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Carotenoid Profiles and Consumer Sensory Evaluation of Specialty Carrots (*Daucus carota*, L.) of Various Colors

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Five different colored carrots were analyzed for their carotenoid profile and underwent sensory evaluation to determine consumer acceptance (n = 96). Four major carotenoids were identified and quantified by use of HPLC methods. High β -carotene orange carrots were found to contain the greatest concentration of total carotenoids. Except for the white, all the carrots are a significant source of bioavailable carotenoids. Sensory evaluation showed the high β -carotene orange and white carrots to be favored over the yellow, red, and purple carrots in both blind and nonblind treatments (P < 0.01). However, all the carrots were well accepted by the consumer panel. With this information, carrot growers should be encouraged to cultivate specialty carrots to provide sources of both vitamin A precursors and phytochemicals.

KEYWORDS: Carrot; carotenoid extraction; carotenoid profile; sensory evaluation; Daucus carota

INTRODUCTION

Consumption of fruits and vegetables is associated with a decreased risk of cancer and other chronic diseases. Fruits and vegetables provide a complex mixture of nutrients and nonnutrients, such as phytochemicals, that work together to protect against disease (1). Different colors of fruits and vegetables are due to pigmented disease-fighting phytochemicals, which is one reason the Dietary Guidelines recommend choosing a variety of fruits and vegetables (2).

Considering dietary recommendations and better accessibility, the proportion of adults in the U.S. who consume fruit and vegetables at least 5 times daily increased from 19% in 1990 to 23% in 1996 (3). In addition, U.S. carrot availability increased 66% and world availability by 46% between the years 1925 and 1995 (4). The increase in carrot consumption rates may be due to the introduction of prepackaged and precut carrots, as well as the nutritional benefits that carrots provide (5, 6).

Simon et al. (7) have developed unusual carrot strains individually high in β -carotene, lutein, lycopene, and anthocyanins. These various compounds have been implicated as potent phytochemicals (8). β -Carotene has been studied extensively for both its pro-vitamin A activity and its role in disease prevention (9). Associations between the consumption of lutein and prevention of age-related macular degeneration (10) and reduced risk of atherosclerosis (11) have been documented. Dietary lycopene has an inverse association with the risk of various cancers (12-14), cardiovascular disease (15-17), and diabetes (18). Flavonoids, one class being anthocyanins, potentially act as antioxidants (19), antiinflammatory agents (20), anti-thrombotic agents (21), and anti-carcinogens (22).

Carrots are popular in a variety of foods because of their pleasant flavor. Consumer sensory indicators of carrot include terpenoids and sugars (23). In a sensory evaluation of carrot flavor with trained panelists, Simon et al. (24) concluded increasing sugar content played an important role in the overall carrot preference, but the level of terpenoids can mask the overall sweetness. Using four different colored carrots, Alasalvar et al. (23) conducted a sensory evaluation with trained panelists and found the purple carrot to be the sweetest.

Horticulturists have been working on genetic approaches for improving nutrient content and visual appeal of vegetables in hopes of increasing consumer consumption of beneficial phytochemicals. The purpose of this study was 2-fold: (1) to determine the carotenoid profile of five specialty carrots and (2) to determine if carrot color influences consumer perception of taste and other sensory characteristics. The overall results of this study could assist carrot breeders in further developing nutrient-rich carrots. Increased market availability of specialty carrots would also increase consumer consumption of various disease-preventing phytochemicals. From a global perspective, the consumption of these carrots, especially the high β -carotene variety, may improve vitamin A status in countries where deficiency is a major health problem.

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MATERIALS AND METHODS

Carrots. Five carrot populations representing a wide range in color including high β -carotene orange, purple with orange core, yellow, red, and white were sown by the University of California Desert Research and Extension Station in sandy, loam soil in October and were lifted in March the following year. Carrots were refrigerated at 2 °C after harvest and subsequently shipped overnight from California to Wisconsin. Upon arrival, they were immediately returned to 2 °C.

