

Starch digestibility and β -carotene bioaccessibility in the orange-fleshed sweet potato puree-wheat bread

Daniel Mbogo, Tawanda Muzhingi, and Srinivas Janaswamy 

Abstract: Vitamin A is essential for vision, human health, growth, immune function, and reproduction. Its deficiency leads to anemia, xerophthalmia, and growth reduction in children. Foods enriched with naturally occurring carotenes have the potential, in this regard, and orange-fleshed sweet potato (OFSP) stands out tall as it is rich in β -carotene (β C), a provitamin A carotenoid. In view of developing OFSP-based functional foods to address the vitamin A deficiency (VAD) issues, herein, OFSP puree-wheat composite breads have been prepared at 10% to 50% OFSP puree concentrations and bioaccessibility of β C has been estimated. The total β C is found to be 4.3, 9.2, 16.5, 23.3, and 33.6 μ g/g in 10, 20, 30, 40, and 50% OFSP bread, respectively. The corresponding calculated retinol activity equivalents (RAE) are 30.9, 66.4, 119.5, 170.4, and 246.2 RAE/100 g. The efficiency of micellization of all-*trans*- β C, 13-*cis* β C, and 9-*cis* β C after simulated oral, gastric, and small intestinal digestion are 1.4% to 6.4%, 1.4% to 7.2%, and 1.1% to 6.9%, respectively. The amount of micellized β C correlates linearly with the OFSP concentration in the bread. Furthermore, *in vitro* starch digestion decreases with significant reduction in the Rapidly Digestible Starch (RDS) amount coupled with increase in the Slowly Digestible Starch (SDS) and Resistant Starch (RS) fractions. Overall, OFSP-wheat composite bread holds adequate amount of provitamin A carotenoids. The amount of bioaccessible β C coupled with altered starch digestion of the OFSP wheat breads highlight their usefulness as novel functional foods that could address the VAD as well as glycemic issues toward improving human health.

1. INTRODUCTION

Quoting Helen Keller “Of all the senses, sight must be the most delightful.” Vitamin A is essential for vision. It also controls human health, growth, immune system function, and reproduction. Its deficiency sets xerophthalmia, growth reduction in children, and anemia along with increased morbidity and mortality (Sherwin et al., 2012). Indeed, vitamin A insufficiency (VAD) is a global public health concern. It is estimated that yearly 2,50,000 to 5,00,000 children around the globe become blind with about 50% fatality within 12 months of the sight failing (<https://www.who.int/nutrition/topics/vad/en/>). In pregnant women, VAD could even increase the risk of maternal mortality and 19.1 million are projected with the low serum retinol concentration of $<0.70 \mu$ mol/L (Darnton-Hill et al., 2017). Incidentally, the Sub-Saharan Africa (SSA) and South Asia (SA) regions bear the highest burden of the VAD (WHO, 2009). Strategies, such as dietary diversification, food fortification, and distribution of high-dose vitamin A capsules, are being explored to combat the VAD but are far from providing the much-needed solution (Stevens et al., 2015). In this regard, plant-based and biofortified crops-based foods, such as sweet potatoes, that are rich in β -carotene (β C), a provitamin A carotenoid, prominently offer cost-effective and sustainable alternatives (Low

et al., 2020). Sweet potatoes are the seventh most important food crops produced in the world. They serve as an energy supplement, and are good source of β C, anthocyanins, polyphenols, starch, soluble fiber, and minerals. Their intrinsic antioxidant and anticarcinogenic properties further underscore the benefits in serving as a health-promoting staple food source. Generally, there are four colored sweet potatoes known as white, yellow, orange, and purple, and among them orange-fleshed sweet potatoes (OFSPs) possess the highest amount of β C (Tang et al., 2015).

In the SSA and SA regions, OFSP is being grown and promoted as a food-based strategy to address the VAD (Chaudhary, & Sahani, 2017). The recommended dietary allowance (RDA) of retinol activity equivalent (RAE) per day for men and women is 900 and 700 μ g, respectively. The OFSP, like ordinary sweet potato, is traditionally consumed plain as boiled or steamed. However, incorporating OFSP in processed products, such as breads and buns, certainly expands its widespread utility and acceptability, and to address the VAD concerns. Fresh OFSP roots are bulky and perishable and, thus, their wholesome and long-term utility is limited. Processing the roots to chips and/or flour is a practical approach, as they would be shelf-stable and have less storage and transportation hurdles. Though the total amount of β C in the OFSP flour is reduced as compared to fresh OFSPs (Chilungo et al., 2019), baked products, such as breads and buns, incorporated with the OFSP flour retain considerable carotene; but processing, drying, storing, and packaging wilt carotenoid amount (Nzamwita et al., 2017), warranting alternative strategies to maximize carotenoid retention in foods. In this regard, OFSP puree (steamed and mashed OFSP) would be highly desirable. Purees are prepared by peeling, cutting, grinding, and cooking sweet potatoes in water. Most importantly, this process withholds more than 90% of carotenoids. Thus, incorporating the OFSP puree in foods is highly desirable and this ideology has been adapted in this research by incorporating

JFDS-2020-1929 Submitted 10/25/2020, Accepted 1/5/2021. Authors Mbogo and Muzhingi are with Food and Nutritional Evaluation Laboratory, Biosciences for East and Central Africa at International Livestock Research Institute (ILRI), International Potato Center (CIP) Sub-Saharan Africa Regional Office, P. O. Box 25171, Nairobi, Kenya. Author Muzhingi is with Department of Food, Nutrition and Bioprocessing, Schaub Hall, North Carolina State University, Raleigh, NC, 27607. Authors Mbogo and Janaswamy are with Department of Dairy and Food Science, South Dakota State University, Brookings, SD, 57007. Direct inquiries to author Janaswamy (E-mail: Srinivas.Janaswamy@sdstate.edu).

