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Research Article

Red Dragon Fruit Powder as a Basic Ingredient for Functional Foods Rich in Bioactive Compounds, Nutritional Substances and Antioxidants

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Abstract

Objective: The objective of this study was to identify the total bioactive compounds, nutritional value and antioxidant activity in red dragon fruit powder. **Methodology:** Red dragon fruit powder was prepared using fresh red dragon fruit by sorting, bark stripping, cutting and pureeing the fruit into a slurry with a vacuum evaporator. Chemical analysis was performed using a spectrophotometer included total flavonoid, total phenolic acid and total anthocyanin. An approximate analysis was also performed. **Results:** The bioactive contents of red dragon fruit powder were: 210.02 ± 0.83 mg/100 g total flavonoids, 386.09 ± 1.52 mg/100 g total phenolic acid and 81.75 ± 1.43 mg/100 g total anthocyanins. Red dragon fruit powder contained: 11.53% moisture, 4.17% ash, 5.30% crude fiber, 5.68% protein and 0.43% fat. **Conclusion:** Red dragon fruit powder contains highly bioactive compounds (antioxidants) and is a good functional food ingredient with the potential to scavenge 50% of free radicals using 1195.181 ppm red dragon fruit powder. This powder can also be stored for a long time.

Key words: Red dragon fruit powder, flavonoid, phenolic acid, anthocyanin, functional food, antioxidant activity, nutritional substances

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

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INTRODUCTION

Cardiovascular diseases, diabetes, cancer, arthritis and many other diseases may be caused by genetic or age-related factors¹. However, personal lifestyle, including dietary intake and physical activity, along with environmental surroundings, may also contribute to disease risk. Ghiselli *et al.*² mentioned that the free radicals produced by our body induce oxidative stress and are related to several other harmful effects, including coronary heart diseases, neurodegenerative disorders and aging.

Consumption of fresh fruits and vegetables is important to help prevent the action of free radicals³.

For many years, fruits have become an area of investigation due to their bioactive compounds, which are beneficial for human health. The red pitaya fruit (red dragon fruit) is a new promising fruit.

Research shows that ripe red dragon fruit contains numerous total dissolved solids that are rich in organic acids⁴, proteins⁵ and other minerals such as potassium, magnesium, calcium and vitamin C. Various betacyanins and betaxanthins have shown the ability to capture free radicals which in turn contributes to the interest in red dragon fruit consumption as a source of antioxidants⁶. Furthermore, another study mentioned that dragon fruit has strong antiradical properties with other phenolic compounds, but its characteristics have not been reported⁷.

Tenore *et al.*⁸ suggested that red dragon fruit contains phytochemicals that are good for the body, such as polyphenols and flavonoids. Rebecca *et al.*⁹ showed that the total polyphenol content in red dragon fruit flesh is as high as 86.129 ± 17.016 (mg/0.5 g gallic acid), while the total flavonoid content can reach 2.3 ± 0.20 (mg g⁻¹ catechin).

However, the bioactive compound content of red dragon fruit in powder remains unknown. The powder has a long shelf life and a high economic value and could serve as a functional food that can be added to a variety of processed food products. The objective of this study was to identify the total bioactive compounds in red dragon fruit powder.

MATERIALS AND METHODS

Sample preparation: Fruits were obtained from a local Sabisa farm located in Gunung Batu Bogor. The process of powdering the local red dragon fruit involved a vacuum evaporator as follows; the fruit were sorted, washed, drained, halved and peeled manually. The fresh red dragon fruit was then crushed and added to 30% maltodextrin while stirring thoroughly, followed by drying and milling.

Red dragon fruit powder analysis: After processing the red dragon fruit powder, the next step was to conduct proximate analysis, including total ash, fat, protein, carbohydrate and crude fiber, using the AOAC procedure¹⁰.

