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# Artículo de Revisión / Review Article

Biofortified sweet potatoes as a tool to combat vitamin A deficiency: Effect of food processing in carotenoid content

El camote biofortificado como herramienta para combatir la deficiencia de vitamina A: Efecto del procesamiento de alimentos sobre el contenido de carotenoides

Joseana Severo<sup>1\*</sup>. https://orcid.org/0000-0003-0571-1955 Felipe Nardo dos Santos<sup>2</sup>. https://orcid.org/0000-0002-4058-7622 Tarcísio Samborski<sup>3</sup>. https://orcid.org/0000-0002-4632-9763 Rosane Rodrigues<sup>2</sup>. https://orcid.org/0000-0001-8172-2053 Alexandre Furtado Silveira Mello<sup>4</sup>. https://orcid.org/0000-0003-2477-9517

Department of Food Production, Campus Santa Rosa, Federal Institute Farroupilha, Santa Rosa, RS, Brazil.
Department of Agroindustrial Science and Technology, Federal University of Pelotas, Pelotas, RS, Brazil.
Department of Food Production, Campus Santo Augusto, Federal Institute Farroupilha, Santo Augusto, RS, Brazil.
Brazilian Agricultural Research Company (EMBRAPA), Brasilia-DF, Brazil.

\*Corresponding author: Joseana Severo. Federal Institute Farroupilha, Campus Santa Rosa, Department of Food Production. Santa Rosa, RS, 98787-740, Brazil. E-mail: joseana.severo@iffarroupilha.edu.br

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### ABSTRACT

The supply of food products that present adequate nutritional quality is extremely important for maintaining the health of the population. Thus, different techniques have been used to obtain biofortified foods, with the aim of combating malnutrition caused by the absence of essential micronutrients, especially in the poorest populations. This review presents an overview of biofortification, with an emphasis on orange-flesh sweet potatoes (OFSP), and points out the effects of food processing on nutritional compounds. The identification of cultivars and biofortification actions to obtain biofortified OFSP by conventional breeding are presented as affordable strategies to supply  $\beta$ -carotene to alleviate vitamin A deficiency, without having ethical dilemmas related to transgenics. Studies using OFSP have shown promising results in obtaining foods with high levels of carotenoids. However, biofortified species must be validated for crop production viability, target micronutrient bioavailability and bioaccessibility, as well as the effect of processing on nutrients, so that the benefits to human health are effectively achieved.

*Keywords: β*-carotene; Food technology; Ipomoea batata; Plant breeding.

#### RESUMEN

El suministro de alimentos que presenten una adecuada calidad nutricional es de suma importancia para el mantenimiento de la salud de la población. Así, se han utilizado diferentes técnicas para la obtención de alimentos biofortificados, con

el objetivo de combatir la desnutrición provocada por la ausencia de micronutrientes esenciales, especialmente en las poblaciones más pobres. Esta revisión presenta una descripción general de la biofortificación, con énfasis en los camotes de pulpa anaranjada (BDPA), y señala los efectos del procesamiento de alimentos sobre los compuestos nutricionales. La identificación de cultivares y acciones de biofortificación para obtener BDPA biofortificado por mejoramiento convencional se presentan como estrategias accesibles para aportar  $\beta$ -caroteno para aliviar la deficiencia de vitamina A, sin presentar dilemas éticos relacionados con los transgénicos. Los estudios que utilizan BDPA han mostrado resultados prometedores en la obtención de alimentos con altos niveles de carotenoides. Sin embargo, las especies biofortificadas deben validarse no solo por la viabilidad de la producción vegetal, sino también por la biodisponibilidad y bioaccesibilidad del micronutriente objetivo, así como el efecto del procesamiento sobre los nutrientes, de modo que los beneficios para la salud humana se logren de manera efectiva.

Palabras clave: β-caroteno; Fitomejoramiento; Ipomoea batata; Tecnología en alimentos.

## INTRODUCTION

Malnutrition is related not only to a lack of food but also to the low intake of micronutrients essential for the healthy functioning of the human body. Malnutrition is the leading cause of death in the world, with nutritional deficiencies and chronic diseases related to diet causing millions of deaths annually<sup>1,2</sup>. Micronutrient deficiency (vitamins and minerals) is also known as a "hidden hunger," because most affected people do not show signs typically associated with hunger and malnutrition. This occurs when micronutrient uptake or absorption is too low to maintain health and promote the proper development of children, and normal physical and mental functions<sup>1</sup>.

