

# Antioxidant capacity in ice cream and dairy products

## Capacidad antioxidante en helados y derivados lácteos

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**Abstract** Nowadays, there is an increasing global trend in the consumption of foods or products with functional potential to benefit human health. Most of the bioactive compounds or natural ingredients used in functional food products are derived from medicinal plants, fruits, and vegetables. One of the main functions reported is the reduction of the risk of degenerative diseases, which manages to direct research, mainly to the characterization of specific antioxidant compounds. Therefore, this review focuses on exposing the antioxidant power of bioactive compounds and functional ingredients that are used in some dairy products and ice creams, evidencing their benefits on human health through their use in food products.

**Key words:** antioxidant power; ice cream; dairy products; functional ingredients.

**Resumen** En la actualidad, se ha incrementado la tendencia global del consumo de alimentos o productos con potencial funcional de beneficiar a la salud humana. Gran parte de los compuestos bioactivos o ingredientes naturales empleados en los productos alimenticios funcionales derivan de plantas medicinales, frutas y vegetales. Una de las principales funciones reportadas es la disminución del riesgo de enfermedades degenerativas, que logra orientar las investigaciones, principalmente a la caracterización de componentes antioxidantes específicos. Por lo tanto, esta revisión se centra en exponer el poder antioxidante de compuestos bioactivos e ingredientes funcionales que se utilizan en algunos derivados lácteos y helados, evidenciando sus beneficios sobre la salud humana a través de su aprovechamiento en productos alimenticios.

**Palabras clave:** poder antioxidante; helados; lácteos; ingredientes funcionales..

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## Introduction

Increasing knowledge and research on the relationship between food and health, along with the technological need for innovations, has generated new products, some of which have the functional potential to benefit human health (Granato et al., 2018; Gremski et al., 2019). Functional foods (FAs) or health foods provide health benefits. Beyond their nutritional effect, they favor one or more physiological functions in the human body by improving overall fitness and/or reducing the risk of disease (Ramirez-Navas, 2019). The term 'functional food' originated in Japan (FOSHU, foods for specific health uses) (Illanes, 2015).

Vegetable and fruit-based food products and their by-products, cereals, pseudo cereals, legumes, flours, herbal teas, and teas, herbal or spice extracts, cold-pressed oils, among others, are some of the most common foods containing high amounts of antioxidant compounds such as polyphenols, carotenoids, tocopherols, tocotrienols, glutathione, ascorbic acid and enzymes with antioxidant activity (Soukoulis; Fisk; Bohn, 2014). Among the natural ingredients used to impart functional properties, plant extracts are increasingly used, mainly to replace artificial chemicals in foods, and to increase the content of antioxidant compounds, especially in high-fat foods. Traditional medicinal plants can be sources of various bioactive compounds, especially phenolic compounds, and can be used in the development of novel foods.

It is well known that the adoption of a diet rich in natural antioxidants appears to be inversely associated with chronic degenerative diseases such as cardiovascular disease, cancer, obesity, and diabetes, as well as related to the prevention of general inflammatory health implications and improvement of blood lipids such as cholesterol (Soukoulis et al., 2014). A diet rich in phenolic compounds is related to a lower

risk of multiple non-communicable diseases, such as type 2 diabetes and atherosclerosis (de Lima et al., 2018; Granato et al., 2016; Santhakumar; Battino; Alvarez-Suarez, 2018). Some plant vitro digestion, evidencing their potential to be incorporated into foods (Rashidinejad; Birch; Sun-Waterhouse; Everett, 2015). In humans, antioxidants exert cellular protective functions and preserve deoxyribonucleic acid (DNA) against injury and cleavage, reducing the risk of some types of cancer (Amarowicz; Shahidi, 2017).

The development of new food products is becoming increasingly challenging as companies aim to meet consumer demand for a food that is both healthy and appealing. Combining milk and dairy products with biologically active molecules, such as those found in various plant extracts, may be an interesting strategy to produce foods with functional potential.

The aim of this scientific review was to identify the relevant aspects in the determination of antioxidant capacity in functional ice cream. In addition, to establish what type of antioxidant compounds have been used in the elaboration of ice cream and to determine whether ice cream can be considered a functional food.

## Methodology

In this paper a narrative review of the scientific literature was developed. A literature search was conducted on the relevant aspects in the determination of antioxidant capacity in dairy products. Figure 1 shows the search algorithm. The keywords used were dairy products, ice cream, natural ingredients, and antioxidant capacity. The selection criteria established were: 1) dairy and ice cream, and 2) antioxidant capacity. Google Scholar, ScienceDirect, Wiley, Scopus and ACS were used as search engines, where articles, books and book chapters were

found, in a time range of 10 years. Subsequently, the studies found were selected, compiled, and classified to perform a comparative analysis

according to the information reported by the different authors.

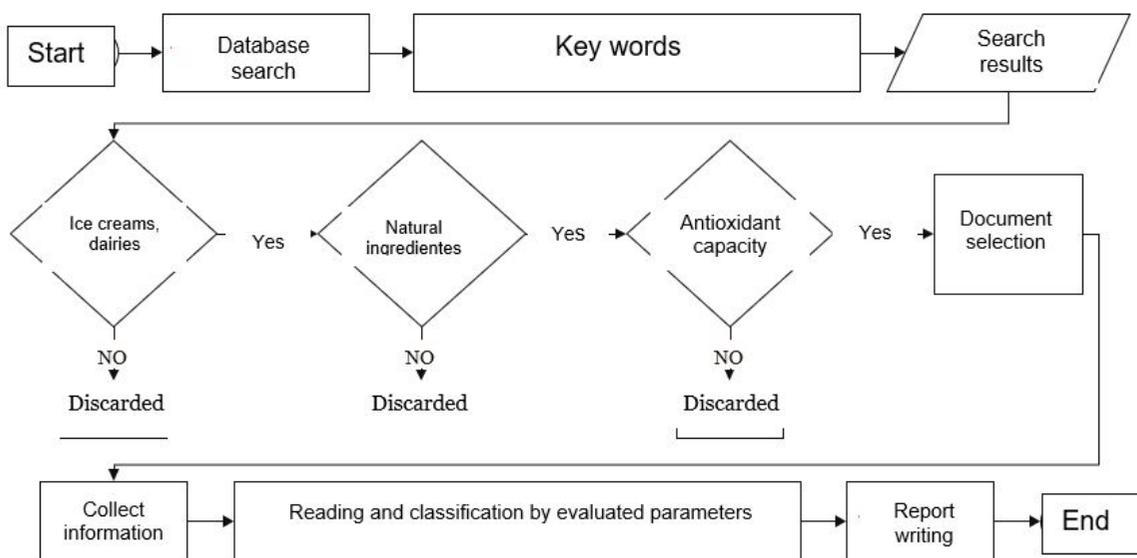


Figure 1

Algorithm applied for bibliographic search.  
Source: Own elaboration.