Total ash content was determined by heating 10 g of dry red dragon fruit powder in a silica dish at 130°C for 24 h in an oven (Memmert 600, Germany). After 24 h, the flour was placed and soaked in the oven (Furnace 5500, Germany) at 550°C for 8 h.

The Kjeldahl method was used to determine total nitrogen using a Tecator Kjeltec System 1002, (Sweden). The percentage of nitrogen is converted into protein by multiplying by a factor of 6.25.

The fat content was determined by directly extracting the dry red dragon fruit powder with petroleum ether via intermittent extraction using a Soxhlet extractor. The residue in the flask after removal of the solvent extraction is the fat content of the sample.

Carbohydrate and crude fiber determinations of the total carbohydrates in the red dragon fruit powder were determined using the AOAC method. Discoloration was determined using a spectrophotometer system MD 2010 Plus equipped with a diode array UV PDA detector (Jasco Corporation, Kyoto, Japan).

Bioactive compound analysis: Flavonoid content was assessed by colorimetric assay¹¹. An aliquot of 50 μ L of extract and a calibration solution of quercetin (20, 40, 60, 80 and 100 mg L⁻¹) were added to a 5 mL flask containing 2 mL of ddH₂O. First, 0.15 mL of NaNO₂ aqueous solution (5 g/100 mL) was added to the flasks. After 5 min, 0.15 mL of a solution of AlCl₃ (10 g/100 mL) was added. After 6 min, 1 mL of 1 M NaOH was added to the mixture. Then, the reaction mixture were diluted to volume with the addition of 1.2 mL of ddH₂O and mixed well. Absorbance was determined at 510 nm. Total flavonoid content was expressed as mg quercetin equivalents (QE)/100 mg fw.

The anthocyanins were quantified by spectrophotometry. The concentration of anthocyanins was determined by applying the Lambert-Beer law. The spectra recorded using a UV-28O $_2$ diode array spectrophotometer (UNIC, USA) were measured at 25 °C and 530 nm against the solvent. For that purpose, 10 mm quartz cells were used.

The phenolic content was measured using the method of Singleton and Rossi 12 , with some modifications. Briefly, an aliquot (20 μ L) extract and the calibration solution of gallic acid (20, 40, 60, 80 and 100 mg L $^{-1}$) was added to a 25-mL volumetric flask containing 9 mL of double-distilled water (ddH $_2$ O). The HPLC by column chromatography (401.2 cm, i.d.)

on Sephadex G-25 (Sigma Steinheim, Germany) was used to measure the total phenolic content. One milliliter of Folin and Ciocalteu phenol reagent was added to the mixture and stirred. After 5 min, 10 mL of Na_2CO_3 aqueous solution (7 g/100 mL) was added into the mixture. The solution was then immediately diluted to volume with ddH_2O and mixed thoroughly. After incubation for 90 min at $23\,^{\circ}C$, absorbance compared with the blank control was read at 765 nm. Total phenolic content was expressed as mg gallic acid equivalent (GAE)/100 mg fw.

Antioxidant activity: The DPPH (2,2-Diphenyl-1-picrylhydrazil) method is an easy, fast and sensitive method for testing the antioxidant activity of certain compounds or plant extracts 13,14 . Antioxidant activity is the ability of a compound or extract to inhibit an oxidation reaction which can be expressed by the percentage of inhibition. Parameters used to demonstrate antioxidant activity are efficient concentration (EC $_{50}$) or inhibition concentration (IC $_{50}$), i.e., the concentration of an antioxidant substance that can cause 50% DPPH to lose radical character or the concentration of an antioxidant substance that yields 50% inhibition, respectively. Substances that have high antioxidant activity will have low EC $_{50}$ or IC $_{50}$ values 15 .