various levels of OFSP puree in the wheat bread. Several bakeries and supermarkets in Kenya, Rwanda, Malawi, and Mozambique are exploring the commercial success of such food products (Bocher et al., 2017). Breads with 10% OFSP flour contain 0.04 $\mu\text{g}/100\text{ g}$ of βC in the crumb and 0.05 $\mu\text{g}/100\text{ g}$ in the crust that are equivalent of 3.9 and 5.4 RAE/100 g from crumb and crust, respectively (Muzhingi et al., 2016). Instead, OFSP puree substitution of 45% results in 1.36 and 1.05 μg of βC , which translates to 148 and 98 RAE/100 g from the crumb and crust, respectively. These examples accentuate the advantages of OFSP puree fortification in breads so as to gain good amounts of RAE; indeed, bread as the staple food also serves as a sustainable tool to address the VAD.

Generally, processing affects the amount of bioaccessible βC in food products mainly due to exposure to heat, light, and oxygen, to name a few (Berni et al., 2014). Several *in vitro* protocols have been developed to estimate the bioaccessible βC in biofortified staple foods such as raw and processed OFSP (Failla et al., 2009), OFSP flour (Trancoso-Reyes et al., 2016), and cassava genotypes (Thakkar et al., 2007). The micellized βC , for example, after simulated oral, gastric, and small intestinal phase digestion of boiled and highly processed OFSP ranges from 0.6% to 3.0% (Failla et al., 2009). Hence, estimation of bioaccessible βC from the OFSP breads is in need for the large-scale application and effective utilization of the OFSP breads.

Furthermore, diabetes mellitus (diabetes for brevity) affects 425 million people globally with an expected 48% rise by 2045. Its incidence is prone to mortalities by triggering kidney failure, nerve damage, cardiovascular disease, and stroke. The total direct and indirect costs associated with this ailment make it one of the most expensive. The worldwide spending in 2017 was \$727 billion, around 12% of the global health expenditure (<https://www.diabetesatlas.org/>). Diabetes is characterized by the elevated levels of glucose in the blood (hyperglycemia) as the body cannot produce required quantities of insulin and/or cells are unable to respond to insulin. Dietary measures that abate elevated blood glucose are recommended and food products with controlled starch digestion will help to deliberate the progression as well as prevention. In sweet potato, up to 90% of the dry matter is made up of carbohydrates of which starch alone accounts up to 80%. The proportion of amylose and amylopectin is 30% to 40% and 60% to 70%, respectively (Aina et al., 2009; Zhu et al., 2011). The rate of starch digestion, especially Rapidly Digestible Starch (RDS), Slowly Digestible Starch (SDS), and Resistant Starch (RS) are influenced by amylose and amylopectin content along with phytochemicals, dietary fiber, protein, and fat as well as the processing methods and cultivar variations (Ngoc et al., 2017). In addition, several studies signify the role of polyphenols on modulating the α -amylase activity toward reducing the starch digestion (McDougall et al., 2005; Li, Koecher, Hansen, & Ferruzzi et al., 2017). In this respect, a decrease in starch digestion by the OFSP puree-wheat bread could be expected and if so, it certainly expands the utilization of OFSP beyond the vitamin A addressor.

The OFSP puree-wheat composite breads have potential to accost the VAD concerns and stand out as a healthy alternative food product to curb chronic illness such as diabetes. In this regard, it is essential to generate data on the bioaccessibility of βC and starch digestibility of the OFSP puree-wheat breads. The objective of this study was to prepare a series of wheat breads by substituting the wheat flour with 10, 20, 30, 40, and 50% OFSP puree and to determine the βC amount and bioaccessible βC upon simulated digestion along with understanding the role OFSP in modulating

the starch digestion. The obtained results suggest that the amount of βC after digestion is dependent on the amount of OFSP puree substitution, and more importantly starch digestion is moderated, which certainly expands the OFSP utilization beyond the VAD applications and could as well serve as a catalyst to address the diabetes concerns.

2. MATERIALS AND METHODS

2.1 Materials

Required chemicals and supplies were purchased from MilliporeSigma, Fisher Scientific, and VWR. Carotenoid standards (all-trans- βC , 9-cis- βC , 13-cis- βC , and echinenone) were procured from CaroteNature GmbH, Ostermundigen, Switzerland. Home baking wheat flour, instant dry yeast, sugar, dough improver, and table salt, for breadmaking, were from local supermarket.

2.2 OFSP puree

Medium-size OFSP roots purchased from the local supermarket were hand washed and peeled using kitchen potato peeler. They were then cooked by steaming for 30 min in a kitchen steamer. The cooked sweet potatoes were removed from the steamer and allowed to cool at room temperature for 1 hr and were then mashed into puree, packed into 1 kg Ziploc bags, and frozen at $-20\text{ }^{\circ}\text{C}$ until use.

2.3 OFSP puree-wheat composite bread

Breads were formulated using baking flour and OFSP puree. The details are as follows: wheat flour 480 g, vegetable fat 15 g, instant baker's yeast 5 g, salt 4.5 g, sugar 18 g, dough improver 1 g, and water 55 to 220 mL. The breads were prepared by replacing 10, 20, 30, 40, and 50% weight of wheat flour with the OFSP puree. The dough containing pure wheat flour served as the control. Dough mixing, fermentation, and proofing and baking was done using a benchtop bread machine (SKG 2LB Automatic Programmable Bread Maker). The baking processes was based on the literature protocol (Wanjuu et al., 2018) but with some modification. Briefly, ingredients were weighed into a pan of the bread machine and mixed for 15 min with a 5 min rest after 10 min. Dough proofing was allowed for 1 hr during which yeast in the dough produces carbon dioxide allowing the dough to rise to desirable volume. The dough was then baked at $200\text{ }^{\circ}\text{C}$ for 40 min, after which the bread was depanned and allowed to cool to room temperature. The breads were frozen at $-20\text{ }^{\circ}\text{C}$ in air-tight packs until analysis.

