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Provitamin A retention and sensory acceptability of landrace orange maize (MW5021) food products among school-aged children living in rural Malawi

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Abstract

Landrace orange maize is rich in carotenoids and is thought to be a potentially sustainable solution for Vitamin A deficiency. This study evaluated the acceptability of landrace orange maize food products among school-aged children living in rural Malawi. It also determined the retention of provitamin A carotenoids after storage in an uncontrolled environment, followed by milling and cooking. Sensory evaluations of porridge and *nsima* (thick porridge) were carried out in school-aged children ($n = 160$) using a 7-point hedonic scale. Total carotenoid content (TCC) was analysed using a spectrophotometric method and High-Performance Liquid Chromatography (HPLC) was used to identify the specific carotenoid composition of the food products. Sensory evaluation results showed a higher acceptance of landrace orange maize food products in comparison to those commonly prepared from white maize. Plain porridge scored 6.5 ± 1.4 and 5.6 ± 1.9 for orange and white maize, respectively. Similar results were observed with *nsima*. Orange maize *nsima* received a score of 6.8 ± 0.7 while white maize was evaluated at 5.8 ± 1.9. After 10 months of storage and processing, there was 89% retention of total carotenoids with 59% accounted for by evaluation of individual carotenoids. Despite the total degradation of beta-carotene, 42% retention of beta-cryptoxanthin with provitamin A activity was observed. Encouraging the production of landrace orange maize appears to be a useful strategy for providing Malawian farmers with carotenoid-rich foods with high palatability. It presents a cost-effective option to reduce dependence on supplementation and fortification.

Keyword Carotenoids, Sensory evaluation, Landrace orange maize, Provitamin A

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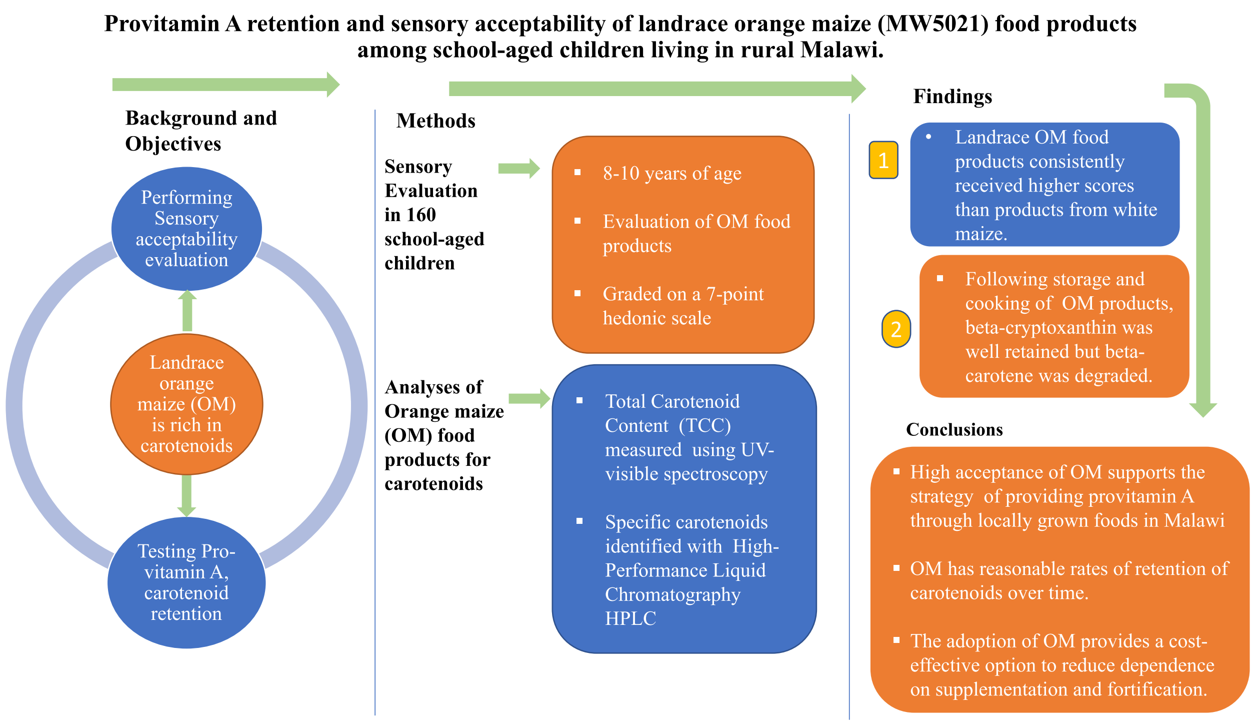
Graphical Abstract



Background

The reintroduction of pigmented landrace maize has recently been adopted as a way of improving diets and promoting food security of Malawians. Landrace maize is a cultivated, genetically heterogeneous variety of maize that has evolved in a certain ecogeographical area and is therefore adapted to the edaphic and climatic conditions (Casañas et al. 2017). It is considered to have a high tolerance to stress and produce intermediate to high yields depending on the growing conditions (Hwang et al. 2016).

