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Original article

Effect of processing and oil type on carotene bioaccessibility in traditional foods prepared with flour and puree from orange-fleshed sweetpotatoes

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Summary Consumption of Orange-Fleshed sweetpotato (OFSP) and products as source of provitamin A is being promoted to tackle vitamin A deficiency (VAD) in sub-Saharan Africa. However, limited information is available on β-carotene retention in foods and delivery after digestion. The study assessed carotene retention and bioaccessibility following *in vitro* digestion on traditional foods having OFSP among the ingredients. Sunflower oil, margarine and beef fat were evaluated on their effect on β-carotene bioaccessibility. Porridge and *chapatis* were prepared with either OFSP puree or flours in the formulations. Carotene retention was highest in *chapatis* (83%) as compared to porridge (65%). Micerallisation efficiency of all-trans β-carotene was comparable between similar products but greater in *chapatis* (62%) than porridge (11%). Sunflower oil had the highest all-trans β-carotene bioaccessibility compared to margarine and beef fat. The results support the promotion of consumption of OFSP-based products as good source of provitamin A to fight VAD.

Keywords Bioaccessibility, *in vitro* digestion, oil type, provitamin A, sweetpotato, β-carotene.

Introduction

Sweetpotato (*Ipomea batatas* Lam) is the fourth most important staple crop in sub-Saharan Africa (FAO, 2004). It is rich source of energy, fibre, minerals and vitamins. Orange-Fleshed sweetpotato (OFSP) contains β-carotene, a precursor for vitamin A (VA) in the body. In sub-Saharan Africa, traditional methods of sweetpotato preparation include boiling, steaming, roasting and drying (Hall *et al*., 1998). Dried OFSP products like chips and flour are the common ingredients for food preparations at the household level (Hall *et al*., 1998). In the recent past, the use of OFSP puree has become important due to its high carotene retention during storage as compared to flour and dried chips. Utilisation of OFSP puree in bread formulation and porridge has been adopted in Kenya.

Although OFSP contains significant amounts of β-carotene, not all of the available quantity is retained after processing. Bechoff *et al*. (2011) reported retention of all-trans-β carotene in porridges and *chapatis* supplemented with 30% OFSP flour by 69–93% and 70–97%, respectively. Significant degradation in β-carotene content of deep fried OFSP products has been reported (Kidmose *et al*., 2006). The carotene losses of the deep fried products were mostly due to longer frying times at high temperatures causing transfer of carotenes into the oil, as they are oil-soluble. These studies have demonstrated carotenes loss during processing and storage of OFSP products. However, there is need to investigate carotene retention of various OFSP products as affected by processing and sweetpotato genotype. In addition, information on β-carotene bioaccessibility of OFSP traditional products is limited. Carotenoids bioaccessibility is defined as the fraction of carotenoids transferred by food to mixed micelles (small aggregates of mixed lipids and bile acids suspended within the ingesta), therefore becoming accessible for subsequent uptake by intestinal mucosa (Burri, 2011). It is important to have high bioaccessibility of β-carotene in OFSP in order to fully have health benefits from the nutrient. Carotenoid

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bioaccessibility is affected by a number of factors such as carotenoid structure, processing, dietary fat and oil. Among the different types of carotenoids, lutein is reported to be more bioaccessible due to the fact that lutein is an oxycarotenoid and therefore more hydrophilic than hydrocarbon carotenoids (Garret *et al*., 1999). High temperature processing of carotene-rich foods disrupts plant cell walls and organelle membranes, facilitating greater access of digestive enzymes to substrates and release of carotenoids for integration into mixed micelles resulting in high bioaccessibility (Hedr′en *et al*., 2002). Bioaccessibility of β-carotene increased from 0 to 20% with addition of fat/oil to digestion (Ekesa *et al*., 2012; Pugliese *et al*., 2013). Similarly, Failla *et al*. (2009) observed an increase in carotenes as a result of addition of triglyceride to carotenoids rich salad. The effect of oil type on *in vitro* carotene bioaccessibility of OFSP products has not been fully explored. Therefore, the current study focused on assessing the bioaccessibility of all-trans β- carotene from OFSP products for improved VA intake and consequently contributed towards eradicating VA deficiency in Africa. The effect of oil type on β-carotene bioaccessibility was also evaluated in this study. Information generated by the study is important to determine processing methods and oil type with optimal carotene delivery after digestion for probable intestinal absorption.

Materials and methods

Chemicals and standards

Reagents for chemical analysis (ethanol, methanol, tetrahydrofuran (THF), hexane, butylated hydroxytoluene, acetone and methyl tert-butyl ether), β-carotene standards (all-trans β-carotene, 13-cis and 9-cis β-carotene isomers) and enzymes (α amylase, pepsin and pancreatin) for *in vitro* digestibility studies were purchased from Sigma Aldrich (St. Gallen, Switzerland). All local food ingredients and materials were sourced in Kenya.

