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# Potential of pumpkin to combat vitamin A deficiency during complementary feeding in low and middle income countries: variety, provitamin A carotenoid content and retention, and dietary reference intakes

Edward Buzigi<sup>a,b,c</sup> , Kirthee Pillay<sup>a</sup> , and Muthulisi Siwela<sup>a</sup>

<sup>a</sup>Department of Dietetics and Human Nutrition, School of Agricultural, Earth and Environmental Sciences, University of KwaZulu-Natal, Pietermaritzburg, South Africa; <sup>b</sup>Health Economics and HIV/AIDS Research Division (HEARD), University of KwaZulu-Natal, Durban, South Africa; <sup>c</sup>Department of Human Nutrition & Home Economics, Kyambogo University, Kyambogo, Kampala Uganda

## ABSTRACT

The risk of child vitamin A deficiency (VAD) in low and middle income countries (LMICs) begins during the age range of complementary feeding (6–24 months), when children are fed complementary foods (CFs) deficient in vitamin A. However, pumpkin, a source of provitamin A carotenoids (PVACs) is widely cultivated in LMICs, but underutilized as a complementary food. Moreover, when consumed by humans, PVACs are bioconverted to retinol, the active form of vitamin A used by the body. This study evaluated the potential of pumpkin toward combating VAD by reviewing varieties of pumpkin cultivated in LMICs and their provitamin A carotenoid (PVAC) content; retention of PVACs in pumpkin during processing it as a CF; and the extent to which a CF prepared from pumpkin may meet the dietary reference intakes (DRIs) for vitamin A for children aged 6–24 months old. Pumpkin may combat VAD because the varieties cultivated have high  $\beta$ -carotene content, it is a provitamin A biofortifiable food crop, and 100% retention of PVACs was observed when processed using home cooking methods. Feeding less than 50 g of cooked pumpkin per day meets 100% of the recommended dietary allowance (RDA) and adequate intake (AI) of vitamin A for children 6 to 24 months old. Consumption of pumpkin may be used to complement vitamin A supplementation, fortification, and diversification of CFs with animal source foods. For better yield of pumpkin in LMICs, nutrition sensitive agricultural programmes such as biofortification and agronomic management of pumpkin need to be promoted and supported.

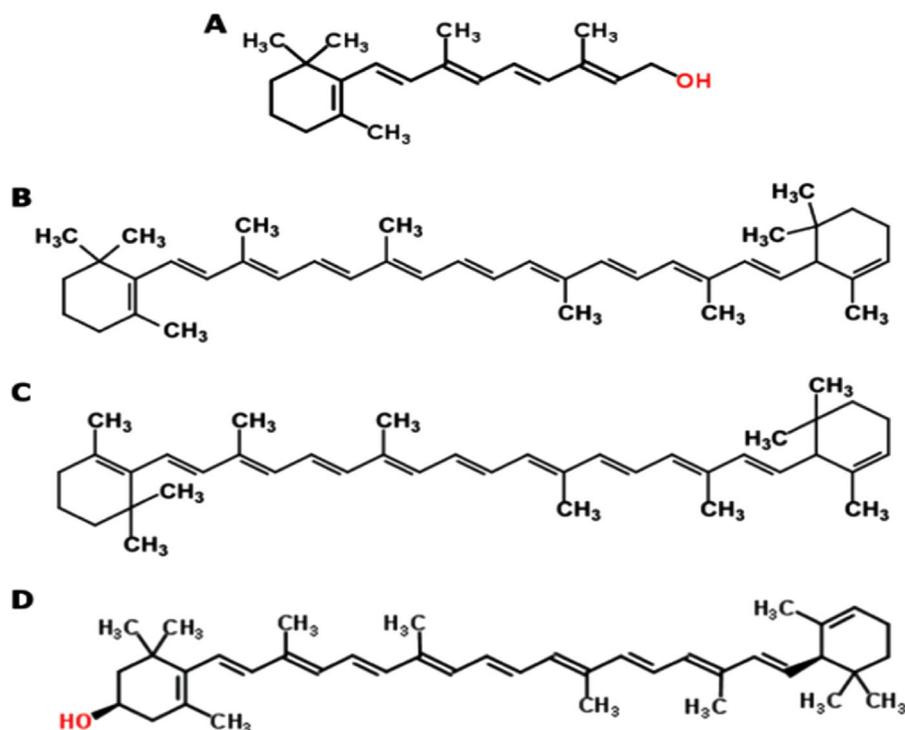
## KEYWORDS

Pumpkin; children 6–24 months; recommended dietary allowance; adequate intake; low and middle income countries

## Introduction

Vitamin A deficiency (VAD), defined as serum retinol below  $0.70 \mu\text{mol/L}$  disproportionately affects children below five years of age from low and middle income countries (LMICs) than high income countries (HICs) (WHO/FAO 2004; WHO 2009a). VAD remains a problem of public health nutrition concern because it is a leading cause of irreversible blindness, and marginal VAD is a risk factor for childhood illnesses such as diarrhea, that leads to high child mortality rates in LMICs (Humphrey, West, and Sommer 1992). By the year 2013, the prevalence of VAD among children below five years in LMICs was 29%, of which sub-Saharan Africa (SSA) and eastern Asia contributed 48% and 44%, respectively (Stevens et al., 2015). The burden of child VAD in LMICs starts during the period of complementary feeding (6–24 months old), when nutritional demands for vitamin A are high (Dewey 2001; Ferraz, Daneluzzi, and Vannucchi 2000). However, infant and young children (IYC) in LMICs are fed complementary foods (CFs) formulated from root tubers and cereals, deficient in vitamin A (Ekesa, Nabuuma, and Kennedy 2019; Gegios et al. 2010;

