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Original Research



Skin Carotenoid Response to a High-Carotenoid Juice in Children: A Randomized Clinical Trial

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ABSTRACT

Background Previous studies have shown an increase in serum carotenoid status among children when fed carotenoids. This study looked at the effect and dose–response of a known amount of carotenoid consumption on change in skin carotenoid status among children.

Methods Participants were children aged 5 to 17 years from Cache County, UT (n=58). Children were randomly assigned to one of three groups: high (n=18) or low (n=18) dose of a carotenoid-rich juice (2.75 mg carotenoids/30 mL juice), or placebo juice (n=22). Children were asked to drink an assigned dose of the juice (30 to 120 mL/day) based on the weight of the child and group assignment, every day for 8 weeks. Skin carotenoids were measured every 2 weeks by resonance Raman spectroscopy. Participants were asked to maintain their usual diet throughout the study. Usual diet was assessed using three averaged 24-hour recalls; diet constancy was measured using food frequency questionnaires administered at baseline, Week 4, and Week 8. Repeated measures analysis of variance was used to assess the group differences in skin carotenoid status over time.

Results The high-dose and low-dose groups had mean \pm standard deviation increases in skin carotenoid status of 11,515 \pm 1,134 and 10,009 \pm 1,439 Raman intensity counts, respectively (both *P* values <0.001, for change in means compared with baseline) at Week 8, although they showed significant change from baseline by Week 2. The placebo group's change of 985 Raman intensity counts was not statistically significant. The difference in change between the 2 experimental groups was not significant at Week 2, 4, 6, or 8.

Conclusions Consumption of 30 to 120 mL (2.75 to 11 mg carotenoids) of a carotenoidrich juice significantly increased skin carotenoid status over an 8-week period among children aged 5 to 17 years. The amount of carotenoids found in this amount of juice is equal to the amount found in approximately 23 to 92 g cooked carrots per day. J Acad Nutr Diet. 2015;115:1771-1778.

W UMEROUS STUDIES HAVE DEMONSTRATED THAT fruits and vegetables (F/V) can be protective against oxidative damage¹⁻³ and people who eat high amounts of F/V have lower risk for mortality and many chronic diseases.⁴ Highly pigmented red, orange, yellow, and green F/V are good sources of carotenoids as well as other antioxidants. Carotenoid status may be a biomarker of antioxidant status and F/V intake.^{5,6}

Carotenoids are fat-soluble pigments and when eaten are transported in the blood in lipoproteins and deposited in the lipid layer of the epidermis of human beings. Levels can be assessed by measuring the concentration of carotenoids in blood or skin. Previous studies in adults⁷ and children⁸ have identified strong correlations (r=0.81 and r=0.79, respectively; both *P* values <0.001) between the amount of

carotenoids measured in the blood using high-performance liquid chromatography and the amount measured in the skin, using resonance Raman spectroscopy (RRS).

Primary carotenoids detected in the skin are lycopene, beta carotene, alpha carotene, beta cryptoxanthin, lutein, zeaxanthin, phytoene, and phytofluene, with lycopene present in the highest amounts.⁹ Carotenoid levels in the diet have been shown to be significantly correlated with skin carotenoid status.^{8,10-13}

Studies have found that providing beta carotene supplements,^{14,15} high-carotenoid fruit,¹⁵ or high-carotenoid vegetables^{14,16,17} increase plasma beta carotene levels quickly in children. Jahns and colleagues¹⁸ found that skin carotenoid status measured by RRS closely followed changes in plasma carotenoid values in adults as total dietary carotenoid intake increased and decreased.¹⁸

The purpose of this study was to examine changes in skin carotenoid status in children, measured by RRS over an 8week period of time when a known dose of carotenoids was consumed in the form of juice with standardized carotenoid content.