2.4 Simulated digestion

Bread samples of 3 g were subjected to simulated oral, gastric, and small intestinal phases of digestion according to the reported protocols (Failla et al., 2009) but with some modification. The samples were grounded using a kitchen blender and each of the sample was digested in triplicate in 6 mL of oral phase containing 3,015 units of α -amylase in a basal solution with KCl, Na_3PO_4 , Na_2SO_4 , NaCl, and NaHCO_3 at concentrations of 24.0, 10.8, 8.0, 10.1, and 0.04 mmol/L, respectively, along with urea, uric acid, and mucin of 0.4, 0.03, and 5 mg/mL, respectively. The pH was adjusted to 6.7 using 1.0 M NaOH and blanketed with nitrogen gas and incubated in a shaking water bath at $37\text{ }^{\circ}\text{C}$ for 10 min. At the gastric phase, 30 mL of 0.9% NaCl solution was added and pH adjusted to 3.5 with 1.0 N HCL. Subsequently, 2 mL of pepsin (10 mg/mL) was added, pH adjusted further to 2.5 with 1.0 N HCL, and the final volume was brought to 40 mL with 0.9% NaCl solution. Later, blanketed with the nitrogen gas and incubated in a shaking water

bath at 37 °C for 1 hr. The intestinal phase was initiated by adjusting the pH to 5.0 with 1.0 N NaOH, and then adding 2 mL (20 mg/mL pancreatin and 10 mg/mL lipase), 3 mL of bile extract (30 mg/mL), and pH adjusted to 6.5 with 1.0 N NaOH. This was blanketed with nitrogen gas and incubated in a shaking water bath at 37 °C for 2 hr. After the small intestinal phase of digestion, 15 mL of digesta was aliquoted in tubes and stored at -80 °C. The remaining digesta was centrifuged (12,000×g, 4 °C, 45 min) and the supernatant was passed through 0.22 µm microfilter to obtain the bioaccessible aqueous fraction. Aliquots of aqueous fraction was then stored at -80 °C to analyze the micellarized carotenoids within a week. The digestive stability was calculated as percentage recovered βC in the digesta after oral, gastric, and intestinal digestion. Efficiency of micellarization was calculated as percentage of βC in the aqueous phase compared to amount in the digesta (Thakkar et al., 2007; Berni et al., 2015).

2.5 Extraction and analysis of β-carotene in test sample, digesta, and aqueous fraction

Carotenoid extraction in bread samples was performed by saponification technique (Kurilich, & Juvik, 1999) but with some modification. Extraction from digesta and the aqueous fraction was done by adding 3.0 mL petroleum ether:acetone (2:1) containing (0.1% BHT in the petroleum ether) to 4 mL sample, mixing on a vortex mixer for 1 min, centrifuging at 800×g for 5 min, and collecting the upper phase. The procedure was repeated three times and petroleum ether fractions were combined and dried at room temperature under a stream of nitrogen gas. It was then reconstituted in 150 µL ethyl acetate:methanol (1:1) and analyzed immediately using the HPLC (Chitchumroonchokchai et al., 2004). Echinone was used as internal standard to estimate the efficiency of recovery of carotenoids during extraction.

The HPLC system consists of Waters 717 plus autosampler, Waters 1525 binary HPLC pump, and Waters 2487 dual λ absorbance detector. A reverse-phase gradient method was used with an YMC C30, carotenoid column (3 µm, 150 × 3 mm, YMC Wilmington, NC). Mobile phase A: methanol/tert-butyl methyl ether/water (85:12:3, v/v/v, with 1.5% ammonium acetate in the water). Mobile phase B: methanol/tert-butyl methyl ether/water (8:90:2, v/v/v, with 1% ammonium acetate in the water). All-trans-βC, 9-cis-βC, 13-cis-βC were identified using absorption spectra and retention time of pure standards, and quantities estimated using the external standard curve of pure carotenoids, after correcting for extraction efficiency based on the recovery of echinenone (Trumbo et al., 2001).

2.6 In vitro starch digestion

The *in vitro* starch digestion of bread samples was measured using the Englyst et al. (1992) protocol with some modifications. The sample weighed in duplicate to be 550 mg of starch equivalent was suspended in 20 mL sodium acetate buffer solution containing 50 mg guar gum and stabilized at 37 °C in a water bath. An enzyme preparation that contained pancreatin (4.5 g in 30 mL) and amyloglucosidase (3.9 mL) was added and immediately put in the shaking water bath set at 37 °C and 150 rpm. During digestion, 0.5 mL aliquots were removed at 2, 5, 10, 15, 20, 30, 60, 90, and 120 min and mixed with 20 mL of 80% v/v ethanol. Megazyme GOPOD assay kit was used to measure glucose, in duplicate, in the mixture and referred as G₂, G₅, G₁₀, G₁₅, G₂₀, G₃₀, G₆₀, G₉₀, and G₁₂₀ (for brevity) in the rest of the discussion. The total starch (TS) in the bread samples was measured using the amyloglucosidase/α-amylase method (AACC, 2009). The G₂₀, G₁₂₀, and TS values were