It is estimated that micronutrient deficiency affects more than 2 billion people, or 1 in 3 persons worldwide<sup>3,4</sup>. Micronutrients related to hidden hunger include iron (Fe), zinc (Zn) and vitamin A (VA). Iron deficiency affects the physical and intellectual development of children, favoring premature birth<sup>5</sup>. VA and Zn deficiencies may alter the immune system; Zn deficiency also affects child growth and is related to various diseases or physiological dysfunctions, including reproductive abnormalities, skin lesions and anemia, diarrhea, anorexia, cognitive impairment and dysfunction, diabetes mellitus, impaired visual function, osteoporosis, cirrhosis of the liver, intestinal disease and some tumors<sup>6,7</sup>. VA deficiency results in 600,000 deaths a year in the world, mostly among children and pregnant women<sup>8</sup>.

The improvement of species, with the objective of increasing the nutrient content important for human health, is an interesting alternative in the reduction of nutritional deficiencies, with consequences for food security, especially in the poorest countries<sup>4,9</sup>. However, the improvement of species through transgenic techniques, aimed to improve the nutritional content of foods, is a highly debated area and full of ethical dilemmas<sup>3,10</sup>.

While conventional food fortification requires the addition of fortifying ingredients to the products, biofortification involves the synthesis or accumulation of nutrients by the raw materials<sup>11</sup>. The biofortification of species has already proven to be an effective, low-cost technique that is being adopted by small farmers around the world, focusing on populations with difficult access to a diversified diet, supplements or commercially fortified foods<sup>4</sup>. The biofortification of orange-flesh sweet potato (OFSP) with  $\beta$ -carotene is considered an affordable strategy in

tackling VA deficiency, since these potatoes are produced and consumed in several countries  $^{12}$ .

In this way, food technology has been improved to ensure that food processing results in quality and quantity products for the population. However, although nutritional losses during processing are often unavoidable, they can be minimized by the use of appropriate processing technologies<sup>13,14</sup>.

This review presents an overview of the main techniques currently used to obtain nutritionally enriched vegetables, with emphasis on the conventional biofortification of OFSP with VA, and discusses the influence of food processing techniques on the nutritional characteristics of foods.

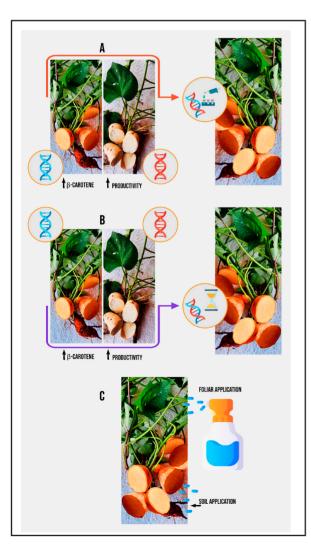
#### Nutritional deficiency and biofortification of species

Food and nutritional security consist of guaranteeing the right of everyone to regular and permanent access to sufficient and quality food without compromising access to other essential needs, based on health-promoting food practices that respect cultural diversity and that are environmentally, culturally, economically and socially sustainable<sup>15</sup>. It means that food security refers not only to the supply of food but to the availability of nutrients in the minimum quantity necessary for maintaining health.

WHO reports indicate that there are 122 countries that suffer from VA deficiency (VAD) in a subclinical state globally. Preschool children are the most affected; 190 million, that is, 33.3% of children who are of preschool age, are part of the population considered to be at high risk on a global scal<sup>16,17,18</sup>. Most preschool children with VAD live in South East Asia with 91.5 million people affected. In India, 62% of children under the age of 5 suffer from VAD. South African countries are also high affected<sup>19</sup>. VA deficiency ranges from 7.8% in China, 30-50% in Congo, 33.5-34.8% in Ethiopia, 57.5% in Bangladesh and 63% in Thailand<sup>20,21,22</sup>. In Latin America and the Caribbean Region, VAD is mostly subclinical (data do not consider Chile, Haiti, Paraguay, Uruguay, Venezuela, and the English-speaking Caribbean). The prevalence of subclinical VAD (serum retinol <20 g/dl) in children under 5 years of age ranges between 6% in Panama and 36% in El Salvador. The problem is severe in five countries, moderate in six, and mild in four. The population affected amounts to about 14.5 million children under 5 years of age (25% of that age group). Schoolchildren and adult women may also have significant VAD<sup>23</sup>. In Brazil, it is estimated that 17.4% of children under 5 years of age are deficient in VA. The highest estimates of VAD are found in the Southeast and Northeast regions of Brazil, with 22.1 and 19.3%, respectively. The Midwest, North and South regions have the lowest levels of VAD, 12.1; 9.8; 10.1%, respectively, which can be considered a moderate public health problem<sup>24</sup>.

Nutritional increment through crop biofortification can be carried out using conventional breeding, through repeated crossings, between genotypes of the same species; by transgenic means, also known as transgenic biofortification; or through plant management in the field, especially by plant fertilization (Figure 1)<sup>3,25,26</sup>.