Sweetpotato genotypes and preparation of dried chips and flours

Two OFSP genotypes namely Vita and Kabode were bought from farmers in Oyugisi, Kenya. After harvest, the roots were washed with water to remove sand and dirt and immediately processed into flour and puree. For flours, sweetpotato roots were cut into 5 mm slices using a chipper and then dried in a solar dryer to a moisture content of 10%. After drying, the sweetpotato slices were milled into flour, packed in brown sack bags lined with polyethylene paper on both sides and stored at −20 °C until used in porridge and *chapati* preparations. For sweetpotato puree, sweetpotato roots were cleaned, trimmed and boiled unpeeled at 100 °C for 1 h. After cooling to room temperature, the cooked roots were pureed using a hammer mill, vacuum packed and stored at −20 °C until usage in porridge and *chapati* making.

Porridge preparation

Hundred grams of 50% OFSP flour/puree and 50% maize flour were weighed and mixed with 100 mL cold water to form a slurry. The slurry was then added to hot water while stirring to form a paste and allowed to cook for 5 min. Hundred percent maize flour porridge was prepared as control. Cooked porridge was allowed to cool, packed in ziplock bags and stored at −80 °C until analysis.

Chapati preparation and effect of oil type study

Hundred grams of 50% OFSP flour/puree and 50% wheat flour blend were weighed into a mixing bowl. A pinch of salt, a tablespoon of pure sunflower oil and water were added to form a dough. The dough was cut into small portions and then rolled to form round flat doughs. The flat doughs were roasted in a pan containing two tablespoons of oil at 175 °C for 2 min. Hundred percent wheat flour *chapati* was prepared as control. Cooled *chapatis* were packed in ziplock bags and stored in −80 °C until further tests were conducted.

Three oil types, namely sunflower oil, margarine and beef fat, were evaluated. The choice of the oils was based on the degree of saturation and their local availability. According to our free fatty acid analysis with gas chromatography, the composition of the oils were as follows: sunflower oil contained mostly polyunsaturated (59%), with 30% monounsaturated and 11% saturated fatty acids; margarine is hydrogenated oil and behaves more like saturated comprised of 46% saturated, 43% monounsaturated and 11% polyunsaturated fatty acids; beef fat comprised mostly saturated (49%), monounsaturated (42%) and polyunsaturated (9%) fatty acids. Either pure sunflower oil, margarine and beef fat were added to OFSP-supplemented *chapatis*. The amount of oil added was 10% (w/w) of *chapati* formulation. OFSP puree and flour without oil were the controls for the experiment. The *chapatis* were prepared and stored as explained above.

In vitro digestion

The *in vitro* digestion of porridge and *chapati* was based on the method reported by Failla *et al*. (2009) with some modifications. Ten-gram sample was mixed with 6 mL of oral phase (pH 6.9) containing 3015 units of α-amylase enzyme and incubated in a shaking water bath at

37 °C for 10 min. The gastric phase was initiated by adding 30 mL 0.9% sodium chloride solution and adjusting pH to 3.5 with 1.0 N HCL. Two millilitres of 10 mg/mL pepsin enzyme was added and pH adjusted to 2.5 and final volume adjusted to 40 mL with 0.9% saline solution. The sample was incubated again at 37 °C for 1 h in a shaking water bath. To initiate the intestinal phase, pH was adjusted to 5.0 with 1.0N NaOH. Two millilitres of pancreatin enzyme (20 mg mL^{-1}) and 3 mL of bile extract (30 mg mL^{-1}) were added to the sample and pH adjusted again to 6.5 with 1.0N NaOH. This was followed by incubating the sample at 37 °C for 2 h. At each stage of incubation, the sample was blanketed with nitrogen to prevent oxidation. After intestinal phase of digestion, about 15 mL of the digesta was transferred into a clean test tube, blanketed with nitrogen and kept in a −80 °C freezer awaiting carotenoid analysis. The remaining material was centrifuged at 4000 ***g***, 4 °C for 1 h. The aqueous micellar fraction was filtered through micro filter (0.45 mm), blanketed with nitrogen and kept at −80 °C until carotenoid analysis. The *in vitro* digestion experiment was done in triplicate for each prepared product.

Carotenoid extraction and analysis

Porridge and *chapati*: Triplicate samples (about 1 g) of OFSP porridge and *chapati* were extracted three times with hexane prior to saponification with 120 μL of 80% KOH and incubating at 85 °C for 10 min. The extract was evaporated to complete dryness under a stream of nitrogen and reconstituted with appropriate volumes of methanol:THFsolution (85:15 v/v) according to extract concentration.

