **Tables 3** and **4** show the untransformed data. Multifactorial ANOVA analysis was used to determine statistical significance of main factors and interaction of sweetpotato varieties with the growing media. The model was specified in GLM according to split plot experimental design. When ANOVA showed statistical significances, means were separated using Tukey's HSD test (P < 0.05). Multivariate analysis was carried out using the XLSTAT software package. For determination of key traits responsible for discrimination based on differences in growing media for all sweetpotato samples and differences according to sweetpotato variety, the multivariate analysis by discriminant analysis was used.

3. Results and discussion

3.1. Genetic differentiation

SSR screening of sweetpotato cultivars was performed on eight loci (91.7% polymorphic loci) where the highest levels (Ht > 0.65) of genetic differentiation were assigned to loci Ib-318, Ib-297, Ib-248, Ib-242 and Ib-286. Locus Ib-255 reflected the lowest informativity through low Ht (0.278), high inbreeding coefficient (Fst = 0.400) and the lowest number of genetic migrants (Nm = 0.375), detected among genotypes. According to parameters of genetic diversity for specific loci, described in **Table 2**, the most effective genetic differentiation was obtained for locus Ib-286, where the lowest proportion of total genetic diversity that separates cultivars was calculated via Fst (0.082) and the highest number of genetic migrants among genotypes and cultivars (Nm = 2.813) was detected.

|	'Janja'	'Martina'	'Lučka'
'Janja'	*	0.072	0.148
'Martina'	0.829	*	0.153
'Lučka'	0.626	0.608	*

Table 2. Pairwise population comparisons of Nei genetic identity (below diagonal) and pairwise population Fst values (above diagonal).

AMOVA was performed through R-statistics (P ≥ 0.01), where Rst is an estimator of genetic differentiation for SSR loci that assumes a stepwise mutation model. Therefore, molecular variance among varieties was 36%, among genotypes 63% and within genotypes 1%, respectively. In contrast, report about evaluation of genetic variability of sweetpotato germplasm, originated from Africa, Asia and USA shows only 23% of genetic variance between different accessions [41]. Therefore, our study indicate the low level of genetic relatedness between cultivars 'Lučka', 'Janja' and 'Martina' compared to the genetic relatedness between different genetic resources from geographically distant genetic origins. First three axes in PCoA cumulatively explain 76.2% of genetic variation within observed genotypes and cultivars (data not shown). Allelic patterns across three sweetpotato varieties (**Figure 3**) showed that the most genetically diverse variety is 'Martina'. Meanwhile, variety 'Lučka' possess the highest number of alleles which are unique and specific for this variety only.

According to Nei's genetic distance and pairwise population Fst analysis, the most related cultivars are 'Janja' and 'Martina'; in contrast, 'Lučka' and 'Martina' show the weakest genetic linkages (**Table 2**).

3.2. Growth, yield and nutritional parameters

Table 3 shows the summary statistics of main factors and interactions and **Table 4** shows the effect of different growing substrate and cultivar on agronomic and nutritional traits
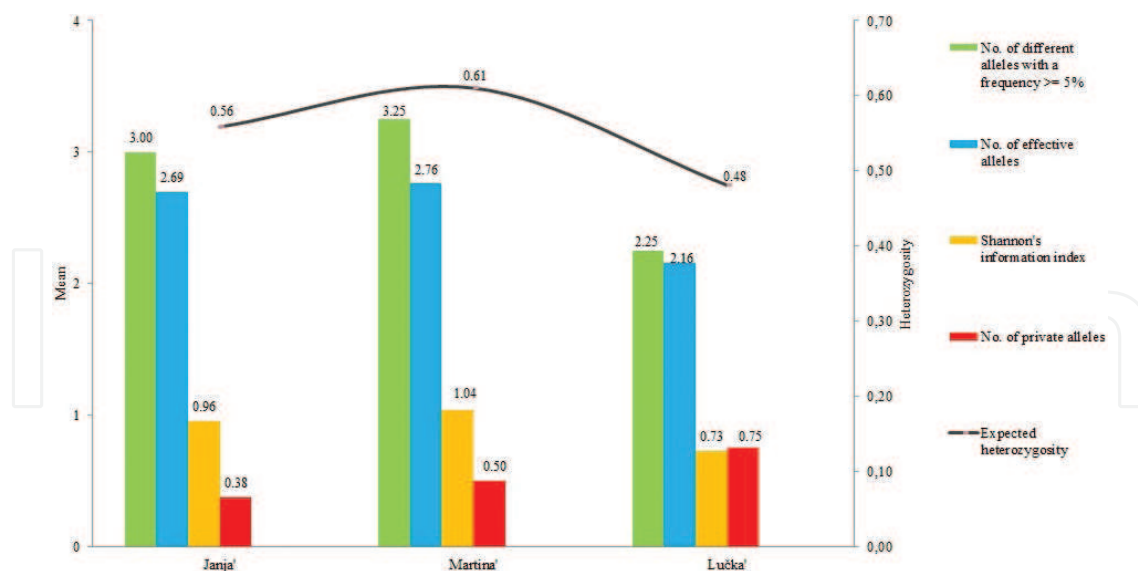
Figure 3. Allelic patterns according to genetic analysis across observed sweetpotato cultivars.