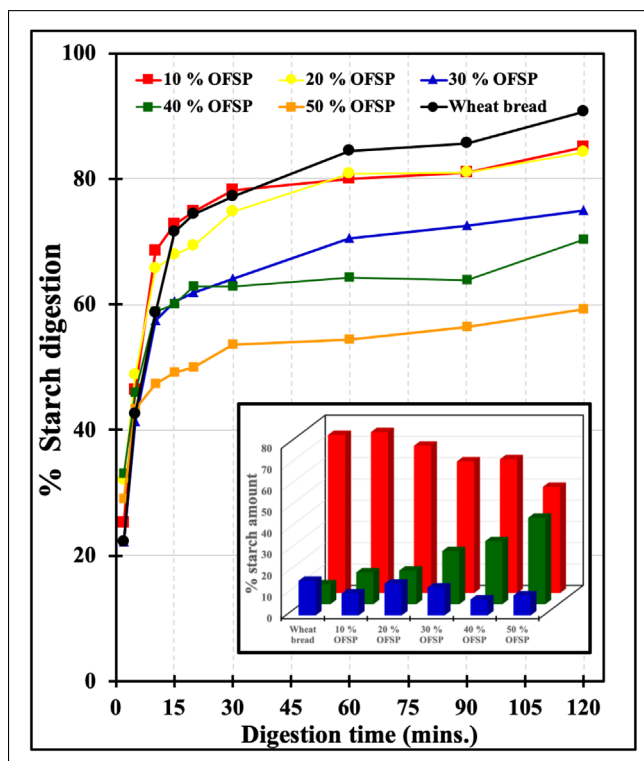


Figure 3-Starch digestion profiles of orange-fleshed sweet potato (OFSP) pure-wheat bread as a function of digestion time. The inset highlights amount of Rapidly Digestible Starch (RDS) (red), Resistant Starch (RS) (green), and Slowly Digestible Starch (SDS) (blue) of the prepared breads.

used to calculate RDS, SDS, and RS amounts using the following equations:

$$\text{RDS} = G_{20} \times 0.9$$

$$\text{SDS} = G_{20} \times 0.9$$

$$\text{RS} = \text{TS} - (\text{RDS} + \text{SDS})$$

2.7 Statistical analysis

Data were expressed as mean values from triplicate measurements. The variation in recovery and micellarization of βC after simulated digestion and starch fractions, among the OFSP bread were determined by the one-way analysis of variance (ANOVA) followed by the Duncan's multiple range test ($p < 0.05$) using Gen-Stat 15th edition statistical software. The correlation analysis between the micellarized βC and total βC in the OFSP breads and micellarization efficiency and total digestible starch was carried out using the Pearson correlation coefficients.

3. RESULTS AND DISCUSSION

3.1 Carotenoid composition and retinol activity equivalent (RAE) of OFSP bread

The three βC isomers namely all-trans-βC, 13-cis-βC, and 9-cis-βC have been detected in the OFSP composite bread whereas only all-trans-βC in the wheat bread (Table 1). The total βC has been found to be 4.3, 9.2, 16.5, 23.3, and 33.6 µg/g fresh weight in the 10, 20, 30, 40, and 50% OFSP bread, respectively. The corresponding calculated RAEs are 30.9, 66.4, 119.5, 170.4, and 246.2 RAE/100 g. Based on RDA for children of 1 to 3 and 4 to 8 years,

Table 1—The β -carotene (β C) concentrations, calculated Retinol Activity Equivalent (RAE),¹ Rapidly Digestible Starch (RDS), Slowly Digestible Starch (SDS), and Resistant Starch (RS) fractions in the wheat bread and OFSP wheat breads with orange-fleshed sweet potato (OFSP) puree of 10, 20, 30, 40, and 50%.

% OFSP	Concentration (μ g/g fresh weight)			RAE	Starch fraction (% w/w)		
	All-trans- β C	13-cis- β C	9-cis- β C		RDS	SDS	RS
Wheat bread	0.013 $\pm$ 0.002 ^a	—	—	0.11	74.4 $\pm$ 1.9 ^a	16.3 $\pm$ 2.9 ^a	9.4 $\pm$ 1.2 ^a
10	3.134 $\pm$ 0.112 ^b	1.014 $\pm$ 0.050 ^a	0.132 $\pm$ 0.005 ^a	30.89	75.7 $\pm$ 6.2 ^a	10.3 $\pm$ 1.2 ^{bc}	15.0 $\pm$ 3.7 ^b
20	6.752 $\pm$ 0.305 ^c	2.199 $\pm$ 0.103 ^b	0.239 $\pm$ 0.009 ^b	66.42	69.3 $\pm$ 2.2 ^b	14.9 $\pm$ 3.5 ^a	15.8 $\pm$ 2.8 ^b
30	12.224 $\pm$ 0.523 ^d	3.814 $\pm$ 0.416 ^c	0.413 $\pm$ 0.044 ^c	119.48	61.9 $\pm$ 0.4 ^c	13.1 $\pm$ 0.6 ^{ab}	25.0 $\pm$ 0.3 ^c
40	17.614 $\pm$ 0.065 ^e	5.148 $\pm$ 0.084 ^d	0.515 $\pm$ 0.018 ^d	170.39	62.9 $\pm$ 1.6 ^c	7.4 $\pm$ 1.8 ^c	29.7 $\pm$ 0.3 ^d
50	25.507 $\pm$ 2.019 ^f	7.306 $\pm$ 0.698 ^e	0.769 $\pm$ 0.084 ^e	246.21	50.0 $\pm$ 1.6 ^d	9.4 $\pm$ 1.7 ^c	40.7 $\pm$ 2.0 ^e

¹ 1 RAE = 12 μ g/g of trans- β C or 24 μ g/g of cis- β C and expressed per 100 g.

Data followed by the same superscript letters in the each column are not significantly different ($p < 0.05$).

