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Aktivitas Antioksidan Sirup Kombinasi Cacing Tanah (*Lumbricus rubellus*) Dan Kulit Buah Naga (*Hylocereus polyrhizus*) Dengan Metode DPPH

Antioxidant Activity of Syrup Made from a Combination of Earthworth (*Lumbricus rubellus*) And Dragon Fruit Peel (*Hylocereus polyrhizus*) With DPPH Method

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Abstract

Dragon fruit peels and earthworms contain an antioxidant activity that can inhibit free radicals. Their utilization can be developed in the form of syrup ingredients. Dragon fruit peel extract has IC_{50} 3.14 gram / 100 ml, while earthworm flour has IC_{50} 120.22. This research aimed to determine the best syrup formula and antioxidant activity from a combination of earthworms (Lumbricus rubellus) and dragon fruit peels (Hylocereus polyrhizus).

The syrup was made in 3 formulas with varying levels of earthworms and dragon fruit peels, namely formula 1 (20%; 10%), formula 2 (15%; 15%), and formula 3 (10%; 20%). Each formula was tested for its physical properties, organoleptic properties (color, smell, and taste), homogeneity, viscosity, pH, specific gravity, and hedonic test to obtain the syrup with the best profile. Then, the best syrup was tested for antioxidant activity by using the DPPH method.

The results showed that the syrup from a combination of earthworm and dragon fruit peel met the syrup quality test (organoleptic, homogeneity, viscosity, pH, and BJ). In this study, formula 2 (15%; 15%) was chosen because it had an attractive taste and appearance, no sediment/homogeneity, low viscosity, stable pH, and the highest hedonic test results. Earthworm syrup and dragon fruit peel have moderate antioxidant activity (IC_{50} 124.9 ppm) than ascorbic acid (IC_{50} 10.24 ppm).

Keywords: *antioxidant; dragon fruit peel (Hyocereus polyrhizus); DPPH; earthworm* (*Lumbricus rubellus); syrup*

Abstrak

Kulit buah naga dan cacing tanah merupakan bahan yang memiliki aktivitas antioksidan yang mampu menghambat radikal bebas, sehingga pemanfaatannya dapat dikembangkan dalam bentuk sediaan sirup. Ekstrak kulit buah naga memiliki IC_{50} 3,14 gram/100 ml, sedangkan tepung cacing tanah IC_{50} 120,22. Penelitian ini bertujuan untuk mengetahui formula sirup terbaik dan aktivitas antioksidan dari kombinasi cacing tanah (*Lumbricus rubellus*) dan kulit buah naga (*Hylocereus polyrhizus)*

Sirup dibuat dalam 3 formula dengan variasi kadar cacing tanah dan kulit buah naga, yaitu formula 1 (20%;10%), formula 2 (15%;15%), formula 3 (10%;20%). Masing-masing formula diuji sifat fisiknya, organoleptis (warna,bau,dan rasa), homogenitas, viskositas, pH, berat jenis, dan uji hedonic untuk mendapatkan sirup dengan profil terbaik. Sirup terbaik kemudian diuji aktivitas antioksidan dengan metode DPPH.

Hasil penelitian menunjukkan sirup cacing dan kulit buah naga memenuhi uji kualitas sirup (organoleptis, homogenitas, viskositas, pH, dan BJ). Pada penelitianini formula 2 (15%;15%) merupakan formula terpilih karena memiliki rasa dan penampilan menarik, tidak terdapat endapan/ homogen, kekentalan yang rendah, pH stabil, dan menunjukkan hasil uji hedonic tertinggi. Sirup cacing tanah dan kulit buah naga memiliki aktivitas antioksidan sedang (IC_{50} 124,9 ppm) dibanding asam askorbat (IC_{50} 10,24 ppm)

Kata kunci: antioksidan; cacing tanah (*Lumbricus rubellus);* DPPH; kulit buah naga *(Hylocereus polyrhizus)*, sirup

Introduction

Free radicals are a form of reactive compound generally known as compounds with unpaired electrons in the outer shell. Free radicals in the human body could cause various degenerative diseases. Free radicals can be counteracted or prevented by giving antioxidants[12]. Antioxidants inhibit oxidation reactions caused by free radicals, which could damage unsaturated fatty acids, cell wall membranes, blood vessels, DNA bases, and lipid tissue, causing diseases. [9]

Dragon fruit is a plant that has antioxidant activity. It could ward off free radicals[2]. According to previous studies[12], dragon fruit has properties, benefits, and high nutritional value. 30-35% of dragon fruit is fruit peel, but it is often only thrown away as waste. According to the research(17), the advantage of dragon fruit skin is that it is rich in polyphenols and a good source of antioxidants.

Earthworms are low-level animals because they do not have a backbone (invertebrates). Earthworms belong to the Phylum Annelida. Annelida comes from the word "Annulus," which means ring. The body of this animal consists of rings or segments[10]. Empirically, earthworms can be used as typhus and antipyretic medicines[3]. It is reported that earthworms have activities as aphrodisiacs [14]. Then, earthworms have antibacterial and antioxidant abilities[16].

