

Natural sweeteners: Sources, extraction and current uses in foods and food industries

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Abstract

Food producers have leaned towards alternative natural and synthetic sweeteners in food formulations to satisfy market demands. Even so, several synthetic sweeteners (e.g., aspartame, saccharin, sucralose) are becoming less popular due to health-related concerns, lower nutritional values, and controversies around their safety. Conversely, natural sweeteners confer favourable customer perceptions due to their association to a healthier lifestyle and higher nutritional values. This article discusses the evidence of natural sweeteners in the available commercial products. A comprehensive review of natural sweeteners is presented, which includes their resources, properties and extraction methods, as well as a discussion on several emerging technologies that offer improvements to the traditional extraction methods. Finally, the progress of natural sweeteners in the food industry is assessed, and the commercial food products containing these natural sweeteners are mentioned.

Keywords: *Natural sweeteners; food industry; steviol glycosides; novel extraction methods; honey; commercial products.*

Nomenclature

ADME: Absorption, Distribution, Metabolism and Excretion

ATP: Adenosine triphosphate

CPCRI: Central Plantation Crops Research Institute

DNL: *De novo lipogenesis*



- 44 FDA: Food and Drug Administration
- 45 F&B: Food & beverage
- 46 EFSA: European Food and Safety Authority
- 47 FAO: Food and Agriculture Organization
- 48 FE: fructose equivalent
- 49 FOS: fructo-oligosaccharides
- 50 GLP-1: glucagon-like peptide-1
- 51 GRAS: generally recognized as safe
- 52 HHP: High-Pressure Processing
- 53 MWCO: Molecular weight cut-off
- 54 MF: Microfiltration
- 55 NF: Nanofiltration
- 56 RE: Rotary Evaporator
- 57 Reb-A: *rebaudioside A*
- 58 SGs: Steviol glycosides
- 59 SSB: Sugar-Sweetened Beverages
- 60 UF: Ultrafiltration
- 61 WHO: World Health Organization
- 62 5-HMF: 5-hydroxymethylfurfural

63

64 **1. Introduction**

65 Among macronutrients, carbohydrates provide 40-80% of the total energy intake. In
 66 foods, carbohydrates can be found as free and non-free sugars; non-free sugars are

naturally present within the cell structure, e.g., sugar in fruit and vegetables, starchy carbohydrates in grains, lactose in dairy products, etc. In contrast, the free sugars do not present naturally but are often added to food, such as monosaccharides (glucose, fructose) and disaccharides. However, excess energy intake is associated with the accumulation of body fat (Onaolapo et al., 2020). More specifically, excess intakes of free sugars not only result in fat accumulation but also compromises micronutrient density and increased the risk of other adverse health conditions, such as diabetes and cardiovascular diseases (Hagger et al., 2017).

Sucrose (common sugar), composed of fructose and glucose in equal ratio, is fundamental in our diet since glucose metabolism is needed to produce adenosine triphosphate (ATP), synthesis of different biomolecules and more importantly, for cellular respiration function (Archer, 2018). Unfavourably, fructose causes dysregulation of carbohydrate and lipid metabolism. This is because fructose metabolism is not regulated by hepatic energy requirements, resulting in excess fructose uptake by the liver and an increase in *De novo lipogenesis* (DNL) (Stanhope, 2016). Consequently, even though sugar is a significant energy source in the human diet, it may also promote dysmetabolic conditions (Lee et al., 2018).

The main disadvantage of common sugar made out of sugarcane juice is that the refined product lacks additional beneficial compounds (e.g., bioactive molecules) that could enhance its nutritional value. In the sugarcane juice refining process, non-centrifugal cane sugars, brown sugar and molasses are produced as by-products. These by-products were proven to contain several bioactive molecules, including flavonoid glycosides and phenolic acids (Singh et al., 2015), which later led several other authors to recommend

the non-centrifugal sugars to substitute the refined sugars due to phenols and flavonoids' favourable dietary effects (Cervera-Chiner et al., 2021; Lee et al., 2018). For the same reason, natural sweetening alternatives are becoming more attractive to consumers.

Although the global sugar demand has generally declined due to the rising concerns around the potential health effects caused by elevated sugar intakes, the Food and Agriculture Organization (FAO) data suggest that the growth in sugar intake will remain strong in developing countries. By 2028, it is estimated to increase by 32 Mt, compared to the approximated value of 203 Mt in 2008 (OECD/FAO, 2019).

Currently, processed foods play a substantial role in excess sugar intake. Steele et al. (2016) reported that ~90% of the total average sugar intake comes from ultra-processed foods (e.g., fruit juices), concentrated syrups, soft and sports drinks, bakery products, among others, which often consist of elevated sucrose indexes ranged from 50 to 1000 g/L (Raganati et al., 2015). In addition, the most popular drinks (e.g., energy drinks, soda, fruit juices), depending on their beverage type, contain sugar contents of between 100-135 g/L (Health, 2014). Past studies have pointed out that a greater intake of sugar-sweetened beverages (SSBs) is related to a 30% increase in developing type 2 diabetes (Wang et al., 2015), and one SSB serving (250 mL) per day increased the type 2 diabetes incidence by 18% (Imamura et al., 2015).

The increasing demand has led researchers to explore new natural and synthetic sweeteners as sucrose alternatives (Saraiva et al., 2020). When extracted with the beneficial compounds from their sources, the natural sweeteners (glucose, fructose, and sucrose) are classified as nutritional choices. New possibilities have been proposed and studied to cater to the demand, including honey, xylitol, erythritol, maltose, maltodextrin,

113 stevia, molasses, maple syrup, coconut sugar, agave nectar and date sugar (Valle et al.,
114 2020).

115 Artificial sweeteners, also known as sugar substitutes or non-sugar sweeteners, are
116 synthetic substances used to replace sugar during the sweetening process of several
117 products. These additives are identified as intensive sweeteners because they display a
118 higher sweetening power than conventional sugar. The use of synthetic sweeteners has
119 become popular in the last 40 years. Currently, they are used in a wide range of
120 processed foods, such as sweets, preserves, dairy products and beverages. Six of these
121 high-intensity sweeteners have been approved as food additives by the Food and Drug
122 Administration (FDA), including aspartame, neotame, saccharin, acesulfame-k, sucralose
123 and advantame (Mooradian et al., 2017).

124 Aspartame, the first FDA-approved sweetener, was used in solid foods since 1981, in
125 beverages since 1983, and it currently has no usage restriction in food and beverage
126 (F&B) preparation since 1996. However, the use of aspartame is still controversial.

127 Aspartame consumption generates concerns among health representatives and the
128 general consumers since it is widely used by big F&B companies such as *Coca-Cola* in
129 many of its products (FDA, 2019). The consumption has been associated with different
130 health effects, including oxidative stress in blood cells (even at the recommended dose
131 of 40 mg/kg per day), interference of neuronal cell function, hepatotoxicity and kidney
132 disfunction (Ardalan et al., 2017; Choudhary & Pretorius, 2017). Moreover, its
133 consumption is not recommended for people suffering from phenylketonuria since they
134 cannot metabolize phenylalanine (a chemical component used in the aspartame
135 synthesis). On the contrary, some evidence support that aspartame consumption does

not influence blood pressure, glucose and lipid profiles, suggesting that aspartame has no adverse effects on human metabolism and constitutes a safe option for type 2 diabetes patients (Choudhary, 2018; Gupta, 2018), particularly neotame, an isomer of aspartame. Similar advantages also claimed for advantame, an N-substituted derivative of aspartame and vanillin (B. Chen et al., 2020; Dhartiben & Aparnathi, 2017). Both neotame and advantame are sweeter than aspartame, ~13,000 and ~20,000 times sweeter than common sugar, respectively. The neotame was FDA-approved as a general-purpose sweetener in 2002 (Gupta, 2018), whereas the advantame was approved in 2014 (Khan & Aroulmoji, 2018). Despite their relationship with phenylalanine, neither neotame nor advantame present a bitter taste (Carocho et al., 2017).

Another artificial sweetener, sucralose, is also a non-caloric sweetener (does not break down in the body) and ~600 times sweeter than sugar. Sucralose is stable at different temperature and pH conditions, making it an optimal option for many industrial applications. However, some studies pointed out that sucralose interferes with digestive processes and may increase glucose and insulin levels in the organism, boosting the risk of weight gain and diabetes (Khamise et al., 2020). Regardless of this evidence, ADME (i.e., absorption, distribution, metabolism, and excretion) research on sucralose shows that it is mainly eliminated through faecal excretion and is not absorbed or digested in the human body (Magnuson et al., 2017)

As for saccharin, its usage in Canada had been banned since 1977. The USA FDA had also considered its prohibition since evidence suggests its role in inducing bladder cancer in rats. However, in several subsequent studies, the relationship between saccharin and bladder cancer in humans was not demonstrated. After that, the use of this product has

159 been approved by the FDA and the European Food and Safety Authority, EFSA (Ansari
160 et al., 2015). Another common synthetic sweetener is acesulfame-K, which is recognized
161 as a thermostable component and used in cooking. Acesulfame-K is 120 times sweeter
162 than sugar but bears a bitter taste, and thus, it is commonly used with other sweeteners.
163 It is worth mentioning that acesulfame-K cannot be metabolized, implying a zero caloric
164 intake. Acetoacetamide, a breakdown product of acesulfame-K, may be toxic at high
165 concentrations; nonetheless, the human exposure at low concentration is negligible.
166 (Krishnasamy, 2020).

167 In general, artificial sweeteners have very low-calorie content and intense sweetness,
168 making them attractive to both consumers and food manufacturers. However, these non-
169 caloric sweeteners have minimal nutritional value. They also may result in incomplete
170 activation of food reward pathways, leading to sweet cravings and food-seeking, which
171 may cause excessive caloric intake and weight gain (Mooradian et al., 2017).
172 Furthermore, some experimentation in animals has demonstrated that these artificial
173 sweeteners influence specific metabolic syndromes. For example, glucose tolerance may
174 be reduced in response to the changes in the microbiome after a moderately prolonged
175 artificial sweeteners consumption (Green & Syn, 2019). Although notable field authorities,
176 including FDA and EFSA, have approved some artificial sweeteners, the available
177 evidence to support their industrial use and consumption is still inconclusive. Moreover,
178 many contradictory findings on safety and health implications were reported, making their
179 use is controversial.

180 Presently, both sucrose and artificial sweeteners can be replaced by natural sweeteners.
181 Prevailing market trends suggest that natural food products are more appealing to

consumers, who identify natural products as healthier options. The current trend indicates that consumers are willing to try natural sucrose alternatives (Mora & Dando, 2021). For example, Stevia-sweetened beverages have more positive consumer perceptions than the common SSBs (Olivo, 2019). Thus, natural sweetener usage may represent a new and substantial commercial opportunity for many companies. Natural sweeteners also present positive consumption effects, such as improving metabolic health, preventing weight gain and lowering blood glucose. Other advantages are; (1) low glycemic potency, as presented in honey and agave nectar, could be advantageous for people on low glycemic index diets, (2) low fructose contents, as found in maple syrup (Edwards et al., 2016), and (3) containing biomolecules with nutritional and health benefits (e.g., vitamins, phytohormones and minerals) (Valle et al., 2020). It was reported that the general composition of honey, maple and agave syrup consists of at least 3% dietary fibre, 1.4% proteins, <2% minerals (potassium, calcium, and magnesium), and polyphenols with potential antioxidant activity (Edwards et al., 2016). Similarly, dark molasses and blackstrap molasses consist of high antioxidant activities, at 4.89 and 4.56 mmol/100g, respectively. The substitution of 130g refined sugar with 337g blackstrap molasses in viable products would increase its antioxidant content by ~10.7 mmol (Eggleston, 2019). Stevia, another important natural sweetener that constitutes mainly steviol glycosides, has been widely used in Paraguay and Japan. Stevia has low-calorie with a zero glycemic index; it helps lower blood glucose levels and maintains dental health since it does not contain carbohydrates. It has been widely used in bakery products due to its stability at elevated temperatures (Singh et al., 2020). Because of its attractive benefits, *Cargill Inc.*, in collaboration with *Coca-Cola*, *Whole Earth Sweetener Co.* and *PepsiCo*, requested

stevia to be FDA-approved as a Generally Recognized As Safe (GRAS) ingredient. After exhaustive attempts, stevia was authorized in 2004 by the United Nation's FAO and the World Health Organization (WHO) Committee on Food Additives (Merillon & Ramawat, 2017). Not until 2008, the US FDA granted GRAS recognition for the purified steviol glycosides (Perrier et al., 2018). Ever since, this sweetener has been employed in several commercial beverages, such as Sprite®, produced by the *Coca-Cola* company (Ismail et al., 2020).

