



Pharmaceutical and Therapeutic Potential of Miraculin and Miracle Berry

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ABSTRACT

Richadella dulcifica or *Synsepalum ducificum* also known as miracle fruit is a kind of cherry plant, which originates from West Africa. The glycoprotein, miraculin in miracle berry is responsible for its unique function (suppression of sour taste). It acts by binding to the sweet receptor cells of the tongue, thus suppressing the response to sour taste in the central nervous system and this effect can be sustained up to 50 minutes to 4 hours. It could be harnessed as a novel or alternative additive in the food, cosmetic and pharmaceutical industries and could be an abundant source of hydrophilic and lipophilic antioxidant-rich phytochemicals. In miracle berry, more than ten phenolics were identified. Also, contains oxidizable vitamins, essential and non-essential amino acids. Its application is interesting, particularly, it is known to exhibit anti hyperuricaemic activity, improves dysgeusia, and expressed in transgenic plant. This review is meant to enhance the fruits' acceptability and also promotes its utilization in reducing the risk of chronic diseases generally associated with lipid metabolism. This article provides an overview of *Synsepalum ducificum*, commonly known as Miracle berry and its unique taste modifying properties by miraculin, a glycoprotein with particular reference to its history, the proposed mechanism of action, functions, limitations and potential applications. In addition, it focuses on the Indian varieties of miracle fruit and their formulations. The method of extraction of miraculin from the berry in laboratory scale is also discussed with emphasis on the storage condition(s).

Keywords: Miracle fruit, Miraculin, Taste modifier, *Richadella dulcifica*, *hyperuricaemia*.

Introduction

The Miracle berry *Richardella dulcifica* or *Synsepalum dulcificum* also called miracle fruit^{2,3} is indigenous to tropical West Africa and belongs to the family Sapotaceae. The Miracle Fruit plant was introduced by the U.S. Dept. of Agriculture at the Federal Experiment Station in Puerto Rico⁴. The fruits of miracle berry are red in colour where it derives its name red berry and their fruits are collected in the month of December to June. It has a big seed surrounded by a thin layer of berry flesh with a faint cherry-like flavour.^{4,5}

Miracle berry acts as a taste modifier. The glycoprotein miraculin in miracle berry is responsible for this unique function by binding to the sweet receptor cells of the tongue, thus suppressing the response of a sour taste in the central nervous system.⁶ This effect would last until the miraculin is diluted and eliminated by saliva. With the taste modification function, miracle berry has a great potential in food application as an alternative sweetener to mask undesirable sour tastes in food products.⁷⁻¹⁰

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Methodology

The review was systematically conducted by searching the databases of MEDLINE, PubMed, the Web of Knowledge, Google Scholar, and Science Direct for original research articles and books using relevant search terms or their combinations: "Miraculin, miracle berry, miracle fruit, *Richardella dulcifica*, *Gymnema sylvestre*, *Synsepalum dulcificum*, and taste-modifier." Our search was not limited by date but to all relevant publications available in the English language.

Miracle berry phytochemicals

Besides Miracle Fruit's unique potential to make sour food taste sweet, the pigment from the red-coloured skin could be a natural colour food ingredient. Miracle fruit is rich in antioxidants. These phytochemical antioxidants have been confirmed to possess health-promoting functions in preventing various chronic diseases, such as cardiovascular diseases, obesity, diabetes and certain cancers. Miracle berry also contains hydrophilic and lipophilic phytochemicals.^{11,12} Anthocyanin and flavanol pigments in miracle fruits¹³ and leaves¹⁴ are identified as cyanidin-3-monoglucoside, cyanidin-3-monogalactoside, delphinidin-3-monoarabinoside, cyanidin-3-monoarabinoside, and delphinidin-3-monogalactoside using paper chromatography and spectral analysis. Rutin, gallic acid, ferulic acid, quercetin, epicatechin, myricetin, kaempferol, syringic acid are phenolics, whereas also contain leutin, vitamin A, C and E with tocopherols and tocotrienols.¹¹ Miracle berry also contains essential amino acids (leucin, methionine), non-essential amino acid (Glutamic acid, glycine).¹⁴ Miracle berries leaves, and roots are rich in carbohydrate, fibres, tannins, cardiac glycosides, polyphenols, flavonoids and minerals.¹⁵