of sweetpotato. Measurements of agronomic traits showed that among growing substrate (factor A) vermiculite had the greatest impact on the vine length (144.4 cm). Between sweetpotato cultivars (factor B), significantly longer vine length was observed for 'Lučka' (147.1 cm). For thickness of vine-base significant differences were found for factor growing substrate, but not for cultivar. The thickness of vine-base (10.7 mm) was significantly higher in expanded clay. Number of branches was significantly higher for sweetpotato grown in peat (13.0), while between cultivars 'Martina' (10.9) and 'Janja' (10.6) had significantly more branches than 'Lučka'. Both, growing substrate and cultivar, had significant impact on weight of above ground part. The weight of above ground part was significantly higher for sweetpotato grown in peat (1402.4 g). Cultivar 'Martina' produced significantly higher weight of above ground part (1177.5 g), that is, more than double as 'Lučka' (463.7 g). Significantly higher number of leaves plant^{-1} was observed for 'Martina' (113.1), and between growing substrate in peat (131.1) and perlite (123.9). Both yield components, number of tubers plant^{-1} and tubers weight plant^{-1} were the lowest for sweetpotatoes grown in garden soil. Comparison between cultivars showed that 'Janja' had the highest yield. Mukhtar et al. [19] reported similar findings for vine length, number of branches and number of leaves plant^{-1} when tested two local sweetpotato cultivars with orange and white flesh.

Analyses of nutritional traits included TPC, AOP and AA of tubers. Data showed significant differences ($P \leq 0.001$) between the growing substrate and the cultivars in all three traits (**Table 3**). The TPC ranged from 36.2 to 65.1 mg GAE 100 g^{-1} FW, AOP from 0.18 to 0.56 mg TE g^{-1} FW and AA from 13.7 to 23.5 mg 100 g^{-1} FW (**Table 4**). Significantly lower TPC was determined in peat (41.2 mg GAE 100 g^{-1} FW). Between cultivars significantly higher TPC was observed in 'Lučka' (60.1 mg GAE 100 g^{-1} FW). Cultivar 'Lučka' with orange flesh color showed significantly higher TPC compared to the other two white flesh colored cultivars, which is in agreement with previous studies on other cultivars [3, 11, 42]. Similar to TPC, higher AOP was found for sweetpotato grown in perlite, expanded clay and vermiculite (for all >0.44 mg TE g^{-1} FW). Tubers of cultivars