Polyols, including erythritol, mannitol, and xylitol, are natural sweeteners since they occur naturally in fruits and vegetables, but they also can be synthesized from mono or disaccharides (Edwards et al., 2016). Xylitol is a five-carbon polyol, obtained from xylose hydrogenation and the sweetest polyol but only provides 2.4 kcal per gram (making it 5% less sweet than sugar). Xylitol is beneficial to promote salivation, as well as the reduction of bacterial load and cavities. It is estimated that the xylitol market is worth 670 million USD and continues to increase, as it is being used in several commercial product formulations, including soft drinks, gum, candy, and baked products (Carocho et al., 2017). However, consumers are still not familiar with xylitol as much as stevia or other artificial sweeteners (sucralose, aspartame and saccharin); thus, making xylitol a less preferred sucrose-alternative (Mora & Dando, 2021).

Considering the benefits and current attention given to the natural sweeteners in the sweetened food market, this review aims to provide evidence related to their implementations and recent uses. Traditional natural sweeteners discussed are the common nutritive alternatives and possess promising commercial potentials, including honey, molasses and blackstrap molasses, maple syrup, coconut sugar, agave nectar,

date syrup and steviol glycosides and sorghum syrup. Most of these sweeteners are readily introduced in several commercial products, frequently marketed as healthier choices and accepted by the general consumers for their positive attributes. This review also compiles the information on the natural sweeteners' main extraction routes, production and purification. Equally important, their physicochemical properties are presented. Finally, an overview of the current uses and applications in the food industry is provided.

2. An overview of sources, properties, current and promising extraction methods of the natural sweeteners

2.1. Honey

2.1.1. Physicochemical and bioactive properties

Honey is likely to be the most consumed natural sweetener worldwide. Its composition and properties may vary depending on the botanical source, harvest reason, climate and the geographical region where it is produced. Nonetheless, its typical chemical composition consists of 60–85% carbohydrates, 12–23% water, minerals, vitamins, proteins, organic and amino acids, enzymes and several bioactive substances (e.g., phenols and flavonoids) (Machado De-Melo et al., 2018). Due to its unique properties, honey is useful in both nutritional and therapeutic applications. Its moisture content, typically ranging from 13 to 25%, is the primary determinant for honey shelf-life and the optimal value for preservation is 17%. The water content heavily influences the proliferation of microorganisms in honey. Honey with high water content is susceptible to osmophilic yeast spawning, whereas not in common sugar due to its osmotic pressure



(unsuitable for yeast growth). Moisture also affects other properties, such as colour, crystallization and viscosity (Machado De-Melo et al., 2018). In theory, honey with low water content tends to be difficult to be processed. Water availability for microbial growth is defined by water activity, a_w (Reid, 2020). In honey, sugar binds with water molecules making it unavailable for microorganism growth (Machado De-Melo et al., 2018). Hence, it is a better-quality criterion than water content (Reid, 2020); e.g., a_w values of honey is ~ 0.49 – 0.65 and bacterial growth requires an a_w of ~ 0.9 , while yeast and moulds need an a_w of ~ 0.7 and ~ 0.8 , respectively. Therefore, honey generally inhibits self-fermentation and self-spoilage (Machado De-Melo et al., 2018).

A small number of enzymes, including glucose-oxidase, diastase and invertase, are present in honey chemical composition. *Glucose-oxidase* is predominantly produced in the bees' hypopharyngeal glands. This enzyme degrades glucose into gluconolactone, which turns into gluconic acid and after converted into hydrogen peroxide. Hydrogen peroxide gives honey its microbial resistance. Elevated concentrations of hydrogen peroxide protect honey from bacterial degradation until the sugar concentration is high enough to preserve the honey through osmotic pressure. *Glucose-oxidase* is light-sensitive and deactivated at only 60°C (Machado De-Melo et al., 2018). On the contrary, *diastase* or *amylase* is the most thermal resistant enzyme; used to determine the honey's freshness. *Diastase* is responsible for starch and dextrins hydrolysis; however, its function in honey has yet to be understood since the nectar does not contain starch. Nonetheless, it is believed that *diastase* participates in pollen digestion (Machado De-Melo et al., 2018). Another important enzyme is *invertase* (α -glucosidase), which is crucial in transforming honeydew into honey. *Invertase* hydrolyzes sucrose into fructose and glucose, and its

activity is maintained even after the extraction. Unfortunately, due to the fructose inhibition property, the activity is lowered over time in storage. Furthermore, since *invertase* has a higher sensitivity to thermal processes, it has been proposed to be the primary indicator for honey quality control (Machado De-Melo et al., 2018).

Unlike the other natural sweeteners, honey presents various functional properties, the most important being antioxidant and antimicrobial activities (Dzugan et al., 2018). As discussed, honey is proven to delay or prevent food spoilage caused by its oxidative reactions. Moreover, *in vitro* studies have shown that honey ingestion inhibits the oxidation of human serum lipoproteins. Many honey components are classified as antioxidants, such as glucose oxidase, catalase, organic acids, carotenoids, ascorbic acid, amino acids, proteins, phenolic compounds and melanoidins (Maillard reaction products). Different pathways have been suggested to describe the honey's antioxidant activity, such as reducing reactive oxygen and nitrogen species, and inhibiting super oxidant production enzymes. Based on the presence of multiple bioactive compounds in its composition, a recent research has suggested that honey can also be used as a protective agent against pathologies that cause liver damage, radiation and inflammation (Machado De-Melo et al., 2018).

2.1.2. *Extraction and promising methods for honey processing*

Honey is extracted from either combs or apiaries. Initially, a beekeeper must place the beehive (by brushing the frames or using a fume board, bee blower, or escape board) in another apiary. The hive is then cleared for the extraction to begin. Several traditional honey extraction methods are available, but they often reduce the honey quality. The most common commercial honey production uses an extractor method (radial or

tangential), which essentially applies centrifugal force to the uncapped frames. Here, the wax-sealed honey is removed with a knife (MacFawn, 2018). After the extraction, honey may still contain undesirable compounds such as beeswax and pollen, which are removed to increase its quality and shelf-life (MacFawn, 2018). This process variation depends on the operation scale, but the general schematic is described in **Figure 1**.

Figure 1. Traditional honey production process.

The conventional extraction method starts with the removal of suspended particles. This step is combined with a preheating process (up to 40 °C) in a large scale production. The strained honey is filtered with pressure filters (e.g., polypropylene microfilter, a pore size of 80 µm) at 50-55 °C and followed by indirect heating in a tubular heat exchanger between 60–65 °C for 25–30 min. In the final stage, rapid cooling is applied to preserve its natural colour, flavour, enzyme content, and other biological substances.

Microwave heating and infrared processing, well-known methods in the food industry, have been proposed as promising extraction practices to meet better-quality honey demand. Water and dissolved sugars in honey are highly receptive to microwave interactions (Reynolds, 2019) and exhibit good absorption in the thermal radiation region. This will reduce the heating periods in both methodologies, thus bringing several added advantages over the conventional method.

- i) *Thermosonication* combines both thermal treatment and ultrasonication, two methods that have been proven successful in honey treatment. For

instance, Chong et al. (2017) demonstrated that enhanced honey quality (water activity, viscosity and colour intensity) was obtained using an ultrasonication bath with heating.

ii) *Membrane processing* is a non-thermal alternative technique. To date, membrane-based technologies have been successfully applied in many food sectors (Castro-Muñoz et al., 2020). In honey processing, different ultrafiltration (UF) membranes, characterized by MWCO of 20, 25, 50, and 100 kDa, could eliminate microorganisms; however, the desirable enzymes were also removed. This ultrafiltered honey is typically used in the pharmaceutical industry.

iii) *High-pressure processing (HPP)* is another non-thermal processing alternative. Akhmazillah and co-workers reported a Manuka honey, HPP-processed at ambient temperature, presented higher total phenolics content than its thermally processed counterpart. This method proved to improve the honey's nutritional value (Akhmazillah et al., 2013).

2.2. Molasses and blackstrap molasses

2.2.1. Physicochemical and bioactive properties

Molasses is a broad term that refers to the concentrated sugarcane or sugar beet juice. This sweetener is derived from the sucrose crystallization process, where crystallization inhibitors are accumulated in the residual syrups. These syrups are called molasse and are mainly used as livestock feed and substrate for ethanol production (Palmonari et al., 2020). Although their composition depends on the syrup's recovery stage, the molasses'

approximate compositions correspond to 17-25% water, 30-40% sucrose, 4-9% glucose and 5-12% fructose. Several compounds, such as amino acids and vitamins, are also present in small percentages. These molasses are acidic, with pH values ranging from ~5 to ~7 for cane molasses and pH of ~5.8 for blackstrap molasses. Also, salt content in these molasses can prevent hydrolysis, thus providing buffering capacity to stabilize the flavours (Mordenti et al., 2021).

Molasses are classified depending on their source and constituents. For example, blackstrap molasses, heavy and dark viscous syrups, are the residual when sucrose recovery is not feasible by the standard physical methods in a sugarcane manufacturing process or raw sugar refinery (Mangwanda et al., 2021; Mordenti et al., 2021). Similarly, high-test molasses are the resulting high concentration syrups from the clarified cane juice and have approximately 85° Brix. In contrast to the blackstrap molasses, high-test molasses are intentionally produced and not as a by-product. It has several advantages over the blackstrap molasses, such as (1) containing fewer sugar decomposition sub-products, (2) higher sugar contents as it is only subjected to a lower temperature process, and (3) possesses an intense aromatic flavour. Another molasses type is sulfured molasses, in which sulfur dioxide is added to bleach out the initial dark colouration. Sulfured molasses present higher ash contents (Mordenti et al., 2021); however, their physicochemical properties differ considerably since the compositions are highly varied.

Additionally, molasses have interesting properties for food processing, e.g., masking undesired flavours. These sweeteners may present a caramel flavour (in high-test molasses) and heavy liquorice-like nuances in the other forms. Molasses are valued as a colouring agent, especially in baked goods, where it gives golden, dark and brown

colours, enhancing the goods' visual presentation. Molasses also exhibit humectant and colligative properties, reducing water activity and extending the shelf-life of baked products. Since molasses retain the high-added-value components (e.g., phenolics) from their sources, it has been reported that the extracts promote resistance to infections and auto-inflammatory activities. Furthermore, molasses are considered a non-expensive antioxidant source (Chen et al., 2015).

2.2.2. Extraction and promising methods for molasses and blackstrap molasses processing

It is estimated that ~70% of fructose production is obtained from sugarcane (grown in tropical climates), while ~30% is produced from sugar beet (grown in relatively temperate zones). As the process by-product, 35 million tons of molasses are produced annually. **Figure 2** illustrates the sugar production process and its derived blackstrap molasses.

Figure 2. Sugar production process and its derived blackstrap molasses.

In the process, the sugar canes are first washed before the juice is extracted. The extracted juice goes through a purification stage, where it is clarified to remove the contaminants before undergoing an evaporation process (concentration step) to reduce the water content, becoming a syrup. In the crystallization step, the goal is to maximize the sucrose recovery from the syrup, and due to the process limitation, ~80% sucrose is retained in a large quantity of molasses. Even though the crystallization process involves recirculation steps to decrease the purity of the mother liquor (syrup), it still contains high

sucrose levels. When the mother liquor purity cannot be lowered further, it is separated from the process to prevent non-sucrose compounds accumulation and referred to as blackstrap molasses (Mangwanda et al., 2021).