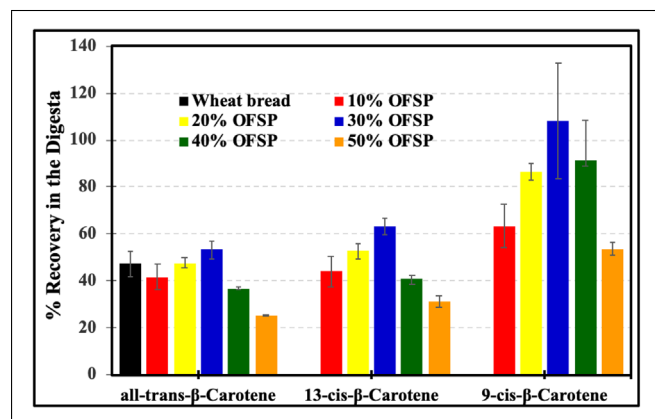


Figure 1—The percentage recovery of all-trans- β -carotene, 13-cis- β -carotene, and 9-cis- β -carotene in the orange-fleshed sweet potato (OFSP) composite bread after simulated oral, gastric, and small intestinal phases of digestion. Data represent mean $\pm$ standard deviation from six samples.

pregnant and lactating women and women of 14 to 18 and 19 to 50 years (Trumbo et al., 2001), the percentage contribution of 100 g of 40% OFSP bread is found to be 57, 43, 23, 23, 14, and 13%, respectively. These values further boost to 82, 62, 33, 33, 21, and 19%, in the same order, with the 50% OFSP bread.

3.2 Digestive stability and micellarization efficiency of β C in the OFSP composite bread

The three β C isomers detected in the OFSP breads have been recovered after the simulated oral, gastric, and small intestinal phases of digestion. The percentage recovery of all-trans- β C is significantly different ($p < 0.05$) in the OFSP breads and appears to increase until 30% OFSP substitution but declines at higher amounts (Figure 1). The proportion of 13-cis- β C and 9-cis- β C isomers increases during digestion, which is attributed to isomerization of all-trans to cis isomers. The amount of β C incorporated into micelles ranges from 0.03 to 0.61 μ g/g of the OFSP bread and are in line with the OFSP percentage. The efficiency of micellarization of β C ranges from 1.4% to 6.4% for all-trans- β C, 1.4% to 7.3% for 13-cis- β C, and 1.7% to 6.9% for 9-cis- β C (Figure 2). Overall, the micellized β C quantity is positively correlated ($r = 0.8927$; $p < 0.05$) with the total β C in the OFSP bread.

In this research, wheat bread was formulated by incorporating varying levels (10, 20, 30, 40, and 50%) of OFSP puree into the bread dough, which significantly increased the β C content in the bread and resulted in higher RAE. For example, with 40 and 50% OFSP substitution as high as 170 and 246 RAE/100 g of bread has been observed. These values agree with the reported 148

RAE/100 g of 45% OFSP puree–bread prepared in a commercial bakery (Muzhingi et al., 2016); observed subtle differences could be due to variations in the OFSP variety used in respective studies. The efficiency of micellarization of carotenoids during the *in vitro* digestion has been shown to correlate well with the micellized carotenoids from *in vivo* human studies and, thus, *in vitro* digestion would be a useful tool to assess the bioaccessibility of carotenoids in food products. In this study, micellarization efficiency of the all-trans- β C is observed to be in the range 1.4% to 6.4% and is slightly higher than the 0.6% to 3% of boiled OFSP cultivars (Failla et al., 2009) and 0.5% to 1.1% of heat-processed OFSP (Bengtsson et al., 2009). Interestingly, micellarization efficiency is found to increase by about 50% with the addition of fat (Bengtsson et al., 2010). Similarly, OFSP flour blended flat breads, *chapatis*, baked with oil also display increased micellarization efficiency of β C (Bechoff et al., 2011; Chilungo et al., 2019). Presence of fat along with severe maceration during OFSP puree preparation might aid to break the matrix containing the β C leading to higher efficiency in the OFSP puree–wheat bread. Further research is warranted to decipher the role of fat on the micellarization efficiency of β C.

3.3 *In vitro* starch digestibility of the OFSP composite bread

The *in vitro* starch digestion profiles are shown in Figure 3. The percentage of starch digested from the wheat bread and 10% to 40% OFSP breads is comparable during the first 20 min but varies for the higher substitution of 50% OFSP bread. All the bread samples follow similar starch digestion pattern but variations are quite apparent during the 60 to 120 min digestion. At 60 min, 71, 64, and 54% of starch has been digested in the 30, 40, and 50% OFSP breads, respectively. The corresponding amounts of RDS, SDS, and RS are listed in Table 1. In the wheat bread, they are 74.4, 16.3, and 9.4%, respectively. However, with the presence of OFSP, these amounts change appreciably. For example, in the 50% OFSP bread they are 50.0, 9.4, and 40.7%, respectively. After 120 min, the RS fraction for the wheat bread, 10, 20, 30, and 50% OFSP bread is 9, 15, 16, 25, 30, and 41%, respectively, suggesting that partial substitution of wheat flour with the OFSP puree significantly improves the RS content in the wheat bread.