	Vine length (cm)	Thickness of vine-base (mm)	Number of branches	Weight of above ground part (g)	Number of leaves plant^{-1}	Number of tubers plant^{-1}	Tubers weight plant^{-1} (g)	TPC (mg GAE 100 g^{-1} FW)	AOP (mg TE g^{-1} FW)	AA (mg 100 g^{-1} FW)
Factor A (growing substrate)										
Perlite	107.3	6.9 c	8.5 ab	724.6 b	123.9 a	15.8 a	982.2 ab	54.1 a	0.50 a	19.7 a
Peat	129.8	9.3 ab	13.0 a	1402.4 a	131.1 a	11.0 a	1517.6 a	41.2 b	0.32 c	16.7 b
Expanded clay	133.8	10.7 a	11.0 ab	934.9 ab	91.1 ab	14.1 a	1198.6 a	54.5 a	0.44 ab	16.6 b
Vermiculite	144.4	9.3 ab	9.9 ab	663.9 b	68.0 b	14.8 a	1358.3 a	53.2 a	0.44 ab	16.2 c
Garden soil	134.1	8.9 b	5.7 b	242.9 c	40.1 c	6.8 b	323.4 b	55.0 a	0.38 bc	16.3 c
P	Ns	**	*	***	***	**	**	***	***	***
Factor B (cultivar)										
'Janja'	119.5 b	9.3	10.6 a	738.0 b	90.6 ab	14.9	1168.0	46.0 c	0.37 b	15.9 b
'Lučka'	147.1 a	8.5	7.6 b	463.7 c	68.9 b	10.1	976.7	60.1 a	0.45 a	20.4 a
'Martina'	123.0 b	9.3	10.9 a	1177.5 a	113.1 a	12.4	1082.3	48.7 b	0.43 a	15.0 c
P	**	Ns	*	***	*	Ns	Ns	***	***	***
Interactions										
A × B	Ns	*	Ns	*	Ns	Ns	Ns	***	***	***

Ns, not significant. Mean values with different letters (a, b, c) in a column are significantly different according to the results of Tukey's HSD test (P < 0.05). TPC, total phenolic content; AOP, antioxidant potential; AA, ascorbic acid.

*Level of significance: P ≤ 0.05.

**Level of significance: P ≤ 0.01.

***Level of significance: P ≤ 0.001.

Table 3. Statistics of main factors and interactions for selected agronomic and nutritional traits of sweetpotato.

'Lučka' and 'Martina' had significantly higher AOP, 0.45 and 0.43 mg TE g^{-1} FW, respectively. These results are lower as reported by Tang et al. [11] in their study on different sweetpotato cultivars grown in China. Significantly higher AA was observed in tubers of sweetpotato grown in perlite (19.7 mg 100 g^{-1} FW), while between cultivars significant higher AA was observed in 'Lučka' (20.4 mg 100 g^{-1} FW) (**Table 3**). These data are higher as reported by Suárez et al. [42] on 30 sweetpotato cultivars from Canary Islands, where average values varies from 10 to 14 mg 100 g^{-1} FW.

Significant interactions of growing substrate (factor A) × cultivar (factor B) were observed (**Table 3**) for thickness of vine-base, weight of above ground part, AOP, TPC and AA. Interactions showed that different cultivars showed different response on growing substrate (data not shown). For example, cultivar 'Martina' had significantly higher thickness