Molasses are desirable for high-value biotechnological processes and auxiliary sucrose extraction. Unfortunately, molasses contain many contaminants, making the extraction process challenging and requiring purification before further processing (Sjölin et al., 2019). To date, several alternative methods for further molasses processing have been implemented, such as:

i) Carbonation: The increased sucrose recovery is obtained by ion-exclusion chromatography. Nonetheless, the obtained molasses cannot be used for other applications due to the impurities bound irreversibly to the compound, rendering it useless. Thus, the molasses are first diluted (to decrease its viscosity) and subsequently carbonated at high pH to precipitate the divalent cations, the major impurities. This procedure can be accomplished with different salts (e.g., potassium carbonate and sodium carbonate) or carbon dioxide. The resulting mixture is further diluted with an organic solvent, which precipitates the organic polymers, polysaccharides and proteins. Afterwards, the supernatant and precipitated fractions are separated using filtration or centrifugation, and the sugar is recovered from the supernatant solution using ion-exclusion chromatography.

ii) Ceramic nanofiltration (NF) and ultrafiltration (UF) membranes: Membrane-based process is often seen as a reliable separation technique. For sugar beet molasses, 10 kDa UF and 200 Da NF ceramic membranes have been selected to remove the high-molecular-mass compounds, including polyphenols and the



small-molecular-mass contaminants (like salts). Purified sucrose is collected as the permeate in UF and as the retentate in NF. The main drawback is the severe membrane fouling caused by starch, pectin and proteins (Sjölin et al., 2019). This phenomenon is widely acknowledged as one of the main limitations when processing complex mixtures (Pichardo-Romero et al., 2020).

iii) Bentonite addition: This process is an additional filtration step that decreases the non-sugar compounds in molasses, easing the sucrose extraction process. In principle, bentonite-based solutions are added to the molasses with citric acid to adjust the pH. The blends are heated in a water bath at 60 °C and mixed for 30 min before being filtered. This method was reported to yield a reduction in turbidity and colour whilst minimally decreased the sucrose content (Djordjević et al., 2018).

2.3. Maple syrup

2.3.1. Physicochemical and bioactive properties

Maple syrup is a natural sweetener produced from different Canadian maple tree species, and the most common is *Acer saccharum Marsh* (Garcia et al., 2020). Maple syrup contains phenolic compounds that account for its antioxidant, anti-mutagenic and human cancer anti-proliferative properties. The properties of maple syrup, such as pH, colour, sugars, depending on the cultivation area. Maple syrup can be categorized into four types: golden, amber, dark and very dark (Brochu et al., 2019). Recent studies addressed that a darker colour is related to higher phenolic compounds (e.g., protocatechuic acid, coniferyl alcohol, vanillin) and mineral contents (Nimalaratne et al., 2020).

The inhibition of the carbohydrate hydrolyzing enzymes, such as α -glucosidase, is beneficial for type 2 diabetes personnel since α -glucosidase has shown inhibitory activity against glucose absorption in the intestine (Wan et al., 2012). Additionally, ethyl acetate-based extracts from this sweetener have the potential to aid in treating Alzheimer's disease. Notably, the extracts decrease the oligomerization and aggregation of the primary peptides (e.g., β -amyloid and *T*-peptides) that cause Alzheimer (Hawco et al., 2015). Besides that, the acetate-based extracts also exhibit anti-inflammatory effects.

2.3.2. Extraction and promising methods for maple syrup processing

The traditional maple syrup production techniques involve changing sap into syrup. Usually, the sap is collected in buckets, followed by water evaporation using direct heat between 9-56 h. A standardized process tends to follow these steps: extraction, filtration, evaporation and filtration, as described in **Figure 3**. It is estimated that ca. 44-gallons sap are needed to produce 1-gallon syrup (Nimalaratne et al., 2020; Snyder et al., 2019). Alternatively, a vacuum pump and tubing may extract more sap than gravity tubing or sap buckets (Moore et al., 2020).

It is also reported that reverse osmosis can enhance the sap concentration process efficiency. For example, using a commercial polyimide membrane (from GE Sepa®) crossflow filtration process increased the sap concentration due to the amplified system's osmotic pressure (Weaver et al., 2020). The method also required a shorter evaporation time. However, reverse osmosis involves maintenance and filter modules replacement, which translated into downtime issues and additional costs (Ali et al., 2021). Reverse osmosis and evaporation coupling method was suggested to overcome the limitation.

Figure 3. Schematic representation of the traditional maple syrup production.

2.4. Coconut sugar

2.4.1. Physicochemical and bioactive properties

Coconut sugar is a natural sweetener rich in carbohydrates. The sugar obtained from Suttiphuan coconut palm or *Cocos nucifera* L comprises ~15% sucrose (Muriel et al., 2019). The sugar provides a better digestion rate and lower glycemic index (GI) of between 35 to 42. The sugar contains ~4 kcal per gram, high in vitamin C, B1, B2, B3, and B6. It also presents a low glass transition temperature (Srikaeo & Thongta, 2015), typically associated with its fructose, glucose and sucrose components (Asghar et al., 2020). It is known that a coconut tree produces sap throughout the year. The sap, collected from unopened spadixes, generally presents a pH value of 7.0–7.3, along with high antioxidant activity and phenolic content (Asghar et al., 2020).

2.4.2. Extraction and promising methods for coconut sugar processing

Conventional coconut sugar processing involves sap collecting and storing at -2 °C. The sap is then heated at 115-120 °C for 3-5 h; before the nectar is crystallized into granules, oven-dried, and subsequently sieved. However, the protocol overcooks sugar, affecting its physical and chemical properties, e.g., reducing the vitamin C and B3 concentrations (Asghar et al., 2020). The temperature also affects the colour quality and pH, directly influences its antioxidant activity (Karseno et al., 2018). The standard process involves

collection and filtration, evaporation, crystallization, sieving/filtration and drying (as illustrated in **Figure 4**).

Figure 4. Schematic representation of the traditional coconut sugar production.

The sap collection is commonly performed by tapping the palm's spathe (for approximately ten-day) and collected into pots. The collected sap is filtered with deodorizing aids at 2-8 °C before being pasteurized at 90-95 °C (Karseno et al., 2018). Additionally, the sap is heated up to 140 °C for 30 min to increase the sugar crystalline properties. The product is then cooled, producing a varied mass and mixed for 10 min to dissolve the remaining lumps before drying at room temperature (Muriel et al., 2019). The steps are described below:

i) Collection techniques: Tapping and coconut sap collection are conducted at atmospheric temperature for 8-12 h using a container on the palm tree. Sap fermentation is a critical issue, where it turns white and the pH decreases from 7 to 6. The sap is preferred to be collected every hour at a steadily low temperature to avoid the occurrence. Insects and other particles could also contaminate the sap. To prevent the issues, Kasaragod-based Central Plantation Crops Research Institute (CPCRI) designed and built a coco-sap chiller, a closed system that regulates the temperature between 2-3 °C, maintaining its hygienic and freshness. The system can collect coconut sap with a zero fermentation possibility (Hebbar et al., 2015).

ii) Filtration: A filter funnel and chiffon fabric have been used in the traditional process to extend the utilization of the sap (Muriel et al., 2019).

iii) Evaporation: Generally, the syrup is concentrated between 100–120 °C for 3-5 h (Srikao & Thongta, 2015). Coconut sugar producers actively seek new technologies to concentrate and preserve the sugar better; **Table 1** tabulated several alternatives explored to date. The rotary evaporator method (RE) has been promising by providing precise and efficient distillation that conventional methods cannot achieve. RE is conducted under vacuum and allows water to evaporate at a lower temperature (the lowest of all methods). Other techniques are categorized as open heat evaporation, often conducted at high temperatures and consumed more time and energy. Yet, it is the most common technology on an industrial scale (Asghar et al., 2020).

Table 1. Alternative techniques used to concentrate the coconut sugar.

iv) Drying: The concentrated sap is further dehydrated to produce a dried product. Herein, the sap is heated until it crystalized and then gradually cooled. Since stickiness is an issue in the traditional drying processes, several advancements have been proposed, such as spray and vacuum drying (Nurhadi et al., 2018). For example, the lyophilization method can obtain final dried sugar with good functional (e.g., protein solubilities, emulsification and foaming capacity) and physicochemical properties (e.g., colour and sensory characteristics). In comparison, the spray drying method can

produce a final product with good product features and functional properties (Nurhadi et al., 2018).

2.5. Agave nectar

2.5.1. Physicochemical and bioactive properties

Agave nectar or agave syrup is a natural sweetener obtained from its fructan hydrolysis. The nectar, its main carbohydrate reserve in the form of fructans, is stored in the agave core (*piña* in Spanish) of the *Agave* ssp. plant. *Agave* genus, growing in arid and semi-arid regions, follow a crassulacean acid metabolism and a photosynthetic adaptation to the periodic water supply. These plants are commonly found in Northern and Central America; however, nearly 55% of the species are widespread in Mexico and considered agave's diversity core and origin. Due to its high sugar content, agave's consumption and socio-economic impacts are dated back to the pre-Columbian era (Pérez-López & Simpson, 2020). Among all the recorded species, *Agave tequilana* weber var. azul is the main crop for raw material extraction of agave syrup and Tequila production. The standardized extraction techniques and regulations have supported blue agave syrup's increasing popularity and acceptance as a sweetener in Mexico. It is conditioned that the agave syrup must not contain any food additives, glucose, molasses, dextrin, starches, among other compounds (NOM-003-SAGARPA-2016, 2016).

As for its composition, fructans are fructose-bound polymers with a dextrose equivalent between 3 and 29, linked to a single glucose molecule. These carbohydrates are classified accordingly to their link formation type, distinguished into three categories:



inulin, fructans with linear $\beta(2\rightarrow1)$ glycosidic bond structure; *levan-type*, fructans with linear $\beta(2\rightarrow6)$ glycosidic bond structure; and *graminan*, fructans with both $\beta(2\rightarrow1)$ and $\beta(2\rightarrow6)$ glycosidic bond branched structure (Witzel & Matros, 2020). These fructans are involved in the agave syrup production from *Agave tequilana* Weber var. azul, which contains a fructooligosaccharides mixture with branched structures $\beta(2\rightarrow1)$ and $\beta(2\rightarrow6)$ glycosidic bonds, identified mainly as graminan type of carbohydrates (Miramontes-Corona et al., 2020). The agave nectar composition presents nearly 95% total soluble solids (TSS), of which 90% corresponds to fructose concentrates, followed by glucose and sucrose (the lowest). The high content in fructose causes the syrup to have a low glycemic index (between 17-27), lower than other sweeteners (e.g., honey = ca. 55). Agave nectar is also sweeter than most syrups with high glucose or sucrose levels (e.g., honey and maple syrups). Therefore, lower amounts of agave nectar are needed to reach the desired sweetness and translated into a lower caloric intake (Barajas et al., 2017). The benefit favours agave nectar as the alternative to regular refined sugar, suitable for obesity and disease prevention (such as diabetes) (Ozuna et al., 2020).

Additionally, the pH values are often close to 4 with and 22% average humidity. This analysis could serve as a potential tool for authenticity analysis and adulterants identification for agave syrup commercialization (Barajas et al., 2017).

2.5.2. Extraction and promising methods for agave nectar processing

The standardized industrial agave syrup production follows Tequila's procedure, except for the additional fermentation process, distillation and purification steps. Approximately 10% of the agave harvest is used for syrup production. The main operating units are shown in **Figure 5**. The process begins with harvesting a mature 5-7 years old blue agave

plant, which stores high carbohydrate contents in the plant's *piña*, resembles a pineapple after the leaves are removed. High-quality *piña* (weight up to 68 kg) contains about 25-30% w/w sugars. The second stage involves milling and crushing the *piña* until juicy fibres are obtained. Subsequently, the juice is extracted by hot water washing in a diffuser, and the fibres are discarded. A filtration process is later performed to remove solid particle residues from the raw agave juice. The filtered juice is then thermally hydrolyzed by heating at 80 °C for 8-12 h and refiltered; however, the process could last longer (36 – 48 h) in a traditional technique that uses brick-wall kilns. After reducing the water content in the second filtration, the juice is vacuum evaporated at 90 °C for glycosidic activities denaturation, resulting in the final syrup product (Maldonado-Guevara et al., 2018).

Figure 5. Major processing operations involved in agave nectar production.

Over the last decade, many modern and improved production processes have been introduced to cater to demand growth. In agave syrup, enzymatic treatments, which involve hydrolysis and extraction steps, would enhance fructans hydrolysis efficiency, reduce energy waste and simplify the syrup production process. However, enzymatic hydrolysis could only be achieved through microbial enzymes (Barajas et al., 2017). In theory, inulinases, which catalyze the $\beta(2\rightarrow1)$ fructan hydrolysis, can be found in bacterial, yeast, and filamentous fungal strains; thus, significant variability is expected within their physicochemical characteristics. Since the process involves a high temperature, thermostable enzymes are needed. Exo- and endo-inulinase from

Aspergillus niger with maximal activity at pH 4.3 is suitable for large-scale production. The enzymes are commercially used and display a negligible influence on the flavour and aroma properties of the final product (Ilgin et al., 2020).