Digestive fractions: Intestinal digesta and aqueous micellar fractions were extracted three times with 1:3 acetone:hexane with 0.1% w/v butylated hydroxytoluene, dried using nitrogen evaporator (Organomation Associates, Berlin, MA, USA) and then reconstituted with methanol:THF solution (85:15 v/v). The reconstitution volumes for intestinal digesta and filtered micellar fractions were 1000 and 150 μL, respectively.

All extraction procedures were done under yellow light to minimise carotenoid degradation. Carotenoids were analysed by reversed phase HPLC equipped with auto sampler injector, degasser, pump and Waters 9562-UV-visible photodiode array detector operating at 450 nm (Waters Corporation, Milford, MA, USA). Separations were carried out on a 3-μm, 150 × 3.0 mm, Semibore column (YMC, Wilmington, NC, USA). The mobile phase consisted of methanol:methyl tert-butyl ether:water (85:12:3, v/v/v), plus 1.5% ammonium acetate(w/v) (phase A), methanol:methyl tert-butyl ether: water (8:90:2, v/v/v), plus 1% ammonium acetate(w/v) (phase B). The gradient procedure at a flow rate was 0.4 mL min^{-1} and injection volume of the sample was 30 μL. The gradient program was as follows: 100% solvent A for 2 min, followed by 9 min 90% solvent A and 10% solvent B, then 12 min 45% solvent A and 55% solvent B, a 15 min hold at 5% solvent A and 95% solvent B, then a 2 min gradient back to 100% solvent A. Standard curves of pure all-trans β-carotene, 13-cis β-carotene and 9-cis β-carotene were used to quantify the carotenoids.

Moisture content determination

Porridge and *chapati* samples were analysed for moisture content at the same time as for carotenoid analysis. Determinations were made by drying triplicate 5 g samples in an oven (Gallenkamp, Leicestershire, UK) at 105 °C to a constant weight (minimum 24 h) (AOAC, 1984).

Statistical analysis

All experiments were performed with three replications. The statistical analysis of collected data was performed by ANOVA using Genstat version 6.0 to determine differences among the treatments. Significant differences among treatments were obtained by Tukey's HSD multiple rank test at $P < 0.05$.

Results and discussion

β-carotene content of OFSP products

The HPLC chromatogram of carotenoids in OFSP-supplemented porridge and *chapati* is shown in Fig. 1. Identification of carotenoids was achieved by retention time and absorption spectra collected between 250 and 550 nm with those of authentic standards. From the method developed in the study, 13-cis β-carotene, all-trans β-carotene and 9-cis β-carotene were identified by peak retention times occurring at 21, 23 and 24 min, respectively. Quantification of carotenoids was achieved using a calibration curve with the correlation coefficient of ≥0.998. Total β-carotene content was expressed as summation of the three carotenes. As shown in Fig. 1, all-trans β-carotene and its isomers 13-cis and 9-cis β-carotene were the main carotenes found in the OFSP products. It is worthwhile to mention that the control products, maize flour porridge and wheat flour *chapati* did not contain any detectable level of provitamin A carotenes, hence were not included in subsequent experiments.

All-trans β-carotene retention during processing

The carotene contents of the products before cooking are shown in Table 1. Results of carotene analysis of the products expressed in μg g^{-1} fresh weight (FW)

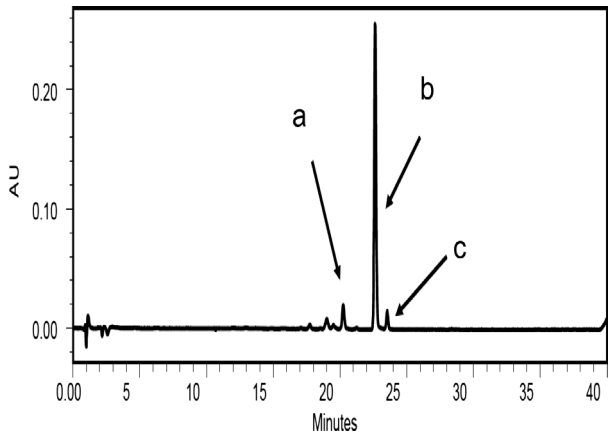

Figure 1 Chromatograms (abs 450 nm) of major provitamin A carotenoids found in OFSP-maize flour porridge and OFSP-wheat flour *chapati*. Peak identification: 13-cis β-carotene (a), all-trans β-carotene (b), 9-cis β-carotene (c).

