Pol. J. Environ. Stud. Vol. 22, No. 1 (2013), 167-173

Original Research

Comparison of Vitamin C and β-Carotene in Cruciferous Vegetables Grown in Diversified Ecological Conditions

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Received: 18 May 2012
Accepted: 16 August 2012

Abstract

The aim of this study was to compare vitamin C and β-carotene contents in cruciferous vegetables (*Brassica oleracea*) grown under diversified ecological conditions for three consecutive years. The hypothesis to be verified was that statistically, vitamin C and β-carotene contents in cruciferous vegetables from the closest areas around a steelworks, organic farms, and randomly purchased in retail differed significantly, independent of the climate and agro-technical conditions. The study was done on three species of cruciferous vegetables: '*Stone head*' white cabbage, '*Langedijker*' red cabbage, and '*Dolores F1*' Brussels sprouts. All these species of cruciferous vegetables were characterized by significantly higher vitamin C and β-carotene contents (Brussels sprouts) when grown on organic farms *vis-à-vis* vegetables grown around a steelworks or available in retail in Kraków, which may be evidence of efficient organic growing technology, and may indicate another way to improve the quality of food.

Keywords: bioactive compounds, conventional farming, organic farming, red head cabbage, white head cabbage, Brussels sprouts

Introduction

Cruciferous vegetables are among the most important dietary vegetables consumed in Europe and all over the world owing to their availability at local markets, cheapness, and consumer preference. Phytochemicals from cruciferous vegetables induce detoxification enzymes, scavenge free radicals, alleviate inflammation, stimulate immune functions, decrease the risk for cancers, inhibit malignant transformation, and regulate the growth of cancer cells [1].

Fruits and vegetables are good dietary sources of natural antioxidants for humans, containing many different antioxidant components that provide protection against harmful free radicals. Consumption of fruits and vegetables has been strongly associated with the reduced risk of chronic diseases, such as cardiovascular disease, cancer, diabetes, Alzheimer's, cataracts, and age-related functional decline, and has been shown also to provide other health benefits [2-4].

At the present time, there is no consistent epidemiological evidence to support a role for cruciferous vegetables as chemopreventive agents against cancers other than lung, colorectal, breast, prostate, and pancreatic cancer. One exception may be gastric cancer. A systematic literature review published by the World Cancer Research Fund in 2006 evaluated five cohort studies and identified a general direction of effect, which tended to be protective against stomach cancer for higher consumption of cruciferous vegetables, although in no one study was the indi-

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vidual risk estimate statistically significant. The actual available data only reveal the conclusion that a frequent intake of vegetables of the cruciferous family lowers risk and may lead to a weaker metastasis of tumors in some persons [1, 5].

Many consumers believe that organically grown foods are of better quality, healthier and more nutritious than conventionally grown foods. There is a considerable number of scientific data indicating that organic vegetables and fruits contain more compounds with antioxidant properties compared to products from conventional farms, which is decisive for their greater biological value, but according to the latest review, the health benefits of consuming organic compared to conventional foods are unclear [6, 7].

The aim of the present study was to compare vitamin C and β-carotene in cruciferous vegetables cultivated in areas around a steelworks, on organic farms and those bought in retail for three consecutive years. The hypothesis to be verified through research was that statistically, vitamin C and β-carotene in cruciferous vegetables from areas around a steelworks, organic farms, and random retailers differ significantly independent of the climate and agro-technical conditions.

Materials and Methods

Material

This study was done on three species of cruciferous vegetables: the white head cabbage '*Stone head*', the red head cabbage '*Langedijker*', and the '*Dolores F1*' Brussels sprout. The experiment lasted three years, from 2005 to 2008. Vegetables came from three different sources:

- 15 conventional farms from five different locations around ArcelorMittal Poland SA steelworks (Małopolskie Voivodeship), each of the farms producing vegetables for commercial purposes, located directly east of the emission source (western winds prevail in that territory)
- Five organic farms holding "Agro Bio Test" certificates (Małopolskie and Świetokrzyskie voivodeships, located 50 kilometers from the ArcelorMittal Poland SA steelworks and sources of possible contamination)
- unknown method of cultivation (conventional/organic/integrated), of recognizable varieties, obtained from five different retailers in Kraków (Małopolskie Voivodeship)

Seedlings of the above vegetables were all grown by Poland, a Kraków-based cultivation and seed production firm, and were planted at the turn of June and July for three consecutive years in the above locations. The last group of tested vegetables was purchased randomly from five different retailers in Kraków.

