

Development of Functional Drink Using *Hibiscus rosa-sinensis* Leaves

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ABSTRACT

This study aimed to develop a functional drink based on *Hibiscus rosa-sinensis* and evaluate the chemical, and sensory properties. The functional drink was made in a traditional method using hibiscus leaves juice and then diluted to several concentrations. The experiment was carried out using a completely randomized design with 5 levels of concentration of hibiscus leaves juice (5, 10, 15, 20, and 25% v / v). Results showed that there was no difference in preferences between concentrations and the 25% treatment concentration had the best value of various parameters. The DPPH radical scavenging activity in the 25% of hibiscus leaves juice was $34,90 \pm 1,85\%$; total phenolic content was $119,25 \pm 1,91$ mg-GAE/g and total tannin was $41,31 \pm 2,25$ mg-TAE/g. This study concludes that 25% of hibiscus leaves juice can be used to formulate an acceptable functional drink from *Hibiscus Rosa-Sinensis* Leaves.

Keywords: Antioxidant, *Hibiscus rosa-sinensis*, Functional drink.

1. INTRODUCTION

Indonesia is a country with a tropical climate with a geographical location right on the equator. This position causes Indonesia to have a high level of biodiversity. The potential location of the country is described as providing certain characteristics of natural resources in the form of tropical rain forest ecosystems. The hibiscus plant (*Hibiscus rosa-sinensis* Linn.) is a type of plant that is widespread in almost all parts of Indonesia. The parts of the hibiscus plant consist of flowers, leaves, stems, and roots [1].

Hibiscus contains antioxidants, flavonoids, tannins, saponins, polyphenols, taraxeryl acetate, peroxidase, calcium oxalate, and terpenoids. Antioxidants have been recognized by the public as active substances that are beneficial to the health of the human body. The function of antioxidants is to overcome or neutralize free radicals to inhibit the aging process and can prevent damage from the disease [2].

Flowers, leaves, and roots of hibiscus flowers contain flavonoids. Besides, the leaves also contain saponins, tannins, and polyphenols, flowers that contain polyphenols, and the roots contain saponins and tannins [3]. The benefits of hibiscus (*Hibiscus rosa-sinensis* L.)

itself include anti-bacterial, antioxidant, anti-tumor, anti-hypertensive, and wound healing [4].

The activity of bioactive compounds in the form of polyphenols, flavonoids, tannins, and saponins in hibiscus leaves has an important role in the healing process of disease. The role of flavonoids is to improve blood circulation throughout the body and prevent blockage of blood vessels, anti-inflammatory and as an anti-pain (analgesic) while saponins are chemical substances that are useful in influencing collagen (early stage of tissue repair) by inhibiting the production of excessive scar tissue [5]. Fresh hibiscus leaves extract contains phenol activity of 526.15 mg GAE extracted using acetone solvent and 305.33 mg GAE extracted with distilled water. Whereas dry leaves contain phenol activity of 1307.33 mg GAE extracted using acetone solvent and 921.67 mg GAE extracted with distilled water [6].

In Indonesia, boiled and squeezed hibiscus leaves are often used as traditional herbal ingredients that are believed to cure fever, reduce heat, heal wounds, and canker sores. In vivo and invitro experiments on the use of the leaves, flowers, and roots of flower plants have been widely carried out. Based on the research conducted, it is known that hibiscus plants can be used to nourish the body and cure various diseases because of the content of

compounds in them, one of which is to prevent Diabetes mellitus (DM type 2) [7].

To increase its utilization and usefulness, hibiscus has the potential as a functional drink, especially the leaves when viewed from the content and properties it provides. Functional food is food or drinks which contain ingredients that have certain physiological functions and can improve health status and prevent certain diseases. One of the components of functional food that has a physiological function for the body is antioxidants. Daily intake of antioxidants can reduce the chances of developing degenerative disease symptoms [7]. This study aimed to develop a functional drink based on *Hibiscus rosa-sinensis* and evaluate the chemical, and sensory properties.

2. MATERIALS AND METHODS

2.1 Materials

The materials used in this study were hibiscus leaves, sugar, and water. The materials used for analysis were buffer solution, DPPH, (1,1-diphenyl-2-picrylhydrazil), 70% methanol, Ciocalteu folin, Na₂CO₃, tannic acid, gallic acid, and distilled water.