and μg g^{-1} dry weight (DW) are presented in Tables 2 and 3, respectively. It is clear that all-trans β-carotene was the predominant carotene in all the products accounting for >70% of total carotene. This was followed by 13-cis β-carotene while 9-cis β-carotene isomer was the least. Significant differences were observed among the products in terms of all-trans β-carotene content ($P < 0.05$; Table 2). Between OFSP flour and puree, products made from flour had more all-trans β-carotene content than those made with puree. For instance, the all-trans β-carotene contents in vita flour *chapati* and kabode flour *chapati* were 37.91 μg g^{-1} FW and 36.22 μg g^{-1} FW, respectively while vita puree *chapati* and kabode puree *chapati* had 17.46 μg g^{-1} FW and 11.64 μg g^{-1} FW, respectively. A similar trend was observed when the porridges were supplemented with either OFSP puree or flour. The total β-carotene contents of vita flour and kabode flour porridge were significantly higher than vita puree and kabode puree porridge ($P < 0.05$; Table 3). The results might be related to high moisture content of porridge and puree-based products compared to flour although puree-based *chapatis* had low moisture content probably due to high roasting temperature. With reference to the moisture content of the products, the dry weight basis (Table 3) gave a different trend whereby the carotene content of porridge was higher than *chapati*. The results imply that considering the fresh weight basis, we benefit more of the nutrient in products with low moisture content. In the same line, high moisture content foods are nutrient dense and more beneficial nutritionally on dry weight basis. Thus, we might obtain more β-carotene from OFSP *chapati* and porridge on fresh weight and dry weigh basis, respectively. Ideally, we consume porridge on fresh weight basis so for the same amount of β-carotene, more of the porridge will have to be consumed (as is) compared to *chapati*. The carotene content of puree-based products did not significantly improve on dry weight basis compared to flour-based products. This was due to high moisture content of puree which might have contributed to excessive dilution of total carotene when mixed with non-OFSP flour. It is seen from the results that there were no significance differences ($P \geq 0.05$) in all-trans β-carotene content among the products containing either OFSP puree or flour (Tables 2 and 3). Overall, on fresh weight basis, OFSP-supplemented *chapatis* had high all-trans β-carotene content compared to porridge. The findings are in accordance with Bechoff *et al.* (2011), who reported higher all-trans β-carotene content from *chapatis* than porridge supplemented with 30% OFSP flour. The relatively low all-trans β-carotene of porridges might be related to its degradation associated with longer cooking time. The average cooking time of porridge was 5 min while *chapati* was 2 min, thus, allowing more carotene degradation in porridge. The low moisture content of *chapatis* might have also contributed to higher carotene content as compared to porridge with higher moisture content on wet weight basis.

Table 1 Contents of all-trans, 13-cis and 9-cis β-carotenes (μg g^{-1} on a Fresh Weight Basis) in the OFSP puree/flour and maize/wheat flour blends before cooking

Sample	13-cis	All-trans	9-cis	Total	Moisture content
50% vita puree+50% maize flour	5.26 ± 0.57^{a}	21.62 ± 2.18^{a}	0.51 ± 0.12^{a}	26.20 ± 3.20^{a}	60.8 ± 1.09^{b}
50% kabode puree+50% maize flour	3.37 ± 0.50^{a}	17.31 ± 1.67^{a}	0.42 ± 0.10^{a}	22.39 ± 2.70^{a}	60.6 ± 0.66^{b}
50% vita flour+50% maize flour	4.85 ± 0.06^{a}	25.30 ± 0.34^{a}	0.57 ± 0.08^{a}	30.89 ± 1.50^{a}	6.5 ± 0.07^{a}
50% kabode flour+50% maize flour	4.06 ± 0.20^{a}	23.56 ± 0.63^{a}	0.76 ± 0.11^{a}	27.50 ± 1.65^{a}	6.5 ± 0.17^{a}
50% vita puree+50% wheat flour	5.64 ± 0.30^{ab}	21.17 ± 0.67^{a}	0.59 ± 0.01^{a}	28.14 ± 1.27^{a}	60.6 ± 0.61^{b}
50% kabode puree+50% wheat flour	4.04 ± 0.20^{a}	14.37 ± 0.30^{a}	0.45 ± 0.23^{a}	18.33 ± 1.24^{a}	59.6 ± 0.43^{b}
50% vita flour+50% wheat flour	8.38 ± 1.14^{bc}	44.50 ± 1.20^{b}	1.40 ± 0.20^{b}	55.62 ± 2.12^{b}	6.6 ± 0.17^{a}
50% kabode flour+50% wheat flour	8.42 ± 0.70^{c}	43.15 ± 5.50^{b}	1.50 ± 0.11^{b}	51.55 ± 4.87^{b}	6.3 ± 0.04^{a}

Data represent mean ± SEM; $n = 5$. Letters with different superscripts represent significant differences ($P < 0.05$) between samples within individual species and total carotenoid using Tukey's test.