## Free radicals

A free radical (FR) can be defined as "an atom or group of atoms with one or more unpaired electrons that exist in free form" (Zeb, 2020). RLs are generated by various processes including homolytic, heterolytic cleavage or redox reactions. The main oxidants are singlet oxygen, ozone, hydrogen peroxide, nitrous acid, hypochlorite, nitric or nitrous oxides, etc. (Lushchak, 2014). These oxidizing species are called reactive oxygen species (Prasad; Gupta; Tyagi, 2017). The RL formed can be lipid, superoxide anion, hydroxyl, hydroperoxy, peroxy, nitric oxide and nitrosyl cation as shown in Figure 2 (Zeb, 2015a, 2020).

FR chain reactions start with the formation of the radical, called the initiation reaction. Then, the RL propagates by attacking

other molecules (Ahmadinejad; Geir-Møller; Hashemzadeh-Chaleshtori; Bidkhori; Jami, 2017). The latter reactions are called termination, with the formation of neutral species or non-radical products, as shown in Figure 3. The substrate (R) for FRs can be a lipid, a protein, carbohydrates, an organic compound, inorganic compound, or a metal atom. RL formation occurs as a normal process of cellular biochemistry. However, an increase in the level of RL will cause an imbalance in the biochemical pathways, generating oxidative stress, which is responsible for food aging or spoilage. Therefore, antioxidant is necessary to overcome oxidative stress (Zeb, 2018, 2020). When reactive oxygen species (ROS) are generated in excess, or in case of limited antioxidant concentrations, there may be a shift in redox potential towards oxidative stress (Gupta; Finelli; Agarwal; Henkel, 2020).

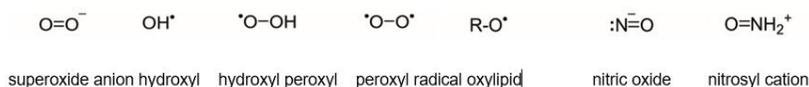


Figure 2

Chemical Structures of Some Important Free Radicals.  
Source: Zeb (2015a, 2020).

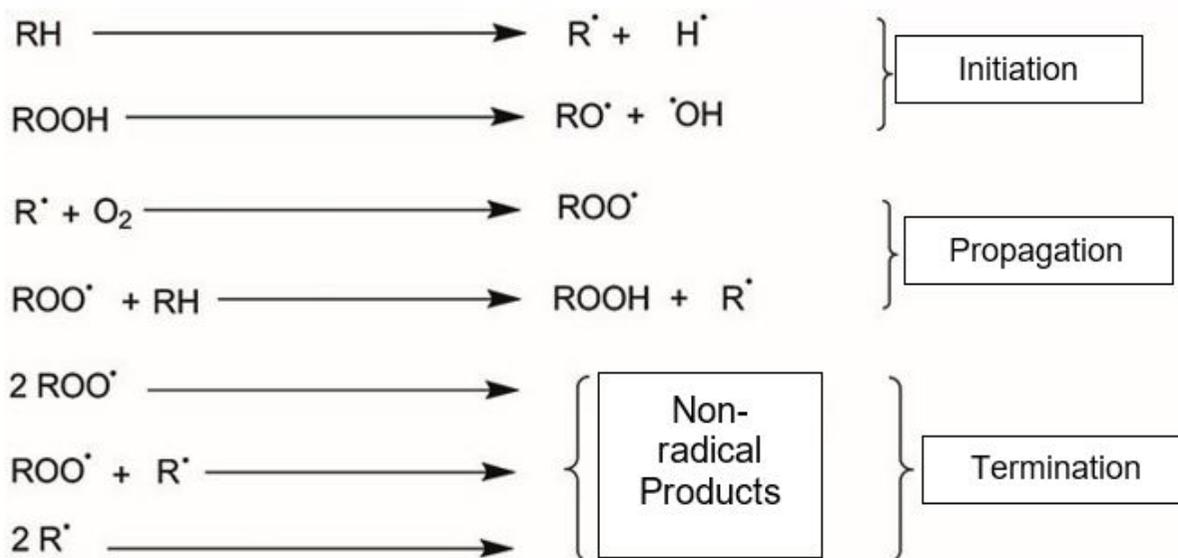


Figure 3

*Free Radical Chain Reactions.*  
Source: Zeb (2020).