Extraction and Analysis of Carotenoids. On the day of analysis, each carrot type was homogenized with a food processor, extracted and analyzed for carotenoids. Under fluorescent gold lighting, carotenoids were extracted according to methods developed by Horvitz et al. (25) with slight modifications depending on carrot type. Triplicate analyses of processed carrots were done using the following extraction method: internal standard, β -apo-8'-carotenyl decanoate synthesized in our lab using published methods (26), was added to 0.5 g of sample and ground using a mortar and pestle; 2.5 g of sodium sulfate was added to absorb water and form a paste. The amount of internal standard added varied dependent upon carrot type, for example, 0.91 nmol for white, 3.1 nmol for yellow, 9.3 nmol for red, and 18.3 nmol for the purple, orange, and high- β -carotene orange. Carotenoids were extracted into alternating 10-mL washes of dichloromethane and acetone, which were filtered into a 100 mL volumetric flask. A 1-mL aliquot of the filtrate was dried under argon, redissolved in 100 μ L of 50:50 (v/v) dichloroethane/methanol and analyzed using HPLC. A 25-µL aliquot was injected onto a Waters Resolve C₁₈ 5- μ m column, 3.9- \times 300-mm (Milford, MA) equipped with a guard column. The Waters HPLC system (Milford, MA) consisted of a 600 solvent delivery system, 717 autosampler, and 996 photodiode array detector. A gradient system was developed to optimize separation of carotenoids. The HPLC mobile phase consisted of 95:5 (v/v) acetonitrile/water with the modifiers ammonium acetate (10 mM) and triethylamine (0.1%) as solvent A, and 85:10:5 (v/v/v) acetonitrile/methanol/ dichloroethane, with the same modifiers, as solvent B. At 2 mL/min, the gradient procedure was as follows: (1) 100% solvent A for 3 min, (2) a 12-min linear gradient to 100% solvent B, (3) a 3-min hold at 100% solvent B, (4) a 1-min linear gradient back to 100% solvent A. The detector was set in scan mode, 210 to 550 nm, during the analysis and the wavelength of detection for quantification against authentic standards was 450 nm, which is a compromise for λ maxima of 426 nm for $\beta\mbox{-apo-8'-carotenyl}$ decanoate, 445 nm for lutein, 472 nm for lycopene, 444 nm for α -carotene, and 453 nm for β -carotene. To optimize the detection of carotenoids in the white carrots, the entire filtered extract of 1-g of sample was concentrated with a rotoevaporator, transferred to a test tube, and dried under argon. A slight change in the gradient system was used in the analysis of the yellow carrots to optimize the separation of lutein from zeaxanthin.

Identification and Quantification of Carotenoids. Carotenoids were identified by comparing their retention time and spectra with respective standards purified by HPLC in our lab immediately prior to use. Lutein was graciously obtained from Kemin Industries (Des Moines, IA). β -Carotene was purchased from a local General Nutrition Corporation outlet as softgels. α -Carotene was extracted from the high β -carotene carrots and purified twice with HPLC. Lycopene was extracted and purified from tomato paste. Purity was assured by spectral analysis on a UV-vis spectrophotometer and photodiode array HPLC as described above. β -Apo-8'-carotenyl decanoate was injected externally for comparison to the internal recovery to determine extraction efficiency. Calibration curves were created for each HPLC purified standard and used to quantify each carotenoid by comparing peak areas to the curve.

Sample Preparation. Carrots were stored in a sealed plastic bag under dark and chilled conditions at 2 °C. The carrots were washed, peeled, cut into similar-size carrot sticks (7–10 g) and stored in a plastic bag with \sim 5 mL of tap water to preserve freshness 1–2 days before they were sampled by panelists. After 2 days, the carrots were discarded and new ones were prepared. The samples were removed from the plastic storage bags and presented to each subject in small plastic cups labeled with the order of administration.