The syrup is a concentrated preparation in water from sugar or sugar substitutes with or without additives, fragrances, and active substances as medicine[1]. According to previous studies (5), syrup contains at least 50% sucrose and usually 60-65%.

It is stated in the previous research that the results of the antioxidant activity test for earthworm flour had a value of IC_{50}120,222 ppm, and ascorbic acid of 5.404 ppm, while the single-use red dragon fruit peel ethanol extract had an antioxidant activity with an IC value of 3.14 grams / 100 ml[9].

One of the strategies to increase the use of dragon fruit peel as an antioxidant was making its syrup made from a combination of dragon fruit peel and earthworms. Generally, syrup formulation could maintain quality, extend shelf life, and optimize its utilization. Next, the mixture was expected to increase the antioxidant activity of earthworm syrup. This research was important to determine the best formulation combination of earthworm syrup and dragon fruit peel with the best antioxidant activity by implementing the DPPH method and to contribute to scientific improvement in universities, especially in health technology related to the development of traditional medicine.

Research Method

Tools and Materials

The tools used were a magnetic stirrer, pyrex glassware, analytical balance, pH meter, test tube, micropipette, measuring flask, UV Vis spectrophotometer filter paper, and 1 mL blue tip.

The materials used were dragon fruit peel and earthworms obtained from the Mojosongo area, Boyolali Regency, Central Java, water, glucose, sodium benzoate, DPPH solution, and methanol p.a.

Procedure

In this research, 100 ml of syrup was made. The concentrations of earthworms and dragon fruit peel were added for F1 (20%; 10%), F2 (15%; 15%),

and F3 (10%; 15%). The syrup formula that combines earthworms and dragon fruit skin can be seen in Table 1.

Fresh earthworms F1 (20 g), F2 (15 g), F3 (10 g), and dragon fruit peel F1 (10 g), F2 (15 g), F3 (20 g) were prepared. Earthworms and dragon fruit peels were sorted, washed, and drained. Then, the earthworms and dragon fruit skin were boiled in 100 ml water at 90^{o}C for 15 minutes. Lift

60g of glucose was added to the solution of earthworm and hot dragon fruit peel in some steps until it was dissolved. Then, it was filtered. After that, 0.15 g of sodium benzoate was added and stirred until homogeneous.

The tablet formulation of syrup made from a combination of earthworm and dragon fruit peel can be seen in the table below:

Table 1. The formula of Syrup Made from a combination of Earthworms and Dragon Fruit Peel

	Formula		
Ingredients	**1**	**2**	**3**
Earthworms	20 g	15 g	10 g
Dragon Fruit Peel	10 g	15 g	20 g
Glucose	60 g	60 g	60 g
Sodium benzoate	0,15 g	0,15 g	0,15 g
Aquadest	100 ml	100 ml	100 ml

1. Syrup Ingredients Evaluation

There were five tests performed in this evaluation. The first test was the organoleptic test. The test analyzed the changes in color, smell, and taste. The second was the homogeneity test. The test was performed by observing the ingredients to determine whether there were particles/deposits in the syrup solution. The third was the pH test. The pH value measurement aimed to determine the syrup's pH value; its height used a pH meter. The fourth was the viscosity test by using a stormer viscosity meter. The fifth was the density Test. The determination of specific gravity was performed using a pycnometer.

2. Hedonic Test

The syrup was tested on 25 respondents. Then, the respondents were asked to fill out a questionnaire that included their taste, aroma, and appearance [3].

3. Antioxidant Activity of Syrup Made from a combination of Earthworm and Dragon Fruit Peel Using the DPPH method

The 25 mg of earthworm fruit syrup and dragon fruit peel were weighed as the sample solution. It was dissolved in absolute methanol, homogenized, and then added to the volume of up to 25 ml. Next, the dilution was performed again by making five solution concentrations (1; 1,5; 2; 2.5; and 3 ppm). Each concentration was piped 1 mL of sample solution with a micropipette and input into a test tube, then 4 ml of the 50 µM DPPH solution was added. The mixture was homogenized and left for 30 minutes in a dark place; a UV-Vis spectrophotometer measured the absorption at the 514 nm wave. The antioxidant activity of the sample was determined by the amount of DPPH radical absorption inhibition by calculating the

percentage (%) of DPPH absorption inhibition using the formula (Molyneux, 2004).

Finding and Discussion

Syrup Ingredients Evaluation

Organoleptic test

The findings showed that the ingredient obtained was pink. It had a distinctive aroma of worms and dragon fruit skin. It also had a sweet taste.

Homogeneity Test

In the homogeneity test, all syrup formulas that were not tested had lumps and sediment deposits in the solution. It occurred because there was no difference between the tested ingredients and the active substances used in the syrup ingredients of this research.