Conventional biofortification consists of repeated crosses of plants of the same species until more nutritious cultivars are obtained. Therefore, biofortified foods have higher levels of certain micronutrients when compared to conventional food. This process is also known as conventional breeding or conventional genetic breeding. Among the advantages of conventional biofortification is the fact that it uses the intrinsic



characteristics, without the introduction of exogenous genes. However, it takes longer in the selection and obtaining of a biofortified lineage, and it also demands intellectual property registration<sup>3,27</sup>.

Biofortification is an approach widely used for the HarvestPlus program, a global action focused on nutritional safety and concentrating on the development of biofortified cultivars aimed at reducing hidden hunger in the world. The program encourages the development and production of biofortified species with the aim of reducing malnutrition regarding minerals in regions such as South Asia, Sub-Saharan Africa and Latin America<sup>8</sup>. Currently, improved cultivars are being tested and grown in over 40 countries<sup>28</sup>. According to Bouis et al.<sup>2</sup>, biofortified crops are currently cultivated and consumed worldwide by more than 20 million people and using a structured program, which is both public and private, they are expected to reach 1 billion people by 2030.

In Brazil, food biofortification projects began in 2009, under the coordination of the Brazilian Agricultural Research Corporation (EMBRAPA). They currently have the participation of Universities, Federal Institutes, and other research entities, in addition to associations of producers and non-governmental organizations. Embrapa has a prominent position in the launching of new biofortified cultivars in South America, actively working on the biofortification of different crops, such as rice, sweet potatoes, beans, cowpea, cassava, corn, wheat and pumpkin<sup>28,29</sup>.

Food fortification is also an approach used in different countries which, according to the nutritional deficiencies of each population, adds micronutrients to food ingredients and products. Although Brazil practices the fortification of wheat and corn flours with Fe and folic acid<sup>11</sup>, VA fortification of foods does not occur. Studies that seek to fortify foods with VA, with the maintenance of its stability, deserve attention from governments and the scientific community.

OFSP can be obtained by plant breeding conducted by public or private companies or naturally selected by growers over the years. In Brazil, the research with biofortified sweet potatoes started in 2009<sup>29</sup>, using imported clones that were obtained through conventional breeding. Since then, Embrapa has been actively working to launch varieties obtained by conventional breeding that, in addition to the high levels of β-carotene, maintain the characteristics and yield of the species. In 2011, Embrapa Clima Temperado (Pelotas, RS- Brazil) launched the cultivar BRS Amélia, an OFSP selected from local accessions that have undergone viral cleaning and agronomic characterization, presenting high productivity and significant levels of carotenoids. However, the carotenoid content observed in the BRS Amelia cultivar is lower than that observed in the Beauregard cultivar," a cultivar obtained through conventional biofortification, developed by the Louisiana Agricultural Experiment Station (USA). Vizzotto et al.<sup>30</sup> reported total carotenoid contents of 10.26 mg 100g<sup>-1</sup> in roots of cv. Amelia and 21.79 mg 100 g<sup>-1</sup> in roots of cv. Beauregard. OFSP cv. Beauregard was tested and

**Figure 1:** Crop biofortification by transgenic (A), conventional breeding (B), and plant fertilization (C).

recommended by Embrapa Hortaliças (Brasília, DF- Brazil) in Brazil, presenting  $\beta$ -carotene content higher than 10 times that found in white flesh cultivars<sup>31</sup>.

Currently, studies on biofortification of other species are also being conducted. In the fight against hypovitaminosis A, the first version of Golden Rice (GR1) was obtained from the incorporation of the psy (phytoene synthase) and crt (phytoene desaturase) genes of an ornamental flower (Narcissus pseudonarcissus) in the common rice genotype, which allowed the recovery of the  $\beta$ -carotene biosynthesis route in the endosperm, leading to the accumulation of more than 1.6  $\mu$ g g<sup>-1</sup> of carotenoids by dry weight (d/w) of the grain<sup>32,3</sup>. Further studies achieved total carotenoid accumulations of 37  $\mu$ g g<sup>-1</sup> in rice grains (d/w), of which 31  $\mu$ g g<sup>1</sup> was  $\beta$ -carotene (d/w), due to the use of genes from other species such as maize<sup>31</sup>. Diretto et al.<sup>33</sup> developed a variety of potato (Solanum tuberosum) containing phytoene synthase, phytoene desnaturase and lycopene β-cyclase from a bacterium (*Erwinia herbicola*) and obtained 114 mg  $g^{-1}$  (d/w) total carotenoids content and 47 mg g<sup>-1</sup> (d/w) of  $\beta$ -carotene. Through conventional biofortification, Chávez et al.<sup>27</sup> selected 2.457 cassava root genotypes from more than 20 countries, obtaining  $\beta$ -carotene levels ranging from 0.102 to 1.040 mg  $100g^{-1}$  fresh weight (f/w). Further, some genotypes presented protein contents ranging from 5.75 to 8.31% and mean Fe and Zn contents of 17.1 and 7.5 mg kg<sup>-1</sup> (f/w), respectively, suggesting the potential of this culture for biofortified genotype selection research.