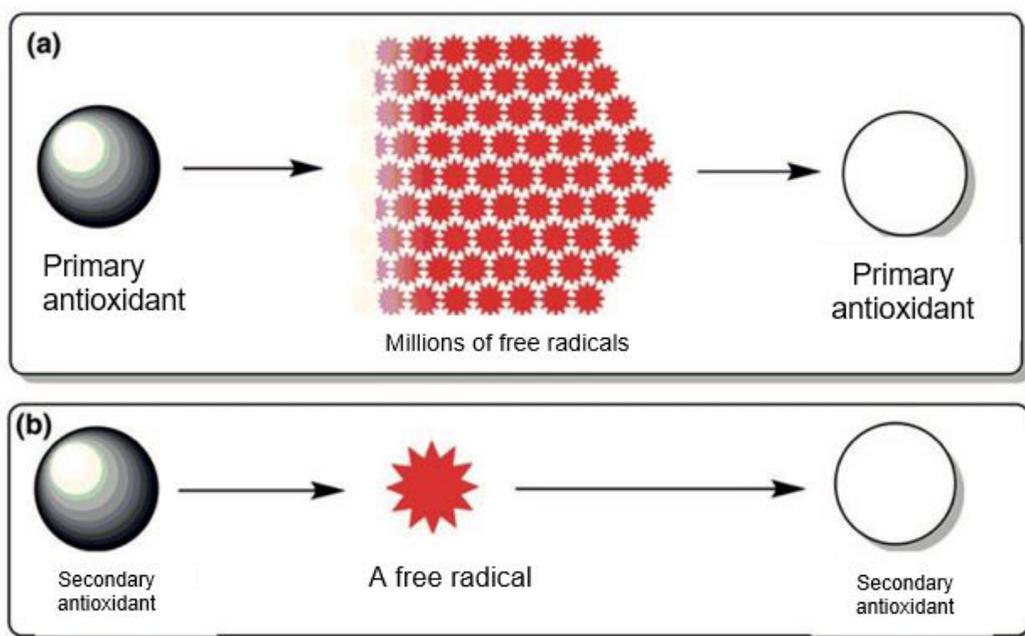
## Antioxidants

An antioxidant is a chemical substance that inhibits, stops, or controls the oxidation of a substrate. It can be an organic or inorganic compound. The substrate is known as an oxidizable substrate. However, the definition of antioxidant varies in relation to its applications. In Food Science, the term 'antioxidant' is implicitly limited to free lipid radical chain inhibitors. However, the wide distribution of FR demanded that the definition be more precise, comprehensive, and widely accepted. Thus, antioxidant was defined as "substances present in relatively low amount in food, which inhibit, delay or control oxidation of food components, prevent spoilage and prolong the shelf life of food" (Zeb, 2020). Inhibition of oxidation resulted in its retardation, a process that prolongs the shelf life of food (Byrd, 2001; Halliwell, 1995, 2009; Khan; Liu; Wang; Sun, 2020).

Antioxidants are classified as primary and secondary. This classification is based on their mechanism of action. For example, primary

antioxidants neutralize LR by hydrogen atom transfer (HAT) or by a single electron transfer mechanism (SET). These antioxidants are very efficient and are usually required in very limited amounts to neutralize a large amount of RLs (Figure 4). The high catalytic properties of these are one of the main reasons for their diversity in nature. These antioxidants are easily regenerated, phenolics are included in this category (Zeb, 2020).

On the other hand, secondaries have been characterized by the neutralization mechanism of pro-oxidant catalysts. Examples include pro-oxidant metal ion chelators (e.g, Fe and Cu), such as ethylene-di-amino-tetra-acetic acid (EDTA) and citric acid (CA). The  $\beta$ -carotene can neutralize reactive species such as singlet oxygen. These antioxidants usually quench an RL and are therefore easily depleted (Figure 4). More recently, a third class called tertiary has been included. These antioxidants repair damaged biomolecules such as DNA or proteins. However, very little is known about their role in food (Zeb, 2020).



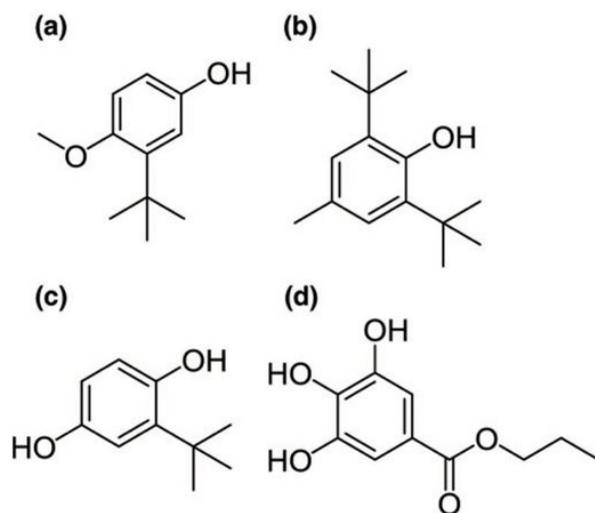
**Figure 4**

*Representative reactions of primary and secondary antioxidants in foods.*

Note. (a) Reaction of the primary antioxidant with many free radicals, and (b) reaction of the secondary antioxidant with a free radical  
Source: Zeb (2020).

Both primary and secondary antioxidants can be synthetic or from natural sources. Synthetic primary antioxidants include butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate (PG), and tert-butylhydroquinone (TBHQ) (Figure 5). These have been used to control lipid oxidation and off-flavor development by quenching RLs in foods. In addition, they contain one or more

hydroxyl or phenol groups, and are very effective (Shahidi, 2015). However, attention has been focused on natural antioxidants mainly from plants and foods due to the harmfulness and carcinogenic properties of synthetic antioxidants evaluated in animal models (Augustyniak et al., 2010; Chen; Pearson; Gray, 1992; Kumar; Singh; Sharma; Kishore, 2019; Zeb, 2020).



**Figure 5**

*Chemical Structures of Synthetic Antioxidants Used in Foods.*

Note. (a) butylated hydroxyanisole (BHA), (b) butylated hydroxytoluene (BHT), (c) tert-butylhydroquinone (TBHQ) and (d) propyl gallate (PG).  
Source: Zeb (2020).

Phenolic compounds (PC) are natural antioxidants present in plant foods. They are generally biosynthesized from phenylalanine or tyrosine, through the shikimic acid pathway. They range from simple compounds to conjugated or complex compounds. The hydroxyl group on the benzene ring is responsible for the antioxidant properties of CF. Therefore, more than two hydroxyl-containing compounds are called polyhydroxyphenolic compounds.

When there are more than several phenols in a compound, it is called a polyphenolic compound. CFs are widely studied in different plant foods such as vegetables, fruits, cereals, seeds, berries, tea, onion bulbs, wine, and vegetable oils (Dimitrios, 2006; Zeb, 2020). Table 1 presents the phenolic composition of some common foods.