Maize is a rich source of carbohydrates and also contains reasonable amounts of protein, vitamins, minerals and other bioactive components such as carotenoids (Hwang et al. 2016). The list of carotenoids found in orange maize includes: lutein, zeaxanthin, beta-cryptoxanthin and beta-carotene (Nuss & Tanumihardjo 2010). Apart from being naturally occurring pigments, carotenoids have important health roles in the human body such as pro-vitamin A activity (beta-carotene and beta-cryptoxanthin), critical for growth and development. Carotenoids exhibit strong antioxidant activity and are thought to inhibit some forms of cancer, prevent macular degeneration, decrease the risk of cataract formation, prevent cardiovascular disease and are capable of immuno-enhancement (Bungau et al., 2019; Burt et al., 2010; Dutta et al. 2005; Gammone et al. 2017). Due to the importance of these compounds, they have been used in supplementation and fortification of foods. However, the use of isolated carotenoids is limited due to their poor solubility in water, limited bioavailability and quick release (González-Peña et al. 2023). New technologies with nanoparticles and microencapsulation may help overcome these limitations (Osanlou et al. 2022; González-Peña et al. 2023). Until these products are available and affordable, consuming foods rich in carotenoids is the best approach for providing these nutritionally important compounds. At harvest, carotenoids are present in high concentrations in landrace orange maize but conditions of storage and methods of food processing are critical factors that affect their content and bioavailability over time (Schieber & Weber 2016). Due to the unsaturated nature of carotenoids, they are highly susceptible to degradation through oxidation when exposed to light, oxygen and heat. This causes colour changes in the food products and reduces bioactivity (Moura et al. 2015; Schieber & Weber 2016). Schieber and Weber (2016) describe the co-oxidation of carotenoids by type II lipoxygenases (LOX), which are present in most plant tissues. Beta-carotene seems to be affected most by oxidation. This oxidation depends on the concentration of the substrate for LOX enzymes, that is, unsaturated fatty acids with a 1-*cis*, 4-*cis* configuration.

Acceptability is critical for introducing or reintroducing products to consumers. Consumption of any food is dependent on its sensory characteristics and the quality of products made from it. Along with colour, acceptance is also influenced by several demographic and personal factors such as age, sex, familiarity with the product, taste, and socioeconomic status (Talsma et al. 2017). Most of the crops known to have vitamin A carotenoid precursors, such as biofortified maize, are coloured yellow to orange. The landrace maize used in this study had a deep orange colour.

A negative perception of orange maize in Sub-Saharan African (SSA) countries like Malawi, is common. It is thought that yellow and orange maize is food for the poor and only fit to be provided as food aid in the time of emergency (Groote et al. 2011). In recent years, sensory acceptance and processing effects have been published for biofortified maize (Pillay et al. 2011; Taleon et al. 2017). However, little is documented regarding landrace orange maize reintroduced in Malawi, especially among children. This innovative approach for increasing carotenoids in the diet targeted children, with the aim of enhancing the intake of pro-vitamin A to reduce deficiencies and promote optimal growth and development. The objectives of this study were to assess the acceptability of landrace orange maize products among school-aged children. In addition, carotenoid retention in commonly eaten foods was determined using maize that had been stored under home conditions for 10 months, then processed and cooked.

Methodology

Study area

The study was conducted in Dedza district (in central Malawi) and Thyolo district (in southern Malawi). The two districts were originally chosen to be part of the ProFarmer initiative because they have high rates of malnutrition and were readily receptive to taking up the production of orange maize. Specifically, Traditional Authority (T/A) Kachere and T/A Nnaseta of Dedza and Thyolo districts respectively were the study sites. The areas were project sites of the ProFarmer project and had farmers who had been cultivating orange maize for at least three years (referred to as primary farmers), and adjacent areas where additional farmers were cultivating orange maize for the first time (referred to as "secondary farmers"). Eighty households, with children of the stated age group, in each area, were purposively sampled from a list of over 2000 orange maize producers kept by local agriculture extension workers.