Figure 1: Miracle berry¹

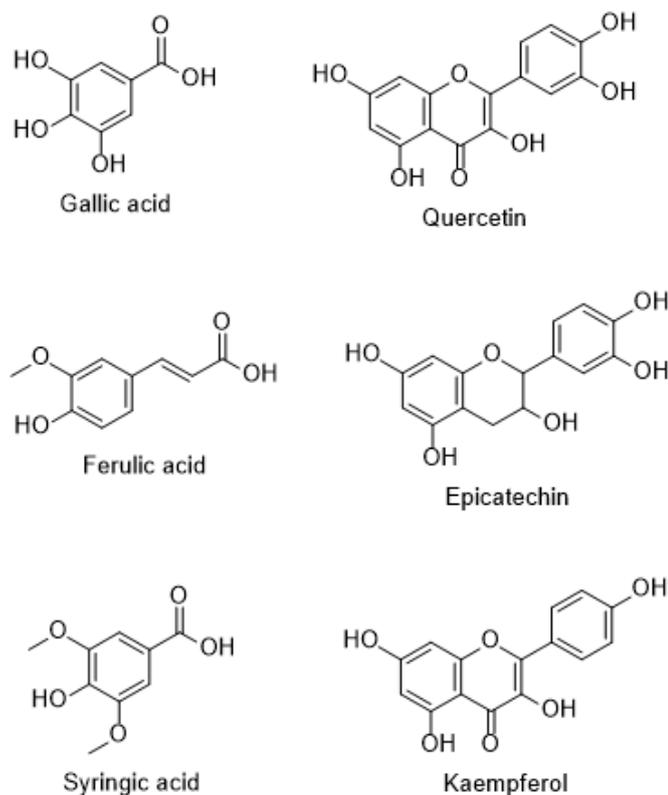


Figure 2: Molecular structures of phenolics present in Miracle berry



Figure 3: Molecular structures of essential amino acids present in Miracle berry

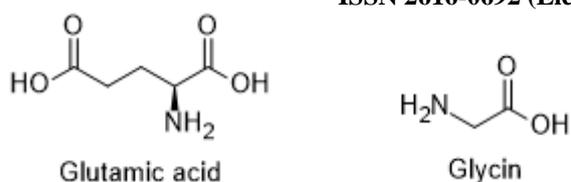


Figure 4: Molecular structures of non-essential amino acids present in Miracle berry

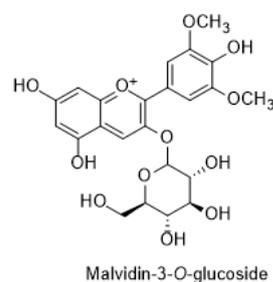
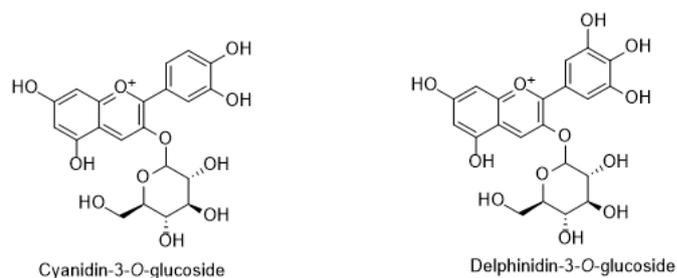


Figure 5: Molecular structures of anthocyanins present in *Synespalum ducificum*

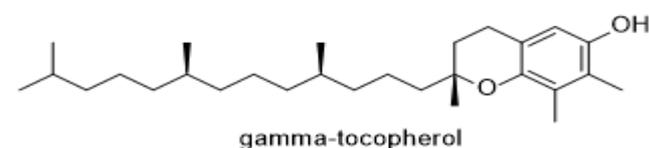
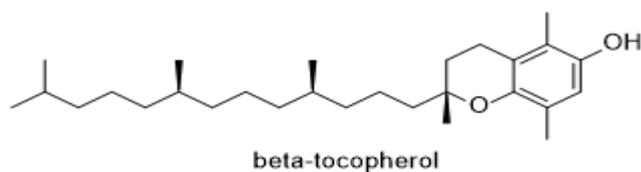
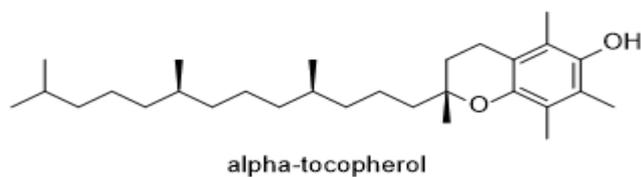
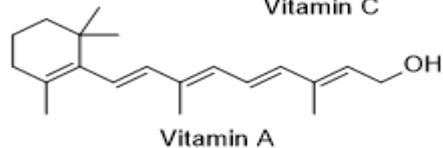
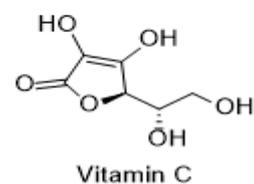