**Table 1**

*Main sources of phenolic antioxidants from plant foods*

Food	Antioxidants	Ref.
<b>Fruits</b>		
Cherries ( <i>Prunus salicifolia</i> )	Hydroxycinnamic acid, anthocyanins	Manach, Scalbert, Morand, Rémésy and Jiménez (2004); Yanishlieva–Maslarova and Heinonen (2001)
Black grapes ( <i>Vitis vinifera</i> )	Anthocyanins, flavonols	Manach <i>et al.</i> (2004); Yanishlieva–Maslarova and Heinonen (2001)
Citrus Fruits ( <i>Citricus</i> )	Flavanones, flavonols, phenolic acids	Beecher (2003); Manach <i>et al.</i> (2004); Citrus fruits ( <i>Citricus</i> ) Flavanones, flavonols, phenolic acids Yanishlieva–Maslarova and Heinonen (2001)
Plums ( <i>Prunus domestica</i> ), apples ( <i>Malus pumila</i> ), pears ( <i>Pyrus communis</i> ), kiwifruit ( <i>Actinidia chinensis</i> )	Hydroxycinnamic acids, Catechins	Manach <i>et al.</i> (2004); Yanishlieva–Maslarova and Heinonen (2001)
<b>Vegetables</b>		
Eggplant ( <i>Solanum melongena</i> )	Anthocyanins, hydroxycinnamic acids	Manach <i>et al.</i> (2004)
Chicory ( <i>Cichorium intybus</i> ), artichoke ( <i>Cynara scolymus</i> )	Hydroxycinnamic acids	Manach <i>et al.</i> (2004)
Parsley ( <i>Petroselinum crispum</i> )	Flavones	Beecher (2003); Manach <i>et al.</i> (2004)
Rhubarb ( <i>Rheum rhabarbarum</i> )	Anthocyanins	Manach <i>et al.</i> (2004)
Sweet potato leaves ( <i>Ipomoea batatas</i> )	Flavonols, flavones	Chu, Chang and Hsu (2000)
Yellow onion ( <i>Allium cepa</i> ), Curly lettuce ( <i>Lactuca sativa</i> ), Beans ( <i>Phaseolus vulgaris</i> )	Flavonols	Manach <i>et al.</i> (2004)
Watercress ( <i>Nasturtium officinale</i> )	Phenolic acids, flavonoids, flavonols	Bergman, Varshavsky, Gottlieb and Grossman (2001)
Spinach ( <i>Spinacia oleracea</i> )	Flavonoids, p-coumaric acid	Bergman <i>et al.</i> (2001)
<b>Cereals</b>		
Oats ( <i>Avena sativa</i> ), Wheat ( <i>Triticum durum</i> ), Rice ( <i>Oryza sativa</i> )	Ferulic and caffeic acids	Manach <i>et al.</i> (2004); Yanishlieva–Maslarova and Heinonen (2001)
<b>Herbs and spices</b>		
Rosemary ( <i>Salvia rosmarinus</i> )	Carnosic Acid, Carnosol, Rosmarinic Acid, Rosmanol	Ibañez <i>et al.</i> (2002); Yanishlieva–Maslarova and Heinonen (2001)
Sage ( <i>Salvia officinalis</i> )	Carnosol, carnosic acid, lateolin, rosmanol, rosmarinic acid, rosmarinic acid	Yanishlieva–Maslarova and Heinonen (2001); Zheng and Wang (2001)
Oregano ( <i>Origanum vulgare</i> )	Rosmarinic acid, phenolic acids, flavonoids	Exarchou <i>et al.</i> (2002); Yanishlieva–Maslarova and Heinonen (2001)
Thyme ( <i>Thymus</i> )	Thymol, carvacrol, flavonoids, luteolin	Exarchou <i>et al.</i> (2002); Yanishlieva–Maslarova and Heinonen (2001); Zheng and Wang (2001)
Ginger ( <i>Zingiber officinale</i> )	Ginger and derivatives	Yanishlieva–Maslarova and Heinonen (2001)

Food	Antioxidants	Ref.
<b>Teas</b>		
Black ( <i>Camellia sinensis</i> )	Flava-3-ols, flavonols	Beecher (2003); Manach <i>et al.</i> (2004)
Green ( <i>Camellia sinensis</i> )	Epicatechin, epigallocatechin, epicatechin gallate, epigallocatechin gallate	Luo <i>et al.</i> (2020)
Matcha green tea ( <i>Camellia sinensis</i> )	Flavonoids, polyphenols, vitamin C	Jakubczyk <i>et al.</i> (2020)
<b>Alcoholic beverages</b>		
Red wine	Flavan-3-ols, flavonols, anthocyanins	Beecher (2003); Manach <i>et al.</i> (2004)
Cider	Hydroxycinnamic acids	Manach <i>et al.</i> (2004)
<b>Other beverages</b>		
Orange juice ( <i>Citrus sinensis</i> )	Flavanols	Manach <i>et al.</i> (2004)
Coffee ( <i>Coffea</i> )	Hydroxycinnamic acids	Manach <i>et al.</i> (2004); Sánchez-González, Jiménez-Escrig and Saura-Calixto (2005).
Chocolate ( <i>Theobroma cacao</i> )	Flavanols	Beecher (2003); Manach <i>et al.</i> (2004)

## Antioxidant capacity

In this work, it will be exemplified using FCs. The mechanism of the antioxidant activity of a phenolic compound could be through a HAT mechanism or single electron transfer through proton transfer (SET-PT) (Zhang; Tsao, 2016), sequential electron transfer with proton loss (Lee *et al.*, 2020), or transition metal chelation (TMC) (Zeb, 2020). Foods, especially vegetables, are rich in antioxidant (phenolic) compounds. In the scientific literature, reviews are found on the distribution, synthesis and analysis techniques used for the determination of antioxidant compounds, antioxidant capacity and bioactivity of phenolic acids in foods, beverages, and spices. In the different papers found, the authors summarized synthetic and phenolic antioxidants from natural sources, their mode of action, degradation, health effects and toxicological properties. Most conclude that natural phenolic antioxidants are safer than synthetic antioxidants, and their sources in foods should be explored in detail (Escarpa; Gonzalez, 2001; Heleno; Martins; Queiroz; Ferreira, 2015; Shahidi, 2015; Zeb, 2020).

The antioxidant functions of foods are usually measured in the *in vitro* system, using different analytical techniques. There are several antioxidant analysis techniques, such as azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) (ABTS), ferric reducing antioxidant

power (FRAP), cupric reducing antioxidant capacity (CUPRAC), oxygen radical absorbance capacity (ORAC), Trolox equivalent antioxidant capacity (TEAC), diphenyl-1-picrylhydrazyl (DPPH), total antioxidant activity (TAA) and photochemiluminescence (PoCL) (Zeb, 2020) (see Table 2).