Gibson et al. 2010). To prevent VAD during the period of complementary feeding, the 2009 World Health Organization (WHO) recommends that caregivers feed their children vitamin A fortified foods, and foods diversified with animal source foods (ASF) because they are rich sources of vitamin A (WHO 2009b). However, fortified foods and ASFs are not affordable and sustainable to the rural and urban poor from LMICs (Faber 2005; Ferguson and Darmon 2007). Several LMICs introduced bi-annual vitamin A supplementation (VAS) programmes for children 6 months to 5 years old (Wirth et al. 2017); however, it has been established that VAS programmes do not prevent VAD (Mason et al. 2015). This is plausible because the liver is unable to store the high dose of  $60,000 \mu\text{g}$  retinol given to children during VAS (200 times the recommended dietary allowance (RDA) for a child 12–24 months old), therefore, the excess vitamin A, retinol is destroyed by the liver and excreted (Blomhoff, Green, and Norum 1992). Moreover, the rise in serum retinol resulting from 6 monthly VAS is small, short-lived, and lasts only for 1–3 months (Pedro et al. 2004). Therefore, daily sustainable dietary intake of provitamin A carotenoid (PVAC) rich foods would provide



**Figure 1.** Structural formula of retinol (A) (Ross et al. 2014),  $\beta$ -carotene (B),  $\alpha$ -carotene (C) and  $\beta$ -cryptoxanthin (D) (Rodriguez-Amaya & Kimura, 2004).

a more practical solution for meeting vitamin A requirements (Ruel, Quisumbing, and Balagamwala 2018; Ruel and Alderman 2013). Pumpkin (*Cucurbita spp*) is cultivated in LMICs but underutilized (Ndegwa 2016; Sharma and Ramana Rao 2013), yet it is a potential source of affordable and sustainable provitamin A carotenoids (PVACs) (Norshazila et al. 2012). Three common PVACs are  $\beta$ -carotene,  $\alpha$ -carotene, and  $\beta$ -cryptoxanthin (Rodriguez-Amaya and Kimura 2004). When PVAC rich foods are eaten, the PVACs are bioconverted into retinol, the active form of vitamin A used by the human body (Tang 2010). Figure 1 shows the structural formulas of PVACs and retinol (Rodriguez-Amaya and Kimura 2004; Ross et al. 2014).

The aim of this study was to review (i) the varieties of pumpkin cultivated or marketed in LMICs and their PVAC content. Furthermore, PVACs are likely to be degraded during preparation or processing provitamin A rich foods before they are ready for human consumption (Bechoff et al. 2017). To this end, this study also (ii) reviewed cooking or processing methods that have a high retention rate of PVACs. A successful complementary feeding strategy should meet the dietary reference intakes (DRIs) for the target nutrient for IYC in the age range of complementary feeding (Lutter and Dewey 2003). The recommended dietary allowance (RDA) is the intake that meets the nutrient needs of almost all (97–98%) individuals in a group, while the adequate intake (AI) is the observed average or experimentally determined intake by a defined population or subgroup that appears to sustain a defined nutritional status, such as growth rate, normal circulating nutrient values, or other functional indicators of health (Trumbo et al. 2001). To this end, this study also (iii) reviewed the extent to which a CF prepared from pumpkin may meet the vitamin A RDA and

AI for IYC in the age group of complementary feeding (6–24 months).

When humans ingest PVAC rich foods, 12  $\mu$ g, 24  $\mu$ g, and 24  $\mu$ g of  $\beta$ -carotene,  $\alpha$ -carotene and  $\beta$ -cryptoxanthin in PVAC rich foods are bioconverted by the liver to 1  $\mu$ g of retinol, respectively (Institute of Medicine 2001). Therefore, based on the Institute of Medicine (2001) bioconversion rates of PVACs to retinol, 1  $\mu$ g of retinol is equivalent to 12  $\mu$ g, 24  $\mu$ g and 24  $\mu$ g of  $\beta$ -carotene,  $\alpha$ -carotene and  $\beta$ -cryptoxanthin, respectively. The retinol activity equivalent (RAE) for PVACs and DRIs for IYC, 6–24 months are shown in Table 1.

## Methods

### Literature search

Potentially relevant publications were searched on ScienceDirect, ISI Web of Knowledge, and Google scholar databases using the following search words: (Nutrient OR carotenoids OR provitamin A carotenoids or  $\beta$ -carotene OR  $\alpha$ -carotene OR  $\beta$ -cryptoxanthin) (content OR composition) of (pumpkin or *Cucurbita species*) (cultivated OR grown OR marketed) in (low and middle income OR developing) countries.

### Selection of studies for evaluation

Studies were eligible for inclusion if they met the following inclusion criteria: (i) study was conducted in a LMIC as defined by the World Bank (World Bank 2018); (ii) Pumpkin under study was cultivated in a LMIC or purchased from a market in a LMIC (iii) outcome of interest

**Table 1.** Retinol activity equivalent for provitamin A carotenoids, and vitamin A dietary reference intakes for children 6–24 months old (Institute of Medicine 2001; Trumbo et al. 2001).