Figure 6: Molecular structures of vitamins present in *Synespalum ducificum*

Health benefits of Miracle berry

The value of the fruit as a taste modifier has possible benefits for human health. Various studies have proven that the fruit and leaf extracts of miracle berry help in reducing the blood glucose level in alloxan-induced albino rats.^{16,17} Shi *et al.*,¹⁸ described that the butanol extracts of miracle fruit may be an effective treatment for acute gouty arthritis.¹⁸ This extract attenuated oxonic acid potassium salt-induced hyperuricaemia in mice by lowering serum uric acid levels and activating hepatic xanthine oxidase. Miracle fruits have tremendous therapeutic properties as shown in Figure 7.

Extraction and purification of miraculin

Inglett *et al.*,⁵ modified the extraction procedure of Adom and Liu,¹⁹ which described two-extraction methods for the pulp and seeds of Miracle Fruit. The first method was Sequential extraction which includes two steps i.e. double extraction for free compounds and Alkaline extraction for bound compounds. The second method was direct extraction method. Before analysis dried pulp-seeds were separated into pulp and seeds after freeze-drying. The skin, pulp and seeds were ground individually to fine powder.^{5,19}

Dioso *et al.*,¹⁷ performed the extraction method of fruits and leaves of this plant. They macerate leaves and fruits of miracle plant with ethanol and then evaporate at room temperature.¹⁷

He, *et al.*,³⁰ modified the extraction method for miraculin from miracle berry as described by Theerasilp and Kurihara.²² In this method, lyophilized pulp powder was suspended with water and homogenized. The homogenate was centrifuged and the sediment was homogenized in 0.5 M sodium chloride solution. Then again centrifuged and the colourless supernatant was freeze-dried.^{22,30}

Miraculin has been purified by using various methods like solvent extraction method from polar to non-polar and from non-polar to polar in succession,⁹ ion-exchange chromatography technique,¹⁰ immobilized-metal affinity chromatography (IMAC),³¹ and reverse micelle system.³⁰ The solvent extraction method is tedious and the purity of miraculin is relatively low. High purity miraculin was obtained by ion-exchange chromatography but this is time-consuming, inefficient and costly.¹⁰ IMAC is a simple and efficient method, but this method is difficult to scale-up beyond laboratory scale and considered to be expensive.³² It has been proved that reverse micelle extraction method is an efficient method to extract miraculin from miracle berry due to its low energy requirements and also shows great potential for large-scale application and use in continuous separation of biological substances.³²

Mechanism of taste modifying activity of miraculin

The flesh of the berry contains aglycoprotein, miraculin, with a molecular weight of about 43,000 D. Miraculin is actually not a sweetener, but has the property of making sour products taste sweet²⁰⁻²³. It is a glycoprotein consisting of 191 amino acid residues with two glycosylated polypeptides i.e. Asn-186 and Asn-42, cross-linked by a disulphide bond.²⁴⁻²⁶ It consists of approx. 14 % sugars, including glucosamine, mannose, fucose, galactose, and xylose. The taste-modifying effect of miraculin begins a few seconds after consumption, though several minutes of chewing the berry's pulp may be necessary to sufficiently coat the taste buds. The duration of the taste-modifying effect typically lasts 30 minutes to 2 hours.²⁷⁻²⁸

Miraculin binds tightly to the lingual epithelium's microvilli plasma membrane of sweet-taste receptors (hT1R2-hT1R3) without activating them. Miraculin does not activate these receptors until subjected to an acidic pH, generally, between pH 3.0 and 6.0.²⁹ The possible mechanism of miraculin in the modification of taste is shown in Figure 9.

Limitations and Preservation of miracle berry

Miracle berry or *Richadella dulcifica* is highly perishable. It is thermolabile and inactivated below pH 3 and above pH 12. Not used in cooking and in processed foods because it degrades at high temperature. These limitations can be overcome by preservation techniques using a polysaccharide chitosan coating.³³ Also, the miracle berry can be freeze-dried at -20° F for approximately 3 months before use without significant degradation.³⁴

The miraculin protein itself very stable and can be stored in pure form or in solution for 6 months without loss of function³⁵ and long-time storage can be done in Ziploc bags.