Agave syrup also possesses prebiotic effects by stimulating colon bacteria growth, making it suitable for developing nutraceutical products. Other benefits may involve enteric infection protection and immune response stimulation (Catry et al., 2018). Therefore, many current investigations are focused on studying new techniques and protocols to extract and characterize the fructans. Ávila-Fernández et al. (2011) developed a partially thermal acid hydrolysis technique. They demonstrated a relationship between agave fructans hydrolysis and the number of fructo-oligosaccharides (FOS) in the mixture through a fructose equivalent (FE) parameter. The FE defines the percentage of total sugars converted into reduced sugars and could characterize the novel bifunctional product containing prebiotic FOS molecules and sweetening features in the syrup. The study also documented the possibility to remove the resulting monosaccharides through *P. pastoris* cultures. This technique was concluded to be an economical and efficient method at a laboratory scale to develop potential sugar-free prebiotic FOS products (Cervantes et al., 2020).

2.6. Date syrup

2.6.1. Physicochemical and bioactive properties

Date syrup, identified as a dark brown substance, is the main derived product from dates, one of the main fruit trees (*Phoenix dactylifera* L.) in the Middle East region. It is mainly



produced by Iran (20%), Egypt (17%), Iraq (15%) and Saudi Arabia (14%) (Hashemi et al., 2018). Since ancient times, dates are deemed essential to human nutrition and food preparation in the desert regions. Favouring the rapid market growth, it is estimated that over 8 million tons of dates were harvested in 2017 (Ben Yahmed et al., 2021). The date fruit's significances rely on its rich carbohydrates' composition (70-80% w/w), dietary fibre (8.7%), amino acids, proteins (1.8%), vitamins, salts and minerals. Its main physicochemical compositions are moisture content of 16% and total sugar of 79.5% (94% corresponds to inverted sugar, including glucose and fructose). Due to the complex non-sugar molecules mix, the syrup presents high viscosity (17 P at 20 °C) and 4.1% of colouring matter. Moreover, the inverted sugar compounds influence the product's acidic components (pH 3.8), contributing to its resistance against microorganisms (Ghnimi et al., 2017).

The most important properties of date syrup are its potential health benefits, which are related to its high nutritional profiles, i.e., high content of unsaturated fatty acids (such as oleic, linoleic, palmitoleic and linolenic acids) and a combination of 15 minerals, including potassium, iron, magnesium and calcium. The syrup also contains fluorine and selenium, which display good teeth protection against decay and stimulate immune function. It also contains at least six vitamins, including B1 thiamine, B2 riboflavin, nicotinic acid, A and C (Ibrahim et al., 2020).

2.6.2. Extraction and promising methods for date syrup processing

Date syrup extraction is considered one of the oldest practices in sweeteners production, and it is naturally produced while the fruit is stored under hot conditions. Industrial production involves six main steps: pretreatment, extraction, filtration, clarification,

purification and concentration (see **Figure 6**). In principle, the pretreatment step consists of soaking, pitting and pulping of the fruit. Once the pulp is obtained, the extraction is done by mixing the pulp with water and heating until the sugars are released. The syrup is then filtered to eliminate the remaining solids, followed by clarification and purification processes before evaporation to concentrate the syrup (Ben Yahmed et al., 2021; Bertuzzi, 2018).

Figure 6. Major processing operations used in date syrup production.

The applied thermal extraction process contributes to its characteristic dark colour. However, a clear syrup has a better acceptance as a flavouring agent in the food industry (Nasabi et al., 2017). Several different clarification techniques have been explored to eliminate this dark colour characteristic, such as cation/anion exchangers (Nasabi et al., 2017), membrane processes (Zhang et al., 2021), ion exchange adsorption (Ahdno & Jafarizadeh-Malmiri, 2017). Traditionally, thermal extraction usually degrades specific nutritional components and darkens the product, resulting in the need for an additional clarification step and thus increases the production cost. Other new improvement techniques have also been studied, and the sonication method is the most promising. Sonication disrupts cell tissues and organic matter, improving the extraction process by enabling a more efficient solvent penetration into the cells and thus allowing a greater release of the targeted molecules. Another sonication advantage is microorganism inactivation, caused by the disruption of microorganisms' membrane cells (Hamza et al.,



2021). For instance, the ultrasonication extraction at high-intensity ultrasonic waves (ca. 20 kHz) and low temperature (ca. 15 °C) resulted in the highest extraction efficiency at a shortened extraction time while significantly reduced the total microbial counts. These features play an essential role in producing the highest quality final product (desired colour and nutritional profile preservation) and requiring fewer operation units, consequently reducing the production time and cost.

2.7. Stevia (steviol glycosides)

2.7.1. Physicochemical and bioactive properties

Rebaudiana has gained scientific and industrial attention for broad applications as a natural sweetener in commercial food products for its nutritional value and a potential sucrose alternative (Bursać Kovačević, Maras, et al., 2018). *Stevia Rebaudiana* is a perennial herb plant native to South America, notable for its intensely sweet taste and potential pharmaceutical and medicinal applications (Lemus-Mondaca et al., 2015). Diterpene glycosides compounds, specifically stevioside and rebaudioside (shown in **Figure 7**), are responsible for Stevia's sweetness. *Stevia*

Figure 7. Chemical structures of a) *Stevioside* and b) *Rebaudioside A* contained in Stevia plants.

Among the 230 species of *stevia* plants, *Stevia Rebaudiana* and *Stevia phlebophylla* present a sweet taste, contributes by a high concentration of Steviol glycosides (SGs), representing ~4-20% of the dry *stevia* leaves weight (Bursać Kovačević, Maras, et al., 2018). SGs display 40 to 450 times stronger sweetness intensity than sucrose. The SGs amount in the plant varies depending on the climatic, environmental and growth conditions (Aghighi Shahverdi et al., 2018). As previously mentioned, sucrose consumption is controversial due to health concerns related to long-term weight gain, inducing metabolic syndrome, and in the case of saccharin, glucose intolerance and dysbiosis (Yoneda et al., 2017). Therefore, SGs' sweetness intensity, low-calorie content, cardiotonic, anti-cancer and anti-inflammatory properties increase *stevia*'s commercial value in the food market (Mathur et al., 2017). Today, SGs extraction methodologies must be revisited to improve these biologically active compound extraction yields (Bursać Kovačević, Barba, et al., 2018).

2.7.2. *Extraction and promising methods for steviol glycosides processing*

Conventional SGs extraction methods include solvent, Soxhlet, heating-under-flux and cold extraction. These traditional techniques present several drawbacks, such as time-consuming, lack of thermoregulation and utilize high amounts of organic solvents (e.g., methanol, ethanol, acetone, chloroform and even petroleum ether) (Castro-Muñoz et al., 2020; Jentzer et al., 2015). Consequently, other potential SGs extraction techniques have been studied (as reported in **Table 2**).



Table 2. SGs extraction yields from *Stevia Rebaudiana* using different methods.

Adapted from (Castro-Muñoz et al., 2020).

The highest extraction yield was achieved using an ultrasonication extraction of *Rebaudioside A*, producing a 35% yield (35g SGs in 100g) (Gasmalla et al., 2017). The extraction required 60% v/v isopropyl alcohol as the solvent and only used 360 Watts for the 12 minutes process. Nonetheless, the volatile organic solvent is a critical concern due to its high volatility, inflammability, toxicity and represents a negative impact on health, safety, environmental and production costs (Yoneda et al., 2017). Solvent-free SGs extraction methods such as pressure-driven membrane processes (MF, UF and NF) are successfully proven to facilitate the extraction of various high-value-added components, such as antioxidants, sugars, carbohydrates, pectin, phenolic compounds. Membrane processes rely on transmembrane pressure as the driving force for selective separation, while the membrane serves as a physical barrier that selectively permeates the targeted compounds (Castro-Muñoz et al., 2016). The classification of pressure-driven membrane processes is based on membrane pore sizes, as specified in **Table 3**.

Table 3. Classification of pressure-driven membrane processes based on the membrane pore size and their application in sweetener extraction.

Membrane processes constitute a practical option for recovering valuable molecules and present numerous advantages, including no harmful solvent requirement. Water may be

used as the main solvent in several membrane processes, directly lowers the environmental impact and simplifies the process conditions (Vian et al., 2017).

To date, different authors have evaluated the pressure-driven membrane performances in SGs extraction from *Stevia Rebaudiana* dried leaves (Castro-Muñoz et al., 2020). All the tested membranes have yielded >90% recovery efficiency, where the highest yield (ca. 91 %) was obtained at 6 bar for 90 min using MF membranes with a pore size of 0.2 µm (Castro-Muñoz et al., 2020). Integrated membrane processes also acquired the highest recovery, as demonstrated by Díaz-Montes et al. (2020) in purifying *rebaudioside A* (Reb-A) from *Stevia rebaudiana* aqueous extracts using a two-step UF process. Each UF membrane has a different molecular weight (100 and 1 kDa, respectively). The results revealed that the 100 kDa membrane removed most of the total solids and carbohydrates (ca. 42% and 41%, respectively). Meanwhile, the 1 kDa membrane was able to retain ~98% of phenolic compounds. The authors concluded that the two-step UF system effectively extracted up to 38 mg Reb-A per 22 g dry leaves. In addition, the process exhibited a 93% Reb-A recovery efficiency from 49 mg presented in the crude extract. Accordingly, it demonstrated that the membrane process could surpass the standard SGs extraction yields and purity.

2.8. Sorghum syrup

2.8.1. Physicochemical and bioactive properties

Sorghum bicolour is among the most widely produced cereals globally, with wheat, rice, maize and barley (de Morais Cardoso et al., 2017) and used to produce syrup for



beverage and food industries. The physicochemical properties of sorghum syrup are comparable to that of sugarcane syrup since both are extracted from the same *Poaceae* species family (Asikin et al., 2018). This cereal is an essential diet in semi-arid regions, such as Africa and some parts of Asia. Phenolic compounds isolated from the plant displayed therapeutic effects in preventing several conditions, such as cancer, obesity and cardiovascular diseases (de Morais Cardoso et al., 2017). Moreover, sorghum consists of other bioactive compounds, such as carotenoids, proteins and vitamins, extracted from specific parts of the grain and summarized in **Table 4** (de Morais Cardoso et al., 2017).

Table 4. Bioactive compounds contained in *Sorghum bicolor* at the specific anatomic structure (de Morais Cardoso et al., 2017).

2.8.2. *Extraction and promising methods for sorghum syrup processing*

Sorghum syrup is produced by boiling the sweet juice extracted from the Sorghum plant stalks. The conventional steps involved in its production are presented in **Figure 8**:

Figure 8. The conventional process for Sorghum syrup production (Ratnavathi & Chavan, 2016).

Sorghum canes, composed of 70% water and 10-15% fibre, are milled with a three-roller power mill, where the natural juice is extracted. This juice represents ~50-60% of the stalk's weight (Ratnavathi & Chavan, 2016). The juice is then concentrated in continuous flow evaporators until the temperature reaches the boiling point (~100 °C) and heavy-density syrup is obtained. Additionally, this step may also be performed with batch evaporators. One important note is that the evaporators are preferably be built of stainless steel or copper for an efficient heat transfer. Finally, it is cooled, and the quality of standardized syrup is monitored in this stage. Herein, enzymes such as isomerase are added to prevent crystallization during storage and must be added at a temperature below 65 °C to avoid denaturalization (Asikin et al., 2018; Ratnavathi & Chavan, 2016). **Table 5** reports the standard physicochemical properties of the resulting sorghum syrup, which may vary depending on both extraction and concentration steps' operating conditions.

Table 5. Standard physicochemical composition of sorghum syrup.

Sorghum syrup contains 5-hydroxymethylfurfural (5-HMF) and should be present in low concentrations, and generally, a higher 5-HMF content is associated with a darker colour, lower density and lower viscosity. During storage, 5-HMF concentration can be evaluated to monitor the syrup quality. More importantly, 5-HMF can produce a negative health impact due to its genotoxic effects. The 5-HMF concentrations and accumulations are commonly influenced by different factors, including production methods, processing temperatures and prolonged storage periods. To improve the extraction and quality of

sorghum syrup (to reduce the 5-HMF concentration in the final product), several promising juice extraction methods and optimal storage conditions have been studied, including hydrolysis by amylolytic enzymes, active charcoal purification, centrifugation, filtration, sucrose hydrolysis by invertase and chitosan absorption followed by evaporation. These techniques have been reported to reduce 5-HMF accumulations and prevent crystallization during storage (Ospankulova et al., 2020).