The importance of glycemic response to starchy foods in the management of diabetes is well documented (Jenkins et al., 1988). Sweet potato is a low to medium glycemic index food and thus stands out as a useful food product for diabetes management (Allen et al., 2012). The dietary fiber fraction RS in the OFSP breads is significantly high and agree well with the findings on sweet potato incorporated steamed rice bowl cake (Chou, & Li, 2018). Indeed, an inverse relationship between RS of sweet potato and glycemic

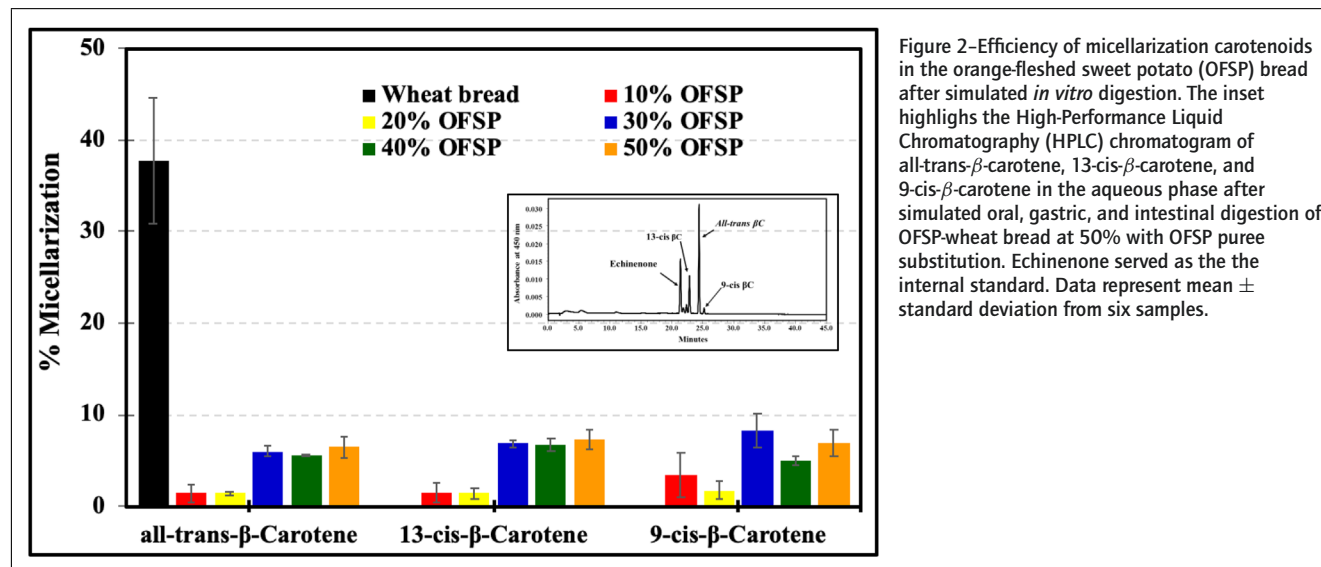


Figure 2-Efficiency of micellization carotenoids in the orange-fleshed sweet potato (OFSP) bread after simulated *in vitro* digestion. The inset highlights the High-Performance Liquid Chromatography (HPLC) chromatogram of all-trans- β -carotene, 13-cis- β -carotene, and 9-cis- β -carotene in the aqueous phase after simulated oral, gastric, and intestinal digestion of OFSP-wheat bread at 50% with OFSP puree substitution. Echinone served as the internal standard. Data represent mean $\pm$ standard deviation from six samples.

index has been observed (Odenigbo et al., 2012). In this regard, the OFSP breads prepared in this research appear to suit the needs to improve gut health. Several reports highlight the role of bioactive compounds in the starch matrix and their favorable impact on RS, SDS, and glycemic index. In the present case, β C and other phytochemicals present in the OFSP appear to modulate the starch digestion. The decreased RDS and increased RS amounts suggest that OFSP wheat breads could be a healthy alternative to counter glycemic issues and improve gut health. Interestingly, postharvest storage of sweet potatoes is found to influence the short-range starch ordered structures, molecular weight distribution, amylose amount, and polyphenol content (Lu et al., 2020). These changes do regulate the sweet potato starch digestion. Furthermore, since sweet potato starch is composed of granules with various sizes, its functional properties get influenced (Ye et al., 2020), so could be expected of the OFSP food products. What remains is scouting for suitable sweet potato cultivar(s) along with appropriate postharvest storage conditions that not only possess high β C amount but also suitable starch digestion properties.

4. CONCLUSION

Since vitamin A cannot be synthesized by the human body, its deficiency is a major health nutrition problem. In this regard, OFSPs offer a potentially viable and simple solution. OFSPs are rich source of β C and their inclusion in daily foods would indeed help to raise the vitamin A level as required. Herein, a series of wheat breads have been prepared by incorporating 10, 20, 30, 40, and 50% OFSP puree in the wheat flour. The total β C is found to be 4.3, 9.2, 16.5, 23.3, and 33.6 μ g/g, respectively, with the RAE of 30.9, 66.4, 119.5, 170.4, 246.2 RAE/100 g, in the same order. These high RAE values could certainly aid to combat the VAD issues. The bioaccessibility of β C in the OFSP breads correlates highly with the total β C amount. Interestingly, incorporation of the OFSP puree in wheat bread alters starch digestion with increased RS amounts. Such starch digestion profile could be beneficial to evade diabetes. Overall, sweet potato appears to readily signify the dictum "Let food be thy medicine and let medicine be thy food." Further research to understand the β C uptake by intestine cells, bread sensory attributes along with glycemic studies on larger cohort is planned.

ACKNOWLEDGMENTS

Support is from Norman E. Borlaug International Agricultural Science and Technology Fellowship Program & CGIAR Borlaug Fellowship Program (FX17BF-10777R.033) and USDA National Institute for Food and Agriculture (SD00H648-18).

AUTHOR CONTRIBUTIONS

DM conducted experiments and drafted the original manuscript. TM contributed to research discussion, writing, and reviewing. Research conception was by SJ along with gaining funding, writing, reviewing, and editing.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest to declare.