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METHODS

Participants

A total of 67 healthy Cache County, UT, schoolchildren aged 5 to 17 years were recruited through local elementary and secondary schools during December 2012. The Institutional Review Board at Utah State University reviewed and approved the research protocol. A letter of information was sent via e-mail to parents of a local elementary school. Older children were recruited by word of mouth and from siblings of elementary schoolchildren. Researchers recruited a representative sample of approximately 8% of study children from each grade kindergarten through 12, 47% of whom were boys. The race/ethnicity of the participant population reflected the race/ethnicity of the local school-age population in Cache County, UT (80% white, 17% Hispanic, and <3% Asian and Pacific Islander). Parents of children completed a qualifying online survey. In the case that the child met the study inclusion/exclusion criteria, the parents were mailed/emailed and asked to complete a Center for Human Nutrition Studies health history questionnaire that includes past and present medical history, past and current medication use, and nutritional supplement use. Children were excluded if they had a health history or habits that were known to affect carotenoid levels.¹⁹ These exclusions included major illness during the 2 weeks before the study began, use of carotenoid supplements (multivitamins were allowed), use of topical self-tanning lotion, chronic disease such as asthma or type 1 diabetes, and sun exposure for >2 hours/day without use of sunscreen. Three children were excluded from the study due to a reported history of asthma and one due to rheumatoid arthritis.

Researchers obtained parent consent and child assent from participants in person. Participants were asked to maintain their normal lifestyle, including activity, multivitamin use, dietary pattern, and physical activity habits for the duration of the study. Participants received \$10 at clinic visits 2, 4, 6, and 8 and a \$20 bonus for completing the study, for a total of \$60.

Protocol

Qualifying children had a BioPhotonic Scanner (NuSkin, LLC) score, indicating skin carotenoid status, between 11,000 and 32,000 Raman intensity counts. The range was limited to eliminate subjects with abnormally high or low counts and to

stay within 2 standard deviations from the average score for school-aged children of approximately 22,000 Raman intensity counts (information provided by the manufacturer, NuSkin, LLC). Participants completed nine clinic visits scheduled 7 days apart at the Center for Human Nutrition Studies clinic at Utah State University. During the baseline clinic visit, height was measured using a Seca 223 digital stadiometer and weight was measured using a Detecto 758C digital scale. Height and weight (kg/m²) were used to calculate body mass index (BMI). BMI was categorized based on age and sex percentiles.

Participants were assigned sequentially into a randomgenerated matrix by the study coordinator. They were assigned to one of the three treatment groups (see Figure 1): Carotenoid-Rich Active-High (AH) (n=18), Carotenoid-Rich Active-Low (AL) (n=18), or Placebo (P) (n=22). Participants then received either a high dose of juice (30 mL twice per day if participant's weight was <39 kg or 60 mL twice per day if \geq 39 kg) or a low dose of juice (15 mL twice per day if participants weight was <39 kg or 30 mL twice per day if ≥39 kg), according to group assignment. Participants were asked to drink their prescribed amount of juice with a meal or snack for 8 weeks. The carotenoid-rich juice is commercially available under the name g3 Juice (NuSkin, LLC) and contained 2.75 mg carotenoids per 30 mL. The lot of juice that was used in the study was validated by a clinical laboratory (NuSkin Enterprises). Bottles were labeled with the same three-digit number on all of the placebo juice and a different number on the carotenoid-rich juice. Juice was measured and distributed by trained research assistants during weekly clinic visits. Research assistants were informed which bottle number to give to participants but did not know what that number represented. To check compliance, participants were asked to bring all juice containers (glass bottles) distributed to them at the prior week's clinic visit back to the clinic each week. Bottles were weighed using a Mettler Toledo balance to the nearest gram and recorded. Participants, parents, and clinic workers were blinded to the juice assignment. This was done by using juices made to have the same color and viscosity and numbering the juice bottles with one of two random three-digit numbers.

The following occurred at the baseline and at Week 2, 4, 6, and 8 clinic visits. Skin carotenoid status was measured by a trained research assistant using the BioPhotonic Scanner, a

	Carotenoid-Rich Juice				Placebo Juice
		\checkmark	И		\downarrow
Group Assignment:	Active-high (AH) Group Active-low (AL) Group				Placebo (P) Group
	\checkmark	Ы	Ľ	Ы	\downarrow
Weight of child:	>39 kg	<39 kg	>39 kg	<39 kg	>39 kg <39 kg
	\downarrow	\downarrow	\checkmark	\checkmark	\downarrow \downarrow
Juice consumed :	120 mL/d	60 mL/d	60 mL/d	30 mL/d	120 mL/d 60 mL/d

Figure 1. Juice assignments for 58 participants aged 5 to 17 years in a carotenoid dose-response study.

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