Fertilization is also an important approach because it is unrelated to the controversial issue of transgenics. The application of fertilizers enriched with micronutrients and macronutrients is considered a technology with minimal negative environmental impact. Most micronutrients and macronutrients are not susceptible to leaching because they are heavily trapped in the soil. The disadvantage is that these elements accumulate over time and cause toxicity if large quantities are applied repeatedly<sup>26</sup>. Flores et al.<sup>34</sup> reported in their study that fertilization with Ca<sup>2+</sup> and NO<sup>3-</sup> results in an increase in the levels of lycopene and  $\beta$ -carotene in pepper. However, this technique does not work for some minerals (e.g. Fe), besides being dependent on the cultivar, and it is not possible to target only the edible parts of the plant<sup>3</sup>.

Other micronutrients are also being investigated due to nutrient deficiencies in populations, such as vitamins  $B_{g'}B_6$  and E, which could be incorporated in the improvement of already biofortified species, combining efforts in the development of species that are "nutritionally-complete" and more resistant to climatic adversities<sup>9,35,36,37</sup>.

Futhermore, the nutritional increment of species can occur through natural or induced situations with exposure to biotic and abiotic stresses, such as water deficiency, flooding, exposure to UV radiation and attack of pathogens, resulting in an increase in compounds from the special metabolism of plants<sup>9,25,38,39</sup>.

#### OFSP as a tool to combat VA deficiencies

The sweet potato (SP) (*Ipomoea batatas Lam.*) is a root vegetable of great importance because of its robustness, low

implementation cost and versatility for consumption<sup>40</sup>. SP originated in Latin America and nowdays is cultivated in more than 100 countries; it is now a staple food for many indigenous populations in Central and South America, Africa, Hawaii, South and West Asia, among other regions<sup>41,42</sup>.

In Brazil, SP is the sixth place most planted vegetable in the country, with a 2019 production of 805.412 tons, in an estimated area of 57.290 hectares. However, when compared to other Asian countries, such as China, which in 2019 produced 52 million tons, the production and consumption of this crop are low, with great potential for expansion<sup>43</sup>.

Uniform and elongated roots, good pest resistance, broad climate adaptation, and availability of natural genetic sources of genotypes rich in certain micronutrients are some of the motivations for the launching of new SP cultivars. However, besides good agronomic performance, essential for the producers, new genotypes should also be accepted sensorially by consumers<sup>44</sup>.

Brazil has a high genetic diversity of SP, and is able to find roots of various shapes and sizes and different colors<sup>45</sup>. Orange, yellow, purple and white SP varieties can differ not only in their skin or flesh color but also in their nutritional composition and profile of bioactive compounds<sup>46,47</sup>. The variation between different cultivars may be influenced not only by genotypes but also by other factors, such as climatic conditions and cultivation site<sup>46,48</sup>.

SP presents complex carbohydrates and a glycemic index (70) lower than other sources of carbohydrates such as potatoes (80), sweet corn (80) and rice (81)<sup>49</sup>; it is also a good source of antioxidants, Zn, potassium, sodium, magnesium, iron, phenols,  $\beta$ -carotene and vitamin C<sup>8,30,46,47,48</sup>. In addition to vitamins and minerals, SP may have significant levels of bioactive compounds depending on the cultivar. Anthocyanins are significantly present in purple sweet potatoes, and carotenoids in orange sweet potatoes<sup>30</sup>.

The orange coloration, characteristic of some OFSP genotypes, occurs due to the presence of carotenoids. Some carotenoids are precursors of VA, especially  $\beta$ -carotene. These compounds are considered potentially bioactive due to antioxidant activity<sup>30,42,48</sup>, which in turn is related to the inhibition of free radical injury and can reduce the occurrence of various degenerative diseases<sup>47</sup>.

VA, a term used for a group of compounds (retinol, retinoic acid, and retinyl esters), is a fat-soluble micronutrient. It is essential in small amounts for normal functioning, such as growth, and the development of the visual system, maintenance of epithelial cell integrity, cell differentiation, lysosomal membrane stabilization, immune function, and reproduction, including genetic regulation in humans<sup>8,50,51</sup>.  $\beta$ -carotene has also been associated with beneficial effects on human health, such as improving immunity and reducing the risk of degenerative diseases such as cancer and cardiovascular diseases<sup>52</sup>.

In the human body  $\beta$ -carotene is converted by the enzyme  $\beta$ -carotene 15,15<sup>-</sup>-dioxygenase (EC: 1.13.11.63),

through oxidative cleavage, incorporation of 2 oxygen atoms, generating all-trans-retinal, catalyzing the first step in the conversion of carotenoids to VA<sup>53</sup>. Deficiency of this enzyme can cause hypercarotenemia and VA deficiency in the human body.