Preparation of maize flour samples

All the maize used in this study was cultivated in Dedza district (central region of Malawi) by farmers of the Pro Farmer project during the 2017–2018 season and was harvested in April 2018. Both white and orange maize was stored indoors in white polythene sacks under conditions that replicated those used by the local population. No attempt was made to control temperature, light or oxygen exposure. The maize was processed in February 2019 after 10 months of storage. Some of the grains of maize were dehulled and some were not dehulled before being dry-milled into flours of both the grand mill (dehulled) and whole grain. The whole grain orange maize flour and whole-grain white maize flour were used to prepare porridge and the grand mill flours were used to prepare *nsima*. After milling, the flours were kept for 24 h at room temperature before being used to produce the orange and white maize foods. These flours together with the prepared food products were also sampled for carotenoid analysis.

Preparation of the food products

Food products from maize were freshly prepared by local women at both the Dedza and Thyolo sites. The women were requested to prepare both white and orange maize products as they would at home. All the women were given: 3 kg of the grand mill and 2 kg of whole grain flour (both orange and white maize) and bottled water (4.5 L). Six different recipes were used to prepare the products. They included: 1) orange maize porridge with no added sugar (2) white maize porridge with no added sugar (3) orange maize porridge with added sugar (4) white maize porridge with added sugar (5) orange maize *nsima* (6) white maize *nsima*. In the two sweetened recipes, sugar, approximately 6 tablespoons per 4.5-L pot, was used.

Porridge and *nsima* were prepared by cleaning a pot and filling it with bottled water. The water was heated on an open flame and orange flour or white flour was added to make porridge. Additional flour was added to the porridge to achieve the appropriate consistency. *Nsima* was prepared similarly to the porridge but using grand mill flours. When the porridge/*nsima* had cooked for an average of 30 min it was ready to be served.

Sensory evaluation of the food products

Panellists

The participants in the sensory acceptance testing were school-aged children, 8–10 years old, from participating farming households. The farmers were contacted and briefed on the project two weeks before the sensory evaluation day and consent was requested for their children to take part in the activity. The boys and girls came from families that generally consume maize porridge and *nsima* (thick porridge). The rationale for using children rather than adults in the sensory assessment was that school-aged children are among the most vulnerable groups with regard to vitamin A deficiency and their acceptance of the food is important for increasing consumption.

On the day of the tasting, the farmers brought their children to the school, which acted as a centre for sensory evaluation. The families were briefed again about the project before formally requesting both parental consent and the children's assent to participate. In cases where parents could not read or write, oral consent was obtained by the study staff. Before the tasting began, all the children received a detailed explanation of how to carry out the evaluation.

Sensory evaluation

Sensory analyses were performed on two separate dates, both in mid-February 2019. In Dedza the testing was held at Maonde Primary School while in Thyolo it was done at Mikombe Primary School. In both cases, 40 children of primary farmers and 40 children of secondary farmers participated. Data from 8 children were not included in the analyses. Four children were under 8 years of age, 2 children did not understand the instructions and 2 children left before completion of the testing. The final sample size included 152 children, 75 children in Dedza and 77 in Thyolo.

Cooked samples were coded and provided to participants in a single-blinded fashion. The prepared products were provided to the children consecutively (singularly) until all six samples were tested. As they tasted each sample, a questionnaire covering three specific attributes was given to the child to score: colour, taste and sweetness. Before the tasting, the children were reminded of the sensory attributes, and after tasting each sample they were asked to rinse their mouths with clean bottled water before tasting the next sample. New spoons, disposable cups and plates were used for each sample. Ideally, "texture" is included in sensory evaluations (Kohyama 2020). However, it was felt that this concept was difficult to explain to the children, particularly when many of the samples had similar textures. Therefore, this attribute was not included in the evaluation.

The children were instructed to score the food samples using a 7-point hedonic scale whereby, (1) indicated 'dislike very much' and (7) meant 'like very much'. Since the tasting was being conducted with children who might not understand the meaning of the numerical coding, the hedonic scale was modified with facial pictures for each code. Children with challenges in scoring were assisted when necessary. All instruction was in the local Chichewa language.

Carotenoid analyses

Immediately following food preparation, samples of the porridge and *nsima* were collected using zip lock bags and put in tightly closed dark containers and placed in a mobile cooler box. Samples were transported from the field to Chancellor College Chemistry Laboratory and stored at (-80 °C). Before analysis, all food and flour samples were freeze-dried for 48 h in containers covered with tin foil to avoid exposure to light. In a dark room, dried samples were then ground using a mortar and pestle.