Age group (months)	RDA <sup>a</sup> (μgRAE/day)	AI <sup>b</sup> (μg/day)
0–6		400
7–12		500
13–24	300	

RDA<sup>a</sup> Recommended Dietary Allowance; AI<sup>b</sup> Adequate Intake.

RAE is Retinol activity equivalent (Vitamin A, retinol).

RAE =  $\beta$ -carotene (μg)/12 +  $\alpha$ -carotene (μg)/24 +  $\beta$ -cryptoxanthin (μg)/24 (Institute of Medicine 2001).

was PVAC content. The titles and abstracts of all potentially relevant publications were reviewed to evaluate the relevance of the information; full texts were scrutinized if any potentially relevant information was identified in a retrieved abstract.

### Data extraction

For each article identified, we extracted information on study characteristics including authors, publication year, and the country where the study was conducted, and the method of analyzing the PVAC content of pumpkin.

### Results and discussion

We present details of studies, including authors, year of publication, country where pumpkin was cultivated or marketed, method of PVAC analysis, and variety of pumpkin and corresponding PVAC content in Table 2.

#### Pumpkin varieties in low and middle income countries

Three common varieties of pumpkin were identified to be cultivated or marketed in LMICs i.e. *C. pepo*, *C. moschata* and *C. maxima*, as shown in Table 2. These findings are in agreement with a study by Minguez-Mosquera and colleagues, who reported that a large number of pumpkin varieties (*Cucurbitaceae*), each of which contain different amounts of carotenoids, are cultivated worldwide (Minguez-Mosquera, Hornero-Mendez, and Perez-Galvez 2002). However, in Brazil, Azevedo-Meleiro and Rodriguez-Amaya (2007) showed another variety “*Tetsukabuto*” (*C. maxima* × *C. moschata* hybrid) after a cross breed between *C. maxima* and *C. moschata*. The findings of Azevedo-Meleiro and Rodriguez-Amaya (2007) are consistent with those from a high income country, Austria, where Murkovic, Muller, and Neunteufl (2002) cross bred *C. maxima* and *C. moschata* to form “*C. maxima* × *C. moschata* hybrid”. Cross breeding findings by Azevedo-Meleiro and Rodriguez-Amaya (2007) and Murkovic, Muller, and Neunteufl (2002), may inform that pumpkin is a potential food crop for provitamin A biofortification by conventional plant breeding. Pumpkin crossbreeding showed that the  $\beta$ -carotene content of *C. maxima* alone was low at 1540 μg/100g, compared to the *C. maxima* × *C. moschata* hybrid at 3050 μg/100g (Azevedo-Meleiro and Rodriguez-Amaya 2007), a finding that confirms the definition of

biofortification, which involves conventional breeding of food crops to increase the nutrient density of their edible portions (Meenakshi et al. 2010; Prasad et al. 2015). Moreover, other studies have also argued that it is possible to biofortify pumpkin for higher  $\beta$ -carotene content (Paris 2011; Prasad et al. 2015). Over 90% of childhood VAD in LMICs is found in SSA and Asia (Stevens et al., 2015). However, based on Table 2, several LMICs from SSA (Buzigi, Pillay, and Siwela 2020b; Pepping, Vencken, and West 1988) and Asia (Koh and Loh 2018; Norshazila et al. 2014; Pandey et al. 2003; Usha, Lakshmi, and Ranjani 2010), cultivate pumpkin. Therefore, these findings may indicate that there is an existing opportunity to promote and support the utilization of pumpkin as a CF to combat childhood VAD in SSA and Asia.

Provitamin A biofortification in most LMICs, including those from SSA has targeted mainly three food crops i.e., maize, orange-fleshed sweet potato (OFSP) and cassava (Saltzman et al. 2013). However, pumpkin cross breeding studies by Azevedo-Meleiro and Rodriguez-Amaya (2007) and Murkovic, Muller, and Neunteufl (2002), suggest that pumpkin is a promising food crop that could be added to the provitamin A biofortifiable food crops in countries where it is cultivated, and which have high prevalences of VAD such as those from eastern Asia and SSA (Stevens et al., 2015). It is worth noting that pumpkin was added to the provitamin A biofortified food crops list in Brazil (Saltzman et al. 2013). However, there is a need to evaluate its efficacy on vitamin A status. Previously, VAD was reported in Brazilian children aged 6–24 months (Ferraz, Daneluzzi, and Vannucchi 2000), a group suitable for formulating a CF from biofortified pumpkin to evaluate its efficacy on vitamin A status.