## Methods for assessing TAC

Several methods have been proposed in the literature for the assessment of total antioxidant capacity (TAC) (see Table 2), these are summarized in Table 3, and each varies according to the principle and methodology, as well as their sensitivity to different antioxidants. Therefore, results may vary depending on the type of test used (Gupta *et al.*, 2020; Vassalle *et al.*, 2004). Electron transfer assays measure the reducing capacity of the substrate (antioxidant), hydrogen transfer assays measure the hydrogen donating capacity of the substrate. It is clear that hydrogen atom donation is essential in the radical chain reaction stage of lipid peroxidation, therefore, hydrogen transfer assays are relevant to the measurement of chain-breaking antioxidant capacity. In many cases, the antioxidant capacity, or radical trapping capacity, of a compound is related to the ease of hydrogen atom donation, not necessarily to the redox potential of the compound (MacDonald-Wicks; Wood; Garg, 2006).

**Table 2***Important representative in vitro antioxidant activity measurement methods used for foodstuffs*

Methods	Food	Ref.
Oxygen Radical Absorbance Capacity (ORAC)	Guava	Thaipong, Boonprakob, Crosby, Cisneros-Zevallos and Hawkins-Byrne (2006)
Total Antioxidant Radical Trapping Potential (TRAP)	Fruits	Ruiz-Torralba, Guerra-Hernández and García-Villanova (2018).
Trolox equivalent antioxidant capacity (TEAC)	Legumes, fruits	Koley et al. (2018); Thaipong et al. (2006).
2,2-diphenyl-1-picrylhydrazyl (DPPH)	Guava, leafy vegetables, fruits	Ruiz-Torralba et al. (2018); Thaipong et al. (2006); Zeb (2015b).
Ferric Reducing Antioxidant Power (FRAP)	Guava, fruits	Koley et al. (2018); Ruiz-Torralba et al. (2018); Thaipong et al. (2006); Zeb (2015b).
Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) ABTS	Guava, grapes, bakery products	Koley et al. (2018); Ruiz-Torralba et al. (2018); Sridhar and Charles (2019).
Total antioxidant activity (TAA)	Red Beans	Thaipong et al. (2006)
Sage ( <i>Salvia officinalis</i> )	Sage ( <i>Salvia officinalis</i> )	Sage ( <i>Salvia officinalis</i> )
Cupric reducing antioxidant capacity (CUPRAC)	Legumes	Koley et al. (2018)
Peroxy radical scavenging activity (PRSA)	Beans	Amarowicz and Shahidi (2017); Koley et al. (2018).

**Table 3***Methods for assessing antioxidant capacity compared by key criteria*

	Principle		
	ROS/RNS Removal	Transfer of hydrogen atoms	Electron transfer
	Superoxide	ORAC	Total Phenols
	Hydrogen peroxide	TRAP	TEAC
	Hydroxyl	Crocin	FRAP
Example of	Oxygen singlet, peroxy nitrite	Oxidation of LDL	DMPD Cu (II) reduction DPPH
Biologically relevant?	No	Yes	No
Simple to measure?	No	No (except ORAC)	Yes
Instrumentation readily available?	No	Yes (except TRAP and ORAC)	Yes
Reproducible?	Undetermined	Undetermined	Undetermined
Suitable for hydrophilic and lipophilic antioxidants?	No	No (except ORAC)	No (except TEAC)

DMPD 5,5-dimethyl-1-pyrroline-N-oxide, DPPH 2,2-diphenyl-1-picrylhydrazyl, FRAP Ferric reducing antioxidant power, LDL Low density lipoproteins, ORAC Oxygen radical absorbance capacity, TRAP Total radical trapping antioxidant potential, TEAC Trolox equivalent antioxidant capacity. Source: MacDonald-Wicks et al. (2006).

## Dairy applications

Consumers are aware of the effect of diet not only on their well-being, but also on the health promotion that food can confer when combined with good habits and a balanced diet. In this scenario, dairy companies can find multiple opportunities to develop and manufacture

different types of foods: ice creams, desserts, yogurts, fermented milks, among other preparations. In practice, most global companies have paid attention to the development of "natural" foods, that is, with minimal or no use of synthetic chemical compounds (stabilizers, colorants or preservatives) (Caleja et al., 2016;

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Granato; Nunes; Barba; Barba, 2017; Granato *et al.*, 2018; Karnopp; Oliveira; de Andrade; Postinger; Granato, 2017; Ramos *et al.*, 2017; Reis; Martins; Vasconcelos; Morales; Ferreira, 2017).

Among the main technological challenges in developing new dairy products with plant extracts, it is noted that optimization of multiple factors (price, availability and chemical characterization of ingredients, and logistics among others), scaling up and optimization of processes to launch a new product to the market, and optimization of product formulation (i.e., sensory aspects and shelf life) are required (Granato *et al.*, 2018).

Although the term 'extract' is often used in many foods science and technology studies, two important factors emerge that need to be considered: 1) plant extracts should be derived from water, ethyl alcohol or their binary mixtures because the consumer should not receive any extracts obtained using toxic organic solvents. Solvents such as dichloromethane, hexane, ethyl ether, chloroform, and methyl alcohol should be avoided, and extracts should be derived from herbs with traditional and recognized uses (Granato *et al.*, 2017). If these two factors are not considered, toxicological testing (*in vitro* and *in vivo*) should be performed by the industry or research team to ensure that the herbal extract is safe for human consumption. The raw material should be free of Salmonella and have low counts of coliforms, Staphylococcus aureus, yeasts, and moulds.

Then the solvent and extraction conditions (temperature, time, plant: solvent ratio, agitation, etc.) must be defined. In a sequence, the liquid (aqueous extract) or freeze-dried extract (alcoholic or water/EtOH mixture) should be added into the dairy food, and analyses (i.e., physicochemical, rheological, sensory,

functional properties, etc.) should be performed. Another important remark should be made here: since these dairy products are new, it is desirable to make appropriate comparisons with commercial products (as similar as possible) (Granato *et al.*, 2018).

## Milk

FCs are present in milk and milk products, either as natural metabolites (which arrived due to cattle feeding) or added in the process. O'Connell and Fox (1999) examined the importance and application of phenolic antioxidants in milk and milk products. Phenolic antioxidants are not synthesized in animal bodies, however, they come from a plant-rich (high CF) diet. It has been reported that the effects of dietary FC on milk quality and composition are significantly affected (Zeb, 2020).