Sensory Evaluation. An untrained panel of students and faculty (n = 96, 34 male, 62 female) aged between 18 and 56 years, with an average carrot consumption of 2.3 per week, volunteered for the study. To qualify, panelists had to be nonsmokers, have no allergy to carrots, and agree to wear a blindfold during their first evaluation. A signed consent form was presented before the study began. Individual evaluations were performed in the Nutritional Sciences' research kitchen illuminated by normal lighting. The Social and Behavioral Science Institutional Review Board at UW-Madison approved the study design and informed consent form.

Carrot order was determined according to a randomized complete block design (27) so that each carrot type was randomly sampled first, second, third, fourth, and fifth, the same number of times and every volunteer ate each carrot independent of other volunteers. To test if color influences carrot sensory perception, volunteers were asked to wear a blindfold the first time they sampled the carrots and without a blindfold the second time. During their first visit, a trained questioner used a hedonic scale (27) to measure the degrees of how much the volunteers liked each carrot's flavor, sweetness, crispiness, and overall acceptance. Because the volunteers were not trained, the questioners were only looking for how much the volunteers *liked* each attribute. For example, in regards to sweetness, they were asked if they liked the sweetness, not how sweet is the carrot. To avoid confusion between the difference between carrot flavor and overall taste, the questioners told each subject that carrot flavor was in regards to familiarity to carrot, whereas overall taste represented the degree of preference for the vegetable itself. Each carrot stick was placed into a small plastic cup and handed to the volunteer in the proper order. The volunteers sampled each carrot and gave a number between 1 and 9, representing how much he or she liked or disliked each attribute, which was then marked on the ballot with scaled increments for each attribute. A different ballot was used for each carrot color. Number 1 was equivalent to the answer liked extremely, number 5 was neither like nor dislike, and 9 was equivalent to dislike extremely. After a number was given, the questioner repeated the answer to ensure the subject understood their choice. Thus, if the volunteer did not like the sweetness (i.e., it was too sweet or not sweet enough) they might give a score between 6 and 9. Volunteers were offered tap water between each carrot sample. At least 3 days later, subjects were asked to return to the research kitchen to repeat the process without a blindfold. During both visits, volunteers were free to comment on each carrot sample and comments were recorded on the bottom of each ballot.

Statistical Analysis. Statistical analyses were performed with SAS software (version 8.2; SAS Institute, Cary, NC) and R software (version 1.71). Mixed-effect models were generated for flavor, sweetness, crispiness, and overall. The logarithm transformation of the 1 through 9 indexed score was fitted as the response variable; the effects of treatment, whether blind or nonblind, color of carrot and their interaction (treatment \times color) were fitted as fixed terms and the participants were fitted as a random effect. Fisher's Least Significant Difference (LSD) method was used to compare the least-squares means of different colors of carrots.

RESULTS

Carotenoids. Extraction efficiencies for the colored carrots ranged from 76–105% and 64–79% for the white carrots. Lower extraction efficiencies were obtained in the white carrots, as the entire extract was used and losses probably occurred between the evaporation flask and test tube. These losses were corrected by the recovery of internal standard in the final calculations. The coefficient of variation for carotenoids run on the same day was 4%. All samples from the same carrot type were run in the same day. Four major carotenoids were identified and quantified by comparing UV spectra and HPLC retention times of standards to those found in the carrots. The mean (\pm SD) values for the carotenoids in the various carrots and typical orange for comparison are listed in **Table 1**. A typical chromatogram is shown in **Figure 1**.

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carrot type	α-carotene	β -carotene (β C)	lycopene	lutein	total
high- β C orange	3.1 ± 2.4	18.5 ± 2.8	1.7 ± 0.83	0.44 ± 0.07	28.3 ± 0.8
orange ^b	2.2 ± 0.8	12.8 ± 3.3	nd ^d	0.26 ± 0.08	15.2 ± 4.1
purple	4.1 ± 1.2	12.3 ± 5.1	nd	1.1 ± 0.73	17.5 ± 7.0
red	0.11 ^c	3.4 ± 0.89	6.1 ± 0.6	0.32 ± 0.26	9.8 ± 1.4
yellow	0.05 ^c	0.18 ± 0.17	nd	0.51 ± 0.27	0.71 ± 0.38
white	nd	0.006 ± 0.003	nd	0.009 ± 0.002	0.014 ± 0.001

concentrations of carotenoids (mg/100 g carrot)

^a Data are expressed as mean ± SD of three determinations on a fresh weight basis. ^b Typical orange carrots were not used in the sensory evaluation but are shown here for carotenoid comparison. ^c Carotenoid values were found in only one of the three carrots. ^d nd, not detected.