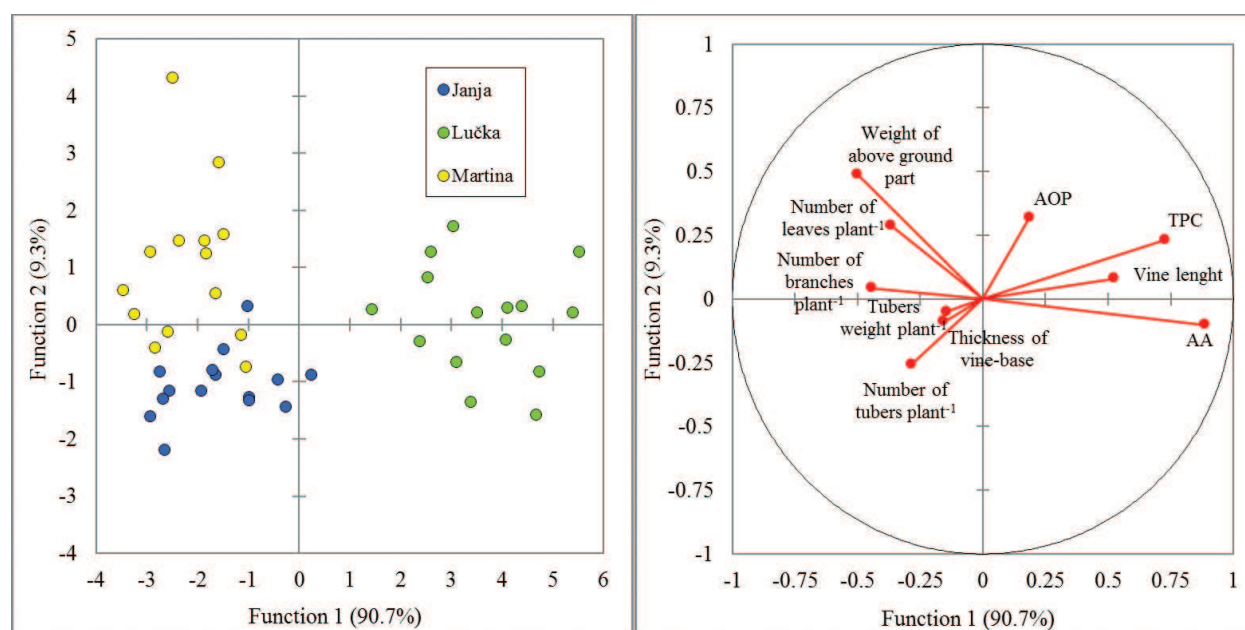| Factor A (growing substrate)	Factor B (cultivar)	Vine length (cm)	Thickness of vine-base (mm)	Number of branches	Weight of above ground part (g)	Number of leaves plant^{-1}	Number of tubers plant^{-1}	Tubers weight plant^{-1} (g)	TPC (mg GAE 100 g^{-1} FW)	AOP (mg TE g^{-1} FW)	AA (mg 100 g^{-1} FW)
Perlite	'Lučka'	116.0 ± 22.3	7.0 ± 1.0	6.3 ± 3.5	224.7 ± 42.1	80.7 ± 21.5	13.7 ± 8.0	853.0 ± 497.3	61.3 ± 0.4	0.48 ± 0.07	23.5 ± 1.2
Peat		160.0 ± 21.8	10.3 ± 2.9	11.7 ± 4.6	979.7 ± 235.7	100.0 ± 50.7	9.3 ± 1.5	1279.7 ± 494.7	49.0 ± 7.2	0.35 ± 0.04	20.4 ± 1.0
Expanded clay		144.7 ± 37.1	9.3 ± 1.5	8.3 ± 2.1	483.3 ± 160.6	63.3 ± 15.3	8.3 ± 3.2	1126.7 ± 559.4	65.1 ± 1.1	0.56 ± 0.01	19.6 ± 1.0
Vermiculite		160.3 ± 28.7	9.3 ± 0.6	7.7 ± 2.9	441.0 ± 78.6	61.3 ± 18.6	12.0 ± 2.6	1136.3 ± 512.5	60.5 ± 1.9	0.50 ± 0.03	20.1 ± 1.0
Garden soil		154.7 ± 28.2	6.3 ± 2.3	4.0 ± 0.0	189.7 ± 35.8	39.0 ± 14.4	7.3 ± 1.5	488.0 ± 45.7	64.5 ± 2.4	0.35 ± 0.01	18.5 ± 0.9
Perlite	'Martina'	92.7 ± 4.6	6.7 ± 1.2	10.7 ± 3.5	1205.0 ± 153.9	165.0 ± 59.6	14.7 ± 9.3	1044.3 ± 579.6	52.2 ± 1.3	0.60 ± 0.03	17.2 ± 0.9
Peat		116.7 ± 19.6	7.3 ± 1.2	16.0 ± 2.0	1992.7 ± 238.5	156.7 ± 40.4	12.7 ± 1.2	1639.0 ± 412.7	38.5 ± 1.5	0.18 ± 0.02	14.5 ± 0.7
Expanded clay		136.0 ± 26.9	11.0 ± 2.0	10.3 ± 2.1	1583.3 ± 840.1	131.7 ± 40.7	12.3 ± 1.2	929.7 ± 208.9	49.6 ± 1.6	0.43 ± 0.04	14.4 ± 0.7
Vermiculite		130.3 ± 8.7	9.7 ± 1.5	11.3 ± 2.3	834.7 ± 84.0	73.3 ± 14.4	14.3 ± 5.1	1502.7 ± 763.2	45.6 ± 0.6	0.56 ± 0.03	13.7 ± 0.7
Garden soil		126.5 ± 33.2	11.5 ± 0.7	6.0 ± 0.0	272.0 ± 53.7	38.5 ± 2.1	8.0 ± 5.7	293.5 ± 200.1	57.0 ± 3.8	0.34 ± 0.11	14.7 ± 0.5
Perlite	'Janja'	113.3 ± 15.3	7.0 ± 2.6	8.7 ± 2.1	744.0 ± 271.8	126.0 ± 80.6	19.0 ± 8.7	1049.3 ± 575.4	48.8 ± 2.6	0.43 ± 0.05	18.4 ± 0.9
Peat		112.7 ± 10.8	10.3 ± 3.5	11.3 ± 0.6	1235.0 ± 486.3	136.7 ± 51.1	11.0 ± 4.6	1634.0 ± 957.2	36.2 ± 2.9	0.42 ± 0.03	15.3 ± 0.8
Expanded clay		120.7 ± 9.5	11.7 ± 1.5	14.3 ± 3.1	738.0 ± 162.4	78.3 ± 12.6	21.7 ± 7.4	1539.3 ± 516.7	48.8 ± 1.3	0.32 ± 0.02	15.8 ± 0.8
Vermiculite		142.7 ± 11.0	9.0 ± 2.6	10.7 ± 3.8	716.0 ± 115.9	69.3 ± 12.9	18.0 ± 7.8	1436.0 ± 225.5	53.4 ± 4.4	0.27 ± 0.04	14.9 ± 0.7
Garden soil		108.3 ± 17.6	8.7 ± 1.2	8.0 ± 4.0	267.0 ± 49.8	42.7 ± 10.5	5.0 ± 2.0	181.3 ± 115.6	42.9 ± 0.7	0.43 ± 0.04	15.2 ± 0.8