The specific signs of VAD include: xerophthalmia and the risk of irreversible blindness; increased morbidity and mortality; increased risk of anemia; and retarded growth and human development. However, these nonspecific adverse effects may also be caused by other nutritional deficits, making it difficult to attribute symptoms specifically to VAD without biochemical proof<sup>50</sup>. Health-promoting effects related to the consumption of OFSP have been reported, namely: antioxidant, anti-inflammatory, immunomodulatory, anticancer/antitumor, antimicrobial, antiulcer, antidiabetic, antiobesity and hepatoprotective<sup>47</sup>. In addition, Jones et al.<sup>54</sup> report that consumption of OFSP may reduce the incidence and duration of cases of diarrhea in children, besides reducing VAD.

However, studies that demonstrate UL (Upper Intake Level), NOAEL (no observed adverse effect level) and LOAFL (lowest observed adverse effect level) should be taken into account<sup>55</sup>. A UL of about 3,000 µg/day is tolerable for VA. While, a NOAEL of 4,500 µg/day, represents a conservative value in light of the evidence about no adverse effects at or below that level. The LOAEL, on the other hand, was established considering the hepatotoxicity reported at VA supplement doses of 14,000  $\mu$ g/day<sup>56</sup>. It is likely that increased blood levels of retinoic acid compounds are responsible for the teratogenic effects of VA. High dose VA supplements can cause teratogenic effects especially if consumed shortly before conception or during the first trimester of pregnancy<sup>55</sup>. A study carried out with asbestos-exposed smokers, showed that supplementation with VA can trigger undesirable and non-protective if ingested in excessive doses<sup>57</sup>.

VA can be obtained in a balanced diet containing foods of animal origin and plant origin. In animal foods, VA is linked to a fatty acid. Animal liver and fish oils are the best sources of this vitamin. However, these foods have restricted availability for a large part of the population, due either to cost or consumption habits, and foods of plant origin are thus the main source. A total of 82% of VA consumed in poorer countries is obtained from  $\beta$ -carotene,  $\alpha$ -carotene and  $\beta$ -cryptoxanthin<sup>8</sup>; unsaturated tetraterpenoids containing at least one  $\beta$ -ionone ring residue responsible for the activity of pro-VA from foods of plant origin. Carotenoids in the body are less effective. Isotope dilution studies, evaluating β-carotene conversion in healthy and well-nourished individuals, show variable conversion rates<sup>8</sup>. The amount of VA required depends on age, sex, genetic factors and lifestyle. The conversion of carotenoids to VA is estimated at 6  $\mu$ g  $\beta$ -carotene to 1  $\mu$ g of VA, while for other carotenoids with pro-VA activity the estimated conversion is 12  $\mu$ g to 1  $\mu$ g of VA<sup>50,58</sup>.

In Brazil, according to RDC MS No. 269/2005, the recommended dietary allowance (RDA) for children aged 0-10 years is 375-500  $\mu$ gRE (Retinol Equivalent); for adults it is 600  $\mu$ gRE, and for pregnant women 800  $\mu$ gRE<sup>58</sup>. It is estimated that the consumption of 25 to 50 g of OFSP cv. Beauregard would supplement the recommended dietary allowance (RDA) of pro-VA for adults<sup>59</sup>.

# Food technology in the supply of biofortified foods obtained with OFSP

Post-harvest retention of micronutrients in biofortified foods is a crucial step in the biofortification program, as it forms the link between the levels required for population health and bioavailability<sup>60</sup>.

Although the processing and obtaining of food products from biofortified SP may increase consumption possibilities and generate income through agroindustrialization in small rural farms<sup>12</sup>, the nutritional quality of the products should not be neglected, avoiding the addition of large amounts of sugar, salt or fats, as well as the use of processing techniques that promote significant losses of nutrients.

Vitamins are sensitive compounds that can be degraded by different factors, such as temperature, oxygen, light, humidity, pH, duration of treatment, etc.<sup>14,11</sup>. Although there are studies that correlate industrial processing and nutritional quality, knowledge of this subject is dispersed and insufficient due to the complexity of each micronutrient and its interaction with the food matrix. In this way, processing technologies must be constantly adapted in order to minimize these losses.

Processing may affect products differently. While carotenoids are easily degraded by environmental conditions, such as atmospheric oxygen, processing and storage, losses are also due to physical damage such as peeling and cutting, geometric isomerization, and enzymatic and non-enzymatic oxidation<sup>60,61</sup>. On the other hand, it is known that food processing can also increase nutrients bioaccessibility. Emerging technologies could improve the bioaccessibility of bioactive compounds. It can be stated that, in general, a combination of high-pressure processing and thermal treatment in the presence of oil could be beneficial for carotenoid bioaccessibility<sup>62</sup>.