#### Provitamin A carotenoid content of pumpkin and varieties

##### Provitamin A carotenoids tested

The PVAC content of various varieties of pumpkin (*Cucurbitaceae*) is presented in Table 2. All the studies reviewed tested for  $\beta$ -carotene content in pumpkin. However, the majority of studies did not test for  $\alpha$ -carotene (Azizah et al. 2009; Kim et al. 2012; Koh and Loh 2018; Muzzaffar et al. 2016; Pandey et al. 2003) and  $\beta$ -cryptoxanthin content (Azevedo-Meleiro and Rodriguez-Amaya 2007; Azizah et al. 2009; Bergantin et al. 2018; Koh and Loh 2018; Murkovic, Muller, and Neunteufl 2002; Muzzaffar et al. 2016; Pepping, Vencken, and West 1988; Provesi, Dias, and Amante 2011) in pumpkin. However, pumpkin is a potential source of all PVACs (Hidaka, Anno, and Nakatsu 1987). Neglecting to assess for  $\alpha$ -carotene and  $\beta$ -cryptoxanthin may lead to under estimation of the PVACs content from pumpkin, yet such findings may be helpful when developing food composition tables and preparing foods for special groups, including IYC (Hotz et al. 2012; Williamson 2005).

All studies tested for the content of  $\beta$ -carotene and at least one other provitamin A carotenoid. These studies consistently showed that  $\beta$ -carotene was significantly higher

**Table 2.** Provitamin A carotenoids content of pumpkin varieties cultivated in low and middle income countries.

Reference	Country	Method	Variety	Provitamin A carotenoids mean content Expressed as µg/100g of edible portion		
				β-carotene	α-carotene	β-Cryptoxanthin
(Kim et al. 2012)	Korea	HPLC	<i>C. pepo</i>	148	NT	ND
			<i>C. moschata</i>	570	NT	ND
			<i>C. maxima</i>	1704	NT	65
(Carvalho et al. 2012)	Brazil	HPLC	<i>C. moschata-A</i>	24422	6706	NT
			<i>C. moschata-B</i>	14195	7299	NT
			<i>C. moschata</i> "Menina Brasileira".	6670	2680	NT
(Azevedo-Meleiro and Rodriguez-Amaya 2007)	Brazil	HPLC	<i>C. moschata</i> "Goianinha"	5670	2380	NT
			<i>C. maxima</i>	1540	ND	NT
			<i>C. maxima</i> × <i>C. moschata</i> hybrid 'Tetsukabuto'	3050	TR	NT
			<i>C. pepo</i>	540	ND	NT
(Pandey et al. 2003)	India	HPLC	<i>C. moschata</i>	234-1485	NT	NT
(Provesi, Dias, and Amante 2011)	Brazil	HPLC	<i>C. moschata</i>	1945	1260	NT
			<i>C. maxima</i>	1338	43	NT
			<i>C. moschata</i>	17220	3995	NT
(Carvalho et al. 2014)	Brazil	HPLC	<i>C. moschata</i>	1980	NT	NT
(Azizah et al. 2009)	Malaysia	HPLC	<i>C. moschata</i>	1980	NT	NT
(Koh and Loh 2018)	Malaysia	HPLC	<i>C. maxima</i>	4340	NT	NT
(Tee and Lim 1991)	Malaysia	HPLC	<i>C. maxima</i>	578-1170	75.6	NT
(Pepping, Vencken, and West 1988)	Tanzania	HPLC	<i>C. moschata</i>	1170	1100	NT
(Norshazila et al. 2014)	Malaysia	HPLC	<i>C. moschata</i>	2916 – 154760	1260- 10200	NT
(Usha, Lakshmi, and Ranjani 2010)	India	HPLC	<i>C. moschata</i>	485-1079.6	NT	NT
(Buzigi, Pillay, and Siwela 2020b)	Uganda	HPLC	<i>C. moschata</i>	1704	46	NT
(Nakazibwe, Olet, and Kagoro-Rugunda 2020)	Uganda	HPLC	<i>C. moschata</i>	2700-121549	1797-88900	NT

NT; Not tested, ND; Not Detected, HPLC; High Performance Liquid Chromatography

than other PVACs, irrespective of pumpkin variety (Azevedo-Meleiro and Rodriguez-Amaya 2007; Carvalho et al. 2012, 2014; Nakazibwe, Olet, and Kagoro-Rugunda 2020; Norshazila et al. 2014; Pepping, Vencken, and West 1988; Provesi, Dias, and Amante 2011; Tee and Lim 1991). These findings are in agreement with other studies that were conducted in HICs such as Japan (Hidaka, Anno, and Nakatsu 1987), United States of America (USA) (Itle and Kabelka 2009), and Italy (Bergantin et al. 2018). The high concentrations of β-carotene in pumpkin may have implications toward improving vitamin A status because β-carotene has a twofold provitamin A activity compared to other PVACs (FAO/WHO 1967; Institute of Medicine 2001).

Furthermore, this review revealed that there are variations in PVAC composition in pumpkin cultivated within or across countries. The β-carotene content in *C. moschata* varied across countries at 9680 µg/100g, 24400 µg/100g, 1980 µg/100g, and 1170 µg/100g in India (Muzzaffar et al. 2016), Brazil (Carvalho et al. 2012), Malaysia (Azizah et al. 2009), and Tanzania (Pepping, Vencken, and West 1988), respectively. A similar variation was observed in *C. maxima* i.e. 4340 µg/100g, 5670 µg/100g, and 1700 µg/100g in Malaysia (Koh and Loh 2018), Brazil (Azevedo-Meleiro and Rodriguez-Amaya 2007), and Korea (Kim et al. 2012), respectively. These variations are consistent with studies from high income countries. The β-carotene of *C. moschata* was 505 µg/100g, 402 µg/100g, 2600 µg/100g, and 5100 µg/100g in Japan (Hidaka, Anno, and Nakatsu 1987), USA (Itle and Kabelka 2009), Italy (Bergantin et al. 2018), and Austria (Murkovic, Mulleder, and Neunteufl 2002), respectively. These variations may inform that cultivation of pumpkin in different localities may affect its PVAC content. This may be due to differences in the cultivation environments like soil fertility, climate change, and water stress that influence the

productivity of pumpkin (Ferrandino and Smith 2007; Langeroodi et al. 2019; Yavuz et al. 2015) and its PVAC content (Norshazila et al. 2014). Therefore, agronomic management, the caring for plants to overcome cultivation stress factors (Moswetsi, Fanadzo, and Ncube 2017), would be important in improving pumpkin yield.