Table 2 Total β-carotene, all-trans β-carotene, 13-cis β-carotene and 9-cis β-carotene in porridge and *chapati* containing OFSP puree and flour (μg g^{-1} FW)

Genotype	Product	13-cis	All-trans	9-cis	Total	Moisture content
Vita	Puree porridge	3.74 ± 0.40^{a}	16.69 ± 1.78^{ab}	0.359 ± 0.08^{a}	20.79 ± 0.22^{ab}	85.8 ± 0.23^{c}
	Flour porridge	3.36 ± 0.12^{a}	19.63 ± 0.33^{b}	0.459 ± 0.05^{a}	23.44 ± 0.24^{b}	85.7 ± 0.01^{c}
	Puree chapati	4.40 ± 0.06^{ab}	17.46 ± 0.26^{ab}	0.441 ± 0.00^{a}	22.29 ± 1.01^{ab}	33.0 ± 0.22^{b}
	Flour chapati	6.51 ± 0.82^{c}	37.91 ± 1.81^{c}	1.003 ± 0.14^{b}	45.42 ± 0.09^{c}	24.7 ± 0.14^{a}
Kabode	Puree porridge	2.51 ± 0.42^{a}	13.33 ± 1.27^{ab}	0.324 ± 0.07^{a}	16.16 ± 1.67^{ab}	85.2 ± 0.01^{c}
	Flour porridge	3.01 ± 0.14^{a}	17.61 ± 0.78^{ab}	0.561 ± 0.07^{a}	21.18 ± 0.95^{ab}	86.0 ± 0.22^{c}
	Puree chapati	3.06 ± 0.23^{a}	11.64 ± 0.47^{a}	0.345 ± 0.02^{a}	15.05 ± 0.48^{a}	28.2 ± 0.22^{a}
	Flour chapati	6.39 ± 0.50^{bc}	36.22 ± 3.2^{c}	1.175 ± 0.10^{b}	43.78 ± 3^{c}	29.3 ± 0.01^{ab}
Control	Maize flour porridge	ND	ND	ND	ND	-
Control	Wheat flour porridge	ND	ND	ND	ND	–

ND, Not detected.
Data represent mean ± SEM; $n = 5$. Letters with different superscripts represent significant differences ($P < 0.05$) between samples within individual species and total carotenoid using Tukey's test.

Table 3 Total β-carotene, all-trans β-carotene, 13-cis β-carotene and 9-cis β-carotene in porridge and *chapati* containing OFSP puree and flour (μg g^{-1} DW)

Genotype	Product	13-cis	All-trans	9-cis	Total
Vita	Puree porridge	3.74 ± 0.32^{a}	116.1 ± 12.34 ^{cd}	2.49 ± 0.56^{bcd}	144.6 ± 15.48^{d}
	Flour porridge	3.36 ± 0.13^{a}	135.8 ± 2.27^{d}	3.18 ± 0.37 ^{cd}	162.2 ± 1.64^{d}
	Puree chapati	6.54 ± 0.10^{bc}	26.0 ± 0.40^{ab}	0.66 ± 0.01^{a}	33.2 ± 0.45^{ab}
	Flour chapati	8.64 ± 1.10^{c}	50.4 ± 2.40^{b}	1.33 ± 0.18^{ab}	60.3 ± 1.34^{b}
Kabode	Puree porridge	2.51 ± 0.42^{a}	89.1 ± 8.46^{c}	3.93 ± 0.44^{d}	108.0 ± 11.15^{c}
	Flour porridge	3.01 ± 0.14^{a}	123.3 ± 5.43^{d}	2.16 ± 0.51^{abc}	148.3 ± 6.68^{d}
	Puree chapati	4.28 ± 0.32^{ab}	16.3 ± 0.65^{a}	0.48 ± 0.03^{a}	21.1 ± 0.67^{a}
	Flour chapati	9.04 ± 0.71^{c}	51.3 ± 4.55^{b}	1.66 ± 0.13^{abc}	62.0 ± 5.38^{b}
Control	Maize flour porridge	ND	ND	ND	ND
Control	Wheat flour porridge	ND	ND	ND	ND

ND, Not detected.
Data represent mean ± SEM; $n = 5$. Letters with different superscripts represent significant differences ($P < 0.05$) between samples within individual species and total carotenoid using Tukey's test.

Beta-carotene is susceptible to degradation during processing at temperatures above 37 °C. In this study, apparent all-trans β-carotene retention of the products was determined by comparing carotene content in finished products (Table 2) to the initial content in OFSP composites flours and/or puree in product formulation (Table 1). All-trans β-carotene retention of *chapatis* ranged from 76% (kabode puree chapati) to 82.33% (vita flour chapati) while among the porridges it ranged from 65% (kabode flour porridge) to 71% (vita flour porridge) ($P < 0.05$). The findings are comparable to carotene true retention in OFSP *chapatis* and porridges reported by Bechoff *et al*. (2011). Kean *et al*. (2008) reported carotene retention of 52% (yellow maize porridge) and 75% (yellow maize bread). The authors attributed isomerisation of carotene during thermal processing as the main cause for decrease in carotene content. Bechoff *et al*. (2011) explained the lower carotene retention in porridge was possibly due to increased heat damage to carotenoids caused by their greater dispersion in boiling water. Increase in 13-cis and 9-cis β-carotene isomers as a result of high temperature processing was also reported (Chandler & Schwartz, 1988; Bechoff *et al*., 2011). However, the effect of β-carotene isomerisation as a result of high temperature processing was not observed in the current study. Differences in OFSP genotype, ingredient interactions and differences in processing conditions may partially explain the observed differences. Apart from isomerisation of carotenes during thermal processing, Bechoff *et al*. (2018) explained physical losses such as leaching of carotenes into water experienced at an initial step of processing as a factor responsible for carotene degradation in processed products. It is important to mention that the carotene retentions reported in this study were calculated based on fresh