3. Current practices of the natural sweeteners in foods

High sugar consumption is regularly related to the prevalence of type II diabetes, obesity, cardiovascular diseases and dental decay (Carocho et al., 2017). More than ever, consumers are more aware and concerned with their sugar intakes and actively seeking for healthier food options, i.e. reduced sugar, sugar of natural origin, and unchanged flavour. Consumer demands have prompted the food industry to make considerable R&D investments to prepare products with natural-based sweeteners (Olivo, 2019). An overview of current applications is presented in **Figure 9**. This section addresses the progress in commercial food manufacturing using natural sweeteners in various companies.

Figure 9. Current applications of natural sweeteners in commercialized food products.

3.1. Stevia



As mentioned earlier, stevia was one of the earliest sweeteners to gain industrial interest and consumer trust and was considered safe in Japan and Paraguay by 1970. Stevia was recognized in 1994 as a dietary supplement in the USA, but not until 2008 when the FDA granted the GRAS designation to stevia extract (specifically the Reb-A). This was reached in response to the request made by *Cargill Inc.*, teamed with *Coca-Cola Co.* and *Whole Earth Sweetener Co.*, a unit company of *PepsiCo* (Chesterton & Yang, 2016). *SweetLeaf® Stevia sweetener* became the first recognized table sweetener with a safe status by 2008 (SweetLeaf Stevia Sweetener, <https://www.sweetleaf.com>, Access date: 6 April 2021). Since then, various beverages and food formulations contain stevia, including ice cream, bakery products, and spices (Carocho et al., 2017).

Cargill Inc. was one of the pioneering companies to launch stevia-based sweeteners, called *Truvia® Natural Sweetener*, introduced in 2008 as a calorie-free stevia leaf extract (Huber, 2017). The success of the natural sweetener came to such an extent that *Coca-Cola* introduced *Sprite Green®* a year later (Fusaro, 2015). Certainly, *Truvia®*, the sweetener developed by *Cargill Inc.* in collaboration with *Coca-Cola Co.*, was included in the product formulation (Beaufort, 2015). In 2014, *Pepsico* and *Coca-Cola* released competing stevia-based sodas, where *Pepsico* launched *Pepsi True®*, claiming a 30% reduced sugar content. In comparison, *Coca-Cola* launched *Coca-Cola Life®* that contained both stevia and sugarcane with 35% fewer calories than other carbonated drinks in the market (Ahmad et al., 2020). Additionally, to expand its natural line, *Hansen's Beverage Company* announced in 2011 a partnership with *Truvia®* to introduce *Natural Fruit Sticks* and *Tea Stix™*, which were marketed as low-calorie and sugar-free beverages, respectively (Newswire, 2020a).

824 *YoCrunch 100*[®] was another product launched with *Truvia*[®], which was part of the
825 reduced-calorie yoghurts *YoBand*[®] line. The original preparation contained ~200 calories
826 per serving but was not considered a healthy snack by consumers. Hence, *YoCrunch*[®]
827 introduced erythritol as a low-calorie sweetener and gained low customer approval. When
828 *Truvia*[®] substituted erythritol in January 2010, the product regained public acceptance.

829 The brand *Freshens* also worked with *Truvia*[®] to develop sugar-free and sugar-reduced
830 products. At this point, *Freshens*, as the biggest frozen yoghurt company in the U.S.,
831 created a new yoghurt-based formula with *Truvia*[®], presenting a lower caloric content
832 and no added sugar (Newswire, 2020b). Furthermore, ice-creams with stevia has been
833 suggested for diabetic consumers. Rodríguez et al. (2016) formulated ice cream with 10%
834 sorbitol and 0.48% stevia, obtaining the recommended sweetness level for diabetic
835 patients.

836 *Ingredion Incorporated* is yet another company that distributes stevia as a table
837 sweetener and has a product line with a specific Reb-A content directed to different food
838 industry applications. *Ingredion's Enliten*[®] Reb-A sweeteners are marketed as natural-
839 based and non-transgenic sweeteners. Additionally, *Health CO.* has released *Stevia*
840 *FSE*[™], enzymatically processed to eliminate its characteristic sourness (Olivo, 2019).
841 Another company associated with stevia as a low-calorie sweetener is HIP gastroplex,
842 which developed *SUGARLESSe*[™]. This commercial product contains a debittered stevia
843 and other plant-extracted sweeteners, often used in chocolates and marshmallows. Its
844 contributions to the final products are consistency, colour, and sweetness (Bosshardt,
845 2020).

3.2. Honey

Honey, for a long time, was the only sweetener available for human consumption. Its application was not restricted to dietary purposes but also promoted in various applications, including drug in-home treatment for infections and burns. Compared to the other natural sweeteners, honey is self-preservative when it is not diluted. Besides being a natural carbohydrate source, it contains vitamins, proteins, amino acids, and polyphenols with high overall nutritional value (Muhammad & Sarbon, 2021; Vara et al., 2019).

The consumer familiarity with honey has positioned it as an ideal option for consumers who expect minimally processed products. While most natural sweeteners (stevia, cane sugar, agave nectar, maple syrup, etc.) need to be further processed (extraction and purification) to obtain their sweet components, honey is not typically processed after its initial collection (NHB, 2021). The National Honey Board encourages food manufacturers to use honey in their formulations to meet customer demands for a clean label and sustainable ingredients. Current honey applications in food products include bakery and snacks, cereals, beverages, and brewing.

Honey is popularly regarded as a natural energy booster due to its high caloric content. This feature is attractive to cereal and bar manufacturers, where honey serves not only as a sweetness and taste enhancer but also for its functionality (Vara et al., 2019). Its viscosity characteristic acts as a binder that helps maintaining the cereal mixture compactness in cereal bars. Despite the promising attributes provided to cereal and bar products, sugar is still the most used sweetener in the new products. By 2016, 78% of the



cereal and bars released in the market were formulated with sugar, while only 21% contained honey (NHB, 2021).

Several recently released snack bar products include *Oats'n Honey* granola bars from *Nature Valley*TM, part of the *Crunchy bars* line. *Nature Valley*TM philosophy is directed by the use of natural products without artificial sweeteners or high fructose syrup; thus, honey is marketed as the preferred ingredient. However, honey is combined with common sugar in other *Nature Valley* products, such as *Oats'n Dark* chocolate and *Coconut crunchy granola bars* (Nature Valley, <https://www.naturevalley.com/all-products/>, Access date: 1 October 2020). *Kind LLC* has a strong reputation for honey-sweetened snacks and has introduced cereal bars with 65% honey content (Nature Valley, 2021). *Honey Oat Breakfast Bars*, *Oats and Honey with Toasted Coconut* and *Oats and Honey* snack bars stand out among their products (Kind Snacks, 2021).

The cereal industry is yet another sector that has widely featured honey in new formulations. Over the last decade, criticism about the elevated sugar content in cereals, particularly those directed to children, raised health concerns associated with their overconsumption. In response, big cereal companies, like *Kellogg's*[®], have been actively highlighting honey usages in their products as a strategy to regain customer acceptance (Askew, 2015; Garcia et al., 2020). Examples of honey-sweetened cereals include *Honey Monster* by *Halo foods* and *Honey Bunches of Oats*[®], a part of the Post Consumer Brands products introduced in 1989 and a top seller in the US. More recently, *Honey Bunches of Oats* released a new version of its traditional recipe: *Honey Bunches of Oats*[®] *Frosted cereal* with frosted flavours and granola clusters (PR Newswire, 2020a).

Interestingly, the brewing industry also uses honey in the fermentation process for centuries. There is evidence of fermented beverages prepared with wild grapes, hawthorn, rice and honey dated back to 7000 years ago in Northern China; however, honey brewing lost its popularity when wine was introduced during the rise of the Roman Empire (Cabras & Higgins, 2016). In recent years, brewing with honey has recaptured commercial interest with increased consumer demands for craft beer. Honey in brewing presents several advantages to producers, including a 90-98% fermentability and an increased polyphenols level relevant to the beer taste and improves its overall antioxidant activity. Among the latest products, barley-malted and honey-enriched *Salmon Fly Honey Rye*[®] was launched in 2014 by *Madison River Brewing Company, Inc.* The company's website states that honey is used to confer a mild sweet taste that serves to attenuate the flavours of rye and hop (Madison River Brewing Industry, 2014). Moreover, *Genesee Brewing Company*, the Dundee Honey Brown Lager trademark owner, has produced beers with natural honey for over 20 years (Genesee Brewing Company, 2021). Another company, *Apex Predator Brewing*, also recently released *Honey Oat Blonde Ale*[®] in Canada, brewed with local honey and marketed as a clean product (Trade et al., 2019).

3.3. Molasses

Molasses have been traditionally related to a bitter taste and low sweetness, although molasses are most often used in home cooking (in sauces and bakeries) (Bosshardt, 2020). The fundamental research in this field aims to produce lighter versions that display a subtle honey taste whilst the manufacturers focus on wider colour varieties and consistencies for different food applications. Unlike other sweeteners, blackstrap

molasses show a lower glycemic index and contain many antioxidant compounds. It was reported that the molasses exhibited a higher polyphenol content (ca. 17 mg gallic acid equivalent (GAE)/g) compared to the other natural sweeteners. It also presents a broader variety of nutraceutical compounds (Deseo et al., 2020; Valle et al., 2020) and various minerals (Senthilkumar et al., 2016).

Molasses can be used as sweeteners in dairy products, like yoghurts (Noureldin et al., 2020). In 2006, *Danone S.A.* announced a partnership with the Bangladeshi bank Grameen to create a new yoghurt formulation that was nutritious and affordable for low-income households. Since the initial product was not sweet enough for the consumers, the company decided to use locally produced date molasses (Peerally et al., 2019).

As a carbon source, molasses are also used for industrial-scale fermentation, such as in the *Saccharomyces cerevisiae* yeast fermentation for bread and food-grade ethanol production (Wu et al., 2020). Molasses represent a suitable fermentation substrate since they are cost-effective and constitute a good energy source for yeasts, mold and bacteria (UM Group, 2021). The *Lesaffre Group*, specialising in yeast, ethanol, and beverage production through fermentation, uses sugar cane molasses in their yeast propagation (Lesaffre, 2021). Moreover, the fermentation of molasses obtained from sugar cane bagasse, has been effectively conducted via *Aspergillus niger* (Bakhiet & Al-Mokhtar, 2015).

Apart from the food, other molasses application includes the foundry industry (manufacturing of casting molds) (Pribulova et al., 2016). Cheap molasses has effective binding properties that allow them to trap fine dust at a large manufacturing scale. Another

936 advantage is that the molasses are non-pollutant, which means they do not produce toxic
937 emissions during combustion and deemed safer.

939 3.4. *Date sugar*

940 Date fruit has a high nutritional value and is a good mineral source. It has an elevated
941 antioxidant content, a low glycemic index, and potentially a suitable sweetener for diabetic
942 individuals (Farahnaky et al., 2016). Date sugar market was valued at \$553.6 million in
943 2018 and is expected to reach \$850.3 million by 2027. Despite being minimally
944 processed, more cost-effective options have similar qualities, such as coconut sugar, thus
945 limiting the date sugar market growth (PR Newswire, 2020b).

946 Although date sugar contains high fibres, vitamins and minerals, it has only 50-70% sugar
947 compounds that directly influence some of the desired physical characteristics (e.g.,
948 solubility), limiting its applicability in the bakery industry (Kumar et al., 2020). On the other
949 hand, date syrup is considered a good sugar substitute in ice cream and yoghurts since
950 it helps maintain good organoleptic properties. A 10% date syrup in yoghurt exhibited
951 higher scores for the sensory evaluation (taste, sweetness, flavour and acceptability).
952 Additionally, date-syrup-enriched yoghurt showed a higher overall antioxidant activity,
953 hydrochloric acid-soluble minerals and folate concentration. Meanwhile, date-syrup-
954 sweetened bakery products have exhibited “good” and “high” in acceptability rating and
955 a “high” for texture (Kumar et al., 2020).