REFERENCES

- AACC, Approved Methods of Analysis, Method 76-13.01. (2009). Total starch assay procedure-megazyme amyloglucosidase/alpha-amylase method, Megazyme, Bray.
- Aina, A.J., Falade, K.O., Akingbala, J.O., & Titus, P. (2009). Physicochemical properties of twenty-one Caribbean sweet potato cultivars. *International Journal of Food Science & Technology*, 44(9), 1696–1704. <https://doi.org/10.1111/j.1365-2621.2009.01941.x>
- Allen, C.J., Corbitt, A.D., Maloney, K.P., Butt, M.S., & Truong, V.D. (2012). Glycemic index of sweet potato as determined by cooking methods. *Open Nutrition Journal*, 6(1), 1–11. <https://doi.org/10.2174/1874288201206010001>
- Bechoff, A., Poulaert, M., Tomlins, K. I., Westby, A., Menya, G., Young, S., & Dhuique-Mayer, C. (2011). Retention and bioaccessibility of β -carotene in blended foods containing orange-fleshed sweet potato flour. *Journal of Agricultural and Food Chemistry*, 59(18), 10373–10380. <https://doi.org/10.1021/jf1201205y>
- Bengtsson, A., Alminger, M.L., & Svanberg, U. (2009). In vitro bioaccessibility of β -carotene from heat-processed orange-fleshed sweet potato. *Journal of Agricultural and Food Chemistry*, 57(20), 9693–9698. <https://doi.org/10.1021/jf901692r>
- Bengtsson, A., Brackmann, C., Enejder, A., Alminger, M.L., & Svanberg, U. (2010). Effects of thermal processing on the in vitro bioaccessibility and microstructure of β -carotene in orange-fleshed sweet potato. *Journal of Agricultural and Food Chemistry*, 58(20), 11090–11096. <https://doi.org/10.1021/jf1024104>
- Berni, P., Chitchumroonchokchai, C., Canniatti-Brazaca, S.G., De Moura, F.F., & Failla, M.L. (2014). Impact of genotype and cooking style on the content, retention and bioaccessibility of β -carotene in biofortified cassava (*Manihot esculenta* Crantz) conventionally bred in Brazil. *Journal of Agricultural and Food Chemistry*, 62 (28), 6677–6686.
- Berni, P., Chitchumroonchokchai, C., Canniatti-Brazaca, S.G., De Moura, F.F., & Failla, M.L. (2015). Comparison of content and in vitro bioaccessibility of provitamin A carotenoids in home cooked and commercially processed orange-fleshed sweet potato (*Ipomea batatas* Lam). *Plant Foods for Human Nutrition*, 70(1), 1–8. <https://doi.org/10.1007/s11130-014-0458-1>
- Bocher, T., Low, J.W., Muoki, P., Magnaghi, A., & Muzhingi, T. (2017). From lab to life: Making storable orange-fleshed sweet potato puree a commercial reality. *Open Agriculture*, 2, 148–154.
- Chaudhary, R.C., & Sahani, A. (2017). Sustainable remedy of vitamin A deficiency through bio-fortified golden sweet potato. *International Journal of Tropical Agriculture*, 35, 113–119.
- Chilungo, S., Muzhingi, T., Truong, V.D., & Allen, J.C. (2019). Effect of processing and oil type on carotene bioaccessibility in traditional foods prepared with flour and puree from orange-fleshed sweet potatoes. *International Journal of Food Science & Technology*, 54, 2055–2063. <https://doi.org/10.1111/ijfs.14106>