weight of the products, since they are consumed fresh. A different scenario would be observed on dry weight basis as shown in Table 3.

In vitro β-carotene bioaccessibility in OFSP products

The effect of processing on digestive stability and micellarisation efficiency of all-trans β-carotene was evaluated on digesta and filtered micellar of OFSP porridge and *chapatis*, respectively. Digestive stability indicates the percentage of the carotenoids recovered in the digesta following simulated digestion of OFSP products. It was calculated as amount of carotenoids in the digesta expressed as a percentage of the amount of carotenoids in the undigested food product. The percentage of carotenoids transferred from the digesta to the filtered aqueous fraction is defined as the micellarisation efficiency and is used as a measure of relative bioaccessibility. Micellarisation efficiency was calculated as the amount of carotenoids in the aqueous micellar fraction expressed as a percentage of the amount of carotenoids in the digesta. As shown in Fig. 2, percent digestive stability of all-trans β-carotene of porridge ranged from 52% to 72.9% whereas in *chapatis* it ranged from 69% to 87%, and the results were not dependent on sweetpotato genotype. The current findings of carotene recovery after simulated digestion are slightly lower than the recovery values of >100% reported by Kean *et al*. (2008). Despite the differences with published literature, the carotene recovery obtained in this study was >50% indicating good stability of carotene in the products during simulated digestion. As shown in Table 2, puree-based products had low carotene content compared to flour-based products, a similar trend was observed on carotene recovery after digestion. It is documented that in the food matrix, carotenes are found complexed to proteins in chromoplasts forming a proteinaceous complex (Garrett *et al*., 2000). Such complex microstructure has major implications on the release of carotenes during processing, simulated digestion, extraction and analysis. Processing methods that cause more disruption of cell matrix result in high carotene recovery after digestion. This explains in part the high digestive recovery of OFSP flour-based porridge and *chapatis*. The maceration of OFSP roots during chipping step as well as milling of dried chips into flour caused cell rupture and facilitated release of carotene from the matrix, resulting in improved carotene delivery. On the other hand, the processing step of puree did not include severe maceration of the cells such as chipping and milling resulting in low carotene recovery after simulated digestion.

The mean efficiency of micellarisation for all-trans β-carotene varied between products. The all-trans β-carotene micellarisation ranged from 11% to 18% for porridge and from 40% to 62% for *chapati* (Fig. 2). With reference to the OFSP primary product, flour-based products had high micellarisation efficiency compared to puree-based products. The result is similar to the findings by Bechoff *et al*. (2011) who reported high micellarisation efficiency for OFSP flour porridge compared to boiled and mashed OFSP. Bengtsson *et al*. (2009) reported a close relationship between damaging effect of drying on sweetpotato cell integrity and bioaccessibility. Moreover, cell disruption during chipping of roots and grinding of OFSP dried chips facilitated release of carotene from cell matrix and giving great access by enzymes during *in vitro* digestion resulting in improved bioaccessibility of flour-based products. In the same way, the kneading step of *chapati* dough

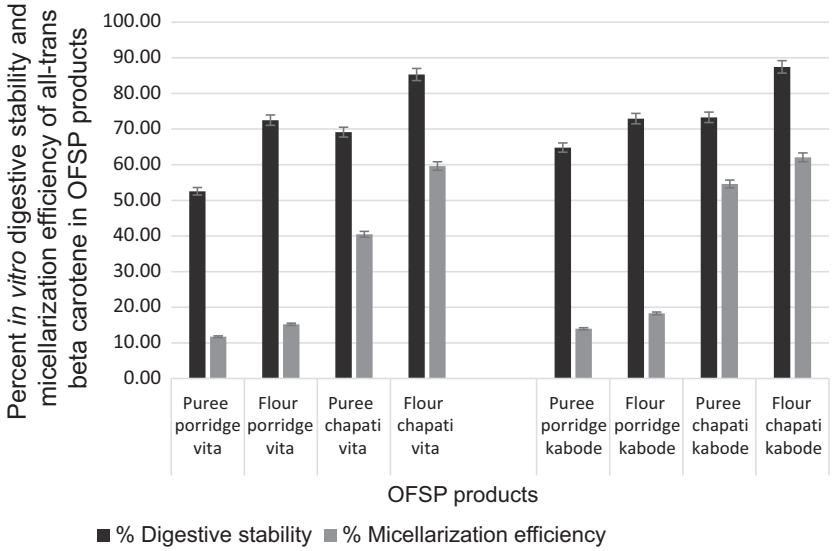

Figure 2 Effect of processing methods on the *in vitro* β-carotene bioaccessibility of OFSP products. Values are presented as mean ± SEM; $n = 3$.