957 3.5. *Agave nectar*

Agave nectar has been recognized as a low-glycemic alternative sweetener, a slow-release carbohydrate with a high prebiotic capacity (Ozuna et al., 2020). Agave also has a high fructose content (~90%) and lower glucose levels, which may be a disadvantage since long-term fructose consumption might lead to insulin resistance and the risk of developing type 2 diabetes (Gardner, 2017). Even so, there have been arguments that the agave composition is safe for diabetic patients (Carranza et al., 2015). For example, Hooshmand et al. (2014) monitored the blood glucose levels, insulin and weight gain of sucrose-fed and agave-nectar-fed mice. They observed a slower weight gain, a decrease in insulin and glucose levels in agave-nectar-fed mice and suggested the agave syrup as a sucrose substitute. The reduced glucose level causes a higher glucagon-like peptide-1 (GLP-1) level, which induces insulin secretion from the pancreas and is responsible for appetite suppression.

Agave nectar is a promising sugar substitute due to its minimal bitterness that does not increase with concentration (McCain et al., 2018). Moreover, agavins or agave fructans (a natural form of fructose with a decreased caloric content) may also be used to replace fat. Cookies prepared with *Agave angustifolia*'s fructans exhibited high water-absorption and water-holding capacities, resulting in an increased water volume and thus expected to induce rapid satiety. Furthermore, fructan addition to the formulation increased the total sugar content, aided in the moisture preservation and produced a better crust colour (Santiago-García et al., 2017).

Previously, agave syrup and native agave dietary fibre were proposed to substitute honey and wheat flour in granola bars. The optimum granola bar formulation, containing a ratio of 3:7 (agave fructans to agave fibre), showed a balance of soluble and insoluble fractions

and exhibited a 72% glycemic index, classified as a moderate level. Meanwhile, agave syrup helped reduce the sugar content in the final product while maintaining its organoleptic properties preferred by the consumers. Since agave products are generated surplus by-products from the tequila industry, their usages in the food industry improved its benefits (Zamora-Gasga et al., 2014).

It is worth mentioning that the non-caloric sweeteners (e.g., stevia) display an increased sweetening power, but their contribution to the overall product consistency is deficient. This is contrary to the synthetic sweeteners that enhance the texture and structure of a pastry but generally lack flavour. A 75% substitution of sucrose with agave syrup, combined with xanthan gum and leavening agents, produced a lower viscosity and increased the thermosetting temperature of batters in muffins. However, higher levels of sucrose substitution with agave syrup produced a darker crust and a paler crumb (Ozuna et al., 2020).

3.6. *Maple Syrup*

Maple syrup is yet another natural sugar suggested as a healthier sweetener substitute. Sucrose is the main carbohydrate, representing ~97% of its composition. It exhibits a high lignans content, including lariciresinol, secoisolariciresinol and phlorizin, which are not found in brown rice syrup, agave syrup, corn syrup, or honey. Maple syrup also presents high phytohormone abscisic acid content, responsible for antidiabetic activity (Mellado-Mojica et al., 2016). Additionally, maple syrup consumption can lower glycemic and insulinemic responses as a result of α -glucosidase activity inhibition, which limits glucose



absorption in the intestine (Mora & Dando, 2021). A lower glycemic response is desirable at the pre-diabetic stages to prevent the transition to an irreversible stage. Besides being sold as a condiment for bakery products, maple syrup is also used to formulate various beverages and snacks. For instance, *King's Row Coffee*, the producer of *Maple water cold brew*[®], markets maple syrup as an antioxidant-rich ingredient while conferring a different taste and aroma in coffees (O'Reilly, 2019). Maple syrup also has been used in craft soda by the *Soda Folk company*, which formulates *Root Beer Soda* with maple syrup, vanilla, water, cane sugar and wintergreen (Morton, 2015). Alternatively, maple syrup is directly sold as a concoction prepared with ginger and sea salt. This product is branded as a natural carbohydrate source with additional vitamins, minerals and amino acids, aimed at consumers who practice sports (Slopeside Syrup Unlapped, 2020). Maple syrup is also included in confectionery products, e.g., *Bixby & Co.* produces *Maine Maple vanilla Bonbons*[®], recognized as the best-selling product in the category (Bixby Co., 2021).

3.7. Other natural sweeteners

3.7.1. Monk fruit

Monk fruit is a high-intensity natural sweetener that originated in China and Indonesia. The commercial extract is obtained from *Siraita gosvernori* species, cultivated in the Chinese province of Guangxi, and it accounts for ~90% of the global production. Traditionally, monk fruit has been used as a natural sweetener and medicinal product for pharyngitis treatment (Świąder et al., 2019). Currently, it is commercially available as table sweeteners. *S. gosvernori* extract is primarily sold in combination with *S. rebaudiana*

and erythritol as a sweetening product (Soejarto et al., 2019). Monk fruit has a GRAS status in the US; however, due to the lack of evaluation in food products, it cannot be used as a food ingredient in the EU (Świąder et al., 2019).

The monk fruit's sweetness is contributed by a group of terpene glycoside compounds called mogrosides. Among the five mogrosides types in monk fruit, mogroside V is found in higher concentrations with ~256-378 times more sweetness intensity than the common sugar (Świąder et al., 2019). Mogrosides from *S. gosvernori* also possess bioactive properties according to the anti-diabetic and anti-cancer effects (Liu et al., 2018; Zhou et al., 2018). Specifically, mogroside V is responsible for the apoptosis and cell cycle arrest of pancreatic tumour cells (Liu et al., 2017) and is associated with free radical scavenging activity (Pandey & Chauhan, 2019). Mogrosides also induce a hypoglycemic response by increasing insulin secretion, preventing lipid peroxidation, and reducing α -glucosidase activity (Gong et al., 2020). Furthermore, mogrosides positively impact blood glucose levels by increasing postprandial insulin levels (Liu et al., 2017). Besides mogrosides, the fresh fruit contains other bioactive compounds, including rutin, kaempferol and quercetin, which account for its antioxidant, antimicrobial and anti-inflammatory properties (Świąder et al., 2019).

Monk fruit is used in a few products, such as sugar, syrup, jam, chocolate and skimmed milk (Pandey & Chauhan, 2019). Mahato et al. (2021) employed a combination of stevia and monk fruit extract to reduce the sugar content in chocolate-flavoured milk. The authors concluded that the combination of 56.27 ppm stevia and 81.90 ppm monk fruit extract exhibited optimum sensory parameters for the overall liking, appearance, aroma, sweetness, mouthfeel and aftertaste. Similarly, sugar-free chocolates have been

developed using monk fruit and fibre extract blends, which has resulted in chocolates with lower fat and complex carbohydrates levels. This new formulation could also be an alternative for people with diabetes (Pandey & Chauhan, 2019).

3.7.2. Yacon syrup

Yacon (*Smallanthus sonchifolius*) is a perennial plant native to the Andean region in South America. The tubers from this plant can be processed into juice (or syrup) and consumed as a sugar substitute. They are mainly composed of fructooligosaccharides (FOSs) and inulin, constituting ~60% of their dry mass (Kamp et al., 2019). FOSs are used as low-calorie sweeteners since human digestive enzymes do not hydrolyze them; thus, they are not metabolized in the gastrointestinal tract (Yan et al., 2019). The FOSs consumption exhibits prebiotic activity by stimulating bifidobacteria and *Lactobacillus spp.* growth in the colon, where these bacteria degrade FOS into short-chain fatty acids (Caetano et al., 2016). At the same time, the inulin has a significant industrial value as a texture modifier in yoghurt, cheese and milk drinks (Yan et al., 2019). The addition of Yacon syrup in yoghurt formulations presented the highest scores for sensory acceptability, appearance, and texture compared to natural yoghurt and yoghurt supplemented with cashew apple extract (Mendes et al., 2019).

The characterization of Yacon syrup has revealed the presence of chlorogenic acid, a phenolic and bioactive compound, known for its therapeutic effects, antioxidant, antibacterial, anti-inflammatory and hepatoprotective activities (Naveed et al., 2018). da Silva et al. (2017) reported a 1202.25 µg GAE/g total polyphenol and a 175.13 µg/g of chlorogenic acid content in Yacon syrup, while its total antioxidant activity was estimated to be ~6.99 µM Trolox/g. Importantly, phenolic compounds in Yacon syrup may be helpful

in chronic disease prevention, including cardiovascular and some cancer types (Yan et al., 2019). Furthermore, Yacon syrup has been recommended as an alternative natural sweetener for diabetic patients. Its extracts have demonstrated inhibitory activity against α -amylase and α -glucosidase, obstructing glucose absorption and, consequently, decreasing postprandial hyperglycemia (Russo et al., 2015). Similarly, Adriano et al. (2020) reported that 40g Yacon syrup consumption after breakfast decreased postprandial glucose and insulin blood levels in adult women.

Its physicochemical properties show water activity of 0.78, pH of 3.71 and 71° Brix. These features directly impact the product shelf-life by preventing the proliferation of undesirable microorganisms (da Silva et al., 2017).

3.7.3. Palm sugar

Palm sugar, a popular natural alternative in Asian countries (Le et al., 2020), is the sap obtained from the flowers of different palm species, including palmyra palm (*Borassus flabellifer*), nipa palm (*Nypa fruticans Wurmb*) and sugar palm (*Arenga pinnata*) (Saputro et al., 2019). Palm sugar has been used in various products, such as sweet soy sauce, beverages and desserts (Saputro et al., 2019). Le et al. (2020) recently studied palmyra palm granulated sugar's physicochemical properties and chemical composition. They revealed an overall A_w value between 0.30-0.48, which is optimum for extended storage times, with a pH value of 6.90. Although the main components of the evaluated samples were ca. 91% sugar and ca. 5.6% reduced sugars, various minerals (potassium, sodium and iron) were also detected. In addition, vitamins E, C and D were found in significant concentrations. Interestingly, palm sugar registered a total phenolic content ranging from

2.77 to 8.94 mg per 100g, depending on the heat treatment used during the sap processing.

In general, palm sap sugars present lower glycemic indexes (Saputro et al., 2019). According to Srikaeo et al. (2019), the predicted glycemic values for cane sugar (~91) are considerably higher than those registered for palm sugars (~70). Furthermore, granulated palm sugar produced from *B. flabellifer* exhibited cytoprotective activity against NIH3T3 fibroblast cells. Le et al. (2020) showed that cells incubated with *tert*-butyl hydroperoxide with granulated palm sugar result in increased cell proliferation compared with those incubated with only *tert*-butyl hydroperoxide (an agent used to induce oxidative stress).

4. Concluding remarks and future trends

Throughout this review, recent uses of natural sweeteners in the food industry were discussed, including current practices in food manufacturing. Their main extraction routes, as well as the key purification techniques, were analyzed. Although the reviewed natural sweeteners may present several health-related advantages, major drawbacks are observed in their extraction processes that directly influence their final physicochemical characteristics. Several emerging techniques to improve the overall production yield and quality are discussed. Promising methods like vacuum drying, spray drying, and pressure-driven membrane processes will likely preserve their natural nutritional properties effectively. Other techniques, including clarification, enzymatic hydrolysis and sonication, boost the development further by significantly decreasing the production costs while satisfying colour, nutritional profile and sterilization parameters. These techniques also reduced the treatment duration, and it is more imperative as these properties are affected

considerably by thermal processes. Moreover, processing parameters and storage conditions are essential to ensure the high quality and purity of the final products.

Their applications in food production highly rely on the organoleptic properties, structure and texture, and the added value they confer to different products. As the food industry explores more sweetening alternatives to meet consumer demands, large-scale applications have been expanded to bakery products, beverages, dairy products (mainly yoghurt) and even fermentation processes. More importantly, the current market trends signify the need for healthier and minimally processed foods. This represents an opportunity for the food companies, and natural sweeteners may play a vital role in the markets. Carob syrup, palm sugar and monk fruit are the rising alternatives that could be used in the near future. Although their current applications in the food industry are still limited, several attempts to include them in food formulations, such as in yoghurt, skim milk and chocolate products, revealed that these sweeteners enhance the final products' organoleptic properties positively. Further studies concerning the cultivation, extraction and safety of these minimally explored sweeteners are needed to widen their commercial use.