Carotenoids were extracted according to the method of Ndolo and Beta (2013) with some modifications. Briefly, 500 mg samples (flour and foods) were prepared by freeze-drying, powdering and sifting (250-micron sieve). Samples were then mixed with 10 ml of water-saturated butanol in tubes covered with black caps and aluminium foil in a fume hood. Each sample was then homogenized for 5 min using a 10 mm diameter vortex. Tubes stood in the fume hood at room temperature for 60 min. Samples were then homogenized for a second time and let stand for another 60 min. About 10 mL of extract was centrifuged (5,000 g, 25° C, 5 min) (Labtech, Mumbai, India). All the procedures were done in the dark and each tube was wrapped in aluminium foil to avoid degradation of the carotenoids during the extraction.

Determination of total carotenoid content (TCC)

Total carotenoid content was analysed using a spectrophotometric method. Supernatants of extracted samples were transferred from the centrifuge tubes into a semi-micro quartz cuvette and absorbance was measured at 450 nm using a PG instrument T90+UV/Visible spectrophotometer (Alma Park, Wibtoft Leicestershire, England). Total carotenoid content (TCC) was calculated using the following equation and expressed as μg lutein equivalent/g sample.

$$C = (10 \times A)/S \times W\left(\mu g/g\right)$$

Where C = lutein content, μg/g; A = absorbance reading, S = regression coefficient (the number that expressed the relationship which is created based on the concentration of lutein standard solutions in μg/mL and the absorbance); 10 = dilution factor (the dilution factor of

10 is based on the total extracted volume of 10 ml) and W = sample weight, g (Ndolo & Beta 2013).

Determination of carotenoid composition

High-performance liquid chromatography (HPLC) was used to separate and identify the individual carotenoids. Supernatants from the calorimetric determination of total carotenoids were used for HPLC analyses. Samples were put in brown vials and kept at -18 °C for analysis on the following day. Stored supernatants were filtered through a 0.45 µm nylon disc filter and underwent analysis. The determination of carotenoid composition was done according to the method described by Abdel-Aal et al. (2007) with some modifications. Briefly, the chromatographic separation and quantification of carotenoids was carried out by HPLC (Agilent, 1200 Infinity Series) equipped with a Diode Array detector, Mumbai, India) and autosampler (Waters 717 Plus, Waters, Milford, MA) using YMC™ carotenoid S-3, 3 µm packing, 4.6 × 100 mm column (Waters, Milford, MA). The column was operated at 35 °C. Twenty µL of the sample was injected by the autosampler and eluted with a gradient system consisting of (A) methanol/methyl tert-butyl ether/Milli-Q water (81:15:4, v/v/v) and (B) methyl tert-butyl ether/methanol (90:10, v/v). The flow rate was set at 1 mL/min. The gradient was programmed as follows: 0–9 min, 100–75% A; 10–12 min 0% A; 12–13 min, 0–100% A; and 13–15 min, 100% A. The separated carotenoids were detected and measured at 450 nm. The carotenoids under study were identified based on the similar retention time of commercial standards which included beta-cryptoxanthin and beta-carotene.

Carotenoid retention was calculated using the formula labelled Eq. 1 below (Taleon et al. 2017). Carotenoid concentration retention after 10 months of storage to that found at the harvest period (Hwang et al. 2016) was compared using the formula labelled Eq. 2 below.

$$\text{Total TCC/Concentration Retention} = (\text{Processed products TCC/concentrations})/(\text{Unprocessed flours TCC/concentrations}) \times 100 \quad (1)$$

$$\text{Carotenoid retention after storage} = (\text{Carotenoid concentration at 10 months})/(\text{Carotenoid concentration at harvest}) \times 100 \quad (2)$$

Statistical analysis

Analysis was carried out using IBM SPSS statistics for Windows, version 25.0 (Armonk, New York, 2017). Descriptive statistics were expressed as Mean ± Standard Deviation. An independent T-test was performed to analyse the acceptance scores between primary farmers' children and secondary farmers' children and between orange and white maize. One-way ANOVA was used to compare means for significant differences in TCC and HPLC respectively between orange maize and white maize and between the cooked samples and raw samples.

Results

Sensory acceptability of orange maize products

Table 1 presents the results of the sensory evaluation of the colour, taste and sweetness of the *nsima* and porridge by children from primary and secondary orange maize producers. Overall, school-aged children of both primary farmers and secondary farmers consistently preferred landrace orange maize products over white maize products. This was observed in all the attributes that were evaluated. Primary farmers' children demonstrated a significant preference for the taste and sweetness of plain orange porridge. A similar trend was noted with regard to colour, albeit, not significant. As expected, sweetened porridge received higher scores than unsweetened porridge and was notably preferred by both primary and secondary farmers' children. This result served as an internal validation that the children understood the instructions, and could differentiate between the tastes of the different samples. There was also a significant preference by the primary farmers' children in all the sensory attributes of orange maize *nsima* (colour, taste, sweetness). Despite a significant preference for orange maize products by both primary and secondary farmers' children, white maize plain porridge, sugar porridge and *nsima* also received high rankings for organoleptic qualities. This is not surprising since white maize is still a significant part of the diet of the farmers' households.