The Kraków region has been an environmentally threatened area for a number of years. According to the Report on the Condition of the Environment in Małopolska Voivodeship in 2009 [8], dust and gas emissions from industrial sources still rank this region among the most polluted ones in Poland, despite the ongoing effort to reduce the pollution load. The ArcelorMittal Poland SA Unit in Kraków (formerly T. Sendzimir Steel Works, TSSW) has remained under the constant scrutiny of the Voivodeship Inspectorate for Environmental Protection (VIEP) since it was counted among the enterprises causing the greatest environmental nuisance due to emission of chemical pollutants [8, 9].

Vegetables came from random suppliers, reflecting the situation of a potential consumer who buys vegetables at the consumer market. It cannot therefore be established what type of vegetable farms they came from.

The subject study was conducted regardless of the climate or agro-technical conditions because it was consumer-type research in nature.

Fresh vegetables were stored in a cold room at +4ºC, from which they were taken directly to a laboratory. Two specimens of white and red cabbages and eight Brussels sprouts, with the biggest and with the smallest diameter, were sampled at each farm and from five different retailers. The plants of each vegetable were cut vertically into four or eight pieces (sub-samples) after removing inedible parts (outer leaves and stalks). Next, the sub-samples of plants were crumbled and mixed thoroughly. The material so prepared provided a representative average sample that was used for analyses of dry matter, vitamin C and β-carotene contents (only in Brussels sprouts).

Analytical Methods

The samples so prepared were examined for the content of dry mass, vitamin C, and β-carotene. The dry matter of the prepared samples of vegetables was determined according to PN-90/A-75101/03. The determination principle comprised determining the decrease in mass upon removal of water from the product during thermal drying at 105ºC under normal pressure conditions [10].

The content of vitamin C (ascorbic and dehydroascorbic acid) was determined using Tillmans Method, as modified by Pijanowski according to the PN-A-04019:1998 standard [11]. The total content of ascorbic acid was determined by reducing dehydroascorbic acid to ascorbic acid with sodium sulfide, precipitating the excess of sulfides using mercury chloride, and determining the total ascorbic acid by 2,6-dichlorophenoloindophenol titration. The amount of β-carotene was measured according to the PN-90/A-75101/12 standard by extracting carotenoid from the test sample using hexane, carotenoid separation on a chromatographic column, and colorimetric determination of β-carotene at a wavelength of 450 nm [12].

For each sample the chemical analyses were done in two or three replicates, the relative error not exceeding 5%. The values presented in the tables were calculated based on 45 repetitions (15 farms × 3 years) for each vegetable species from farms around the ArcelorMittal Poland SA steelworks, on 15 repetitions (5 farms × 3 years) for vegetable species from organic farms, and on 15 repetitions (5 farms × 3 years) for vegetable species from retail stores.

Table 1. Content of dry mass in white head cabbage, red head cabbage, and Brussels sprouts grown in diversified ecological conditions. Mean values±Standard Deviation.

Source of vegetables	Dry mass (g/100g)		
	White head cabbage	Red head cabbage	Brussels sprouts
From closest vicinity of the steelworks	6.63±0.59a	8.16±0.21a	14.8±0.69a
From organic farms	7.55±0.20b	9.33±0.21b	16.0±0.62b
From a market	7.35±0.20b	9.35±0.40b	15.7±0.52b

Differences between values signed with the same small letters are non-significant ($p \leq 0.05$).