Data are mean ± standard deviation (n = 3). TPC, total phenolic content; AOP, antioxidant potential; AA, ascorbic acid.

Table 4. Effect of different growing media and cultivar on selected growth, yield and nutritional parameters of sweetpotato.

of vine-base in garden soil, but significantly lowers in peat. However, cultivars 'Janja' and 'Lučka' had significantly higher thickness of vine-base in peat, but significantly lower in garden soil. Cultivar 'Martina' had significantly higher weight of above ground part compared to other cultivars in all growing substrates, except for garden soil. All three cultivars showed significantly higher weight of above ground part in peat. Interactions between cultivars and growing substrate showed significant differences (P ≤ 0.001) in TPC, AOP and AA. For example, cultivar 'Martina' had significantly higher TPC in tubers grown in garden soil, while cultivar 'Janja' lowers. Cultivar 'Martina' had the lowest AOP in peat, while other cultivars did not show response to this growing substrate. In case of AA all cultivars showed similar response in different growing substrate, except for garden soil.
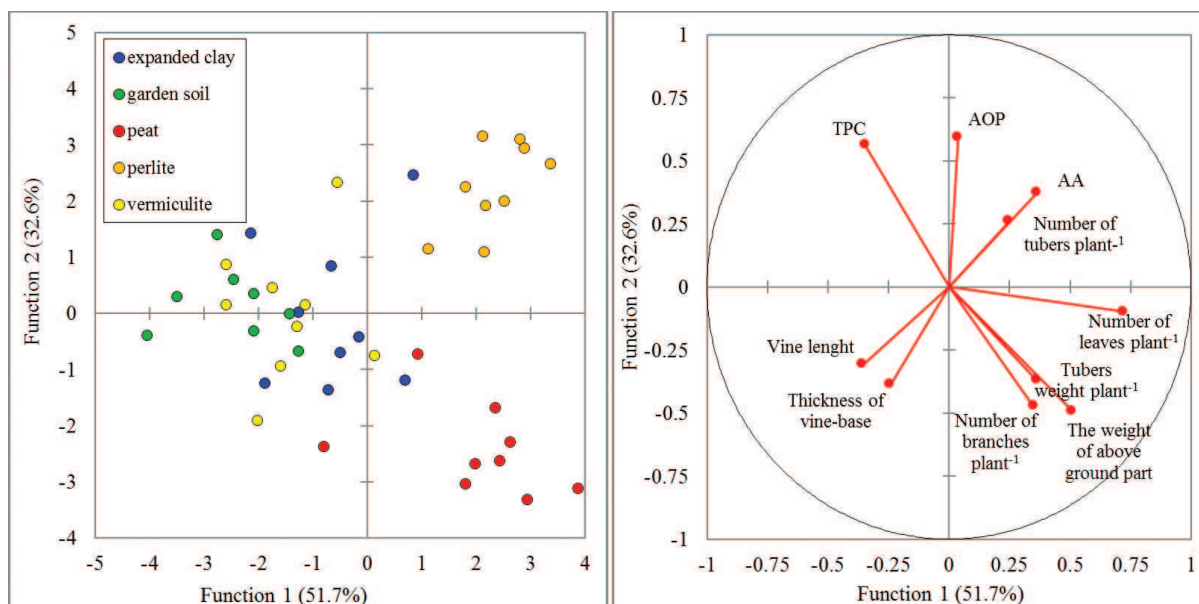
3.3. Multivariate analyses—discriminant analyses