In terms of milk production, quality and animal health, species such as *Trifolium repens* and *Medicago sativa* are preferred for feeding (Aerts; Barry; McNabb, 1999). By following a proper diet, it is possible to find FCs such as thiophenol, phenol, cresols, ethylphenols, ethylphenols, thymol and carvacrol in bovine, goat, and sheep milks (Lopez; Lindsay, 2002). Some of these can also be formed from amino acids. Cow's milk is therefore a good source of several FCs, including equol and phenolic antioxidants (Tsen *et al.*, 2014). These secondary metabolites are formed by the bovine intestinal bacterial flora from the FCs present in the feed. It should be noted that milk lipid concentration was positively correlated with FCs. Kilic and Lindsay (2005) showed that alkylphenol and its conjugates are present in cow, sheep, and goat milk as derivatives, such as sulphate, phosphate and glucuronides.

Propolis extracts have been shown to improve milk quality when supplemented in the diet of dairy cattle (Aguiar *et al.*, 2014). However,

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different FC have different effects on milk quality. It is possible that differences in phenolic profile are caused by variations in diets (Lopez; Lindsay, 2002; Quintero-Anzueta *et al.*, 2021). When cows are fed high levels of any particular type of crop, other FCs can be identified in milk, for example, in those from *Pteridium aquilinum* ptaquilcoside (50 mg/L) has been found (Alonso-Amelot; Castillo; Smith; Lauren, 1996) or in clover genistein (30 µg/L), equol (300 µg/L) and daidzein (5 µg/L) (King; Mano; Head, 1998). Some feeds or plants such as grape pomace produce no effect on phenolic composition, but an induced modification in the fatty acid profile of milk (Ianni *et al.*, 2019). Similarly, in the diet of dairy goats, where pistachio by-products were used as fodder, beneficial effects on the fatty acid profile of milk were generated (Ghaffari; Tahmasbi; Khorvash; Naserian; Vakili, 2013; Sedighi-Vesagh; Naserian; Ghaffari; Petit, 2015; Zeb, 2020).

Due to the antioxidant nature of FCs, they are used as food additives. Milk supplemented with grape seed polyphenols and apple extracts has been shown to suppress flavour characteristics (Axten; Wohlers; Wegrzyn, 2008). Apple extracts were also reported to have major flavor profile implications for product development due to their high level of bitterness. O'Connell and Fox (1999) studied the effects of different FCs on the heat stability of milk. They found that some did not disturb the heat stability of milk, such as guaiacol, thymol, chlorogenic acid, vanillin, BHA, propyl gallate and BHT. Quinic acid showed a significant reduction in the stability of skimmed milk. Caffeic acid, vanillic acid and ferulic acid were found to improve heat stability.

## Yogurt

As yogurts and other fermented milks are very versatile and can be consumed flavored or unflavored, as part of a hot or cold meal, the development of different types of products

(i.e., stirred, Greek, skimmed, whole, probiotic, symbiotic, prebiotic, etc.) is a viable way to increase the market. In this regard, multiple research studies have indicated that the use of herbal preparations (basically aqueous extracts) not only increases the bioactive compounds and antioxidant activity of yogurts and fermented milks, but also considerably increases the sensory acceptability of these novel foods (Granato *et al.*, 2018). Various CF additions have been carried out in yoghurt. For example, extracts of four grape varieties and CF-rich grape callus have been added as functional ingredients (Karaaslan; Ozden; Vardin; Turkoglu, 2011), where it was found that grape callus culture has the potential to be used as a food supplement.

On the other hand, Chouchouli *et al.* (2013) showed that grape seed extract fortification in yoghurt improves antioxidant activity and produces a slow decrease in plain and total phenolic content with storage time, both in whole and nonfat yoghurts. However, polyphenols were still present in all yogurts after 32 days of storage. A similar study reported that grape peel extract was 20–25 % effective in whipped yogurt (El-Said; Haggag; Fakhr El-Din; Gad; Farahat, 2014).

## Cheeses

FCs, when added to cheese curd, showed high retention coefficient values (El-Said *et al.*, 2014). Retention coefficient values were highest in flavone, followed by hesperetin, and lowest in cheese with added catechin, compared to the control. The selected FCs showed three times higher values of antiradical activity than the control with FCs, suggesting beneficial effects. Similarly, it happened with the CFs present in *Matricaria recutita* L. (chamomile) extract, which were incorporated into cottage cheese (Caleja *et al.*, 2015). It was found that these FCs improved the antioxidant activity and storage stability of the cheese.

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## Dairy desserts

Bandyopadhyay, Chakraborty and Raychaudhuri (2008) evaluated the antioxidant activity of beetroot (*Beta vulgaris*), mint (*Mentha spicata* L.) and ginger (*Zingiber officinale* L.) in sandesh, a dairy dessert (a coagulation product of a milk protein mass called chhana, which is itself a heat and acid coagulated milk product analogous to cottage cheese). The authors also compared the antioxidant activity of the product with the synthetic antioxidants TBHQ (2-(1,1-dimethylethyl)-1,4-benzenediol), BHA (2-tert-butyl-4-hydroxyanisole and 3-tert-butyl-4-hydroxyanisole) and BHT (2,6-bis(1,1-dimethylethyl)-4-methylphenol) to evaluate the efficacy of natural antioxidants in reducing lipid oxidation. Beetroot, mint, and ginger have good antioxidant activities, comparable with the synthetic antioxidant TBHQ and the combination of BHA and BHT.

However, ginger has the highest antioxidant activity, closest to TBHQ and BHA + BHT, but peppermint showed better effectiveness in inhibiting lipid oxidation.

In terms of antioxidant activity and lipid oxidation, the combination of ginger and/or mint with beetroot showed better performance than beetroot alone. Sandesh containing beetroot, ginger, a combination of beetroot with ginger or mint, or a combination of mint with ginger was more acceptable to the panelists than the control. According to the authors, herbal sandesh can be easily introduced into the market, with the attraction that the natural antioxidants have no adverse health effects.

## Ice creams with antioxidants

Ice cream and frozen desserts are popular all over the world. The main consuming countries are New Zealand, the United States, Canada, Australia, Belgium, Finland, and Sweden. The popularity of ice cream stems from several

characteristics, such as partial freezing, the cooling and refreshing sensation that occurs when the product is consumed, its sweet taste and the lack of a preconditioning aroma. It comprises several related products, which differ mainly in the relative amounts of ingredients rather than in the manufacturing technology. Ice cream is manufactured as conventional, low-fat, light, fat-free and low-sucrose products. Other frozen desserts include frozen yoghurt, popsicle, sorbet, frozen dairy dessert, frozen confectionery, frozen dairy confectionery, milkshakes, and slush. The nomenclature varies from country to country depending on current legislation (Chandan, 2008).

