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SWEET NUTRACEUTICALS IN PLANTS

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Abstract: The paper discusses the natural substances of plant origin – nutraceuticals – characterized by varying intensity of sweet taste and classified into different chemical compounds such as glycosides, terpenoids, proteins, acids, or aldehydes. Among them, it discusses the main characteristics and origin of the following: steviosides, mogrosides, osladin, glycyrrhizin, thaumatin, mirakulin, monelin, mabinlin, pentadin, brazzein, curculin, sweet acid, A, cynarine, and perillaldehyde. It presents the characteristics of products containing these sweet nutraceuticals, such as stevia (*stevia rebaudiana*), *Siraitia grosvenorii* (Swingle) C. Jeffrey (also known as *Luo Han Guo*), polypody ordinary (*Polypodium vulgare*), Licorice (*Glycyrrhiza glabra*), *Thaumatococcus daniellii* Benth, *Richardella dulcifica*, *Dioscoreophyllum cumminsii*, *Capparis masaikai*, *Pentadiplandra brazzeana*, *Curculigo latifolia*, Scots pine (*Pinus sylvestris*), artichoke (*Cynara scolymus*), and color perilla (*Perilla nankinensis*).

Key words: plants, nutraceuticals, natural substances, intensive sweeteners.

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Introduction

The term "nutraceuticals" has been known for over 25 years and was initially defined as food additives of specific dietary significance, later as bioactive constituents isolated from various plant and animal resources, and more recently – as constituents that are naturally present in food which is labelled as "functional food". In any case, nutraceuticals are perceived as having a beneficial influence on health, resulting from: a) the direct influence on certain functions of the organism, b) the enabling of the modification of content or the ways of producing food, due to which the product acquires specific nutritional properties and, as a result, can be recommended in special nutritional cases. This group features food products used in order to reduce body mass, labelled as low calorie. Products from this group with a sweet taste profile require the use of a sugar replacement, which is eliminated from their content. Instead of synthetic sweetening constituents, an alternative solution could be in the form of plants that include sweet nutraceuticals, which can be used in a minimally processed form (dried, extracts) or as clean derivatives – isolated, sweet substances.

Fig. 1: The leaves of Stevia rebaudiana [2].

Plants as sources of sweet glycosides

The leaves of *Stevia rebaudiana* – Bertoni – a perennial shrub from the *Astraceae* family (Fig. 1), sometimes called the "sweet herb of Paraguay", originating in Brazil and Paraguay [1].

In South America, the plant is known under the names: Caa-che, Azuca-caa and Cua-eh, meaning "a sweet plant". The sweet substances are found in the leaves which feature sweet diterpene glycosides. *Stevia* leaves are gathered twice a year. Out of a single hectare of the plants, about 1200 kg of dried leaves can be collected, which yield from 60 to 70 kg of steviosides [3].

In Brazil, Korea and Japan, stevia leaves, stevioside and the highly refined extracts are officially used as a low-calorie sweetener. In the USA, powdered stevia leaves and extracts of this plant have been used as a dietary supplement since 1995. In 2000, the European Commission rejected the proposal to classify stevia or stevioside as a new food product (Novel Food) due to the lack of sufficient, critical scientific data regarding the possible toxic effects of stevioside, particularly its main aglycone – steviol. In summer 2009, French health authorities issued permission to use a sweetener acquired from the stevia plant in the version containing 97% of rebaudioside A.

The fruit of Siraitia grosvenorii (Luo Han Guo). *Siraitia grosvenorii* (Swingle) C. Jeffrey belongs to the family of *Cucurbitaceae*. It is a perennial climbing vine. Its leaves are in the shape of a heart and are 10-20 cm long. The fruit is round, 5-7 cm in diameter. The fruit's smooth surface is yellow-brown or green-brown and delicately fuzzy (Fig. 2).

Fig. 2: The plant and fresh fruit of *Siraitia grosvenorii* [2].