- Chitchumroonchokchai, C., Schwartz, S.J., & Failla, M.L. (2004). Assessment of lutein bioavailability from meals and a supplement using simulated digestion and Caco-2 human intestinal cells. *Journal of Nutrition*, 134 (9), 2280–2286. <https://doi.org/10.1093/jn/134.9.2280>
- Chou, C. & Li, M. (2018). A research of effect of three sweet potato varieties and addition on resistant starch content and physical characteristics of steamed rice bowl cake. *Journal of Food and Nutrition Research*, 6 (9), 551–556. <https://doi.org/10.12691/jfnr-6-9-2>
- Darnton-Hill, A., Neufeld, L., Vossenaar, M., Osendarp, S. & Martinez, H. (2017). *Large-scale food fortification: An overview of trends and challenges in low-and middle-income countries in 2017*. Micronutrient Forum.
- Englyst, H.N., Kingman, S., & Cummings, J. (1992). Classification and measurement of nutritionally important starch fractions. *European Journal of Clinical Nutrition*, 46 (S2), S33–S50.
- Failla, M.L., Thakkar, S.K., & Kim, J.Y. (2009). In Vitro bioaccessibility of β -carotene in orange-fleshed sweet potato (*Ipomoea batatas*, Lam.). *Journal of Agricultural and Food Chemistry*, 57 (22), 10922–10927. https://doi.org/10.1096/afscbj.30.1_supplement.lb433
- Jenkins, D.J., Wolever, T.M., Buckley, G., Lam, K. Y., Giudici, S., Kalnuskus, J., Jenkins, A. L., Patten, R. L., Bird, J., & Wong, G. S. (1988). Low-glycemic-index starchy foods in the diabetic diet. *American Journal of Clinical Nutrition*, 48 (2), 248–254. <https://doi.org/10.1093/ajcn/48.2.248>
- Kurilich, A.C., & Juvik, J.A. (1999). Quantification of carotenoid and tocopherol antioxidants in Zea mays. *Journal of Agricultural and Food Chemistry*, 47 (5), 1948–1955. <https://doi.org/10.1021/jf981029d>
- Li, M., Koecher, K., Hansen, L. & Ferruzzi, M.G. (2017). Phenolics from whole grain oat products as modifiers of starch digestion and intestinal glucose transport. *Journal of Agricultural Food Chemistry*, 65 (32), 6831–6839. <https://doi.org/10.1021/acs.jafc.7b02171>
- Low, J. W., Ortiz, R., Vandamme, E., Andrade, M., Biazin, B., & Gruneberg, W. L. (2020). Nutrient-dense orange-fleshed sweet potato: Advances in drought-tolerance breeding and understanding of management practices for sustainable next-generation cropping systems in Sub-Saharan Africa. *Frontiers in Sustainable Food Systems*, 4, 50. <https://doi.org/10.3389/fsufs.2020.00050>
- Lu, P., Li, X., Janaswamy, S., Chi, C., Chen, L., Wu, Y., & Liang, Y. (2020). Insights on the structure and digestibility of sweet potato starch: Effect of postharvest storage of sweet potato roots. *International Journal of Biological Macromolecules*, 145, 694–700. <https://doi.org/10.1016/j.ijbiomac.2019.12.151>
- McDougall, G.J., Shpiro, F., Dobson, P., Smith, P., Blake, A., & Stewart, D. (2005). Different polyphenolic components of soft fruits inhibit α -amylase and α -glucosidase. *Journal of Agricultural and Food Chemistry*, 53 (7), 2760–2766. <https://doi.org/10.1021/jf0489926>
- Muzhingi, T., Mbogo, D., Low, J.W., Magnaghi, A., Heck, S., & Gule, S. (2016). Effect of baking on the β -carotene content of orange-flesh sweet potato (*Ipomoea batatas*) puree bread and OFSP flour bread. *FASEB Journal*, 30 (S1), lb433–lb433.
- Nalleli-Trancoso-Reyes, N., Ochoa-Martínez, L. A., Bello-Pérez, L. A., Morales-Castro, J., Estévez-Santiago, R., & Olmedilla-Alonso, B. (2016). Effect of pre-treatment on physicochemical and structural properties, and the bioaccessibility of β -carotene in sweet potato flour. *Food Chemistry*, 200, 199–205. <https://doi.org/10.1016/j.foodchem.2016.01.047>
- Ngoc, L.B.B., Trung, P.T.B., Hoa, P.N., & Hung, P. (2017). Physicochemical properties and resistant starch contents of sweet potato starches from different varieties grown in Vietnam. *International Journal of Food Science and Nutrition*, 2 (1), 53–57.
- Nzambwita, M., Duodu, K.G. & Minnaar, A. (2017). Stability of β -carotene during baking of orange-fleshed sweet potato-wheat composite bread and estimated contribution to vitamin A requirements. *Food Chemistry*, 228, 85–90. <https://doi.org/10.1016/j.foodchem.2017.01.133>
- Odenigbo, A., Rahimi, J., Ngadi, M., Amer, S., & Mustafa, A. (2012). Starch digestibility and predicted glycemic index of fried sweet potato cultivars. *Functional Foods in Health and Disease*, 2 (7), 280–289.
- Sherwin, J.C., Reacher, M.H., Dean, W.H., & Ngondi J. (2012). Epidemiology of vitamin A deficiency and xerophthalmia in at-risk populations. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 106, 205–214. <https://doi.org/10.1016/j.trstmh.2012.01.004>
- Stevens, G.A., J. E. Bennett, Q. Hennocq, Y. Lu, L. M. De-Regil, L. Rogers, ... M. Ezzati. (2015). Trends and mortality effects of vitamin A deficiency in children in 138 low-income and middle-income countries between 1991 and 2013: A pooled analysis of population-based surveys. *Lancet Global Health*, 3, e528–e536. [https://doi.org/10.1016/S2214-109X\(15\)00039-X](https://doi.org/10.1016/S2214-109X(15)00039-X)
- Tang, Y., Cai, W. & Xu, B. (2015). Profiles of phenolics, carotenoids and antioxidative capacities of thermal processed white, yellow, orange and purple sweet potatoes grown in Guilin, China. *Food Science and Human Wellness*, 4, 123–132. <https://doi.org/10.1016/j.fshw.2015.07.003>
- Thakkar, S.K., Maziya-Dixon, B., Dixon, A.G., & Failla, M.L. (2007). β -Carotene micellization during in vitro digestion and uptake by Caco-2 cells is directly proportional to β -carotene content in different genotypes of cassava. *Journal of Nutrition*, 137 (10), 2229–2233. <https://doi.org/10.1093/jn/137.10.2229>
- Trumbo, P., Yates, A.A., Schlicker, S., & Poos, M. (2001). Dietary reference intakes: Vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium, and zinc. *Journal of the Academy of Nutrition and Dietetics*, 101 (3), 94–301. [https://doi.org/10.1016/S0002-8223\(01\)00078-5](https://doi.org/10.1016/S0002-8223(01)00078-5)
- Wanjau, C., Abong, G., Mbogo, D., Heck, S., Low, J., & Muzhingi, T. (2018). The physicochemical properties and shelf-life of orange-fleshed sweet potato puree composite bread. *Food Science and Nutrition*, 6 (6), 1555–1563. <https://doi.org/10.1002/fsn3.710>
- WHO. (2009). *Global prevalence of vitamin A deficiency in populations at risk 1995–2005: WHO global database on vitamin A deficiency*. World Health Organization.
- Ye, F., Li, J., & Zhao, G. (2020). Physicochemical properties of different-sized fractions of sweet potato starch and their contributions to the quality of sweet potato starch. *Food Hydrocolloids*, 108, 106023. <https://doi.org/10.1016/j.foodhyd.2020.106023>
- Zhu, F., Yang, X., Cai, Y.Z., Bertoff, E., & Corke, H. (2011). Physicochemical properties of sweet potato starch. *Starch*, 63 (5), 249–259. <https://doi.org/10.1002/star.201000134>