Carotenoid content and concentrations

Table 2 describes the total carotenoid content of orange and white maize flours and the impact of food preparation on total carotenoid retention. The maize had been stored from the time of harvest in April 2018 until analysis in February 2019. Regardless of the method of processing whole grains or husked flour, the total carotenoid content of orange corn flour was significantly higher than that of white corn flour. Using the same protocol, a previous study reported that the total carotenoid content of this landrace orange maize at harvest was 59.5 mg/kg (Hwang et al. 2016). After 10 months of uncontrolled storage, 53.1 mg/kg was detected, indicating approximately 89% retention.

Overall, raw orange maize had a significantly higher total carotenoid content than the cooked orange maize sweetened porridge (Table 2). Similarly, raw whole grain

Table 1 Acceptance of maize products determined by children of primary and secondary farmers

Food products	Attribute	Farmer's children	Orange maize acceptance scores	White maize acceptance scores
Plain Porridge	Colour	Primary farmer's children	6.5 ± 1.4[a]	5.6 ± 1.9[b]
		Secondary farmer's children	6.1 ± 1.5[a]	5.2 ± 2.0[b]
	Taste	Primary farmer's children	6.0 ± 1.5[a*]	4.9 ± 2.0[b]
		Secondary farmer's children	5.0 ± 1.8[a]	4.3 ± 2.3[b]
	Sweetness	Primary farmer's children	5.5 ± 1.9[a*]	4.2 ± 2.3[b]
		Secondary farmer's children	4.6 ± 2.0[a]	4.2 ± 2.1[a]
Sugar Porridge	Colour	Primary farmer's children	6.6 ± 1.1[a]	6.2 ± 1.2[a]
		Secondary farmer's children	6.7 ± 1.0[a]	6.1 ± 1.5[b]
	Taste	Primary farmer's children	6.5 ± 1.0[a]	6.3 ± 1.2[a]
		Secondary farmer's children	6.6 ± 0.8[a]	6.5 ± 0.9[a]
	Sweetness	Primary farmer's children	6.5 ± 0.9[a]	6.4 ± 1.3[a]
		Secondary farmer's children	6.6 ± 0.9[a]	6.4 ± 1.0[a]
Nsima	Colour	Primary farmer's children	6.8 ± 0.7[a*]	5.8 ± 1.9[b]
		Secondary farmer's children	6.3 ± 1.3[a]	5.6 ± 2.0[b]
	Taste	Primary farmer's children	6.2 ± 1.2[a*]	5.1 ± 2.1[b]
		Secondary farmer's children	5.4 ± 1.8[a]	5.0 ± 2.1[a]
	Sweetness	Primary farmer's children	5.9 ± 1.6[a*]	4.3 ± 2.2[b]
		Secondary farmer's children	5.2 ± 1.8[a]	4.4 ± 2.1[b]

Values represent the mean ± SD of scores of a 1–7 Hedonic Scale. One indicating "dislike" and 7 indicating "like very much". a, b = Values with different letters are significantly different between orange versus white maize in a row at $p \leq 0.05$.* = indicates significant differences between primary and secondary farmers' children for a specific attribute in a column. $n = 76$

Table 2 Total carotenoid content of white and orange maize flours and prepared foods

Maize type	Level of processing	Unit product	Carotenoid Content (mg/kg)
Orange maize	Raw flour	Whole flour	53.1 ± 0.0[a]
	Products	Plain Porridge	56.8 ± 11.0[a]
		Sugar Porridge	31.3 ± 2.7[b]
White maize	Raw flour	Whole flour	2.6 ± 0.1[a]
	Products	Plain Porridge	6.7 ± 2.3[b]
		Sugar Porridge	0.9 ± 0.4[c]
Orange maize	Raw flour	Grand mill flour	61.1 ± 0.5[a]
	Products	*Nsima*	48.7 ± 8.7[b]
White maize	Raw flour	Grand mill flour	1.5 ± 0.1[a]
	Products	*Nsima*	0.5 ± 0.8[b]

Each value is expressed as mean ± SD of triplicate samples. a, b, c = Same column values with different superscript letters are significantly different at $p \leq 0.05$. $n = 6$

white maize flour had significantly higher total carotenoid content than its sweetened porridge. In both cases, plain porridges had higher total carotenoid content than raw whole grain flours.

