JPSE (Journal of Physical Science and Engineering)



http://journal2.um.ac.id/index.php/jpse EISSN: 2541-2485

Grecoli Candy from Grape and Broccoli to Boost the Body's Immune System in the Middle of the COVID-19 Pandemic

Received 15 October 2021

Revised 19 February 2022

Accepted for Publication 19 April 2022

Published 06 July 2022

F Fathurochman¹, Hartatiek^{1*}, N E A Ermalita¹, L M Rizki², and N Ahmad³

- ^{1.} Department of Physics, Faculty of Mathematics and Natural Sciences, Universitas Negeri Malang, Jl. Semarang 5, Malang, 65145, Indonesia.
- Department of Chemistry, Faculty of Mathematics and Natural Sciences, State University of Malang, Jl. Semarang 5, Malang, 65145, Indonesia.
- ³ Department of Materials, Manufacturing, and Industrial Engineering, School of Mechanical Engineering, Faculty of Engineering, Universiti Teknologi Malaysia, Skudai, Johor Bahru, 81310, Malaysia.

***E-mail:** hartatiek.fmipa@um.ac.id



This work is licensed under a <u>Creative</u> <u>Commons Attribution-</u> <u>ShareAlike 4.0</u> International License

Abstract

Grapes and broccoli have been scientifically proven to have high antioxidant content to increase the body's immune power compared to other fruits or vegetables. An immunomodulator is a compound that can regulate the activity and function of the immune system. The purpose of this study was to find out how to improve the body's immune system amid the COVID-19 pandemic and also to find out how to make food practically consumed to help increase the body's immunity during the COVID-19 pandemic. In this study, grapes and broccoli were extracted and then boiled at a temperature of $\pm 85-100^{\circ}$ C, then mixed with several ingredients, including sucrose, glucose, citric acid, gelatin, and flavourings. The antioxidant test was carried out using the Ferric Reducing Antioxidant Power (FRAP) method. After obtaining the antioxidant value, a spectrophotometric test was carried out to determine the maximum wavelength and absorbance value obtained. For the first sample, the results were 22.88 mmAAE/mL extract. In contrast, the second sample obtained results of 25.01mmAAE/mL extract.

Keywords: Grapes, broccoli, antioxidants, immunomodulators, COVID-19.

1. Introduction

The outbreak of the Coronavirus pandemic or commonly called COVID-19, began to spread in several countries in early 2020. Until the end of this year, the number of cases in all countries has reached 60 million positive cases [1]. This large number makes all citizens in the world have to limit outdoor activities and crowds. That is intended to prevent the spread of the COVID-19 outbreak.

Generally, preventing the spread of COVID-19 includes exercising regularly, drinking more mineral water, avoiding stress, diligently washing hands, and consuming immune-boosting foodstuffs [2]. The immune system influences the human immune system in the human body itself. Adequate nutrition, especially vitamins, minerals, and bioactive compounds found in vegetables and fruit, can maintain the body's immune system [3]. One example of vegetables and fruits that can maintain the body's immune system is Broccoli and Grapes. Because of its wide range of non-enzymatic bioactive compounds, nutritional antioxidants, carotenoids and phenolic compounds, broccoli was one of the top three contributors of vegetable phenolics (10.5%) to American diets [4], [5]. Broccoli also has vitamins C, E, and minerals (Ca, Mg, Se, and K), which is suitable for improving the human immune system. Likewise, grapes contain vitamins C, B1, B2, provitamin A, and minerals, so they have good effects on humans such as antioxidants, anti-inflammatory, antiaging, anticancer and antimicrobial. The grape has been extensively examined for its phenolic component composition and possible health benefits. Mainly it can prevent oxidative stress-related disorders such as cancer, cardiovascular disease, and neurological disease [6].