2.2 Methods

Making functional drinks was by sorting hibiscus leaves. Then each leaf was washed separately, then the kneading process was carried out using plastic gloves with added water with a ratio of 1: 1 of each hibiscus leaves and water (500 grams of leaves and 500 mL of water). After that, the extract was filtered using a filter. Then the extract was taken, and the concentration was made according to the treatment, namely hibiscus leaves extract water (5, 10, 15, 20, and 25%: 200 mL of water). Then heated at a temperature of 70 ° C for 5 minutes. After that, the resulting product was put into a sterile bottle and then analyzed.

2.3 Degree of acidity (pH)

Testing the degree of acidity was carried out using a standardized pH meter with a pH 4 buffer solution then the pH 7 buffer was turned on, left to stabilize. The electrodes are rinsed with distilled water and then dried with a tissue. The electrode is immersed in 10 ml of the sample solution and is left for a while until a reading is obtained then the pH of the sample is recorded.

2.4 Determination of total phenolic content

Analysis of total phenol levels was carried out using the spectrophotometric method [8]. 1 mL of the sample is put into a test tube and 1 mL of 95% ethanol and 5 mL of ion-free water are added. Furthermore, 0.5 mL of Folin-Ciocalteu reagent 50% (v / v) was added to

each sample and then diluted with ion-free water. After 5 minutes, 1 mL of 5% (w / v) Na₂CO₃ was added and diluted again with ion-free water (if it was too concentrated). After that, it was vortexed and stored in a dark room for 60 minutes. The sample was then homogenized (vortexed) again, and the absorbance was measured at 725 nm. The content of total phenolics in each extract was determined using a standard curve prepared for gallic acid and expressed as milligrams of gallic acid equivalents (GAE) per gram of sample.

2.5 Determination of total tannin content

Total tannin content was determined using spectrophotometric methods [9]. A total of 1 mL of sample was added with 9 mL of distilled water. 0.2 mL of the extract was added with distilled water to a total solution of 1 mL of Folin-Ciocalteu reagent and then vortexed again. The absorbance was measured at λ 760 nm after incubation for 30 minutes at room temperature. The results obtained were plotted against the tannic acid curve prepared in the same way and expressed as milligrams of tannic acid equivalents (TAE) per gram of sample.

2.6 Determination of Antioxidant activity (DPPH radical scavenging assay)

Determination of antioxidant activity was carried out by spectrophotometric method [10]. A total of 0.2 mL of hibiscus leaves drink solution was piped using a micro pipette into the vial, then added 3.8 mL of 0.05 μL DPPH solution. The solution mixture is then homogenized and left for 30 minutes in a dark place. The absorption was measured using a Uv-Vis spectrophotometer with a wavelength of 517 nm. Ascorbic acid was used for positive control, as a comparison to determine the antioxidant activity of the sample, the treatment was the same as the sample. The absorbance data obtained, or the amount of antioxidant activity was used to determine the% inhibition.

$$\text{Percent inhibition (\%)} = \frac{(\text{DPPH absorbance} - \text{sample absorbance})}{(\text{DPPH absorbance})} \times 100\%$$

2.7 Sensory Evaluation

The sensory evaluation at this stage was carried out to determine the level of preference for the functional drink from hibiscus leaves. The sensory evaluation was carried out using a hedonic scale. This test was conducted by 20 semi-trained panelists consisting of students of the Agricultural Product Technology Study Program, Faculty of Agriculture, Jambi University. Panelists were asked to taste and give a score. The score for the organoleptic test can be seen in Table 1.

Table 1. The score for the organoleptic test

Score	Overall acceptability
1	Very dislike
2	Do not like
3	Kind of like it
4	Like it
5	Really like

2.8 Data analysis

To find out the effect of the treatment given, the data obtained will be analyzed statistically by using variance at the 5% and 1% levels, if significantly different, then proceed with the Duncan's New Multiple Range Test (DNMRT) at the 5% level.