Biofortified SP can be consumed and processed in different ways: cooked, roasted, dried, steamed, and fried; also, added in the formulation of sweets, bread, cakes, ice creams, etc. However, regardless of the form of presentation for consumption, the nutritional quality of the products should be considered to provide food products that aim to combat malnutrition and micronutrient deficiency<sup>12</sup>. Table 1 presents some studies that evaluated food processing techniques, packaging, shelf-life, and acceptability of foods obtained from biofortified SP.

OFSP roots should be considered a healthy food option to be used in different domestic meal preparations as well as for use by the food industry as a functional ingredient for formulation of added-value food products<sup>47</sup>.

Food product	Processing/Packaging/Sensory	Main reported results	Authors
OFSP cv. Ejumula, cv. SPK004, cv. SPK004/6/6a, cv. SPK004/6a, cv. SPK004/1/1a, cv. SPK004/1a, cv. Sowola 6/94/9b	Boiling, oven drying, sun drying and deep frying	β-carotene retention: Boiling: 77.6%; Oven drying: 88.2%; sun drying: 83.8 to 91.1%; deep frying 78.3%.	Bengtsson et al. <sup>63</sup>
OFSP cv. Guineng 05-6	Boiling, roasting and steaming	Losses in carotenoid content: boiling < steaming < roasting	Tang et al. <sup>64</sup>
OFSP cv. Ex-Igbariam	Boiling, roasting and oven drying	β-carotene retention: Boiling: 38%; Oven drying: 92%; Roasting: 87%.	Ukom and Ojimelukwe <sup>65</sup>
OFSP cv. Pushu 32	Boiling, steaming, microwaving, roasting and frying	Losses in carotenoid content: boiling < steaming < microwaving < roasting < frying	Kourouma et al. <sup>66</sup>
Bread with added OFSP cv. Beauregard flour	Bread	β-carotene retention: bread with 10, 20 and 30% of sweet potato flour resulting in 62.7, 71.4 and 83%, respectively.	Nzamwita et al. <sup>67</sup>
OFSP chips cv. Beauregard	Shelf-life / Packaging	β-carotene retention: Minor losses of carotenoids due to nitrogen and reduction of oxygen content in five package system tested. BOPP (biaxially oriented polypropylene)/metBOPP with nitrogen showing the greatest cost benefit, retained 83% of carotenoids. Shelf-life: up to 207 days without sensorially perceptible changes.	Júnior et al. <sup>68</sup>
OFSP flour cv. Beauregard	Packaging	Minor losses of carotenoids over the period of 3-4 months due to reduction of oxygen content and use of PET (polyester) packaging with metal barrier and aluminum.	Alves et al. <sup>69</sup>
Brownies with added OFSP cv. Gendut	Brownies / Sensory	Increase of fiber content (73%) in brownies with 75% of OFSP puree added. Increase in sensory attributes.	Selvakumaran et al. <sup>70</sup>
Cookies with added OFSP cv. Beauregard	Cookies / Sensory	Increase in sensory and nutritional attributes.	Infante et al. <sup>71</sup>

Table 1. Studies evaluating processing, packaging, and acceptability of foods obtained from biofortified OFSP.

Sourdough panettone with added OFSP flour	Sourdough panettone	Positive effects including a higher moisture and strong yellow color of the crumb, and presence of new volatile compounds.	Pereira et al. <sup>73</sup>
OFSP puree	Microwave-assisted-thermal / Packaging / Sensory	Increase in β-carotene because of the enhanced extractability by a thermal process using microwave- assisted thermal treatment. Sensory evaluation indicated as a desirable baby food up to 18 months. Packaging influenced the flavor and overall acceptance.	Zhang et al. <sup>74</sup>
Bread, cake, coconut sweet and, cocoa sweet with added roasted OFSP cv. Beauregard	Roasting and steaming / Sensory	β-carotene retention: 28.7; 52.2; 50.6; 24.2%, for bread, cake, coconut sweet and cocoa sweet added with OFSP cv. Beauregard, respectively. Acceptability higher than 83.3%.	Santos et al. <sup>75</sup>
Beer with added OFSP cv. Beauregard dried	Beer / Sensory	Increase in β-carotene due to added OFSP in different conditions. 50% of Beauregard sweet potato concentration and 75 min of mashing time showed satisfactory values for physicochemical parameters, increased levels of phenols and acceptance of sensory attributes.	Humia et al. <sup>76</sup>

Bengtsson et al.<sup>63</sup>, Tang et al.<sup>64</sup>, Ukom et al.<sup>65</sup> and Kourouma et al.<sup>66</sup> obtained relevant results evaluating the retention of all-trans- $\beta$ -carotene in biofortified OFSP processed by traditional heat methods.