Carotenoid content of raw flour and cooked products

Table 3 presents the carotenoid content of maize products in the study. The results show that orange maize (whole and grand mill flour) had the highest concentrations of beta-cryptoxanthin. Beta-carotene, present in concentrations of 7.24 μg/g at harvest (Hwang et al. 2016) was undetectable after storage. Carotenoid concentrations were very low in the white maize flours and only zeaxanthin was present at detectable levels.

Table 3 Beta-cryptoxanthin concentration of white and orange maize flours and prepared foods

Maize type	Level of processing	Unit product	Beta-cryptoxanthin Concentration (μg/g)
Orange maize	Raw flour	Whole flour	2.1 ± 0.0[a]
	Products	Plain Porridge	2.2 ± 0.2[a]
White maize	Raw flour	Whole flour	n.d
	Products	Plain Porridge	n.d
Orange maize	Raw flour	Whole flour	2.1 ± 0.0[a]
	Products	Sugar porridge	1.3 ± 0.1[b]
White maize	Raw flour	Whole flour	n.d
	Products	Sugar porridge	n.d
Orange maize	Raw flour	Grand mill flour	2.7 ± 0.1[a]
	Products	*Nsima*	2.1 ± 0.1[b]
White maize	Raw flour	Grand mill flour	n.d
	Products	*Nsima*	n.d

Each value is expressed as mean ± SD of triplicate samples. *n.d.* not detected. a, b = Same column values with different superscript letters are significantly different at $p \leq 0.05$. $n = 6$

To calculate the retention of specific carotenoids, data was compared to the work of Hwang et al. (2016), who determined the concentrations of the same carotenoids in a sample of orange maize of this variety (MW5021) from the same area, at harvest time. Hwang et al. (2016) found that the total content of the four individual carotenoids was 44.26 µg/g and in the present study, 26.4 µg/g was measured. This represents 59% overall retention.

As shown in Table 3, there was also a significant reduction in carotenoid concentration in cooked food products. The beta-cryptoxanthin level was reduced significantly in sweetened porridge and *nsima*. Overall, the orange maize and orange maize products retained significant amounts of carotenoids and, to a limited extent, pro-vitamin A, despite long-term storage, milling and cooking. This is in contrast to white maize, which had no provitamin A detected as expected. In this study, levels of beta-cryptoxanthin in raw whole-grain orange maize were also markedly lower (2.1 ± 0.0 µg/g) than those reported at harvest (5.13 µg/g) (Hwang et al. 2016) which demonstrated 42% retention.

Discussion

Sensory acceptance test

Results from this study demonstrated that landrace orange maize was well accepted in food products (*nsima* and porridge) among school-aged children. Consistently, the novel foods were given higher scores (Table 1). These results are similar to those of Meenakshi et al. 2010 from rural Zambia. They reported that despite negative perceptions, consumers liked orange maize and gave it high marks in organoleptic tests. The high ranking of orange maize products by children is of great importance because these foods are widely consumed by the greater population in Malawi (Flax et al. 2019). A high level of acceptance indicates that there is potential for replacing white maize with orange maize, rich in provitamin A carotenoids. It should be noted that people in Malawi, as well as other countries in southern Africa, commonly use white maize for making *nsima* and porridge (Nuss et al. 2012). Upscaling the production of orange maize to replace white maize can act as a cost-effective, sustainable dietary-based approach to alleviating the problems of vitamin A deficiency, especially among young children (Muzhingi et al. 2008).

The preference for orange maize products over white maize products was seen in all the attributes that were assessed (colour, taste and sweetness) by the children in this study. This agrees with the work of Stevens and Winter-Nelson (2008) in Mozambique where a high preference for orange maize over white maize was also documented. The authors attributed these results to the enhanced aroma and palatability of orange maize products as mentioned by some of the participants. This could be due to higher levels of protein and crude fat in this orange maize compared to white maize (Hwang et al. 2016). It was encouraging to see that this preference was observed in an age group, vulnerable to vitamin A deficiency.

The colour of the carotenoid-containing products also influenced the acceptability of the products. This was observed for the porridges and *nsima* that were evaluated. These results were consistent with those from South Africa, which reported that provitamin A biofortified maize was given higher grades due to its colour over the regular white maize (Awobusuyi et al. 2016). However, some studies have reported conflicting results whereby the colour of the product did not appear to have a major influence on the acceptance of biofortified maize products (yellow/orange), (Pillay et al. 2011) or had a negative effect (Sheftel et al. 2017). In Zambia, the orange colour of the maize was a barrier to acceptance (Sheftel et al. 2017). Resistance occurred because orange maize was associated with yellow maize, which had negative associations with food aid and animal feed. Thus, mothers preferred white maize to orange maize for their children. Interestingly, when taste testing was performed, most children and women in Zambia preferred orange porridge to white, even though barriers were reported regarding the colour. It should be noted that children in the current study most likely did not share the negative association of orange colour with food aid, as in recent years Malawi, for the most part, has produced sufficient food to feed its people (Famine Early Warning Systems Network 2020).