Ice cream is a valuable food containing highly nutritious ingredients for human health because it is composed of milk, which is a source of protein, certain vitamins, and minerals. Therefore, it consists of a complex freezing system, which is made from a mixture of dairy products, with the required percentage of fat and non-fat solids. It also includes the addition of sugar, flavor, color, stabilizer, fruit, and other ingredients. All these components are mixed and processed in a suitable manner to constitute the preparation of ice cream, commonly referred to as 'mix'. It is a colloidal system, characterized by thermodynamic instability.

Cream ice creams are considered frozen dairy desserts, their content includes milk fat, and their composition is regulated and varies from country to country. For example, in the United States, the fat content of ice cream must be 10 %, and may increase in premium ice cream, between 12 % to 14 %. In the UK, the milk solids content of the fat must be no less than 5 % and 2,5 %. Different additives and flavorings (such as nut and chocolate) are usually added to create a variety of flavors. In Colombia, according to NTC 1239 in 2002, the allowed percentage of total fat in dairy products is 10-4 %, and vegetables 6 %. For milk solids, 36 - 27 %.

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Although interest in exploiting frozen dairy desserts, as carriers of these bioactive plant compounds against oxidative stress and inflammation-related diseases, has increased in recent years, there are processing and food quality challenges to overcome. For example, several antioxidant compounds are considered non-nutritive or even anti-nutritive, susceptible, or unstable according to common processing practices, such as for ice cream (heat treatment, aeration and frozen storage), leading not only to a partial loss of their physiological activity but also to food discoloration, development of bad taste and aftertaste, and triggering of lipid oxidation reactions. Therefore, the selection of antioxidants for ice cream prototyping is considered to be a rather difficult and laborious process (Soukoulis *et al.*, 2014).

Functional ice cream is the new line that is gaining space in the market. For example, vanilla extract is the most common product with potential antioxidant activity used for ice cream production (Marshall; Goff; Hartel, 2003; Tai; Sawano; Yazama; Ito, 2011). Typically, the amounts of vanillin range from 5 to 100 ppm (Burdock, 2016). Therefore, vanillin, in addition to its dominant role as a flavoring agent in frozen dairy desserts (Cadena; Cruz; Faria; Faria; Bolini, 2012), can also exert antioxidant activity, improving the resistance of ice cream against light- or oxygen-induced oxidation, which can result in flavor and aroma defects, such as metallic, rancid, and cardboard off-notes (Shiota; Ikeda; Konishi; Yoshioka, 2002). Vanilla extract constituents such as p-hydroxybenzoic acid, p-hydroxybenzaldehyde, vanillic acid and vanillin have been held responsible for its potent antioxidant role (Charles, 2013).

Vanillin can also promote RL scavenging action in food products, providing an effective strategy to control quality loss due to lipid oxidation. Based on the first considerations, the addition of pure or concentrated vanilla bean

extracts or synthetic vanilla flavorings was able to hinder the oxygen uptake of unsaturated fatty acids and phospholipids after 12 months of storage of spray-dried ice cream powders (Pyenson; Tracy, 1950). Similarly, the addition of vanillin to ice cream mixes after heat treatment improves their stability against autooxidation, which is responsible for the development of unpleasant cardboard flavors. This is mainly attributed to the inactivation of xanthine oxidase, known to catalyze the oxidation of vanillin to vanillic acid (Gassenmeier, 2003). However, the potential health benefits associated with vanillin ingestion in ice cream need to be clinically proven (Soukoulis *et al.*, 2014).

Gabbi, Bajwa and Goraya (2018) compared the properties of differently processed ginger-added ice creams (ginger juice and paste from 2 to 8 % w/w, ginger candy from 5 to 20 % w/w and ginger powder from 0,5 to 2 % w/w). Inclusion of juice and paste significantly reduced total solids, fat, protein, and excess, while ginger candy and ginger powder increased solids and crude fiber ( $p < 0,01$ ). Ginger (*Zingiber officinale*) is a spice rich in antioxidants and contains important biologically active components, including pungent components, gingerols and shogaols.

Ginger has been highlighted as a potential functional food for use in new product development. The authors found that antioxidant activity and total phenols increased in all formulations ( $p < 0,01$ ), due to all processed ginger products. Samples with 6 % juice and 4 % paste were the most liked in terms of taste and scored the highest in overall acceptability. Ice cream containing more than 10 % caramel and 1 % powder had lower acceptability due to its higher pungency.

Kavaz, Yüksel, and Dağdemir (2016) produced ice cream with dried Besni grapes

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(*Vitis vinifera* L.) at 5-15 g /100 g, and observed that grape pomace increased viscosity, redness, ash, total phenolics, and flavonoids, but no statistical differences were observed in overall sensory acceptability, taste, or texture. Similarly, Borrin, Georges, Brito-Oliveira, Moraes, and Pinho (2018) manufactured ice cream added with curcumin-loaded nanoemulsion, aiming to replace synthetic yellow dyes in ice cream formulation. The nanoemulsion did not cause significant effects on the physicochemical, rheological, and sensory properties of the test samples.

Green or herbal tea extracts were found to be suitable ingredients to increase the phenolic content of ice cream. In a study by Karaman and Kayacier (2011), the use of black and herbal teas (chamomile, linden, and sage) at two temperatures for ice cream production suggested that antioxidant activity depended on both the type of tea and the preparation temperature, with black or linden tea brewed at 80 °C resulting in the highest antioxidant activity of ice cream. The same authors also highlighted that the herbal tea extracts used can also confer significant antimicrobial effects (against *Listeria monocytogenes*) and viscosity improvements, but in most cases, they are accompanied by moderate deterioration of flavor (astringency, sharpness, and bitterness) and color attributes of the final products (Karaman; Kayacier, (2011); Ozturk; Golec; Karaman; Sagdic; Kayacier, 2010).