facilitated release of all-trans β-carotene from the food matrix making it more accessible to digestive enzymes. Processing temperature is another factor affecting carotene bioaccessibility. Hedr′en *et al.* (2002) explained that high temperature processing disrupts plant cell wall and organelle membrane facilitating greater access by digestive enzymes to substrate and carotene release for incorporation into mixed micelle, hence improved bioaccessibility. Thus, the high processing temperature of *chapatis* (175 °C) compared to porridge (100 °C) might have influenced high carotene bioaccessibility.

In general, the micellarisation efficiency trend corresponded well to digestive recovery further confirming stability of carotene to simulated digestion. The lower levels of 13-cis and 9-cis β-carotene in the products consequently affected their digestive recoveries and apparent bioaccessibility (results not shown). This is contrary to findings by Bechoff *et al.* (2011) who reported improved bioaccessibility of cis isomers compared to all-trans β-carotene. It is important to mention that the current all-trans β-carotene micellarisation efficiencies of OFSP products are in line with previous studies (Thakkar & Failla, 2008; Failla *et al.*, 2009; Bechoff *et al.*, 2011; Lipkie *et al.*, 2013). The percentage of micellarised all-trans β-carotene was 16% for porridge and 73% for *chapati* (Bechoff *et al.*, 2011). Considering the processing method, it is clear that addition of oil in *chapati* greatly improved micellarisation efficiency. Lipkie *et al.* (2013) also observed improved bioaccessibility with addition of 10% oil in porridge prior to simulated digestion. Stir-frying of carotene-rich food was reported to lead to high carotene bioaccessibility (Garrett *et al.*, 2000; Veda *et al.*, 2010). Authors attributed improved carotene micellarisation efficiency to addition of oil, as oil can facilitate incorporation of carotene into the mixed micelle during digestion. The VA activity of OFSP-supplemented porridge and *chapati* to meet nutritional requirements among different groups is well documented by Bechoff *et al.* (2011) and similar estimates can be applied to the products in the current study.

Effect of oil type on β-carotene bioaccessibility

In the present study, the effect of three different oil types, including pure sunflower oil, margarine and beef fat, on *in vitro* bioaccessibility of β-carotene was evaluated and results are presented in Fig. 3. According to Fig. 3, the digestive stability of the products was similar to data presented in Fig. 2 indicating that all-trans β-carotene of OFSP products is not affected by oil type. However, efficiency of transfer of all-trans β-carotene

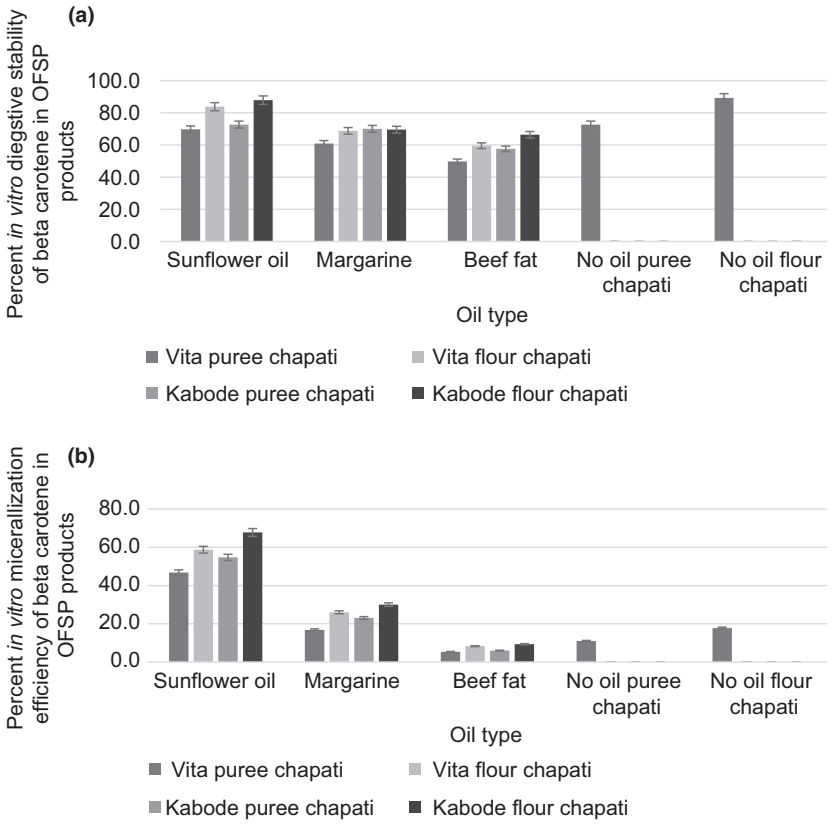

Figure 3 Effect of oil type on bioaccessibility of all-trans β-carotene in OFSP products. Digestive stability (a), Micellarisation efficiency (b). Values are presented as mean ± SEM; $n = 3$.