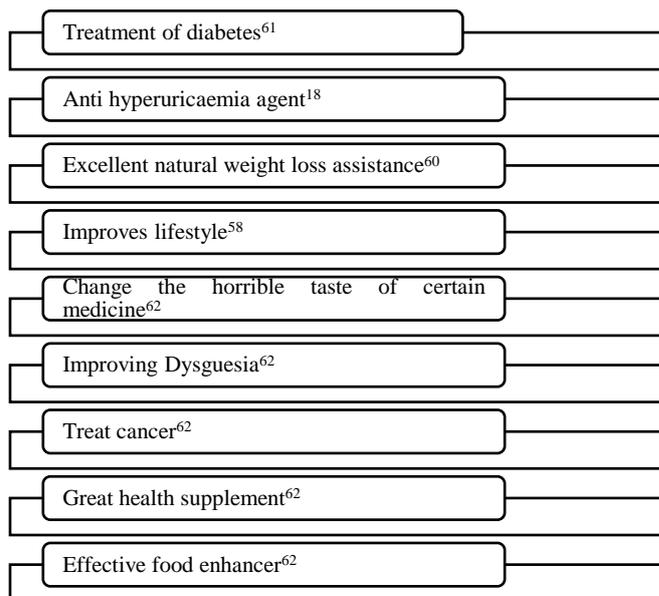


Figure 7: Health benefits of *Synespalum ducificum*

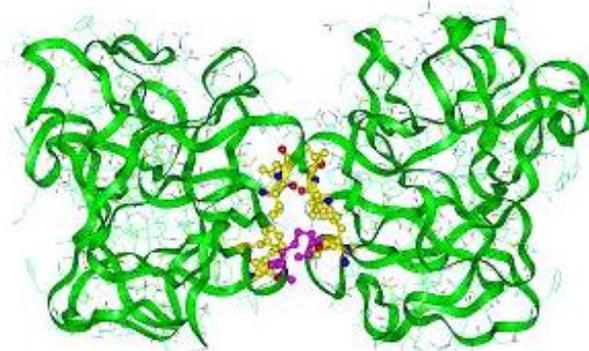


Figure 8: Structure model of Miraculin (Homodimer formed by Disulphide bond)⁶³

1. Regulatory Issue

FDA, the U.S. Department of Agriculture (USDA) does not maintain restrictions on the miracle berry. Growing, selling, and eating miracle berries in the United States is legal.³⁵

2. Miracle fruit: A Promising product

Miracle berry is a promising natural product and miraculin in the fruit enhances its application in the food industry as well as in pharmaceutical industry. In the market, various formulations are available in the form of granules, and tablets.³⁶⁻⁴¹ Various marketed formulations of miracle fruit tablet are in Figure 10.

3. Indian Variety of miracle berry

Indian variety of miracle berry named as *Gymnema sylvestre* with botanical synonyms *Asclepias geminate*, *Periploca sylvestris*, and *Marsdenia sylvestris*. It is native to India and also found in tropical Africa, Australia, Malaysia and Srilanka. It is also known as GUDMAR and Sugar Destroyer. Flowers of *Gymnema sylvestre* are collected in the month of April and November whereas fruits in winter season.⁴²

Gymnema sylvestre, miracle plant having some tremendous activities like anti-diabetic, anti-obesity, anti-inflammatory, anti-microbial, hypolipidemic and anti-arthritic activity⁴⁶. It is also used in the treatment of urinary complaints, stomach problems, piles, chronic cough, breathing troubles, asthma, eye complaints, cardiopathy, constipation, jaundice, and bronchitis. These activities are due to the presence of the bioactive compounds Gymnemic acid, Gymnemenin and Gurmarin.⁴³

Conclusion

Owing to its numerous attributes, miracle berry exploitation has aroused researchers interest in discovering an efficient taste modifier via miraculin. Though miracle berry has interesting and promising uses in the food, pharmaceutical, as well as in cosmetic industry, the fruit wide spread usage is however limited by its availability and perishability. To overcome this limitation, researchers are producing recombinant miraculin in transgenic plants, notably tomatoes. The various phytochemicals in the miracle fruit have been confirmed to possess health promoting functions in reducing various chronic diseases such as diabetes, cancers, acute gouty arthritis, hypertension, cardiovascular diseases and in weight loss. This information enhances the fruits acceptability by more people and thus, generally promotes its utilization and appreciation in human diets.

Conflict of interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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