The FRAP (Ferric Reducing Antioxidant Power) method is one way to measure antioxidant activity. The Fe^{2+} solution is used in the FRAP (Ferric Reducing Antioxidant Power) method. The quickness with which this approach works is well-known. The FRAP method uses an oxidation-reduction reaction as the basis for antioxidant testing. An acidic sample solution containing antioxidants

will undergo a reduction reaction with the Fe^{3+} complex molecule, changing the colour from green (Fe^{3+}) to yellow (Fe^{2+}) [7].

Indonesian people are certainly no stranger to these vegetables and fruits. Because the two foodstuffs are widely consumed by eating directly, making juice drinks, or cooking separately. Therefore, this study offers a new solution for practically consuming these two foodstuffs but still affects increasing human immunity during the COVID-19 pandemic by making Grecoli jelly candy from grapes and broccoli vegetables.

2. Method

This research consisted of two stages. First, make jelly candy. Second, testing the antioxidant activity of jelly candy with the FRAP method using a spectrophotometer.

2.1. Making Jelly Candy

The tools used in making jelly candy from grapes and broccoli include stainless steel knife, blender, filter paper, tunnel drying, stove, pan, thermometer, stirrer, jelly mould, spoon, and baking sheet. Each fruit was cut and washed using running water and then boiled in water at 100°C for 5 minutes. Once boiled, each fruit was drained, mashed with a blender, and filtered to obtain the extract. The results of grape and broccoli extracts were measured with two variations. The first one is 75 mL of each extract. The second one is 100 mL grape extract and 50 mL broccoli extract. After that, each variation was added and added glucose, 30 grams of sucrose, 0.45 grams of citric acid, 15 grams of gelatin and two drops of grape essence to minimize the smell of the developed gelatin. Continuous stirring is carried out in mixing these ingredients until homogeneous and gets a thick texture. If all the ingredients are well mixed, pour the mixture into a petri dish or baking dish and store for 1 hour. Then the petri dish or pan is put in the refrigerator for 24 hours at a temperature of 5°C. After 24 hours, the dough is removed and returned to room temperature for 2 hours to neutralize the temperature.

2.2. Jelly Candy Antioxidant Activity Test

The tools used in this test are 250 mL beaker, 10 mL, 25 mL, 100 mL, 250 mL, 100 mL Erlenmeyer, dropper, 10 mL centrifugation tube, 10 mL test tube, 1 analytical balance, watch glass, 10 mL pipette volume, stirring rod, and spectrophotometer. The materials needed are NaOH, KH₂PO₄, aquades, oxalic acid, FeCl₃, Trichloroacetic acid (TCA), K₃Fe(CN)₆, Ascorbic acid, and ethanol 96%.

The first step is to prepare a sample of jelly candy, a standard curve solution, and several solutions for the FRAP test, including 0.2 M phosphate buffer pH 6.6,1% oxalate solution, 1% solution, and 10% TCA. After all the solutions are ready, the antioxidant activity test is carried out. With as much as 10 mL of jelly candy sample dissolved in 5 mL of 96% ethanol, then 1 mL pipette, added 1 mL of 0.2 M phosphate buffer (pH 6.6) made from a mixture of NaOH solution with KH₂PO₄, and 1 mLK₃Fe(CN)₆. Next, the sample was incubated for 20 minutes at 50°C. After incubation, 1 mL of TCA was added and centrifuged at 3,000 rpm for 10 minutes. After being centrifuged, 1 mL of the top layer was pipetted into a test tube, 1 mL of distilled water, and 0.5 mL of 0.1% FeCl₃ were added. The solution was allowed to stand for 10 minutes, and the absorbance was measured at 720 nm. As a blank, a mixture of oxalate solution was used. Calibration curves were made using ascorbic acid solutions with various concentrations. The FRAP value was expressed in mg equivalent of ascorbic acid per mL extract. After knowing the antioxidant value contained in the jelly candy, the next step is spectroscopy test to determine the candy's wavelength and combine the candy's chemical and physical properties.