The preference for plain orange maize porridge by both secondary and primary Pro-farmers' children implies that the children found the plain orange porridge to naturally taste good, without the need of adding sugar. Similarly, Pillay et al. (2011) observed that the high acceptance of biofortified yellow maize over white maize among young children was highly influenced by its taste. Generally, the attributes of orange maize products were given higher marks by primary farmers' children when compared to secondary farmers' children (Table 1). Possible explanations are that the children of primary farmers were familiar with orange maize products since they have been eating them for some time. The positive effect of familiarity has been documented in numerous studies (Hong et al. 2014; Torrico et al. 2019).

Despite being in the first farming season of orange maize production, secondary farmers' children also showed a preference for orange maize products over white maize products. This indicates high acceptability and the potential for upscaling the production of orange maize in new areas. These results are in line with the findings of Meenakshi et al. (2010) from Zambia, where consumers who tasted biofortified orange maize nshima (thick paste) just once, and for the first time, gave superior scores to those given the white maize nshima. Furthermore, a preference for orange maize over white maize was documented in Zambian consumers who had been eating biofortified orange maize nshima for longer periods in their homes.

Carotenoid content and concentration

This study confirmed that landrace orange maize in Malawi and derived food products have significantly higher total carotenoid content than white maize (Table 2).

These results correspond with a study of maize biofortified with carotenoids where analyses of white maize samples were found to have low concentrations of total carotenoids with little provitamin A activity (Pixley et al. 2013). In the raw maize flour, higher total carotenoid content was observed in orange maize grand mill flour in comparison to the whole orange maize flour. This is most likely because the grand mill flour is composed entirely of endosperm that has a high concentration of carotenoids. The whole orange maize flour included not only the endosperm, but also the bran (Ndolo and Beta 2013; Taleon et al. 2017).

The results from this study show an increase in carotenoid concentrations in plain porridges in comparison to concentrations in the source flours (Table 2). These results are not easy to explain but might be attributed to variability in cooking times or heat exposure. Díaz-Gómez et al. (2017) reported similar findings in a study conducted in Spain. In Gómaez's work, carotenoid content was higher in high carotene maize porridges after cooking than in uncooked flour. It was suggested that the increase in carotenoid content following cooking might be caused by the denaturation of carotenoid-protein complexes allowing for a better extraction from the food matrix. This explanation has also been suggested by others (Palermo et al. 2014).

Orange maize plain porridge and orange maize nsima were the two products with the highest total carotenoid content while orange maize porridge with sugar had the lowest content (Table 2). This is contrary to the findings of Carvalho et al. (2014), who reported no significant differences in the carotenoid content of cooked pumpkins with added sugar compared to those without added sugar. The reasons for a reduction in carotenoid content in the current study are not clear, but could be explained by a relative dilution of the dried samples that included sugar. Additionally, the temperature of gelatinization of starch is elevated in the presence of sweeteners, so exposure to higher temperatures, and possibly longer cooking times, would affect the degradation of carotenoids (Allan et al. 2020).

Beta-cryptoxanthin was the only provitamin A detected in study samples. In previous reports, analyses of food products from recently harvested orange maize identified high concentrations of beta-carotene and beta-cryptoxanthin (Hwang et al. 2016; Nuss & Tanumihardjo 2010). However, beta-carotene was undetectable in the current study samples. It is important to note that beta-cryptoxanthin was identified at levels of approximately 2.2 μg/g. This concentration was notably lower than the 5.13 μg/g measured in samples close to the time of harvest (Hwang et al. 2016). The reduction in carotenoid content was not unexpected and was attributed to exposure to different degradation factors that occurred during the 10-month storage period. This includes exposure to light, heat and oxygen (Schieber & Weber 2016).

Other studies, (Moura et al. 2015; Taleon et al. 2017) have reported that after 6 months of storage, there was significant degradation of carotenoids in biofortified orange maize that was kept in an uncontrolled environment with degradation reaching up to 60%. At 12 months, there was a 90% degradation of beta-carotene in maize stored in storage rooms with controlled temperatures of 22 °C and 37 °C (Moura et al. 2015). This supports the current findings where the loss of carotenoids, in particular beta-carotene, was observed following storage. The sensitivity of beta-cryptoxanthin to degradation factors is considered to be lower than that of beta-carotene (Pillay et al. 2014). This was confirmed in current study results where beta-carotene could not be identified in the unprocessed and processed orange maize or white maize products. Beta and Hwang (2018) reported that over time, beta-carotene degradation was significantly greater than beta-cryptoxanthin.