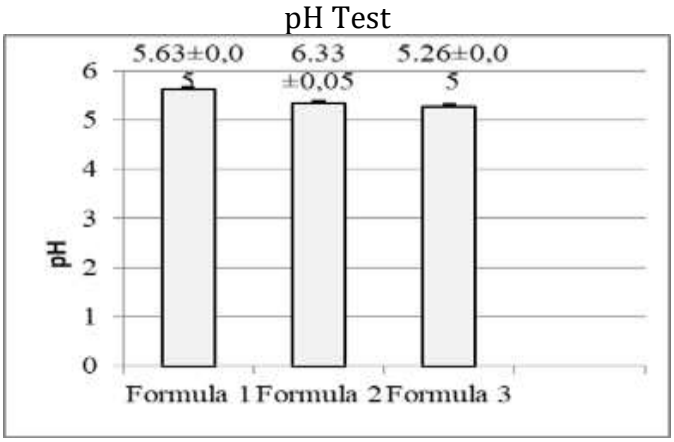

Figure 1. The Graph of Syrup pH Test Result

Based on the observation of the available pH values, it could be seen that all syrup ingredients had a stable pH value, namely F1 (5.633 ± 0.058), F2 (5.33 ± 0.058), and F3 (5.267 ± 0.058). The pH test was an important parameter because the stable pH value of the solution showed that the distribution process of the basic ingredients in the preparation was evenly spread. The recommended pH value for the syrup was between 4 - 7[(1).]

The viscosity test was also performed to determine the consistency of the preparation and to show the thickness of a trial that was measured with a stomachic viscometer. The lowest viscosity of the syrup on observations at the viscosity values of F1 (0.52 ± 0.02), F2 (0.51 ± 0.02), and F3 (0.57 ± 0.03) dpA was in the formula 2 ingredients. The lower the viscosity of the syrup, it was also easier to pour. The viscosity of the syrup was related to the sugar compounds in the syrup ingredients. The previous studies [(5)] propose that syrups contain at least 50% and typically 60-65% of sucrose.

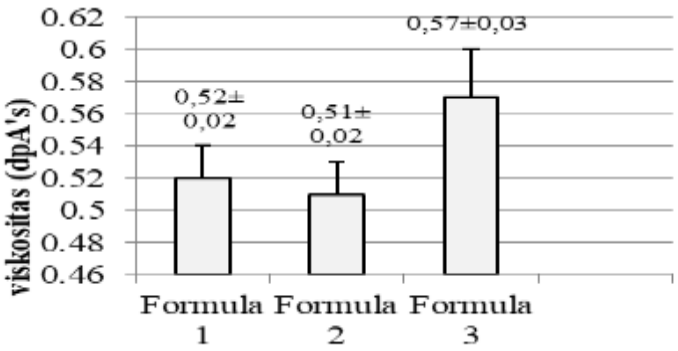

Figure 2. The Graph of Syrup Viscosity Test Results Graph

Specific Gravity

The measurement results of the specific gravity showed the specific gravity values of all formulas. It ranged from 1.15 to 1.23.

2. Hedonic Test for Syrup Made from a combination of Earthworth and Dragon Fruit Peel

The hedonic test was performed by describing the preference value (aroma, appearance, and taste parameters) using 25 panelists. The taste was the most crucial characteristic for evaluating the oral ingredients. Besides, respondents usually measured the color, texture, and smell subjective.

The syrup made from a combination of earth worth and dragon fruit peel was measured by its level of preference for aroma and color using the three formulas with different concentrations of active ingredients.

Figure 2. The Table of Syrup Hedonic Test

Indicators	FI (%)	F II (%)	F III (%)
Like	28	48	36
Dislike	72	52	64
Total	**100**	**100**	**100**

The Best Syrup Formula

The syrup with the best formula can be obtained from the determination of the physical properties test of the syrup ingredients along with the hedonic test. All syrup ingredients meet the appropriate requirements by showing that formula 2 had the lowest viscosity value. Homogeneity and pH testing of all syrup formulas also meet the requirements: homogeneous with a pH between 4-7. The Formula 2 hedonic test has a better acceptance rate. From this test, it can be concluded that Formula 2 was a good and preferable formula to be used for the DPPH test.

C. Antioxidant Activity Test with DPPH

Based on the IC50 value, it was obtained that the best formula (F2) syrup made from a combination of earth worth and dragon fruit peel had moderate antioxidant activity compared to vitamin C as a comparison or positive control in this study. Formula 2 syrup had an IC_{50} of 124.9 ppm, while vitamin C had an IC_{50} value of 10.24 ppm. The previous research stated that the results of the antioxidant activity test of single-use earthworm flour without a combination had an IC_{50} value of

120.222 ppm, while ascorbic acid was 5.404 ppm. Furthermore, the ethanol extract of red dragon fruit peel had an antioxidant activity with an IC value of 3.14 grams / 100 mL[9]. Thus, making syrup ingredients made from a combination of earthworm and dragon fruit peel did not significantly increase the antioxidant activity of earthworm in syrup ingredients.

Conclusion

Syrup formula 2 (15%; 15%) was chosen because it had an attractive taste and appearance, no sediment/ homogeneity, low viscosity, stable pH, and the highest hedonic test results. The syrup made from a combination of earth worth and dragon fruit peel had moderate antioxidant activity (IC50 124.9 ppm) compared to vitamin C IC50 10.24 ppm). Further research must be conducted regarding the combination of ingredients forms and the methods used in making the ingredients.

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