Briefly, the extracts were reconstituted in ethanol (1 mg mL^{-1}) and different concentrations $(200\text{-}0.234\,\mu\text{g mL}^{-1})$ of each extract were used. In a total volume of 1 mL, the test mixture contained 500 mL of extract and 500 mL DPPH (125 μ m in ethanol). The test mixture was shaken and quenched at room temperature in the dark for 30 min. Absorbance was then measured at 517 nm in a DU 7500 spectrophotometer (Beckman Coulter). Quercetin was used as a positive control. The capture capacity of DPPH free radicals was calculated as follows 16:

Percentage antioxidant (%) =
$$\frac{A - B}{A} \times 100$$

Here, A is the absorbance of the negative control (DPPH plus ethanol) and B is the absorbance of the sample (DPPH, ethanol plus sample). Using the correlation between each concentration and the percentage of free radical capture, the EC_{50} was calculated by interpolation. The activity is expressed as EC_{50} (effective concentration of each extract that captures 50% of DPPH radicals).

Data analysis: All of the experimental results were expressed as the Mean ± Standard Deviation (SD) of three independent experiments.

RESULTS AND DISCUSSION

Nutritional substances: Approximately 2.5 kg red dragon fruit was drying in a vacuum evaporator, this yielded 252.7 g of powder. Therefore, the yield was 17%. The result of proximate analysis was as follows: 11.53% moisture, 4.17% ash, 5.30% crude fiber, 5.68% protein, 0.43% fat and 78.01% carbohydrate (Table 1). Nutritional substances, particularly fat, were almost zero, which is good for obese people.

Bioactive compounds: Red dragon fruit powder contained high contents of bioactive compounds, including phenolic acid, flavonoids and anthocyanins (Table 2). Common fruits with significant total polyphenol contents include *Musa* sp. (Banana), with 110 g g⁻¹ total polyphenol; *Ananas comosus* (Pineapple) with 150 g g⁻¹; *Carica papaya* (Papaya) with 260 g g⁻¹; tomatoes with 350 g g⁻¹; cherries with 670 g g⁻¹ and blueberries with 3180 g g⁻¹. In this study, our results show a total phenolic content of 386.09 ± 1.52 mg in 100 g of dry *Hylocereus polyrhizus*. This quantity exceeds the amounts in the aforementioned common fruits, indicating that red dragon fruit powder is a good source of polyphenols that can be integrated into the human diet.

Phenolic compounds are widely known for their beneficial effects, such as preventing hormone-related cancers, potent antioxidant activity and antibacterial properties¹⁷.

Chang *et al.*¹⁸ suggested that phenolic acids and flavonoids were the main contributors to antioxidant activity and anti-LDL peroxidation. The total phenolic content of the pitaya seed was high and was usually correlated with high radical scavenging activity as shown in Table 2.

In addition, high flavonoid content, such as procyanidin B2, epicatechin and epigallocatechin gallate, contributed to the high antioxidative activity against free radicals¹⁹. High phenolic content was usually correlated with high radical scavenging activity²⁰.

Table 1: Nutritional substances of red dragon fruit powder

Nutritional substances	Amount (%)
Water content	11.53±0.7
Ash content	4.17±0.6
Crude fiber content	5.30±0.5
Protein content	5.68±0.3
Fat content	0.43 ± 0.1
Carbohydrate content	78.01 ± 1.1 (by difference)

Table 2: Bioactive compounds of red dragon fruit powder

Sample name	Bioactive compounds	Amount	Unit
Red dragon fruit	Total phenolic acid	386.09±1.52	Mg/100 g
powder	Total flavonoid	210.02 ± 0.83	Mg/100 g
	Total anthocyanin	81.75±1.43	Mg/100 g

Table 3: Comparison of the antioxidant activity of (parameter IC_{50}) red dragon fruit powder versus vitamin C standard, BHT standard and quercetin standard

Sample	Amount	Unit
Red dragon fruit powder	1195.181±16.2	ppm
Vitamin C standard	2.297±0.14	ppm
BHT standard	17.146±0.32	ppm
Quercetin standard	2.229±0.04	ppm

Flavonoids, also known as nature's tender drugs, possess various biological/pharmacological activities, including anticancer, antimicrobial, antiviral, anti-inflammatory, immunomodulatory and antithrombotic activities²¹. Many studies have shown that several flavonoids, including wogonin, luteolin and quercetin, inhibit the expression of proinflammatory molecules in experimental animals; these findings suggest that the modulation of proinflammatory gene expression is a major mechanism for the anti-inflammatory activity of flavonoids²¹⁻²³.