Bengtsson et al.<sup>63</sup> evaluated seven cultivars of OFSP by boiling, oven drying, sun drying and frying, and observed better carotenoid retention (91.1%) using a solar-drying tunnel (temperatures ranging 45-63 °C). Drying using forced ventilation in an oven dryer (57 °C/10h) resulted in the retention of 88.2% of β-carotene. OFSP dried under direct sunlight for 6h to 10h resulted in 83.8% retention. Frying process by immersed sunflower oil (160-170 °C/10 min), showed 78.3% retention, while boiling for 20 min resulted in 77.6% retention. The isomer 13-cis-β-carotene was found in content between 3 and 10.3% over all-trans-β-carotene in the processed products reported by Bengtsson et al.<sup>63</sup>, due to isomerization of the trans-form to the cis-form. Agreeing with Kourouma et al.66, who also reported an increase in cis-isomers due to a conversion of all-trans-β-carotene in boiled, steamed, microwave cooking, roasted and fried OFSP.

Solar drying applied for a long time may result in destructive effects on micronutrient retention due to uncontrolled environmental conditions. However, the energy efficiency of sun-drying can be harnessed if exposure time, sun exposure times, heat intensity, solar irradiance, wind speed, and low relative humidity are controlled<sup>60</sup>.

Ukom et al.<sup>65</sup> reported significant levels of  $\beta$ -carotene retention in OFSP cv. Ex-Igbariam submitted to the oven-drying process (60°C/24h), resulted in 92% retention. Roasting using an electric oven (120 °C/10 min) resulted in 87% retention. However, in this study, boiling in water (98 °C/5 min) resulted in only 38% retention of all-trans- $\beta$ -carotene. Increase in the total carotenoid content was observed in treatments using oven drying (29.25 µg g<sup>-1</sup> f/w) and electric oven (23.53 µg g<sup>-1</sup> f/w), in relation to unprocessed OFSP (13.08 µg g<sup>-1</sup> f/w). This behavior may be associated with evaporation of water due to the roasting process and consequent concentration of the compounds; it was also observed

by Vizzotto et al.<sup>30</sup> in roasted OFSP cv. Beauregard (250 °C/90 min). However, Tang et al.<sup>64</sup> reported that carotenoids were better preserved in boiled water (30 min), when compared to steaming (30 min) and roasting (230 °C/30 min). This result may be associated with high temperature, and the long period of roasted applied (230 °C/30 min), when compared to that reported by Ukom et al.<sup>65</sup> (120 °C/10 min.).

Kourouma et al.<sup>66</sup> evaluated different treatment times of boiling, steaming, microwaving, roasting and frying, and they observed that boiling for 15-35 min, steaming for 15-45 min, and microwaving for 15 min had a relatively high amount of  $\beta$ -carotene content in the range from 97.57 to 135.83 µg/g (d/w), while frying immersed in corn oil at 1-2.5 min exhibited a higher reduction of  $\beta$ -carotene.

Nzamwita et al.<sup>67</sup> evaluated the percentage of alltrans- $\beta$ -carotene retention in breads with 10, 20 and 30% (m/m) of OFSP flour cv. Beauregard, observed levels of 62.7, 71.4 and 83% retention, respectively, after baking. Breads containing 10%, 20% and 30% OFSP flour would have 29.61 and 89.2% (100 g portion), respectively, of the VA RDA required for children between 3 and 10 years old. The formulation with 30% would have half the VA IDR for pregnant and lactating women.

Júnior et al.68 observed minor losses of carotenoids due to nitrogen introduction and reduction of oxygen content, in the shelf-life of sweet potato cv. Beauregard chips packed in five different packaging systems. The packaging system of BOPP (biaxially oriented polypropylene)/metBOPP with nitrogen showed the greatest cost benefit due to the lower cost of the packaging material: which is also the material most used on the market for chips. It retained 83% of carotenoids, and showed no significant sensory alterations during 207 days of storage. Alves et al.<sup>69</sup> observed that the reduction of the oxygen content in the free space of the package, through the application of vacuum combined with the use of oxygen-barrier packaging materials of the order of magnitude of metal barrier PET, are the important factors in the preservation of carotenoids in biofortified sweet potato flour.

Selvakumaran et al.<sup>70</sup> evaluated the substitution of wheat flour by biofortified SP purée, and obtained brownies with higher levels of moisture, fat, fiber and specific volume. Color and texture analyzed showed that the addition of biofortified SP resulted in improved sensory attributes of brownies, where higher amounts of puree (50% and 75%) resulted in higher color scores, texture, taste, and general acceptance.

Infante et al.<sup>71</sup> reported that the introduction of 50% of sweet potato flour cv. Beauregard in cookies made with sorghum flour, resulted in a nutritional (phenolic compounds and carotenoids), and sensorial improvement of the products. Iron bioavailability parameters observed in cookies were similar to the ferrous sulfate control group.

Futhermore, the carotenoids in cookies possibly formed a soluble complex with  $Fe^{+2}$  increasing iron uptake<sup>72</sup>.