This is one of four species of *Siraitia*. Botanic synonyms include *Momordica grosvenorii* and *Thladiantha grosvenorii*. The flower and fruit develop in early spring. The plant can effectively produce fruit for 8 years. Ripe Lu Han fruit is collected in late autumn. The fruit (Fig. 3) is one of several kinds that were believed to be plants of longevity. They feature fleshy, sweet fruit pulp and numerous seeds [4].

Fig. 3: The dried fruit of *Siraitia grosvenorii* [2].

Siraitia grosvenorii Swingle (also known as **Luo Han Guo**) is a traditional plant cultivated mainly in the autonomous region of Guangxi in China (mostly in the area of Guilin mountains). For hundreds of years, the fruit has been used in China as a natural sweetener in the form of dried fruit used in the preparation of soups and hot drinks. The fruit of *Siraitia grosvenorii* is rich in mogrosides, which constitute about 1% of the content of the pulp. This is a group of triterpene glycosides. Five mogrosides were acquired from the plant and labelled from 1 to 5, but the main constituent of *S. grosvenorii* is mogroside-5 called esgoside [4, 5].

The common polypody (*Polypodium vulgare*) (Fig. 4) – is a source of sweet substances included in the rhizomes of the plant. This is a perennial plant that grows in Europe, Asia and America along with mosses in shaded forests and on rocks [6]. It achieves the height of 45 cm and can also be found in Poland.

Fig. 4: The common polypody (*Polypodium vulgare*) [2].

The sweet taste of the common polypody results from the bis-glycoside present in the concentration of about 0.03%. Bis-glycoside is considered a new type of steroid saponin called osladin. It tastes similar to stevioside or glycyrrhizin. Osladin is about 3000 times sweeter than saccharose, but due to its very low concentration in the rhizome of the plant, its use as a sweetener is very limited.

Citrus fruits are known as plants that are sources of bitter substances, such as 7-flavanone glycosides. They are characterised by high concentrations, high availability and the ease of extraction on an industrial scale. Horowitz and Gentili [7] concluded that citrus flavonoids become sweet after catalytic hydrogenation. This fact contributed to the development of a method of synthesis of dihydrochalcone analogues which was achieved by semi-synthetic means from the naringenin acquired from the peel of very bitter grapefruits. The hydrolysis of naringenin is carried out with the use of potassium carbonate, whereas hydrogenation is conducted with a palladium catalyst set in active carbon. They have a sweet taste accompanied by the sensation of freshness, similar to menthol. Their characteristic quality is a sweet taste which lasts much longer than in the case of perfect saccharose. Dihydrochalcones are 2.5 to

7 times sweeter than saccharose and 25 to 40 times sweeter than sodium cyclamate. The factor that limits the use of dihydrochalcones as sweeteners is their low solubility in water (from 0.8 to 3.6 g/l in 25°C) [8, 9].

Naringenin dihydrochalcone is accepted in the USA for the production of chewing gum, toothpaste and mouthwash. It is also recommended as an intense and low-calorie sweetening agent for dietetic products, such as snacks and alcohol-free drinks – in the amounts between 20 to 50 ppm [2, 10].

Plants as sources of sweet terpenoids

The root of liquorice, *Glycyrrhiza Glabra*. Liquorice belongs to the family of *Fabaceae*. It is a perennial plant, characterised by a thick, branching root with numerous stolons which can be even several metres long. The stalk is smooth, widely branching, and it reaches the height of about 1.5 m. Along the stalk, liquorice leaves appear in an alternating arrangement. They are egg-shaped with pointy tips. One branching leaf consists of 7-9 smaller leaves. From the angles of the leaves, clustered, purple inflorescences grow (Fig. 5a).

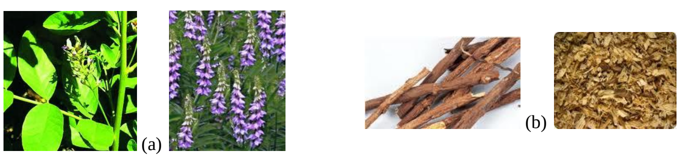

Fig. 5: The plant (a) and roots (b) of liquorice, *Glycyrrhiza Glabra* [2].