Flavonoids show anti-inflammatory activity *in vitro* and *in vivo*. Several cellular mechanisms of action have been proposed to explain their anti-inflammatory activity. In addition to antioxidative activity, they inhibit eicosanoid generating enzymes. Certain flavonoids, mainly flavone derivatives, modulate the expression of proinflammatory molecules, at least partly via inhibition of transcription factor activation.

Flavonoids have different mechanisms depending on their chemical structures. Any single mechanism could not explain all of their *in vivo* activities. They likely have multiple cellular mechanisms that act on multiple sites of the cellular machinery, but the most important contributors to anti inflammation by flavonoids seem to be their effect on eicosanoid generating enzymes and their effect on the expression of proinflammatory molecules²⁴.

In addition, in obese rats, consuming red dragon fruit powder and performing swimming exercises inhibited oxidative stress, primarily MDA and enhanced the immune system by decreasing the TNF- α concentration²⁵.

Moreover, Mann²⁶ suggested that the mechanism of action of flavonoids as antioxidants could be direct or indirect. The direct mechanism is to donate hydrogen ions to neutralize the toxic effects of free radicals. As antioxidants, they can act indirectly by increasing endogenous antioxidant gene expression. This mechanism via the activation of nuclear factor erythroid 2-related factor 2 (Nrf2) results in the increased expression of genes involved in the synthesis of endogenous antioxidant enzymes²⁶.

Antioxidant activity: The antioxidant activity of red dragon fruit powder as expressed as IC₅₀-DPPH is compared with vitamin C standard, BHT standard (butyl hydroxy toluene) and the quercetin standard (Table 3).

The antioxidant activity of red dragon fruit powder *in vitro* compared with the standards of vitamin C, BHT and quercetin was lower because capturing 50% of free radicals required 1195.181 ppm red dragon fruit powder. This is because the standards of vitamin C, BHT and quercetin are components of a single bioactive compound, while red dragon fruit powder as extracted contained 30% maltodextrin filler and various other bioactive components, micronutrients and macronutrients.

However, better results in the *in vitro* assay will not necessarily be shown in an *in vivo* assay²⁷. Other factors that can affect *in vivo* results include differences in absorption ability and the metabolic conversion of bioactive compounds in the digestive system.

The difference in antioxidant solubility would cause coalescence in cellular and tissue structures. The adjacent antioxidants will recharge the neighboring antioxidants in an integrated manner that depends on reaction of stoichiometry and kinetics. The presence of a given antioxidant relies not only on the reaction rates and concentrations in separate parts but also on the ability to interact with the regeneration of various antioxidants²⁸.

CONCLUSION

Red dragon fruit powder represents a major source of antioxidants, which is valuable to any food crop. Further studies are needed for the *in vivo* assay of this powder as a new valuable crop with a significant amount of antioxidants, which could be beneficial for consumers and the pharmaceutical industry.

In addition, red dragon fruit powder can be used as an ingredient in functional foods as a new innovative product and can be stored for a long time. It is important to continue this research by identifying other sources of antioxidants, which are beneficial for human health.

SIGNIFICANCE STATEMENTS

This study discovered the properties of red dragon fruit powder that could be beneficial for human health in every stage of the life cycle. This study will help researchers to uncover critical areas of food technology because the powder has a long shelf life, high economic value and could serve as a functional food that could be added to a variety of

processed food products that many researchers have yet to explore. Thus, this study shows that red dragon fruit can not only be eaten directly, but can also be made as a powder that is rich in bioactive compounds, nutritional substances and potential antioxidant activity.

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