Intra country variations of PVAC content was also observed. In Brazil, *C. moschata* had superior PVAC composition, compared to *C. maxima* i.e. *C. moschata* had 1945 µg/100g and 1260 µg/100g of β-carotene and α-carotene, respectively (Provesi, Dias, and Amante 2011). Consistent findings in Brazil showed that *C. moschata* had a superior PVAC content than *C. maxima* (Azevedo-Meleiro and Rodriguez-Amaya 2007). Similar findings were revealed in a high income country, Austria, which showed that the β-carotene content was higher in *C. moschata* (5100 µg/100g) than in *C. maxima* (4400 µg/100g), when both were cultivated under similar environmental conditions (Murkovic, Mulleder, and Neunteufl 2002). Cultivation environment such as climatic differences in different regions of the same country could be an explanation for the intra-country variations of PVACs in pumpkin. For example, the species *C. moschata* and *C. maxima* cultivated in the southern region of Brazil, where the average temperatures are lower than throughout the rest of the country, showed lower concentrations of PVACs in relation to the same varieties harvested in the northeast and southeast regions of Brazil (Azevedo-Meleiro and Rodriguez-Amaya 2007; Rodriguez-Amaya et al. 2008).

In contrast, in a middle income country, Korea, the β-carotene content of *C. maxima* (1704 µg/100g) was significantly higher than in *C. moschata* (570 µg/100g). Similar findings in two high income countries. i.e., Japan (Hidaka, Anno, and Nakatsu 1987) and Italy (Bergantin et al. 2018),

showed that the  $\beta$ -carotene content in *C. maxima* was higher than in *C. moschata*. However, in Italy,  $\alpha$ -carotene in *C. moschata* was significantly higher at 1756  $\mu\text{g}/100\text{g}$ , compared to non-detectable in *C. maxima* (Bergantin et al. 2018). Such findings are very important in food and nutrition security programmes because they can inform food and nutrition security policy makers, agencies and agricultural officers to identify and choose a variety of pumpkin with superior PVAC composition, so that they can support and promote its cultivation and consumption at community or household level (home gardening), to alleviate VAD (Ruel and Alderman 2013). When such programmes are implemented in the community through home gardening of PVAC rich foods, they improve the vitamin A status of children (Faber and Laurie 2011; Faber and Van Jaarsveld 2007; Faber et al. 2002; Gershon 1985).

Differences in PVAC content could also be due to genetic variation in pumpkin. Carvalho and colleagues, evaluated the variability of  $\beta$ -carotene among biofortified pumpkin genotypes of *C. moschata*, cultivated under similar environmental conditions (Carvalho et al. 2015). The pumpkins showed a large variability in the total PVAC content, varying from 69900  $\mu\text{g}/100\text{g}$  (genotype 25) to 12460  $\mu\text{g}/100\text{g}$  (genotype 26) in fresh pumpkin (Carvalho et al. 2015). These findings are consistent with Ribeiro et al. (2015), who showed that the PVACs in five genotypes of provitamin A biofortified orange-fleshed pumpkin (*C. moschata*) were found to range from 20900  $\mu\text{g}/100\text{g}$  to 65800  $\mu\text{g}/100\text{g}$  (Ribeiro et al. 2015). These findings suggest that, in addition to provitamin A biofortification by conventional breeding (Azevedo-Meleiro and Rodriguez-Amaya 2007; Murkovic, Mulleder, and Neunteufl 2002), pumpkin can also be biofortified by genetic modification. Moreover, the PVAC content in provitamin A biofortified pumpkin by genetic modification (Carvalho et al. 2015; Ribeiro et al. 2015), was significantly higher than those biofortified by conventional breeding (Azevedo-Meleiro and Rodriguez-Amaya 2007; Murkovic, Mulleder, and Neunteufl 2002). Besides, the  $\beta$ -carotene content (22300-52200  $\mu\text{g}/100\text{g}$ ) in biofortified pumpkin (Ribeiro et al. 2015), is higher than the provitamin A biofortified OFSP “Resisto,” a variety with the highest amounts of  $\beta$ -carotene (11987-20525  $\mu\text{g}/100\text{g}$ ) among all varieties of OFSP cultivated in South Africa (Laurie et al. 2012).

### **Retention of provitamin A carotenoids in pumpkin during food preparation**

#### **Home cooking methods**

It is clear that pumpkin is an excellent rich source of PVACs for human consumption (Carvalho et al. 2012; Carvalho et al. 2015; Norshazila et al. 2014; Ribeiro et al. 2015). However, pumpkin flesh cannot be consumed raw and has to be processed into a ready-to-eat CF fit for IYC. Due to the presence of double bonds in their structure, Figure 1 (Rodriguez-Amaya and Kimura 2004), PVACs in pumpkin are susceptible to chemical degradation reactions

(isomerism and oxidation) during preparation and processing (Bechoff et al. 2017).