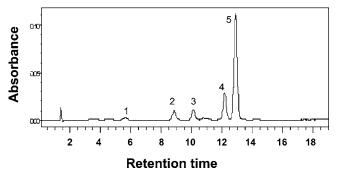


Figure 1. Typical chromatogram of carotenoids found in the high β -carotene orange carrots. Peak identification: (1) lutein, (2) lycopene, (3) β -apo-8'-carotenyl decanoate (used as an internal standard), (4) α -carotene, and (5) β -carotene.

Sensory Evaluation. Including both the blind and nonblinded data, the mean scores of the five carrots for all four organoleptic criteria were below the neither like nor dislike score of 5 (Figure 2). This suggests that the participants generally liked the carrots and gave them low scores with mean values ranging from 2.56 \pm 0.094 for the white carrot crispiness to 4.28 \pm 0.12 for purple with orange core sweetness. Applying Fisher's LSD method to the score means ($\alpha = 0.05$), we found the ranks of the carrots for all organoleptic criteria to be very consistent. The results showed that the carrots fell into two groups: (1) orange and white and (2) yellow, red, and purple carrots. The order for each attribute is as follows:

Flavor: orange < white < yellow, red, purple Sweetness: orange, white < yellow, red, purple Crispiness: white, orange < yellow, red, purple Overall: orange, white < yellow, red, purple

A significant treatment effect was found for flavor, sweetness, and overall acceptance (P < 0.05) but not for crispiness (P > 0.05). **Figure 3** shows that the measured scores were found to be higher when the subjects were treated blind than nonblind. Interestingly, there was no interaction between treatment and the color effects (P > 0.1 for all criteria), and the rank for the carrots was very consistent for both blind and nonblind treatments.

DISCUSSION

The purposes of this study were to analyze the carotenoid content for the carrots used in the sensory analysis and to determine if the average consumer liked the unusual colored carrots and if color influenced their perception. Sensory evaluations using untrained panelists provide reliable data on various foods (28). The sensory evaluation results in this study were consistent in that the high β -carotene and white carrots were

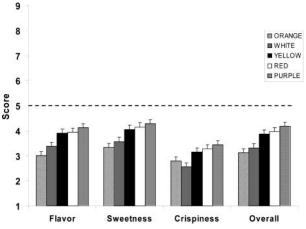


Figure 2. The mean score for combined treatments of the three sensory attributes and the overall acceptance score for five specialty carrots of various colors sampled by a consumer panel (n = 96). Error bars represent the least significant difference value. A score of 1 represents such as extremely and a score of 5 is neither like nor dislike.

favored over the other carrots in all four organoleptic criteria whether the subjects were blind-folded or not. The significantly lower scores found in the nonblind group compared to the blind group suggest that the participants preferred the carrots when they saw them.

The purple carrots tended to be the least favorite in all four criteria in nonblind and blind treatments, but not significantly different from red and yellow. This was contrary to Alasalvar et al.' (23) sensory evaluation; that is, five trained panelists found the purple carrots to be significantly sweeter than three other varieties, even though they had the lowest sugar content. The higher levels of terpenoids in their white and orange varieties may have masked the sweetness giving the purple carrots a higher relative sweetness. We did not evaluate levels of sugar and terpenes in our carrots, therefore the chemical differences accounting for the flavor observations between the studies cannot be compared. Consistent flavor characteristics are difficult to maintain due to the chemical and environmental diversity that influence taste perception (29). Because the genetic background of the carrots differed between the studies, we should not be surprised to find flavor perception differences. As neither carotenoids nor anthocyanins impact flavor, we expect that desirable flavor could be incorporated into carrots of any color with a concerted breeding effort.