Table 2. Content of vitamin C (in fresh and dry mass) in white head cabbage, red head cabbage, and Brussels sprouts grown in diversified ecological conditions. Mean values±Standard Deviation.

Source of vegetables	Vitamin C					
	White head cabbage		Red head cabbage		Brussels sprouts	
	(mg/100g f.m.)	(mg/100g d.m.)	(mg/100g f.m.)	(mg/100g d.m.)	(mg/100g f.m.)	(mg/100g d.m.)
From closest vicinity of the steelworks	35.7±1.74a	460.5±16.0a	35.6±1.78a	458.1± 27.4a	71.1a±8.03	431.5±52.2a
From organic farms	41.2±1.03b	558.2±19.0b	46.2±3.75b	662.6±57.4b	99.1b±8.69	584.9±77.4b
From a market	34.0±1.15a	469.0±40.6a	37.3±2.11a	433.6±48.8a	64.4a±4.76	379.6±31.4a

Differences between values signed with the same small letters are non-significant ($p \leq 0.05$).

Statistical Analysis

To check the significance of differences between the contents of vitamin C and β-carotene in cruciferous vegetables depending on their source, ANOVA single-factor was performed. The significance of differences was evaluated using Duncan's test, with the critical significance level of $p \leq 0.05$. All calculations were done by Statistica v. 8.1 (StatSoft Inc.).

Results

Dry Mass

All three vegetable species under review (Table 1) cultivated on farms holding "Agro Bio Test" certificates were demonstrated to have a clearly higher dry mass content ($p \leq 0.05$) compared with vegetables cultivated near the steelworks. Vegetables from eco-farms and from retailers in Kraków showed a similar dry mass content.

Vitamin C

The three vegetable species grown on farms certified by "Agro Bio Test" contained significantly higher ($p \leq 0.05$) vitamin C, per unit of both fresh and dry weight, in comparison with the vegetables from areas adjacent to the steelworks and from local suppliers (Table 2). The differences in vitamin C content between vegetables from eco-farms vs. vegetables from areas around the steelworks vs. vegetables purchased from random local retailers were: 15 and 21% f.w. and 21% and 19% d.w. for white head cabbage; 30% and 45% f.w. and 24% and 53% d.w. for red cabbage, and 39% and 35% f.w. and 53% and 54% d.w. for Brussels sprouts. The vegetables grown around the steelworks and from local retailers had comparable quantities of vitamin C.

β-Carotene

The results of β-carotene determination (Table 3) showed the content of this component was higher in the organically grown Brussels sprouts, on both fresh and dry weight bases, than in the vegetables from around the steelworks or the vegetables purchased from retailers in Kraków. These differences were statistically significant at $p \leq 0.05$. The content of β-carotene in the Brussels sprouts grown on organic farms was higher by 83% f.w. and 111% d.w. than in the Brussels sprouts grown near the steelworks, and by 99% f.w. and 116% d.w. than in the vegetables from local retailers. The content values of β-carotene in the Brussels sprouts cultivated in the steelworks protection zone or from local retailers were similar.

Discussion

Vitamin C

Cardoso et al. [13] report that organic acerola, which is an excellent source of vitamin C, presented a higher concentration of ascorbic acid and total vitamin C compared to the conventional alternative. Rembiałkowska's research

Table 3. Content of β-carotene (in fresh and dry mass) in Brussels sprouts grown in diversified ecological conditions. Mean values±Standard Deviation.

Brussels sprouts	β-carotene	
	(μg/100g f.m.)	(μg/100g d.m.)
From closest vicinity of the steelworks	166.3±43.8a	923.7±192.1a
From organic farms	303.5±11.4b	1949.8±161.7b
From a market	152.6±28.7a	901.5±82.3a

Differences between values signed with the same small letters are non-significant ($p \leq 0.05$).