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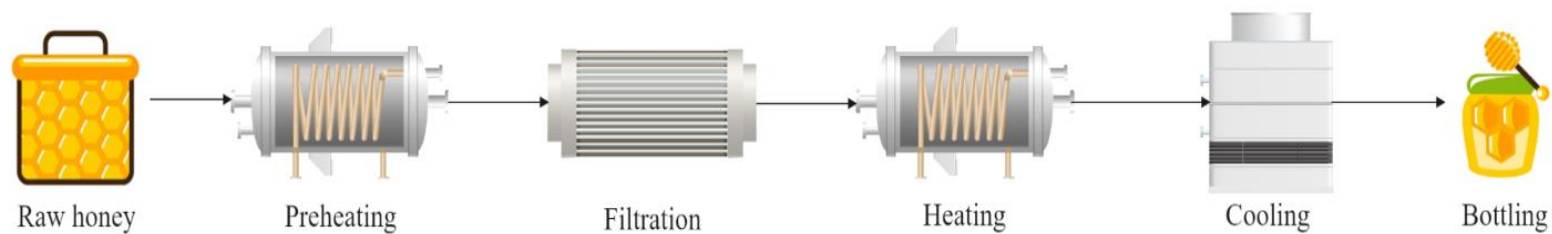
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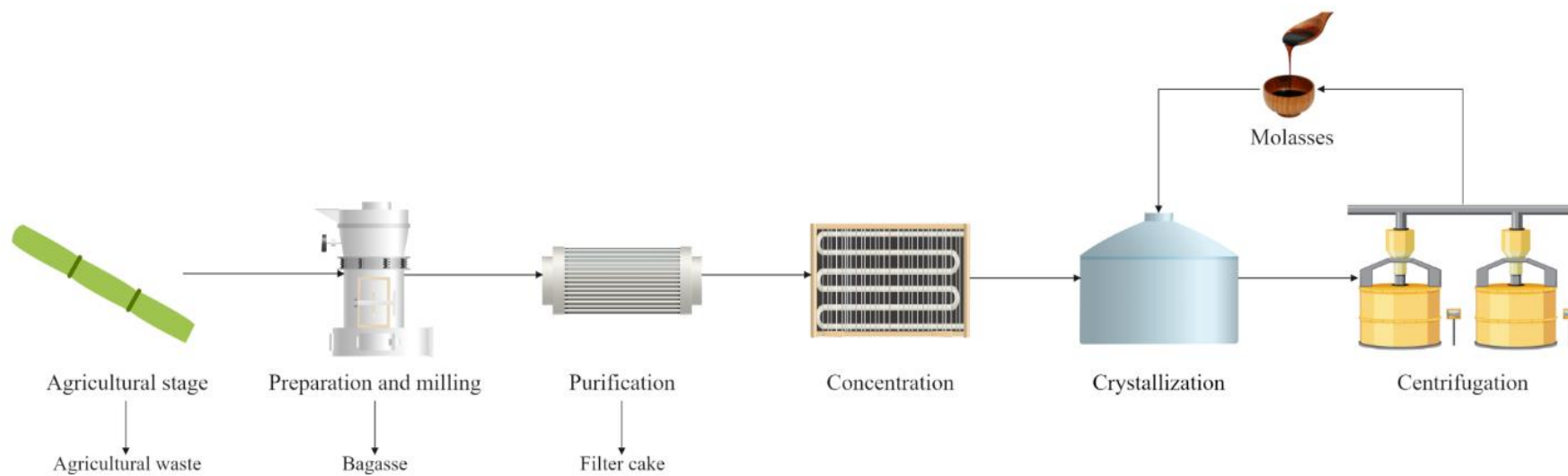
Figure 1. Traditional honey production process.

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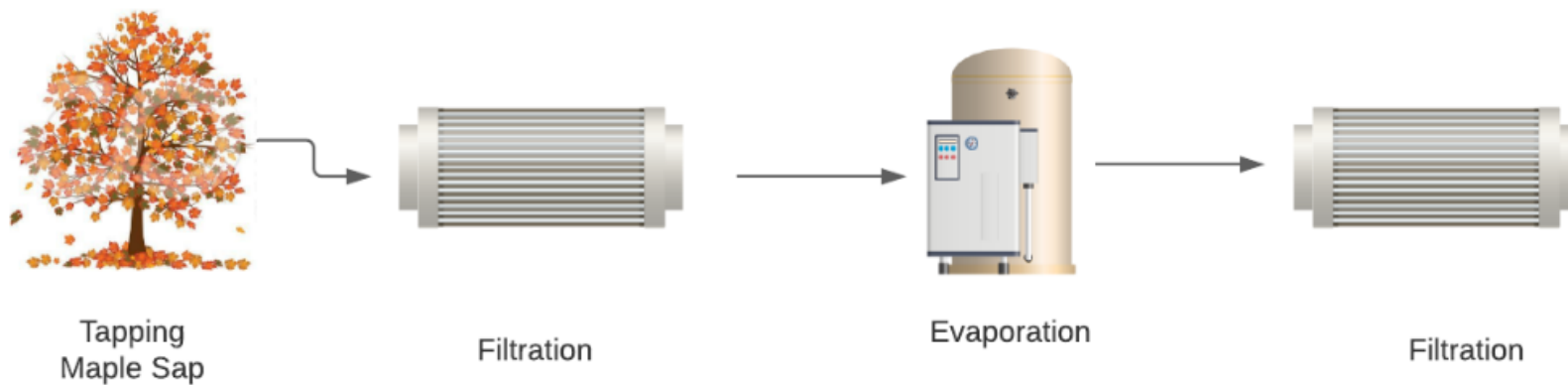
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Figure 2. Sugar production process and its derived blackstrap molasses.

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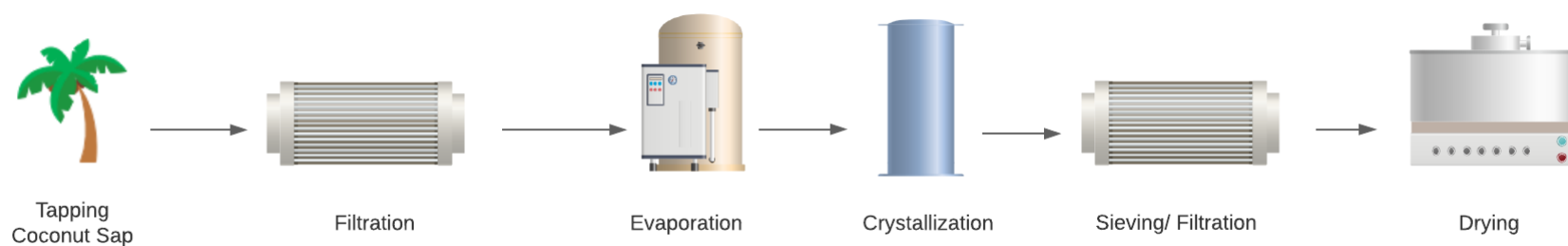


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Figure 3. Schematic representation of the traditional maple syrup production.

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Figure 4. Schematic representation of the traditional coconut sugar production.

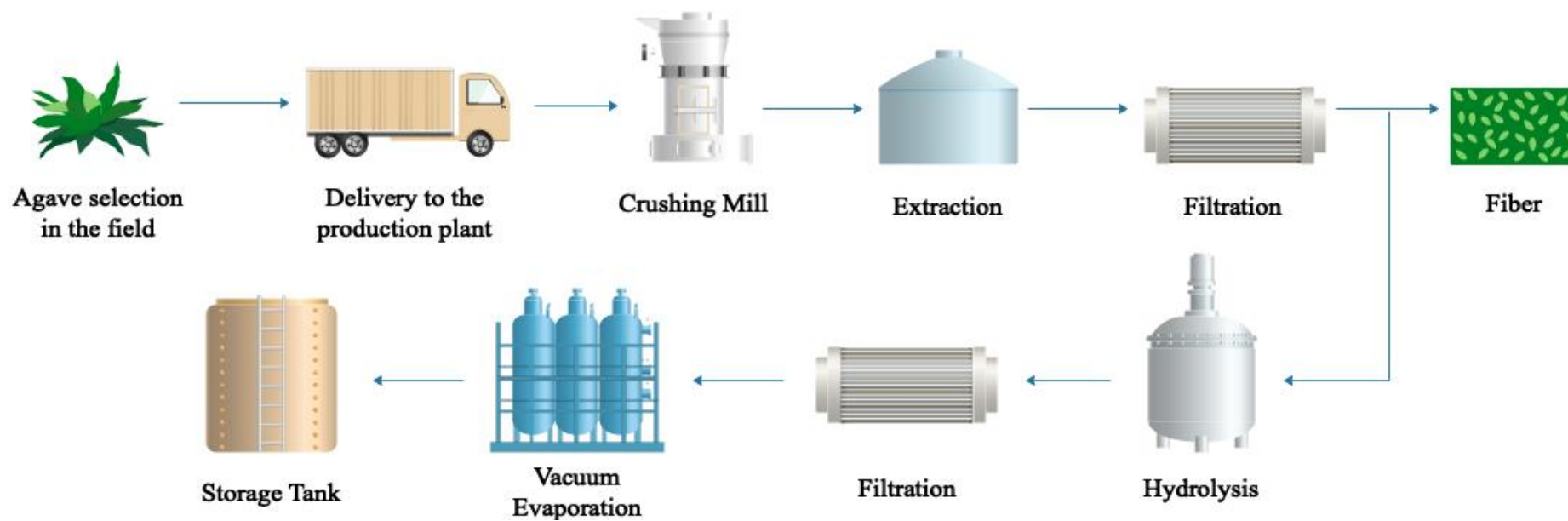
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1713 **Figure 5.** Major processing operations involved in agave nectar production.

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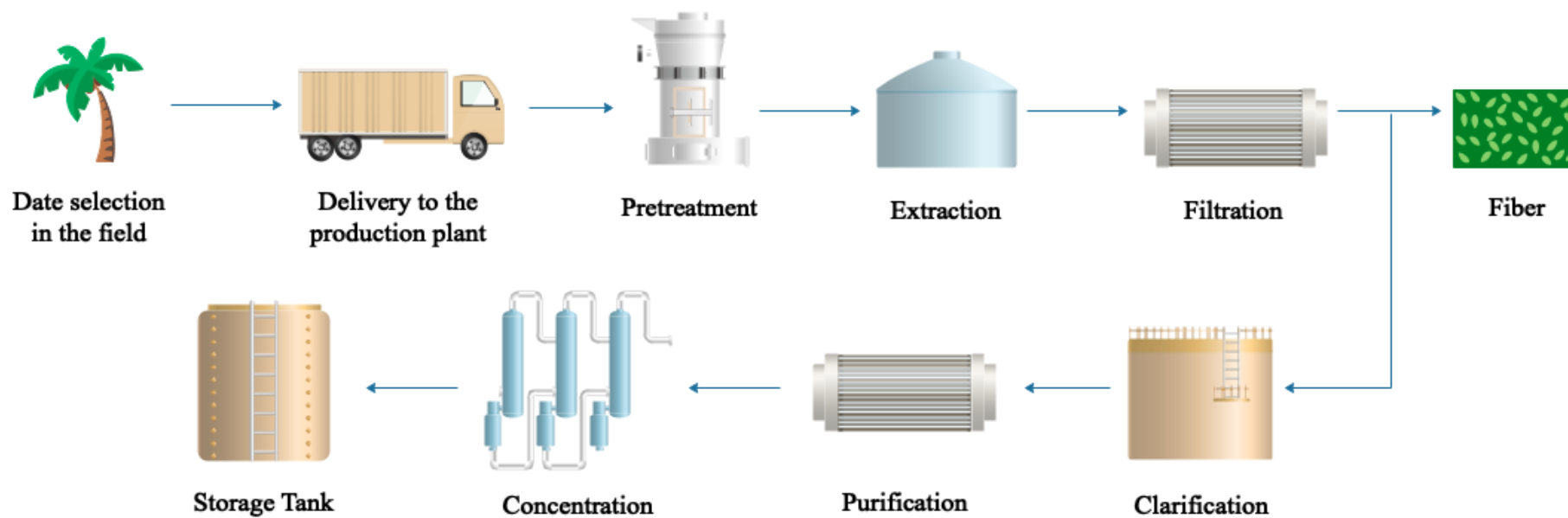
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1720 **Figure 6.** Major processing operations used in date syrup production.