The fruit is a red and brown pod which includes 3-5 seeds [11]. The plant is cultivated mainly for medicinal treatment, but it also grows wildly in Asia Minor, in the region of the Mediterranean Sea and in southern Russia. For the production of sweet glycosides, manufacturers use the root – *Radix Glycyrrhizae*, also called *Radix Liquiritiae* (Fig. 5b).

Liquorice root has a sweet taste, which makes liquorice a frequent sweetening ingredient of fruit teas. The main active constituent of the root of liquorice is glycyrrhizic acid (glycyrrhizin) – a triterpenoid, which is responsible for liquorice root's characteristic sweet taste and aroma, as well as its beneficial properties. Apart from glycyrrhizic and glycyrrhetinic acids, the root of liquorice also includes a lot of other active compounds, which together constitute about 40-50 % of the roots mass. These include numerous flavonoids (liquiritin, isoliquiritin, liquiritigenin, licoflavonol), isoflavones (glabridin, glabren, hispaglabridin A and B, formononetin), saponins, polysaccharides, phytosterols, pectins, ethereal oil, free amino-acids and minerals. These compounds support the functioning of the main constituent – glycyrrhizic acid, creating a complex substance that is biologically more active than pure, isolated glycyrrhizic acid or glycyrrhetinic acid [8, 12].

Plants that include sweet proteins

Thaumatococcus daniellii Benth fruit (Fig. 6) is a source of thaumatin, a substance of protein character. The *Thaumatococcus daniellii* plant grows in the humid and hot areas of West Africa's tropical forests. It is easy to find in the southern parts of Ghana, Ivory Coast and Nigeria. It is also known in Angola, the Central African Republic, Uganda and Indonesia. Also known as "katamfe", it is a single-leaf perennial plant.

Fig. 6: The fruit of *Thaumatococcus danielli* [2].

The intense sweetness and desired properties of *T. danielli* fruit drew the attention of the botanist William Daniell in 1855. The fruit weighs from 6 to 30 g, depending on whether it has 2 or 3 seeds. It is harvested between April and June. Each seed is covered by a dense polysaccharide gel and a soft white shell, which includes thaumatin, a sweet protein. It has been determined that thaumatin proteins are synthesised in the plants as a response to fungal infections [9, 13].

One kilogramme of fruit yields 0.9 g of the protein with a sweetness that is 1600-2500 stronger than that of saccharose (0.9 g of thaumatin equals 15 g of saccharose). By using low concentrations of aluminium salt, the efficiency of the protein is increased to the point of 6 g out of 1 kg of dry fruit. Furthermore, aluminium salts improve the colour, durability, permeability, purity and sweetness of the extract. Water solutions that are used in processes of acquiring thaumatin include aluminium sulphate and sodium disulphate and are considered substances that prevent protein browning. Aluminium is the reason for many human illnesses and thaumatin is used in the production of food in low concentrations, but the amount of metals added to food is insignificant in terms of pathogenicity [14].

The fruit of the exotic *Richardella dulcifica* (previously known as *Synsepalum dulcificum danielli*) (Fig. 7 a-c) is a source of sweet protein called **miraculin**.

Fig. 7: The plant (leaves) and fruit of Richardella dulcifica (a) – the fruit, (b) – the leaves, (c) – the fruit with a seed (cross-section) [2].

Richardella dulcifica belongs to the family *Sapotaceae*. It is a perennial plant that grows in West Africa. *Richardella dulcifica* is not resistant to frost, and it flourishes in the hot, semi-shaded and humid areas of African plains, which are its natural environment [15]. The evergreen tree or bush reaches even up to 5 m in height in its natural habitat, but under farming conditions it is rarely higher than 1.5 – 2 m. It can have a smooth or fuzzy surface. The plant has a single seed (Photo 7a, c). Miraculin is present in the thin layer of pulp which surrounds the seed. The plant gives fruit after reaching about 30 cm of height, so when its 2 – 3 years old.