from the aqueous phase to the filtered micellar phase was significantly affected by oil type. The greatest micellarisation efficiency was found in *chapati* with sunflower oil (67.8%), followed by margarine (30.0%) while beef fat (9.3%) was the least. The micellarisation efficiencies of the control products were significantly lower than *chapatis* with sunflower oil and margarine but higher than *chapatis* with beef fat. Beta-carotene is a lipophilic micronutrient, hence its bioaccessibility is greatly affected by presence of oil during digestion. Sunflower oil, a predominantly long-chain unsaturated fatty acid with low melting point is effective in dispersing and dissolving β-carotene at the digestion temperature, thus facilitating its incorporation into the mixed micelles. On the other hand, margarine and beef fat are more saturated oils characterised by high melting point, hence not as effective at dissolving β-carotene at the digestion temperature. Although margarine contained high all-trans β-carotene content (data not shown), its presence did not enhance all-trans β-carotene of *chapatis*. The high micellarisation efficiencies of the control products compared to beef fat are indicative of inability of beef fat to solubilise β-carotene at the digestion temperature as most of the nutrient was trapped in its bulk structure. The result further confirms that oil type has great impact on carotene bioaccessibility. Apart from dissolving carotene and facilitating incorporation of carotene into mixed micelles, the presence of oils also induces secretion of bile and pancreatic juice in the digestive tract.

Several studies have linked presence of oil to β-carotene bioaccessibility (Failla *et al*., 2009; Ekesa *et al*., 2012; Pugliese *et al*., 2013). Micellarisation efficiency of β-carotene during small intestinal digestion was increased by lipids rich in unsaturated fatty acids: soybean oil > olive > canola > butter (Failla *et al*., 2014). The addition of trioleoylglycerol to spinach homogenate markedly improved the bioaccessibility of β-carotene but not lutein and α-tocopherol (Nagao *et al*., 2013). On the contrary, polyunsaturated fatty acids reduced β-carotene bioaccessibity due to formation of peroxides of the nutrient during digestion (Nagao *et al*., 2013). In a separate study, Berni *et al*. (2014) reported a significant increase in all-trans β-carotene micellarisation efficiency of fried cassava compared to boiled cassava. Bengtsson *et al*. (2010) linked improved micellarisation efficiency to cooking style alongside oil addition. Thus, cooking style involving cell disruption and oil addition improved β-carotene bioaccessibility. However, there is a limit to which dietary oils or fats can improve carotene bioaccessibility. Presence of excess oil relative to digestive enzyme during simulated digestion suppressed carotene bioaccessibility (Nagao *et al*., 2013). The effects of oil type on β-carotene bioaccessibility reported by the present study are relevant *in vivo*. *Chapati* is a staple food commonly consumed in sub-Saharan Africa. The current practice of *Chapati* making involves addition of margarine or shortening as a source of dietary oil/fat. Based on the current results, margarine gave low carotene bioaccessibility. Alternatively, sunflower oil with high carotene bioaccessibility could be used as source of dietary oil in *chapatis*. In Africa, sunflower oil is more available at affordable price and people could take advantage of this to aid in *chapati* formulation for improved carotene bioaccessibility to fully benefit from the nutrient.

In conclusion, the current study has indicated that the type of food product and associated processes may affect carotene bioaccessibility. Processing methods with high cell disruption have higher carotene bioaccessibility. All-trans β-carotene micellarisation efficiency varied between OFSP products and was higher in *chapatis* than porridge but results were not affected by sweetpotato genotype. Addition of oil in OFSP-supplemented products is critical to ensure greater carotene bioaccessibility. Long-chain unsaturated oils, like sunflower oil, have greater carotene bioaccessibility than margarine and beef fat. These results confirm that OFSP can serve as a source of bioaccessible provitamin A carotenoids. Future research needs to assess bioavailability using Caco-2 or a similar cell model for transepithelial absorption. Human studies are warranted to confirm *in vivo* the efficacy towards improving VA status in target populations. There is need for wide screening of OFSP genotypes to ensure development of genotypes with highly bioaccessible carotene.

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Conflict of interest

The authors declare no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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