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Effect of storage on antioxidant activity of freeze-dried potato peels

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ABSTRACT

A study was undertaken with the objective of evaluating the storage stability of the polyphenols and the antioxidant activity in the peel from two varieties of potatoes, Penta and Marcy, grown in Ontario, Canada. Samples were stored at −20, 4 and 25 °C for 0, 2, 4 and 8 weeks. Peel from Penta variety contained higher levels of total polyphenolic compounds, 2.4 mg/g of peel d.w. compare to Marcy, 1.14 mg/g of peel d.w. Chlorogenic and caffeic acids were the major polyphenolic compounds present in both verities. Levels of polyphenolic compounds in the peel were influenced by storage temperature with maximum loss observed at 25 °C. Storage time caused a decline in the levels of polyphenolic compounds up to 4 weeks at all temperatures followed by a significant increase at the end of week 8. There was a parallel increase in the antioxidant activity of the peel samples during storage and until the end of 8 weeks of storage. At all temperatures of storage, there was a significant relationship ($r = 0.43$, $P \leq 0.05$) between the amount of polyphenolic compound in the peel samples and their antioxidant capacity. Results suggest potato peel as a valuable source of polyphenolic compounds requiring proper storage condition to maintain their antioxidant properties.

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1. Introduction

Potato (*Solanum tuberosum* L.) is the world's most widely grown crop and an important part of the human diet (Leo et al., 2008). A significant percentage of the raw potatoes are processed to produce a range of consumer products including French fries and chips (Keijbets, 2008). Industrial processing of potatoes generates large quantities of peel as a by-product that create disposal, sanitation, and environmental problems. Potato peel (PP) is a good source of polyphenolic compounds having antioxidant properties. There is great deal of recent interest in the use of natural antioxidants as functional ingredients in human foods since they protect cell constituents against oxidative damage and limit the risk of various degenerative diseases associated with oxidative stress (Scalbert, Manach, Morand, Remesy, & Jimenez, 2005.). Previous studies have emphasized the use of PP as a source of dietary fiber (Arora & Camire, 1994; Camire & Flint, 1991) as well as antioxidant, either in foods(Kanatt, Chander, Radhakrishna, & Sharma, 2005; Mansour & Khalil, 2000; Rehman, Habib, & Shah, 2004) or in biological systems (Singh, Kamath, Narasimhamurthy, & Rajini, 2008; Singh, Kamath, & Rajini, 2005; Singh & Rajini, 2004; Singh & Rajini, 2008). Utilization of PP as a functional ingredient depends largely on the stability of the antioxidants contained in the peel upon storage. Very little work has been reported relating to the stability of polyphenolic compounds in PP upon storage. The present study was undertaken to determine the effect of storage temperature and time on the stability of polyphenolic compounds and their antioxidant activity in PP.

2. Materials and methods

All solvents, reagents, and standards used in this work were of HPLC grade (purity ≥ 98%) obtained from Sigma-Aldrich, Canada. All reagents and standard solutions were prepared using Milli-Q deionized water (Millipore, Bedford, USA).

2.1. Potato peel samples

Two potato varieties, Penta and Marcy that were harvested in September 2008 were obtained from R.K. Betattie Ltd, Aliston, Ontario, Canada. Uniform shape and size samples without any physical damage were selected. Tubers were washed with cold tap water to remove soil. After cleaning, potato samples were peeled with a mechanical peeler to obtain uniformly thick peel samples. They were weighed and lyophilized (Labconco Co. Freeze-dryer, USA) until constant weight was obtained. After complete dryness, the peel samples were ground to a powder using a kitchen mill (Braun KMM30, Germany) and sieved using 500 μm standard mesh to ensure symmetry of particle size. Freeze-dried powder samples were weighed into tightly closed pouches, sealed and stored at −20, 4, and 25 °C for 8 weeks. Samples were removed for analysis on 0, 2, 4 and 8 weeks, respectively.

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2.2. Extraction of polyphenolic compounds

At each time point, potato peel samples were extracted according to the methods published by (Mattila & Hellström, 2007) with minor modifications. Briefly, to 1 g of freeze-dried samples, a solution containing methanol (containing 2 g/L of 3,5-di-tert-butyl-4-hydroxytoluene) and 10% acetic acid in a ratio of 85:15 was added and ultrasonicated for 30 min. Deionized water was then added and the samples vortexed. The clear upper layer was collected, evaporated under vacuum in a rotary evaporator, redissolved in methanol solution and kept at −20 °C . The residues were redissolved in deionized water containing 1% ascorbic acid and 0.42% ethylenediaminetetraacetic acid-sodium salt (EDTA) and 5 ml of 10 M NaOH, flushed with nitrogen and stirred over night with a magnetic stirrer at room temperature. Samples were then adjusted to pH 2 with concentrated HCl and extracted with a mixture of diethyl ether and ethyl acetate in a 1:1 ratio. Organic layer was collected, evaporated to dryness as before, redissolved in methanol and kept at −20 °C. Concentrated HCl was added to the remaining solution and heated at 85 °C for 30 min. Following cooling, samples were extracted with diethyl ether and ethyl acetate in a ratio of 1:1. The organic layer was collected, evaporated to dryness as before, and redissolved in methanol. All methanol extracts were combined and filtered through a membrane filter (0.45 μm, Pall Corporation, USA) and analyzed for total polyphenolic compounds and antioxidant capacity.