The miracle fruit is commercially cultivated in Thailand, Taiwan, Ghana and the United States (Florida). The existence of the sweet fruit was first documented by the European explorer Chevalier des Marchais during a trip to African countries in 1725. He observed that the locals chewed berries from this bush before eating their meals and added them to food to improve the taste of sour dishes made from corn and sour drinks. The fruit was called agbayun, taami, asaa and ledidi. In Anglo-Saxon countries, the berry of *Richardella dulcifica* is called "the miracle fruit". Out of 10 g of fruit pulp, it is possible to acquire 36 mg of miraculin [15].

The fruit of ***Dioscoreophyllum cumminsii (Stapf) Diels*** is a source of the sweet protein – monellin. The plant belongs to the *Menispermaceae* family. It grows in the dense and humid tropical forests of West and Central Africa [16]. It has long and fuzzy stalks that resemble vines. The leaves consist of three parts that are heart-shaped. They are from 10 to 20 cm long (Fig. 8). Tiny fruit with a seed form in the inflorescence. Around the spiky seeds, there is a layer of gel mass of intensely sweet taste. The substance is of protein character and is called monellin.

The fruit is harvested from September to October. Currently, 3 – 6 kg of pure protein can be acquired from 1 kg of fruit. The fruit of *Dioscoreophyllum cumminsii* is eagerly consumed by the people of Congo. The first description of

Fig. 8: The plant and the fruit of *Dioscoreophylllum cumminsii* (West Africa) [2].

this plant comes from 1895. The inhabitants of Africa call the fruit of *D. cumminsi* wild red berries, Guinea tomatoes or serendipity berries [14].

In the Chinese Yunnan province, one can encounter a plant called ***Capparis masaikai***, which is a source of a sweet substance called mabinlin. *Capparis masaikai* grows in the subtropical region of the Chinese Yunnan province (Fig. 9). The residents of this area often chewed the seeds of this plant to taste its sweetness. Informally, *C. masaikai* is called "*mabinlang*" [13].

Fig. 9: The plant and fruit of *Capparis masaikai Levl (Capparaceae)* [2].

Capparis masaikai Levl. The plant is a part of the *Capparidaceae* family. It is a bush whose vines reach up to 7.5 m in length. Its young extensions are slightly flat, with straight edges and bracts. The leaves are elliptical and elongated in shape, sometimes elliptical and egg-like. The peduncles are short and fuzzy. The inflorescences consist of

3-8 small flowers with a colour range from white to pink. The red-purple fruit is the size of a tennis ball. When ripe, the fruit is dry and elliptical. One piece of fruit consists of at least 10 seeds out of which the sweet protein – mabinlin (mabinlin 1 and 2) – is acquired. The protein predominantly exists in the seed – out of 100 g of seeds, about 1.4 g of mabinlin is acquired [13].

Out of the African plant ***Pentadiplandra brazeana***, two sweet protein substances are extracted – **pentadin** and **brazzein**. *Pentadiplandra brazeana Baillon* is a bush that grows on the outskirts of African forests. It belongs to the *Pentadiplandraceae* family. The plant grows in Angola, the Democratic Republic of Congo, the Central African Republic, the Republic of Congo, Cameroon, Gabon and Nigeria. *Pentadiplandra brazeana* produces round, red berries with about 2 cm in diameter. The berries have up to 5 seeds which are surrounded by a thick layer of pulp (Fig. 10). The pulp features sweet substances called pentadin and brazzein [17].

Due to the pleasant sweetness of *P. brazeana*, the plant was used by breastfeeding mothers to transition their children away from breastmilk because after tasting the fruit, children did not want to return to their mother's milk as it seemed to have no taste. The French that inhabited the northern regions of Gabon nicknamed *Pentadiplandra* as "oubli", meaning "forgetting/oblivion", in connection to the infants' reaction to their mother's milk after tasting the plant's fruit. Out of a single ton of the seeds, about 1 to 2 kg of brazzein is acquired [17].