Pumpkin can be processed into ready-to-eat food by using home cooking methods. Carvalho et al. (2014) showed that the mean  $\beta$ -carotene content increased from 17220  $\mu\text{g}/100\text{g}$  in raw pumpkin to 18480  $\mu\text{g}/100\text{g}$  and 20200  $\mu\text{g}/100\text{g}$  in boiled and steamed pumpkin, respectively. A similar trend was observed with  $\alpha$ -carotene (Carvalho et al. 2014). A study by Carvalho et al. (2014) indicates that either boiling or steaming pumpkin retains over 100% of PVACs,  $\beta$ -carotene and  $\alpha$ -carotene. These findings are consistent with Buzigi, Pillay, and Siwela (2020b) and Azizah et al. (2009), who showed that either boiling or steaming pumpkin retained over 100% of  $\beta$ -carotene and  $\alpha$ -carotene. Moreover, over 100% PVAC retention was also observed after boiling or steaming provitamin A biofortified pumpkin (Carvalho et al. 2015). Studies by Carvalho et al. (2014) and Carvalho et al. (2015) are consistent with several other studies that showed that PVAC retention was over 100% in pumpkin puree prepared from either boiled or steamed pumpkin (Bulatova et al. 2018; Buzigi, Pillay, and Siwela 2020a; Maier et al. 2008; Provesi, Dias, and Amante 2011). The higher retention of  $\beta$ -carotene content observed in cooked pumpkin may be due to the *cis*- $\beta$ -carotene isomers that increase on heating PVAC rich foods (Azizah et al. 2009; Bechoff et al. 2017; Bengtsson et al. 2008; De Moura, Miloff, and Boy 2015; Mugode et al. 2014). Furthermore, maceration and heat processing improve  $\beta$ -carotene bioaccessibility from PVAC rich foods, which is probably due to rupture of the microstructure of plant tissue and subsequent release of PVACs from the complex food matrix (Provesi and Amante 2015; Tumuhimbise, Namutebi, and Muyonga 2009).

In general, pumpkin prepared using water, such as boiled and steamed pumpkin, has higher PVAC retention than foods prepared using cooking oil, such as fried or stir-fried pumpkin (Azizah et al. 2009; Sungpuag et al. 1999; Veda, Platel, and Srinivasan 2010). The possible explanation is that PVACs are fat soluble, and thus the frying process also causes a higher loss by lixiviation of PVACs (Concepcion et al. 2018). However, pumpkin prepared with cooking oil has a higher bioavailability of PVACs after ingestion, because it facilitates PVACs absorption and utilization by the human body (La Frano et al. 2014; Veda, Platel, and Srinivasan 2010).

#### **Pumpkin powder and provitamin A carotenoid retention**

Pumpkin flour/powder is one of the main processed pumpkin products (Ezeji and Ojimekwe 1993; Ravi, Menon, and Anupama 2010; Victor, Oguntuase, and Vincent 2013). To formulate pumpkin powder or flour, it has to be dehydrated before it is milled into powder. In a study involving the drying of pumpkin, the highest retention of PVACs was obtained with dehydration by freeze drying, followed by vacuum-microwave, only vacuum, and lastly the traditional sun drying method (Nawirska et al. 2009). In India, the  $\beta$ -carotene content of raw flesh and powdered pumpkin was 1080  $\mu\text{g}/100\text{g}$  and 4860  $\mu\text{g}/100\text{g}$  of pumpkin, respectively (Usha, Lakshmi, and Ranjani 2010), implying that

processing of pumpkin fresh to pumpkin powder retained the  $\beta$ -carotene content in pumpkin by over 100%. The over 100% retention, could be explained by the dehydration and milling which are characterized by heat treatment and maceration, respectively. It is worth noting that heat processing and maceration improve  $\beta$ -carotene bioaccessibility from PVAC rich foods, which is perhaps due to rupture of the microstructure of plant tissue and later release of PVACs from the complex food matrix (Provesi and Amante 2015; Tumuhimbise, Namutebi, and Muyonga 2009). Another study conducted in Thailand showed that the  $\beta$ -carotene content in pumpkin powder was high at 11,094  $\mu\text{g}/100\text{g}$  (Aukkanit and Sirichokworrakit 2017).