While orange carrots are the most familiar, they were not the first to be cultivated. Orange and white carrots were recorded in the 17th century (6) while other carrots were documented earlier. By selecting carrots rich in orange color, plant breeders have developed carrots rich in β -carotene. The high β -carotene

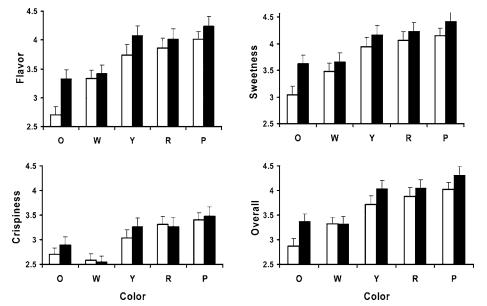


Figure 3. The difference in the mean scores from a consumer sensory panel (n = 96) for five specialty carrots of various colors when the subjects were either blindfolded (black bars) or not (white bars). A score of 1 represents like extremely and a score of 5 is neither like nor dislike. Colors evaluated: O, high β -carotene orange; W, white; Y, yellow; R, red; P, purple with orange core.

carrots were found to contain the greatest total carotenoid concentration, including lycopene, which was not detected in typical orange carrots. The bioavailability of α - and β -carotene has been documented (30-32); thus, the high levels of both carotenes may greatly improve vitamin A status if consumed on a regular basis, especially in developing countries where deficiency is a major health problem (33). In addition, carotenoids are associated with reduced risk for certain diseases. Alasalvar et al. found a variety of phenolics and volatiles in their orange carrots, which have their own disease-fighting capabilities (23). Thus, consumption of high β -carotene carrots could have a profound effect on health in both developed and developing countries. The white carrots were also favored in the sensory evaluation. Though low in carotenoids, Alasalvar et al. found the white carrots to contain the highest concentrations of total volatiles and variable amounts of phenolics (23). Moreover, as carrot fiber is of high quality, white carrots are not completely devoid of nutritional value.

The yellow and purple carrots were cultivated in Afghanistan in the 10th century (6). The carotenoids, lutein and β -carotene, in the yellow carrots are bioavailable (34), and accordingly may provide an alternative source of lutein to other vegetables and egg yolk, which is high in cholesterol and saturated fat. Plant breeders can select yellow carrots with higher lutein along with good flavor and as a result, provide a richer source of lutein. The cultivation and acceptance of yellow carrots could play a role in the prevention of age-related macular degeneration (10). The purple carrots have been cultivated to contain an orange core (6). β -Carotene concentrations in these carrots were not different than typical orange carrots. The high amount of various carotenoids in purple carrots suggests that they have pro-vitamin A activity and may be a good source of lutein. Alasalvar et al. reported these carrots to contain 2.2 and 2.3 times more α - and β -carotene than the typical orange carrots they studied (23), whereas we found this to be true for only α -carotene. In addition, the purple carrots contain the highest levels of phenolics and volatiles compared to orange, yellow, and white carrots (23), suggesting purple carrots have complex nutraceutical components.

The red carrots evolved in China and India around the 18th century (6). We found these carrots to be a significant source

of lycopene as well as β -carotene and lutein. Experiments have shown that the lycopene from the red carrot is bioavailable (25) and could provide a dietary alternative to tomatoes as a source of lycopene. For many individuals, tomatoes are not included in their diets because they are too acidic. Thus, the lycopene in the red carrot could play a role in the prevention of cancer and other chronic diseases if consumed on a regular basis.

Carotene content in carrots varies according to genotype, growing conditions, season, maturity, and storage (*35*, *36*). While a large variation of carotene concentrations were found within each carrot type, this should not discourage health experts from recommending specialty carrots with good flavor as a source of carotenoids for they are popular snacks, capable of being cultivated around the world, and have a long shelf life. With this information, plant breeders should be encouraged to develop these carrots to provide sources of both vitamin A precursors and phytochemicals.

ABBREVIATIONS USED

HPLC, high-performance liquid chromatography; SAS, statistical analyses software; LSD, least significant difference

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