Fig. 10: The fruit of *Pentadiplandra brazeana* [2].

The amazing ability to transform sour taste into sweet is a quality of the fruit of the plant that grows in West Malaysia – ***Curculigo latifolia***, which is a source of the sweet substance called curculin. The fruit is eagerly consumed by the people of Asia, particularly the inhabitants of the Malay Peninsula, who use it to sweeten sour dishes. *Curculigo latifolia* is a perennial plant (Amaryllidaceae). It features bulbous rhizomes. Its l leaves are typically stubby, long and lanceolate (Fig. 11a).

The inflorescences have a head-like shape. The perianth is yellow and has wide segments which are almost identical in size and sometimes joined to form a tube. The fruit is a berry, which does not spontaneously open in the period when seeds are released. The seeds are small and often striped (Fig. 11 a, b). One seed weighs about 1 g. So far, there have been no reports on the successful attempts at acquiring curculin from transgenic plants [18].

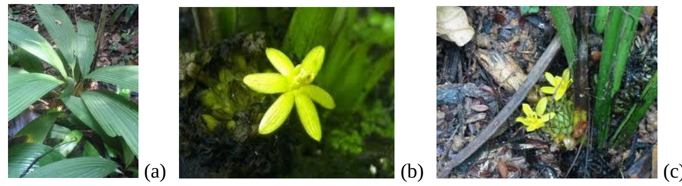

Fig. 11: The plant (leaves) (a), flower and fruit (b, c) of *Curculigo latifolia* [2].

Plants as sources of sweet acids

The Scots Pine (*Pinus slvestris*), which is a type of conifer (Fig. 12), is a resource that is used to acquire a sweet, acidic compound. The sap of this tree was used to isolate 4 stereoisomers of 4-β-10-α-dimethyl-1,2,3,4,5,10-hexahydrofluorene-4,6-dicarboxylic acid, out of which only a single one is sweet – stereoisomer A, while the remaining are tasteless. The free A acid is 1300 to 1800 times sweeter than saccharose, and its sodium salt is between 1600 and 2000 times sweeter than saccharose, but the compounds are also strongly bitter at the same time. There are few studies regarding the toxicity and carcinogenicity of this compound, as well as ones discussing its possible use [14].

The globe artichoke (*Cynara scolymus*) (Fig. 13) is a source of chlorogenic acid and cynarine which, similarly to miraculin, has properties that modify taste. Solutions of various taste profiles (saccharose, citric acid, quinine hydrochloride, sodium chloride) seem to be equally sweet after the preliminary rinsing of the mouth with a globe artichoke extract. The plant has a sweet taste after 4 – 5 minutes from the moment it was consumed [13].

Chlorogenic acid and cynarine can be acquired by extracting the dried leaves of the globe artichoke using alcohol. Cynarine is a didepside of caffeic acid with quinic acid. It is a sweet, colourless and crystalline substance which does not dissolve very well in cold or boiling water, but it is more soluble in ethanol or acetic acid. There have been attempts to use the globe artichoke to derive a sweet taste and to improve the taste of dishes acquired from unconventional sources [14].

The infusions known as sweet tea made from hydrangea – the "ama-cha tea" are made from the **leaves of *Hydrangea aerophylla*** (commonly known under many names, e.g. the bigleaf hydrangea) (the Mountain Hydrangea – Yae-no-Amacha) – include the sweet **phytodulcine**, which is a representative of the natural isocumarins, and is 200-300 times sweeter than saccharose.

Sweet aldehydes

Fig. 12: The Scots Pine (*Cynara scolymus*) [2].