Micronutrient powders (MNP) including those rich in vitamin A have been used in home fortification of home-made foods including CFs in LMICs to increase the micronutrient density of CFs (Chen et al. 2011; De-Regil et al. 2013). In Bangladesh, a cereal CF was fortified with pumpkin powder with  $\beta$ -carotene content of 11000  $\mu\text{g}/100\text{g}$  (Alam et al. 2013). Furthermore, Ravi, Menon, and Anupama (2010) developed a pumpkin MNP with a  $\beta$ -carotene content of 4857.6  $\mu\text{g}/100\text{g}$ , and used it to fortify a CF “instant dhokla mix”. After fortification with this pumpkin MNP, the  $\beta$ -carotene content of “instant dhokla mix” increased by 8.4% (Ravi, Menon, and Anupama 2010), suggesting that the pumpkin MNP improved the  $\beta$ -carotene content of the CF. However,  $\beta$ -carotene content reduced from 4857.6  $\mu\text{g}/100\text{g}$  in the pumpkin powder to 112.9  $\mu\text{g}/100\text{g}$  in the “instant dhokla-pumpkin powder mix” (Ravi, Menon, and Anupama 2010). This may indicate that the cereal, “instant dhokla” diluted the  $\beta$ -carotene content in pumpkin powder on mixing pumpkin powder and the cereal to form “instant dhokla-pumpkin powder mix”. Such a reduction in PVAC content in pumpkin was also observed by Buzigi, Pillay, and Siwela (2020a), after mixing pumpkin with common bean to form common bean pumpkin blend (BPB). It is likely that common bean diluted the PVAC content of pumpkin on blending pumpkin to form BPB (Buzigi, Pillay, and Siwela 2020a). Studies by Ravi, Menon, and Anupama (2010) and Buzigi, Pillay, and Siwela (2020a) may inform food scientists and nutritionists to consider the possible dilution of PVACs in pumpkin on blending pumpkin with other foods deficient in PVACs, when formulating food mixtures enriched with PVACs.

#### **Complementary food from pumpkin and vitamin A dietary reference intake**

When consumed, PVACs in foods are bioconverted to retinol, the active form of vitamin A needed by the body (Tanumihardjo, Palacios, and Pixley 2010). The Food and Agriculture Organization of the United Nations (FAO)/World Health Organization (WHO) of 1967 (FAO/WHO 1967), and the United States (US) Institute of Medicine of 2001 (Institute of Medicine 2001), established recommendations for the bioconversion of PVACs to retinol. The former recommends that 6  $\mu\text{g}$  of  $\beta$ -carotene is equivalent to 1  $\mu\text{g}$  of retinol, whereas 12  $\mu\text{g}$  of all other PVACs each, are equivalent to 1  $\mu\text{g}$  of retinol (FAO/WHO 1967). However, in 2001,

the latter questioned the former, and it recommended that 12  $\mu\text{g}$  of  $\beta$ -carotene is equivalent to 1  $\mu\text{g}$  of retinol, whilst 24  $\mu\text{g}$  of other PVACs each, are equivalent to 1 retinol (Institute of Medicine 2001). Therefore, we discuss the bioconversion of PVACs in pumpkin to vitamin A using the Institute of Medicine (2001) recommendations, and compare it with the vitamin A RDA and AI for IYC, 6 to 24 months old (Trumbo et al. 2001) (Table 1).

#### **Cooked unbiofortified pumpkin and dietary reference intakes**

In Brazil, the mean content of  $\beta$ -carotene and  $\alpha$ -carotene in boiled pumpkin was 18,480  $\mu\text{g}/100\text{g}$  and 4397  $\mu\text{g}/100\text{g}$ , respectively, whilst the mean content of  $\beta$ -carotene and  $\alpha$ -carotene in steamed pumpkin was 20,200  $\mu\text{g}/100\text{g}$  and 4709  $\mu\text{g}/100\text{g}$ , respectively (Carvalho et al. 2014). To this end, boiled pumpkin would contribute 1540  $\mu\text{g}$  and 183  $\mu\text{g}$  of retinol from  $\beta$ -carotene and  $\alpha$ -carotene respectively, giving a total of 1723  $\mu\text{g}$  of retinol per 100 g of unbiofortified pumpkin consumed. Therefore, only 17.5 g, 23.5 g and 29 g of unbiofortified Brazilian boiled pumpkin (Carvalho et al. 2014), would be required to meet the RDA and AI for vitamin A for 13 to 24, 6, and 7 to 12 months old IYC, respectively.

Furthermore, steamed pumpkin would contribute 1680  $\mu\text{g}$  and 196  $\mu\text{g}$  of retinol from  $\beta$ -carotene and  $\alpha$ -carotene, respectively, giving a total of 1880  $\mu\text{g}$  of retinol per 100 g of unbiofortified pumpkin consumed. Therefore, only 16 g, 22 g and 27 g of unbiofortified steamed pumpkin (Carvalho et al. 2014), would be required to meet the vitamin A RDA and AI for a child 13 to 24, 6, and 7 to 12 months old, respectively.

#### **Provitamin A biofortified pumpkin and vitamin A dietary reference intakes**

The  $\beta$ -carotene content of boiled and steamed provitamin A biofortified pumpkin was 10,700  $\mu\text{g}/100\text{g}$  to 65,550  $\mu\text{g}/100\text{g}$  and 11,760  $\mu\text{g}/100\text{g}$  to 65,050  $\mu\text{g}/100\text{g}$ , respectively. Therefore, 100 g of provitamin A biofortified boiled and steamed pumpkin would provide 891  $\mu\text{g}$  to 5463  $\mu\text{g}$  and 980  $\mu\text{g}$  to 5421  $\mu\text{g}$  of retinol, respectively. Hence, only 5.5 g to 34 g and 6 g to 31 g of provitamin A biofortified boiled and steamed pumpkin (Carvalho et al. 2015), would be required to meet the vitamin A RDA for a 13–24-month old child, respectively. Furthermore, only 7.5 g to 45 g and 7.5 g to 41 g of provitamin A biofortified boiled and steamed pumpkin, respectively, would be required to meet the vitamin A AI for a 6 month old infant (Carvalho et al. 2015). The average suggested amount per serving for children aged 6 to 24 months old during complementary feeding is at least 50 g of CF (Lutter and Dewey 2003). However, a less amount of boiled or steamed pumpkin (Carvalho et al. 2015), would meet the Vitamin A RDA and AI for IYC, 6–24 months old compared to the daily recommended amount for this age group.