The discrimination across the original data set of 15 samples originated from 3 sweetpotato cultivars is shown in **Figure 4**. Discriminant analysis was carried out across 10 traits: vine length, thickness of vine-base, number of branches, weight of above ground part, number of leaves plant^{-1}, number of tubers plant^{-1}, tubers weight plant^{-1}, TPC, AOP and AA. The curve defined by the first two discriminant functions (function 1/function 2) represents 100.0% of the total variance for these 10 variables. Function 1 explain 90.7% of the total variance and function 2 9.3% of the total variance. Major contributors to discriminate between different cultivars in function 1 are the AA, number of tubers plant^{-1}, tubers weight plant^{-1} and vine length, respectively; meanwhile the weight of above ground part, TPC, AOP and number of tubers plant^{-1} are major contributors in function 2. The groups of the sweetpotato culti-

Figure 4. Discriminant analysis plot of observations (left) and variables chart (right) performed with the 10 traits: vine length, thickness of vine-base, number of branches, weight of above ground part, number of leaves plant^{-1}, number of tubers plant^{-1}, tubers weight plant^{-1}, TPC, AOP and AA; of the 15 samples originated from 3 sweetpotato cultivars ('Janja', 'Lučka' and 'Martina').

Figure 5. Discriminant analysis of observations (left) and variables chart (right) performed with the 10 traits: given in the legend of **Figure 4** (see also text); of the 15 sweetpotato samples cultivated in 5 different growing substrate (expanded clay, garden soil, peat, perlite and vermiculite).

vars 'Janja', 'Lučka' and 'Martina' were well separated, with the slight overlapping of groups 'Janja' and 'Martina' (one sample of 'Janja' and one of 'Martina' were in the opposite group).

Figure 5 shows the discrimination across the original data set of 15 samples cultivated in 5 different growing substrates (expanded clay, garden soil, peat, perlite and vermiculite). Discriminant analysis was carried out with the same 10 traits as given above. Function 1 explains 51.7% of the total variance and function 2 32.6% of the total variance. Major contributors to discriminate in function 1 between different growing substrate are AA, TPC, weight of above ground part and vine length, respectively; meanwhile the number of tubers plant^{-1}, TPC, AOP and vine length are major contributors in function 2. As seen from **Figure 5**, the sweetpotato samples grown in garden soil, vermiculite and expanded clay are located close to each other and on the other side of the score plot as those grown in perlite or peat. Sweetpotato samples grown in perlite and peat are clearly distinguished from the other growing substrate with slightly overlapping groups of peat and expanded clay (one sample of peat and one of expanded clay are in the opposite group).

4. Conclusions

The present study investigated the genetic differentiation among three new Slovenian sweetpotato cultivars ('Lučka', 'Janja' and 'Martina'). Results showed that the most genetically diverse variety is 'Martina'. Meanwhile, variety 'Lučka' possess the highest number of alleles which are unique and specific for this variety only. Global genetic variance among all three cultivars is 36%. The effect of different growing substrate (perlite, peat, expanded clay, vermiculite and garden soil) was examined for 10 agronomic and nutritional traits of these sweetpotato cultivars. Overall results show

different response of cultivars in different growing substrate. Significant interactions of growing substrate × cultivar were observed for thickness of vine-base, weight of above ground part, AOP, TPC and AA. In conclusion, the discriminant analysis showed that the major traits for distinguishing among sweetpotato cultivars in function 1 are the AA, number of tubers plant^{-1}, tubers weight plant $^{-1}$ and vine length, and in function 2 the weight of above ground part, TPC, AOP and number of tubers plant^{-1}; and between growing substrate in function 1 AA, TPC, weight of above ground part and vine length, and in function 2 the number of tubers plant^{-1}, TPC, AOP and vine length.

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