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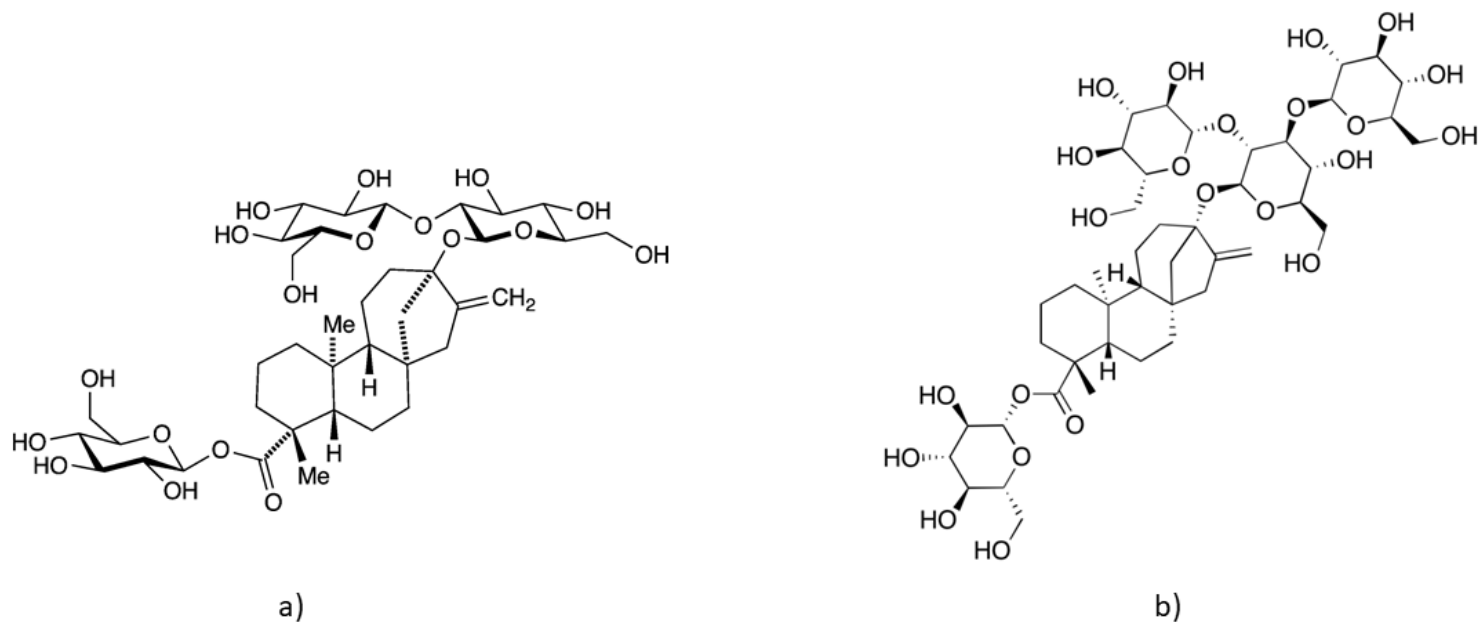
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Figure 7. Chemical structures of a) *Stevioside* and b) *Rebaudioside A* contained in *Stevia* plants.



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Figure 8. Conventional process for Sorghum syrup production (Ratnavathi & Chavan, 2016).

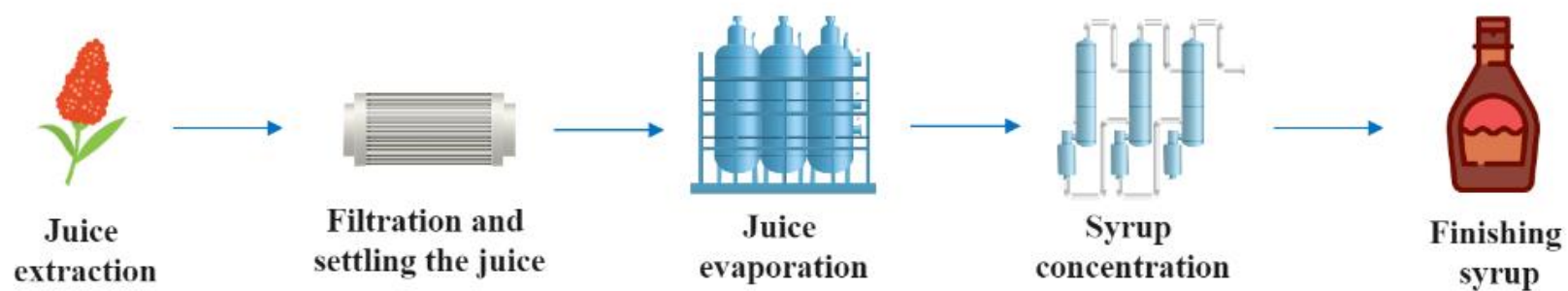


Figure 9. Current applications of natural sweeteners in commercialized food products.

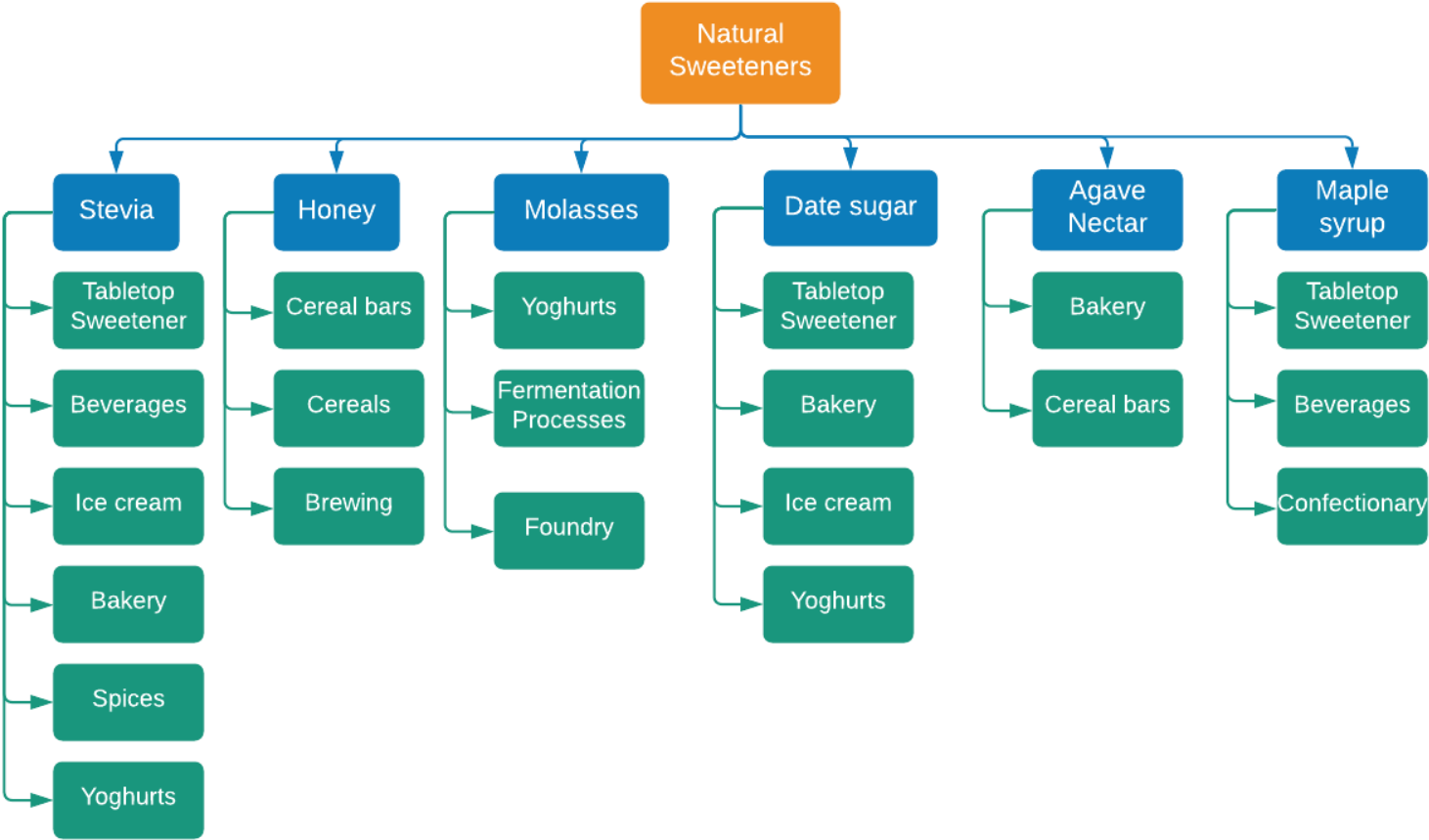


Table 1. Alternative techniques used to concentrate the coconut sugar.

Method	Temperature	Time	Energy
Rotary Evaporation (RE) vacuum	54.8°C	12.2 min	0.35 kWh
Microwave Evaporation (ME)	103.2°C	13 min	0.10kWh
Open-heat Evaporation (OHE)	101.6°C	46.8 min	0.83 kWh

Table 2. SGs extraction yields from *Stevia Rebaudiana* using different methods.

Adapted from (Castro-Muñoz et al., 2020).

Method	SGs	Extraction yield (%)
Accelerated solvent extraction	Stevioside	7.5-9.9
	Rebaudioside A	2.1-3.2
Assisted by ultrasonication extraction	Rebaudioside A	35
Centrifugal partition chromatography	Stevioside	4.2-24.0
	Rebaudioside A	17.2-22
	Dulcoside	6.8-10
Column-chromatographic technique	Steviolmonoside	4.7
	Stevioside	10.7
	Rebaudioside B	7.3
High Voltage electrical discharges	Stevioside	4.5
	Rebaudioside A	2.0
Pulsed electric fields	Stevioside	27.0
	Rebaudioside A	18.0
	Rebaudioside C	6.5
Microwave-assisted extraction	Stevioside	8.6
	Rebaudioside A	2.3
	Stevioside	2.0
	Rebaudioside A	1.5
	Stevioside	4.5
	Rebaudioside A	2.3
Pulsed electric fields	Stevioside	3.7
	Rebaudioside A	2.1
Pressurized fluid extraction	Stevioside	4.7
Rapid solid liquid dynamic extraction	Stevioside	0.5
	Rebaudioside A	1.4
Supercritical fluid extraction	Stevioside	3.7

Ultrasound-assisted extraction	Rebaudioside A	1.8
	Stevioside	4.2
	Rebaudioside A	2.0
	Stevioside	5.1-9.7
	Rebaudioside A	0.4-3.7
	Stevioside	5
	Rebaudioside A	2.2

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Table 3. Classification of pressure-driven membrane processes based on the membrane pore size and their application in sweetener extraction.

Membrane process	Pore size range	Type of targeted components	Pressure requirement (TMP)	Applications for sweetener extraction
Microfiltration (MF)	100-10,000 nm	Macromolecular components (Fungi, yeast, bacteria)	0.1-2 bar	Turbidity reduction of remelt syrup (Li et al., 2016). Pre-clarification of sugarcane juice (Panigrahi et al., 2018).
		High MW compounds (Proteins, virus, polysaccharides, colloids)	0.1-7 bar	Partial removal of sugars in honey and enhancement of spray drying process (Samborska et al., 2018) Decoloration of sugarcane molasses (Guo et al., 2018) Decolorization and turbidity removal of date syrup (Fathi et al., 2013) Separation of <i>Rebaudioside A</i> from aqueous extracts of <i>Stevia rebaudiana</i> (Díaz-Montes et al., 2020). Isolation of glycosides from <i>Stevia</i> (Martínez-Alvarado et al., 2017).
Nanofiltration (NF)	0.5-2 nm	Low MW compounds (Polyphenols, ions, pigments and bioactive compounds)	3- 25 bar	Concentration of Maple sap (Ali et al., 2021) Preclarification of sugarcane juice (Panigrahi et al., 2018). Sucrose purification from Molasses (Sjölin et al., 2019)

Table 4. Bioactive compounds contained in *Sorghum bicolor* at specific anatomic structure (de Morais Cardoso et al., 2017).

Anatomic structure	Compounds
Pericarp	Non-starch polysaccharides, phenolic compounds, carotenoids
Endosperm	Starch, proteins, B-complex vitamins, minerals
Germ	Lipids, fat-soluble vitamins, B-complex vitamins, minerals

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Table 5. Standard physicochemical composition of sorghum syrup.

Compound	Content (mg/100g)
Total sugar content	68, 570
Glucose	10,880
Fructose	5,560
Organic Acids	3,179.67
Minerals	4,607.87
Aconitic acid	2,312.87
Malic Acid	468.09
Citric acid	378.41
Succinic acid	378.41

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