group proved that the content of vitamin C was higher in vegetables and fruits from organic farms [14-17]. Rembiałkowska reports that vitamin C concentration in organically grown cabbage is higher by 10 mg/100g f.w. or by 30% or even by 96% than in cabbage cultivated using conventional technology [18]. Czapski's research [19] on 13 plant species cultivated under varied conditions showed that the vitamin C level in six of them was identical for both technologies, while in the remaining seven species the content of vitamin C was higher in the case of organic cultivation methods. Bartkyavichyute, Evers, and Pither [20] revealed a 15% higher vitamin C level in organic vegetables vs. conventional methods, while Schuphan [21] reported a 75% increase based on 12-year research. Chen [22] and Worthington [23] also claimed the vitamin C content was much higher in plants from organic farms compared with the conventional technology. By contrast, other researchers observed no relationship between cultivation technology and vitamin C content, for example Meier-Ploeger et al. [24], Assano [25], Warman and Havard [26], or Polish authors Jabłońska-Ceglarek et al. [27]. Hoefkens et al. [7] report that significant higher concentrations of vitamin C were found in organic tomato, but significantly lower concentrations in organic carrots and potatoes compared to the conventional alternative.

In a study by Esch et al. [28] the nutritional difference, as determined by vitamin C content, between six sets of conventionally and organically grown fruits were analyzed. There was no significant difference found in five of the six fruits considered. Only organic lemons displayed a significantly higher vitamin C level than their conventionally grown counterparts. A review in 2006 showed that organic foods had significantly higher amounts of antioxidants (vitamin C) and minerals in addition to lower levels of pesticide residues, nitrates, and some heavy metal contaminations than conventionally grown crops. They concluded that because of this, organic crops had a higher nutritional value and a lower risk of causing disease due to contamination [29]. Rossi et al. [30] revealed that organic tomatoes contained less vitamin C than crops grown using a conventional method. However, Crinnion [31] reported that nutrient content also varies from farmer to farmer and year to year, and that reviews of multiple studies show that organic varieties do provide significantly greater levels of vitamin C.

In the study of Koh et al. [32], 27 spinach varieties grown in certified organic and conventional cropping systems were undertaken in order to compare the levels of vitamin C. The mean levels of ascorbic acid were significantly higher in the organically grown spinach compared to the conventionally grown spinach. These results suggest that organic cropping systems result in spinach with higher levels of ascorbic acid.

In the 2 years of the experiment by Hallmann [33], there was a significant effect of cultivation method on the vitamin C content. Organic tomatoes had significantly higher content of vitamin C compared with conventional tomatoes. On the other hand, Pieper and Barrett [34] obtained a lower content of vitamin C in organic tomatoes compared to conventional ones. Similar results were presented by Juroszek et al. [35], who indicated that organic tomatoes showed a tendency to higher content of vitamin C, although the differences between organic and conventional tomatoes were not statistically significant.

In the study of Bizjak Bat et al. [36] the average ascorbic acid content of 32.2 mg/100g in the organically grown fruits was higher than the average content of 23.0 mg/100g in the conventional ones. Higher ascorbic acid content was also found in organically grown fruits [37].

Similar results were obtained by Hallmann and Rembiałkowska [38], who reported that organic bell pepper fruits were distinguished by a significantly higher content of vitamin C compared with conventional fruits.

The aim of the study of Wunderlich et al. [39] was to examine the nutritional quality of broccoli using vitamin C in broccoli as a biomarker. Although the vitamin C content of organically and conventionally labeled broccoli was not significantly different, significant seasonal changes have been observed. The seasonal changes in vitamin C content were larger than the differences between organically labeled and conventionally grown broccoli.

The remarkable difference in results obtained by the above-cited authors could result from different cultivation parameters, dissimilar storage conditions after harvesting, and different varieties of the same species used for their research. Low storage temperature helps preserve vitamin C in vegetables [40]. Vitamin C content can be lowered by exposure to oxygen, high temperature, trace amounts of metals, or heightened ethylene concentration in storage facilities [41].

β-Carotene

The β-carotene content values in Brussels sprouts in this study (Table 3) were lower than literature data available, where the values ranged widely from 430.0 up to 1,020.0 μg/100 g f.w. [42, 43].