3. RESULT

3.1 Degree of acidity (pH)

pH is the degree of acid used to express the level of acidity or alkalinity of a solution. Based on the analysis

Table 2. Physico- chemical properties and Overall Acceptability of *Hibiscus Rosa-Sinensis* Leaves functional drinks

Concentration	pH	Total Phenolic Content (mg GAE/g)	Total Tannin (mg TAE/g)	Antioxidant Activity (% inhibition)	Overall Acceptability
5%	6,28 ^a ± 0,05	49,83 ^a ± 1,70	13,84 ^a ± 2,05	7,08 ^a ± 2,68	2,81 ± 0,93
10%	6,20 ^a ± 0,08	67,80 ^b ± 1,54	26,43 ^b ± 1,41	18,47 ^b ± 1,83	2,91 ± 0,77
15%	6,13 ^{ab} ± 0,09	97,33 ^c ± 0,62	33,74 ^c ± 2,23	20,65 ^{bc} ± 2,48	3,20 ± 0,68
20%	6,00 ^b ± 0,16	107,68 ^d ± 2,13	36,47 ^c ± 2,40	23,44 ^c ± 2,36	3,29 ± 0,91
25%	5,65 ^c ± 0,17	119,25 ^e ± 1,91	41,31 ^d ± 2,25	34,90 ^d ± 1,85	3,05 ± 1,12

of variance, it showed that the pH of hibiscus leaves drink produced significantly different results for each treatment given. The pH value ranges from 5.65 to 6.28 where the higher the concentration of hibiscus leaves, the lower the resulting pH value. This is because the antioxidants of the phenolic compound group function as hydrogen donors. It is explained that the hibiscus leaves have substances rich in hydrogen which can be released by phenolic compounds [11].

3.2 Total phenolic content

Based on the research results, it is known that with the increasing of treatment, the total phenol will also increase. The highest total phenol was produced at 25% treatment with a value of 119.25%. Another study stated that fresh hibiscus leaves extract contained the activity of the phenolic compound 305.33 mg GAE. Meanwhile, dried hibiscus leaves contain the phenolic activity of 921.67 mg GAE extracted with distilled water [6].

Polyphenols have a role as antioxidants that are good for health. Antioxidants from phenolic compounds can

reduce the risk of several diseases such as heart attack and cancer. Phenolic compounds are natural compounds that are widely used today. Its ability as an active biological compound provides a major role for human interests. One of them is as an antioxidant for the prevention and treatment of degenerative diseases, cancer, premature aging, and immune system disorders [12].

3.3 Total Tannin

Based on the analysis of the various levels of tannins produced from the hibiscus leaves drink, they ranged from 13.84-41.31. The value of the tannin content increased with increasing the given concentration which resulted in a significantly different value in each treatment. Hibiscus leaves are known to have a high tannin content. Hibiscus leaves contain tannins, flavonoids, calcium oxalate, taraxeryl acetate, peroxidase, terpenoids and saponins. The main compounds that act as hemostasis are tannins and flavonoids [2].

3.4 Antioxidant activity

Antioxidants are organic compounds that can reduce free radicals in the human body. Many studies show that some plant extracts have phenolic antioxidant compounds, flavonoids that are more effective and safer than synthetic antioxidants. Antioxidants are increasingly being used in the food sector. In food, antioxidants not only act as free radical inhibitors but can also act as preservatives [13].

Analysis of variance showed that the % inhibition produced was significantly different for each treatment given. Where the % inhibition value ranges from 7.08-34.90. % inhibition indicates the antioxidant content of a substance. Where the best treatment was obtained at 25% treatment, namely 34.90 %. Hibiscus leaves act as antibacterial, antioxidant, anti-tumor, antihypertensive, and can even heal wounds [4].

3.5 Overall Acceptability

Overall acceptance is the most important parameter because it relates to the level of acceptance of the product by the panelists. The results showed that the

concentration of hibiscus leaves juice affected the overall acceptance of hibiscus leaves drink. Overall acceptance of hibiscus leaves drink is seen in table 2. The overall acceptance results showed that until the concentration of 25% was still acceptable to the panelists, at 25% there was a slight decrease which was not significantly different, but it might decrease if the concentration was increased. It was because of the drink's thickness produced by the mucilage on the leaves.

4. CONCLUSION

Statistical results showed that there was no difference in overall acceptance and had a significant effect on pH, total phenolic content, total tannin, and antioxidant activity. This study concludes that 25% of hibiscus leaves juice can be used to formulate an acceptable functional drink from *Hibiscus Rosa-Sinensis Leaves*.

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