2.3. Analysis of polyphenolic compounds

2.3.1. Determination of polyphenolic compounds

Potato peel extracts were analyzed for polyphenolic compounds according to the method of (Escarpa & González, 2000). A Waters 2690 Alliance HPLC system (Milford, MA, USA) was used in combination with Waters 996 PDA detector and Waters Millennium32 data management software, 4th edition. Polyphenolic compounds were detected at 280 nm. Two replicates of each samples were ran and the injection volume was 10 μl. Separation was carried out with a Phenomenex Nucleosil 5 μm C_{18} 100 A, 4.6×250 mm analytical column. Gradient elution with Two elution solvents: A (aqueous 0.01 M phosphoric acid), and B (100% methanol) were used at a flow rate of 1 ml/min. The measured polyphenolic compounds were identified based on their retention time that compared with the respective standards; two injections were used for each sample.

2.3.2. Trolox equivalent antioxidant activity (TEAC)

Method as described by (Ozgen, Reese, Tulio, Scheerens, & Miller, 2006) with some change, was used to measure antioxidant activity of potato peels extracts. 7 mM of ABTS, 2, 2-azinobis (3-ethylbenzothiazolin-6-sulfonate) diammonium salt, dissolved in 50 ml of acetate buffer pH 4.6 and 2.45 mM of potassium per sulphate dissolved in the same amount of buffer were mixed and stored at 4 °C in the dark for 12–16 h until a stable oxidative state was reached. This reagent was stable for several weeks when stored in the dark. On the day of analysis, the ABTS solution was diluted with the same buffer to an absorbance of 0.700 ± 0.2 at 734 nm. For the spectrophotometric assay, 3 ml of the ABTS solution and 10 μl of extract were mixed and the absorbance was determined at 734 nm at zero and 1 min after mixing. For the standard curve, Trolox (6-hydroxy-2, 5, 7, 8-tetramethylchromane-2-carboxylic acid) was prepared in ethanol and serial dilutions were mixed with ABTS. Readings were taken after zero and 1 min and a standard curve developed. Readings are measured twice for each sample and results expressed as Mean ± SD.

3. Statistical analysis

Statistical analysis and interpretation of all results was done by using GraphPad Prism version 5.00 for Windows, GraphPad Software, San Diego, California, USA, http://www.graphpad.com. Two-way

Table 1
HPLC analysis of polyphenolic compounds in peel of two potato varieties, Penta and Marcy.