Studies done in vitro, in animal models and in humans have shown that beta-cryptoxanthin is better absorbed than other carotenoids (Burri 2015). When compared to other provitamins A carotenoids, beta-cryptoxanthin rich foods have better bioavailability reaching 72.5% in comparison to beta-carotene rich foods. However, beta-cryptoxanthin appears to be a poorer substrate for beta-carotene 15,15′ oxygenase than beta-carotene (Burri 2015). Despite not being able to detect beta-carotene in this study, the samples that were analyzed were relatively yellow to orange in colour. This could be attributed to the presence of other carotenoids such as lutein and zeaxanthin, which also have yellow pigmentation (Nwachukwu et al. 2016).

Notwithstanding the susceptibility of provitamin A in maize to degradation during storage and processing, the findings suggest a possibility of considerable provitamin

A retention. The retention of provitamin A in different food products subjected to storage and processing conditions is highly variable (Rodriguez-Amaya 1999). Pillay et al. (2014) reported that there were product differences in the retention of provitamin A during processing when biofortified maize was milled into mealie meal (relatively coarse flour) or made into samp, Phutu (thick paste) or cooked like a thin porridge. Milling of biofortified maize into mealie meal resulted in higher retention of provitamin A carotenoids than processing into the sump (Pillay et al. 2014). On the other hand, there was also higher retention of provitamin A in cooked Phutu (thick paste) and cooked samp as compared to the thin porridge.

Beta-cryptoxanthin concentration retention was 77% after processing. This is significantly higher than what Mugode et al. (2014) reported in biofortified maize from Zambia. Beta-cryptoxanthin retention was reportedly between 21–29% in prepared nshima (*nsima*). Differences in storage times, milling procedures, light exposure and cooking/processing of maize may contribute to the varied retention concentrations of the different carotenoids. Because the retention of carotenoids increases with shorter thermal exposures, it is important to minimize processing time and ensure proper storage conditions. Carotenoid-containing grains should be kept away from direct light and stored in tight oxygen-free bags or containers (Dutta et al. 2005).

It should be noted that after prolonged storage in uncontrolled conditions, the retention of 89% of total carotenoids and 77% for beta-cryptoxanthin might be an overestimation. Other studies have reported significantly lower retention rates of carotenoids following 6–12 months of storage (Moura et al. 2015; Taleon et al. 2017). Although laboratory protocols were identical to those used to determine levels at the time of harvest (Hwang et al. 2016), different equipment and materials used may contribute to variability. The use of the spectrophotometric identification method of carotenoids is an accepted laboratory methodology; however, reports differ on its accuracy (Wimalasiri et al. 2017; Nagaraj et al. 2022).

Conclusion

This study demonstrates high acceptance of orange maize by school-aged children and that consumption can increase the provitamin A content of the daily diet in Malawi. Orange maize products are highly palatable, have reasonable rates of retention of carotenoids over time and can be used for preparing local foods, replacing white maize. The adoption of orange maize provides a cost-effective option to reduce dependence on supplementation and fortification. Identifying optimal times to harvest landrace orange maize, using longitudinal analyses of carotenoid concentrations at different levels of crop maturity, would capitalize on the nutritional value of this crop. There is also a need for promoting and evaluating good storage practices such as the use of black polyethene bags to minimize carotenoid loss.

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Authors' contributions

AAK, AHS and MCK were involved in the study conceptualisation and writing of the original manuscript draft. AHS and MCK were responsible for funding acquisition for the study. All the authors were involved in developing data collection tools, data collection, reviewing and editing, and approving the final manuscript.

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Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate

This research was conducted in accordance with the Declaration of Helsinki as part of it involved conducting sensory evaluation with human subjects. The study adhered to the three ethical principles including respect for persons, beneficence, and justice. Ethical approval was sought and given by the Malawi National Committee on Research Ethics in Social Sciences and Humanities (NCRSH); Protocol number P.01/19/345. Informed consent was provided either orally (for those who could not write) or by signing a consent form before conducting the interview. The study investigators made sure that individuals who were invited to participate in the research study were given an adequate description of the study that was clear and complete enough for the individual to judge whether they wanted to participate. Consent was obtained from the children's guardians who were legally, mentally and physically able. Assent was also obtained from the school-going children. An emphasis on voluntary participation in the study was made to the participants. To ensure the confidentiality and privacy of the collected information, the participants were assigned random numbers for identification.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests" in this section.

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