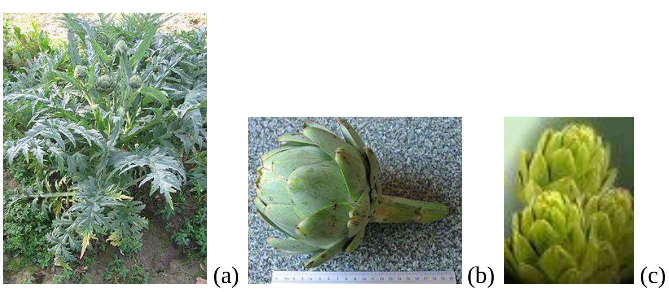

Fig. 13: The globe artichoke (*Cynara scolymus*) (a) – the plant, (b-c) – inflorescence buds [2].

In this group, it is worth mentioning perillaldehyde and perillartine, which are a part of the ethereal oil extracted from ***Perilla nankinensis***. Perillaldehyde is 12 times sweeter than saccharose, whereas the aldoxime of this aldehyde – **perillartin**e (a compound that belongs to the group of natural sweetening compounds that are chemically modified) – is 2000 times sweeter than saccharose. Despite its high degree of sweetness, perillartine is not used due to its significant toxicity. Its use is forbidden, but in Japan, it is added to pipe tobacco to give it a pleasant taste [14]. There are studies that focus on the synthesis of the analogues of perillartine. So far, about 80 new compounds were acquired, out of which the most promising one is the so-called SRJ.V. oxime [(E)-4- methoxymethyl-1,4-cyclohexadiene-1-carboxime], which is about 450 times sweeter than saccharose – a durable substance with a more beneficial taste profile in an acidic environment.

Summary

In the last 30 years, over a dozen substances of sweet taste were isolated from the fruit or seeds of plants of the tropical climate. From the chemical point of view, these include diterpene and triterpene glycosides, as well as peptides and proteins. Steviosides and rebaudiosides are diterpene glycosides acquired from the leaves of the Paraguayan plant known as *Stevia rebaudiana* (*Asteraceae*), and they are about 300 times sweeter than saccharose. The group of triterpene glycosides includes glycyrrhizine, which is isolated from the roots of *Glycyrrhiza gabra* (*Fababeae*). It is 50-100 times sweeter than saccharose. Out of plant resources that are rich in intensely sweetening substances of protein character, it is important to mention the West African fruit of *Thaumatococcus danielli* (*Marabtaceae*), which includes thaumatin (E957) that is about 2500-3000 sweeter than saccharose. This has been the only sweetening substance of natural origin allowed in Poland since 1998. The seeds of the Guinean *Dioscoreophyllum cumminisi* (*Menispermaceae*) include monellin, which is about 300 times sweeter than saccharose, whereas the fruit of the African *Pentadiplandra brazeana* (*Pentadiplandraceae*) includes pentadin and brazzein, which are (respectively) 500 and 500-2000 times sweeter than saccharose. The West African fruit of *Richardella dulcifica* (*Sapotaceae*) is a source of miraculin, which has no taste, but after the acidification has miraculous properties which transform sour taste into sweet taste, and at that point it is about 3000 times sweeter than saccharose. In Malaysia and Vietnam, it is possible to find *Curculigo latifolia* (*Amaryllidaceae*), whose fruit includes curculin, which is 550 times sweeter than saccharose and has the properties to change sour taste into sweet. It is also worth mentioning *Capparis masakai* (*Capparidaceae*), which is a plant that grows in the area of the Chinese Yunnan province. Its fruit includes mabinlin, which is 375 times sweeter than saccharose. In China, there is a plant called *Siraitia grosvenorii* (*Cucurbitaceae*), which is rich in a substance that is 300 times sweeter than saccharose and is called Lu Han.

The discussed natural substances present in plants seem to be an excellent alternative to saccharose and intensely sweetening substances achieved synthetically. The fact that limits their industrial production and the resulting limited use in food is their unfortunately low concentration in plants, which makes production not economically viable due to the need for enormous amounts of resources that would have to be processed. Furthermore, during their growth, the plants require specific climate conditions that are characteristic of very specific regions, which also limits the possibility of their widespread use.

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