Pereira et al.<sup>73</sup> used OFSP flour instead of wheat flour in sourdough panettone, and observed differentiated color and aroma when compared to panettones made using conventional formulations that use flavorings and colorants, without affecting the machinability, with a slight decrease in the time of fermentation.

Total  $\beta$ -carotene content was increased in SP puree evaluated by Zhang et al.<sup>74</sup> due the enhanced extractability by thermal process using microwave-assisted thermal treatment (25 min for preheating at 61 °C, 3.7 min for microwave heating, and 3.8 min for holding at 124 °C). Five types of packages were tested, and vacuum was applied to all of them. Packaging influenced the flavor liking and overall acceptance. Consumers considered the SP puree to be a quality baby food until the end of the 18th month.

Promising results adding OFSP in food products were also reported by Santos et al.<sup>75</sup>. Carotenoid retention of bread, cake, coconut sweet, and cocoa sweet were 28.7, 52.2, 50.6, 24.2%, respectively. Variations are related according to the form of food preparation. In addition, the products showed acceptability higher than 80% (86.3; 83.3; 84.4; 86%, respectively).

In this way, variations in carotenoid contents may be related to the preparation of food (e.g., different equipment, binomial time x temperature, added ingredients, size and handling), genetic variation, and edaphoclimatic factors, may influence the VA composition foods<sup>2,46</sup>. Therefore, biofortified foods should be consumed fresh, and with minimal processing, so that the benefits of the biofortification program are really effective<sup>6,60</sup>.

The results of Humia et al.<sup>76</sup> demonstrated that Beauregard SP is a promising adjunct at brewing process with nutraceutical properties due to its rich composition of bioactive compounds, further, had significant acceptance in sensory analysis.

The efficacy of agronomic biofortification depends largely on the bioavailability of micronutrients<sup>26</sup>. Bioaccessibility and bioavailability depend on the biochemical nature of the nutrient, acting on absorption promotion or delay, as well as determining how efficiently different nutrients will be transported through the blood, stored and used, affecting how they will be processed and used in the human body <sup>29,11,62</sup>.

Food processing is one of the main determinants for bioavailability, because it can have a positive or negative impact by increasing or decreasing bioaccessibility of nutrients and bioactive compounds, respectively<sup>62</sup>. The interaction between constituents present in foods is also a relevant issue and dependent on the nutrient of interest. While folic acid exhibits lower thermal stability; cooking and digestive enzymes weaken the cell walls, disrupting the protein-carotenoid complexes, promoting the release and increased bioavailability of carotenoids<sup>11,14,62</sup>. In contrast, depending on the conditions, heat treatement can reduce carotenoids bioaccessibility, inducing structural changes in carotenoids, mainly isomerization, altering their solubility and consequently their micellarization<sup>62</sup>.

Starch granules may be involved in limiting the bioaccessibility of carotenoids in SP, and carotenoid deposition formed in plant chromoplasts and food matrices also appears to have a large impact on bioaccessibility<sup>77</sup>. It is estimated that about 90% of the pre-formed VA ingested is absorbed by the human body. The efficiency of absorption of pro-vitamin A carotenoids depends on the organ of the plant and the fat content in the food system. It is inferred that an increase in fat intake improves the absorption of VA<sup>8,50,77</sup>. Bengtsson et al.<sup>78</sup> reported an increase of 10-20% in the percentage of accessible all-trans- $\beta$ -carotene in the micellar phase of heat-processed SP with 2.5% (w/w) oil added.

Biofortification alone does not eliminate micronutrient malnutrition, but it can provide an alternative, which combined with integrated approaches, including plant breeding and improvement of agricultural practices, can result in poverty reduction and improved well-being for the poorest and most vulnerable people, at a relatively low cost and showing potential profit<sup>3,29</sup>. It appears that increasing the amount of biofortified SP available to populations at risk for VAD may result in good and sustainable foodbased interventions for preventing this nutritional disease<sup>8</sup>.

#### CONCLUSIONS

The importance of biofortification as a tool to promote the increase in micronutrient content essential for human health and, consequently, in combatting malnutrition, especially in the poorest populations, is evident. Several strategies for biofortification may be considered, such as transgenic, conventional and fertilization, which need to be validated, considering crop viability and the increase of the target nutrient.

Biofortified OFSP is an interesting alternative in the fight against VAD because of its robustness and easy cultivation, demonstrating that agriculture-based interventions could be an economical way to reduce micronutrient deficiencies in humans. Studies have already shown the potential of using OFSP in the nutritional and sensory enhancement of foods, and future studies may provide further knowledge helping to minimize losses during food processing, with emphasis on  $\beta$ -carotene. Therefore, the challenge for the food industry at this moment is producing sensorally acceptable products that preserve the nutrients of interest, considering not only the stability of these compounds during processing but also bioavailability and bioaccessibility, so that the effects on human health are effectively achieved.

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