### Pumpkin powder and vitamin A dietary reference intakes

In India, a  $\beta$ -carotene rich (7010  $\mu\text{g}/100\text{g}$ ) ready to serve soup “Instant Soup Mix (ISM)” was developed from pumpkin powder (Dhiman et al. 2017). Therefore, 100 g of ISM would provide 584  $\mu\text{g}$  of retinol, if consumed. Hence, only 52 g of ISM would be needed to meet the RDA/AI for vitamin A for a child 13 to 24 months old. Furthermore, a cereal based complementary food mix “instant dhokla mix” was fortified with pumpkin flour containing 4860  $\mu\text{g}/100\text{g}$  of  $\beta$ -carotene (Ravi, Menon, and Anupama 2010). This translates to 405  $\mu\text{g}$  of retinol/100g of pumpkin powder consumed. Therefore, 99 g and 75 g of pumpkin powder would be needed to meet the vitamin A AI and RDA for a 6 and 13 to 24 months old child, respectively.

### Strengths and limitations

This review included studies that tested the PVAC content using high performance liquid chromatography (HPLC) (Table 2). HPLC is considered as a gold standard method for PVAC analysis probably because it has a significantly greater power of resolution, reproducibility, and high speed of separation of these carotenoids (Melendez-Martinez, Vicario, and Heredia 2007). Furthermore, we evaluated the contribution of pumpkin toward meeting the RDA/AI for vitamin A using ready-to-eat foods such as boiled and steamed pumpkin. This evaluation would give more valid results compared to other studies which used raw foods to evaluate the contribution of provitamin A rich foods toward meeting DRIs (Faber, Laurie, and Van Jaarsveld 2013; Laurie et al. 2015). It is worth noting that IYC are not fed on raw pumpkin, and using raw pumpkin to evaluate the contribution of PVACs toward meeting vitamin A requirements would be questionable because cooking methods greatly affect the PVAC content of pumpkin (Azizah et al. 2009; Sungpuag et al. 1999; Veda, Platel, and Srinivasan 2010). However, there is evidence that PVAC retention in pumpkin is over 100% after cooking it using common cooking methods for pumpkin such as boiling and steaming, used in LMICs (Buzigi, Pillay, and Siwela 2020b; Carvalho et al. 2012). The main limitation of this study generates from the fact that many of the studies we reviewed did not test for all the PVACs, yet pumpkin is a potential source of the three known PVACs,  $\beta$ -carotene,  $\alpha$ -carotene, and  $\beta$ -cryptoxanthin (Hidaka, Anno, and Nakatsu 1987). This might have led to an underestimation of PVACs in pumpkin. This could subsequently have led to underestimation of the vitamin A contribution from pumpkin.

### Conclusion and recommendations

The commonest varieties of pumpkin cultivated or marketed in LMICs are *C. maxima*, *C. moschata* and *C. pepo*. However, other varieties are emerging due to biofortification. There are few studies from SSA and eastern Asia that evaluated the PVAC content of pumpkin, yet they harbor the highest proportion of children with VAD. Therefore, more studies that evaluate the PVAC content of pumpkin

should be conducted in SSA and eastern Asia. Most studies were limited to testing  $\beta$ -carotene only, yet pumpkin is a potential source of other PVACs including  $\alpha$ -carotene and  $\beta$ -cryptoxanthin. Therefore, future studies should consider testing for all the three known PVACs. Generally, pumpkin contains higher concentrations of  $\beta$ -carotene compared to other PVACs, and is a potential food crop for provitamin A biofortification. However, variations in PVAC content exist in pumpkin across LMICs, suggesting that the cultivation environment of pumpkin for better yield is important. Home based cooking methods such as boiling and steaming have higher retention rates for PVACs in pumpkin. Less than 50 g of both provitamin A biofortified and unbiofortified boiled or steamed pumpkin have the potential to meet 100% of the vitamin A RDA and AI for IYC, 6–24 months old. Therefore, CFs formulated from pumpkin may improve the vitamin A status of IYC in the age range of complementary feeding. It is worth noting that the consumption of pumpkin should not replace the already existing strategies such as VAS; provitamin A biofortification; diversification of CFs with ASF; and fortification used to combat micronutrient deficiencies. However, consumption of pumpkin should be used in combination with the aforementioned existing strategies that aim to combat VAD in LMICs.

### Disclosure statement

The authors declare that they have no conflict of interest.

### Contributor statement

E.B conceptualized the study; E.B searched data bases and performed the selection of studies; K.P and M.S played leadership roles throughout all stages of the study; E.B wrote the first draft of the manuscript; K.P and M.S critically evaluated the review and commented on it; E.B, K.P and MS critically reviewed and edited the manuscript. All authors reviewed and approved the manuscript.

### ORCID

Edward Buzigi  <http://orcid.org/0000-0002-3605-8111>  
Kirthi Pillay  <http://orcid.org/0000-0003-3161-1050>

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