Temp (°C)	Time (weeks)	Phenolic acids content (mg/g d.w. of peel)/variety, Penta						
		GA	HBA	CLA	CAA	CUA	FA	Total
25	0	0.20 ± 0.00 a	0.02 ± 0.00 a	0.76 ± 0.00 a	1.06 ± 0.01 a	0.06 ± 0.00 a	0.28 ± 0.00 a	2.40 ± 0.01 A
	2	0.10 ± 0.08 a	0.01 ± 0.01 a	0.33 ± 0.31 b	0.58 ± 0.53 b	0.04 ± 0.03 a	0.14 ± 0.13 a	1.18 ± 1.09 ACD
	4	0.01 ± 0.00 a	0.02 ± 0.00 a	0.07 ± 0.10 bd	0.38 ± 0.25 c	0.04 ± 0.02 a	0.08 ± 0.05 a	0.59 ± 0.25 BCE
	8	0.01 ± 0.00 a	0.01 ± 0.00 a	0.61 ± 0.10 ce	0.55 ± 0.11 bd	0.05 ± 0.01 a	0.16 ± 0.03 a	1.38 ± 0.24 ADE
4	0	0.20 ± 0.00 a	0.02 ± 0.00 a	0.76 ± 0.00 a	1.06 ± 0.00 a	0.06 ± 0.01 a	0.28 ± 0.00 a	2.40 ± 0.01 A
	2	0.05 ± 0.00 a	0.02 ± 0.00 a	0.55 ± 0.02 a	1.10 ± 0.03 adf	0.05 ± 0.00 a	0.28 ± 0.08 a	1.94 ± 0.12 ABC
	4	0.01 ± 0.00 a	0.09 ± 0.01 a	0.71 ± 0.04 a	0.27 ± 0.01 be	0.07 ± 0.00 a	0.18 ± 0.01 a	1.31 ± 0.07 ABE
	8	0.01 ± 0.00 a	0.01 ± 0.00 a	0.87 ± 0.01 a	0.33 ± 0.00 cg	0.07 ± 0.00 a	0.25 ± 0.00 a	1.51 ± 0.01 ACE
−20	0	0.20 ± 0.00 a	0.03 ± 0.00 a	0.76 ± 0.00 a	1.06 ± 0.01 a	0.06 ± 0.00 a	0.28 ± 0.00 a	2.40 ± 0.01 A
	2	0.15 ± 0.00 a	0.02 ± 0.00 a	0.83 ± 0.02 a	1.05 ± 0.02 a	0.07 ± 0.00 a	0.25 ± 0.00 a	2.36 ± 0.04 ABC
	4	0.01 ± 0.00 a	0.02 ± 0.00 a	0.63 ± 0.00 a	0.88 ± 0.00 a	0.07 ± 0.00 a	0.25 ± 0.02 a	1.85 ± 0.01 ABE
	8	0.01 ± 0.00 a	0.02 ± 0.00 a	0.96 ± 0.01 a	0.92 ± 0.01 a	0.08 ± 0.00 a	0.29 ± 0.00 a	2.27 ± 0.01 ACE
Temp (°C)	Time (weeksks)	Phenolic acids content (mg/g d.w. of peel)/variety, Marcy						
		GA	HBA	CLA	CAA	CUA	FA	Total
25	0	0.12 ± 0.00 a	0.04 ± 0.00 a	0.51 ± 0.00 a	0.37 ± 0.00 a	0.02 ± 0.00 a	0.11 ± 0.00 a	1.14 ± 0.00 A
	2	0.03 ± 0.00 a	0.03 ± 0.00 a	0.27 ± 0.16 b	0.09 ± 0.05 bd	0.01 ± 0.00 a	0.06 ± 0.03 a	0.48 ± 0.27 BCD
	4	0.01 ± 0.00 a	0.07 ± 0.06 a	0.26 ± 0.19 bc	0.17 ± 0.12 bc	0.01 ± 0.01 a	0.05 ± 0.03 a	0.56 ± 0.41 ACE
	8	0.01 ± 0.00 a	0.11 ± 0.00 a	0.43 ± 0.01 a	0.33 ± 0.01 ae	0.02 ± 0.00 a	0.08 ± 0.00 a	0.97 ± 0.02 ADE
4	0	0.12 ± 0.00 a	0.04 ± 0.00 a	0.51 ± 0.00 a	0.38 ± 0.00 a	0.02 ± 0.00 a	0.12 ± 0.00 a	1.14 ± 0.00 A
	2	0.02 ± 0.00 a	0.03 ± 0.01 a	0.41 ± 0.00 a	0.21 ± 0.02 b	0.01 ± 0.00 a	0.04 ± 0.00 a	0.68 ± 0.01 ABC
	4	0.01 ± 0.00 a	0.05 ± 0.02 a	0.33 ± 0.10 a	0.24 ± 0.07 a	0.01 ± 0.00 a	0.061 ± 0.0 a	0.71 ± 0.20 ABD
	8	0.01 ± 0.00 a	0.11 ± 0.04 a	0.41 ± 0.01 a	0.31 ± 0.02 a	0.01 ± 0.00 a	0.08 ± 0.00 a	1.12 ± 0.04 ACD
−20	0	0.12 ± 0.00 a	0.04 ± 0.00 a	0.52 ± 0.00 a	0.37 ± 0.00 a	0.02 ± 0.00 a	0.11 ± 0.00 a	1.14 ± 0.00 A
	2	0.03 ± 0.02 a	0.03 ± 0.00 a	0.46 ± 0.03 a	0.40 ± 0.02 a	0.02 ± 0.00 a	0.11 ± 0.01 a	1.03 ± 0.01 ABC
	4	0.01 ± 0.00 a	0.11 ± 0.00 a	0.37 ± 0.07 a	0.24 ± 0.04 a	0.01 ± 0.00 a	0.10 ± 0.01 a	0.81 ± 0.14 ABD
	8	0.02 ± 0.00 a	0.11 ± 0.00 a	0.41 ± 0.01 a	0.41 ± 0.02 a	0.02 ± 0.00 a	0.12 ± 0.00 a	1.03 ± 0.04 ACD

1) Results are duplicates ± SEM.
2) For each phenolic acid and within the selected temperature, value with the same small letters means no significance difference ($P \leq 0.05$).
3) Values with same capitalized letters shows no significance changes in total amount of phenolic acids within each set temperature ($P \leq 0.05$).
4) (GA) gallic acid, (HBA) hydroxybenzoic acid, (CLA) chlorogenic acid, (CAA) caffeic acid, (CUA) *p*-coumaric acid, (FA) ferulic acid.

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