The use of finely ground green tea (matcha) in ice cream has also been used to increase the antioxidant capacity of the final product but has a negative impact on textural organoleptic properties such as astringency, bitterness, lumpiness, powdery texture, and insufficient air incorporation (Soukoulis *et al.*, 2014). To overcome these drawbacks, Fukuda (2012) developed a new formulation by incorporating non-polymeric catechins and caffeine at a use rate ranging from 0,001

to 0,18. These systems exhibited very good air incorporation capacity and excellent texture and shape retention characteristics without adversely affecting product flavor.

Sanguigni, Manco, Sorge, Gnessi and Francomano (2017) evaluated the effects of a natural antioxidant ice cream on oxidative stress, vascular function, and physical performance in healthy individuals. They produced a natural antioxidant ice cream based on a mixture of cocoa powder with hazelnut and green tea extract. These ingredients are known for their high polyphenol content. The total polyphenol content in the ice cream was 1817 mg GAE/L compared to only 96 mg GAE/L in the control ice cream. The authors suggested that ingestion of an antioxidant ice cream consisting of a mixture of selected foods high in polyphenols strongly improves vascular function and physical performance, probably through an oxidative stress-mediated mechanism. They concluded that ice cream is potentially effective in counteracting oxidative stress in healthy individuals, athletes, the elderly and even in patients suffering from many chronic and degenerative diseases due to powerful antioxidant effects, durability, and stability over time.

On the other hand, the incorporation of fruit preparations (fruit juices, purees, or concentrates) into frozen dairy desserts has been verified as an alternative strategy to increase their phenolic content. The addition of frozen wild blueberry puree (5,33 % w/w) and juice concentrate (4,92 % to 5,33 % w/w) was reported to be a suitable means to provide antioxidant functionality in soy milk-based ice creams without affecting consumer acceptability (Camire; Dougherty; Teh, 2006).

Favaro-Trindade, Bernardi, Bodini, Balieiro and de Almeida (2006) demonstrated

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that ice cream could be an excellent vehicle for providing combined probiotic-antioxidant functionality by blending acerola fruit juice in a mixture with ice cream fermented by *Bifidobacteria*. This allowed retention of live cell counts and ascorbic acid levels under prolonged frozen storage without altering taste or texture. Sun-Waterhouse, Edmonds, Wadhwa and Wibisono (2013) studied the effects of three different varieties of kiwifruit (green, yellow, and red pulp) added to regular ice cream and claimed powerful health benefits due to the presence of several bioactive compounds including ascorbic acid, caffeic acid, catechins, carotenoids (lutein and beta-carotene), salicylic acid and o-coumaric acid. In addition, favorable effects were also obtained in terms of rheological behavior, melt strength and overflow of the final product, where food systems supplemented with red-fleshed kiwifruit were found to be the most acceptable.

Recently, it was shown that the addition of persimmon fruit puree to ice cream contributed to a significant improvement of its total phenolic content and 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity (Karaman *et al.*, 2014). In the same study, by implementing the order of preference by similarity to ideal solution (TOPSIS) technique approach, it was shown that sensory properties accounted for 80 % of consumer preference response compared to 20 % for health claims. It was concluded that fortification of 24 % w/w ice cream with persimmon puree can satisfactorily meet both the criteria for acceptable organoleptic characteristics and antioxidant activity. On the other hand, Teh, Dougherty and Camire (2005) showed that reducing the fat content of ice cream supplemented with blueberry concentrate had no significant effect on the amount of anthocyanins, ferulic acid and CF.

Waste products from the agri-food industry, such as molasses or pomace, are

generally considered good sources of bioactive compounds, including natural antioxidants (Ayala-Zavala *et al.*, 2011). Despite technological obstacles, to overcome the exploitation of food waste (mainly drying, storage and transportation costs), its often-high content of a wide range of bio actives and micronutrients, makes its exploitation for food production purposes attractive (Oreopoulou; Russ, 2007). Significant benefits have been revealed by supplementing ice cream with grape wine corns, a common residue, rich in polyphenols and dietary fibre that is readily fermented by human gut bacteria (Hwang; Shyu; Hsu, 2009; Topping; Clifton, 2001).

Also, the feasibility of using pomegranate (*Punica granatum* L.) peel by-products, rich in CF and PU-FA, to produce functional ice cream has been investigated (Çam; Erdoğan; Aslan; Dinç, 2013; Çam; İçyer; Erdoğan, 2014). According to the findings of Çam *et al.* (2013), the addition of PBP (at 0,1 % and 0,4 % w/w) was accompanied by a significant increase in CF (mainly punicalagin, ellagic acid and its derivatives), which was associated with higher DPPH uptake activity and more effective inhibition of  $\alpha$ -glucosidase activity, the latter reducing the GI of the prepared formulations.

Similarly, increased antioxidant activity was also found after the addition of pomegranate seed oil (PSO) to ice cream (Çam *et al.*, 2013). In a subsequent study, Çam *et al.* (2014) indicated that the antioxidant and  $\alpha$ -glucosidase inhibitory action of PBP encapsulated in spray-dried maltodextrin matrices compared to native PBP was not significantly affected. Moreover, microencapsulation of PBP appeared to be a good strategy to overcome restrictions related to adverse organoleptic properties resulting from the presence of phenolics, such as astringency, unsatisfactory tongue lubrication and puckering of the oral mucosa (Çam *et al.*, 2014).

Finally, the addition of whey protein isolate (WPI) glucosated with D-allose and D-psicose sugars in ice cream has been mentioned as a novel way to provide excellent antioxidant properties along with improved foaming and emulsifying ability (Puangmanee; Hayakawa; Sun; Ogawa, 2008). The authors suggested that the RL scavenging activity for the specific formulations was associated with the conjugation of Maillard reaction products found in WPI with the C-3 hydroxyl group of these sugars.

With these examples, it is evident that consumers want to buy ice cream with fewer artificial ingredients and enhanced bioactivity (i.e., more phenolics and antioxidant activity) (Sloan, 2011).

## Conclusions

Generally, the concentration and total antioxidant activity of different phenolic compounds in the studied fruit concentrates are higher than the data reported for fresh fruits or jams, which is due to the technical process of concentration and the percentage reduction in the moisture content of the product in relation to the starting fruit. These red fruit concentrates may be a suitable raw material to produce FAs rich in natural antioxidants. Long-term consumption of these antioxidants may prevent diseases related to oxidative stress. However, more research is needed on the antioxidant properties of products made from these raw materials and the impact of their intake on the antioxidant defense of consumers.

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