Regardless of their origin, the samples evaluated in our research were stored under the same conditions from harvest to analyses. β-carotene, apart from vitamin C, is one of the most important antioxidants in cruciferous vegetables. Its mean content demonstrates high changeability depending on variety, plant maturation phase, season, climate, agro-technical procedures, and storage conditions after harvest. β-carotene is a high temperature-sensitive component, especially in an acidic environment, and to exposure to oxygen and light. Moreover, outer leaves of cabbage contain 21 times more carotenoids than inner, whiter ones, and the outer, the most colored and β-carotene-saturated leaves were removed from the cabbage chosen for this analysis, which was considered edible. Cabbage heads can differ up to six times in carotenoid content depending on the proportion of leaves exposed to direct light [41]. Storing cabbage in a controlled atmosphere can protect it from serious loss of β-carotene [40].

Hallmann and Rembiałkowska [15] compared the β-carotene level in tomatoes cultivated in various systems, and concluded that tomatoes from organic farms contained more β-carotene compared with conventionally grown ones. Earlier research by Brat et al. [44] reported similar results, but the experiment of Lucarini et al. [45] showed that tomatoes from conventional farms to be richer in β-carotene. According to other foreign research, such as Bartkyavichyute, Evers, and Pithera [20], the differences in β-carotene levels in vegetables cultivated on conventional and organic farms usually did not exceed 15% in favor of eco-farms. Brandt and Mølgaard [46] and Heaton [47] confirmed that it was natural for plants cultivated using organic methods to contain more carotenoids. By contrast, Kumpulainen [48] observed no differences in β-carotene contents in carrots and potatoes from organic and conventional farms.

Søltoft et al. [49] report that the expected higher content of presumed health-promoting carotenoids in carrot roots and human diets was not documented in their study. The plasma status of carotenoids increased significantly after consumption of the organic and conventional diets, but no systematic differences between the agricultural production systems were observed. Jiwan et al. [50] tested the baby foods. The selected for analysis foods were for the same age group (4+ months) and of two types: chicken and vegetable dinners and berry-based desserts. Due to their ingredient composition, carotenoid content and bioaccessibility varied within and between the organic and non-organic foods. The conclusion was that the organic dinners tested were generally not superior to the non-organic foods in terms of carotenoid content and bioaccessibility. Similar conclusions were reached in another study by Cardoso et al. [13] comparing the concentration of vitamin C and carotenoids between three fruits produced by organic and conventional farming in Brazil. There was no evidence of the nutritional superiority of the organically grown fruits.

The study of Hallmann [33] observed a significantly higher content of β-carotene for conventional tomatoes in the 2 years. Different results were indicated by Rossi et al. [30].

Juroszek et al. [35] observed a higher content of β-carotene in organic tomato fruits. Data calculated across two years showed no significant differences between organic and conventionally produced tomatoes for the fruit nutritional parameters, including β-carotene. When matched farm pairs were evaluated on the basis of individual-year data, very few significant differences were found between organic and conventionally produced tomatoes in some farm pairs, either in one or both varieties, for β-carotene.

In the experiment of Hallmann and Rembiałkowska [38] the β-carotene content was significantly higher in organic pepper fruits. Similar results were obtained by Szafirowska and Elkner [51]. An opposite trend was found by Flores et al. [52] who reported a lower level of β-carotene in organic fruits than in conventional fruits.

Conclusions

All species of cruciferous vegetables from organic farms were characterized by significantly higher vitamin C and β-carotene contents (Brussels sprouts) compared to vegetables cultivated near the steelworks or those purchased from local retailers in Kraków. That may confirm the efficiency of organic technologies in agronomy, and may indicate that this is yet another way to improve the quality of health food. Farming techniques may have an impact on the quality of cruciferous vegetables and their higher vitamin C and β-carotene content supports the notion that organic foodstuffs are more wholesome.

Studies investigating different phytochemicals and antioxidant activity in organic and conventional foods have had conflicting results, because it can vary not only among cultivars but also based on the specific substance that is being analyzed.

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