3. Result and Discussion

As mentioned earlier, there are many ways to increase the body's immunity. Immunity is the body's way of fighting and protecting against foreign substances that enter the body, such as bacteria, viruses, and transplanted organs. Several factors can increase the body's immunity, namely avoiding stress, regular exercise, avoiding cigarettes and alcohol, keeping the body from dehydration, and consuming foods that have an immune-boosting effect, such as fruits and vegetables [8]. Nutrition has been investigated and developed into a significant field of study called nutritional immunology as a modifiable element affecting immune function. Appropriate nutrition is also required for the immune system to function correctly [9].

Fruits and vegetables like grapes and broccoli may sound foreign. However, the case is different from Grecoli Candy which is made with the main ingredients of grapes and broccoli. Grapes and broccoli have several vitamins, such as vitamins C and E, minerals, and antioxidant effects. Antioxidants can remove free radical molecules from the body, preventing them from causing disease [10]. That is why grapes and broccoli are excellent for use as Grecoli candy products. Grecoli Candy is a functional food or snack consumed to increase the body's immunity amid this COVID-19 pandemic.

This study uses the FRAP method to determine a material's total antioxidant content based on the antioxidant compounds' ability to reduce Fe^{3+} ions to Fe^{2+} [11], [12]. The ability to reduce Fe ions is then analogous to the strength of the antioxidant [13], [14]. The method used in the standard solution is ascorbic acid. Ascorbic acid was used to compare because it functions as a secondary antioxidant, capturing free radicals and preventing chain reactions [15].

Measurement of antioxidant activity using this FRAP test with the ascorbic acid solution as a standard. The addition of TCA aims to precipitate the potassium ferrocyanide complex [16]. The addition of FeCl₃ also aims to form a green to blue (berlin blue) complex [17], [18]. Power is an indicator of the reduction of an antioxidant compound [19]. In this case, the reducing power is measured by an antioxidant's ability to convert Fe³⁺ into Fe²⁺ [20]. Compounds that have reducing power may be able to act as antioxidants because they can produce electrons or hydrogen atoms so that radical compounds turn out to be more stable. The reaction that occurs is shown in Equation 1.

$$\begin{array}{rcl} K_{3}Fe(CN)_{6} & \rightarrow & K_{4}Fe(CN)_{6} \\ Fe^{3+} + e^{-} & \rightarrow & Fe^{2+} \end{array}$$
(1)

Figure 1 shows the wavelength curve graph indicated by x with the absorbance value indicated by y. In this study, the highest absorbance of the standard solution was obtained at a wavelength of 690 nm. Then the results of the curve graph of the concentration x with the absorbance value y of the comparison solution of ascorbic acid obtained an equation that is y = 0.0081x - 0.0749 with a value of $R^2 = 0.9278$ and to calculate the value of antioxidant activity the absorbance value of the sample is entered into equality. R^2 , which is still 0.9278, indicates an error in the solution being tested, such as a lack of accuracy in adding other solutions or a too concentrated solution. In this study, two samples with different proportions of grapes compared to broccoli were used, i.e. sample 1 with a ratio of 1:1 and sample 2 with a ratio of 2:1.

The FRAP value is expressed in mg ascorbic acid equivalent per mL extract. The final result of the absorbance measurement of the standard solution is shown in Table 1, while the absorbance result of the buffer solution is shown in Figure 2. The vitamin C content in each sample was expressed as Ascorbic Acid Equivalent (AAE). AAE is a general reference for measuring the amount of vitamin C contained in a material. In the calculation, sample 2 has an antioxidant activity of 25.01 mgAAE/mL extract, which is higher than sample 1, which is 22.88 mgAAE/mL extract, which means that each mL of the extract is comparable to 25.01 and 22.88 mg of ascorbate. So that in making Grecoli candy, it is recommended to use a 2:1 dose of grapes and broccoli to increase the body's immune system effectively.

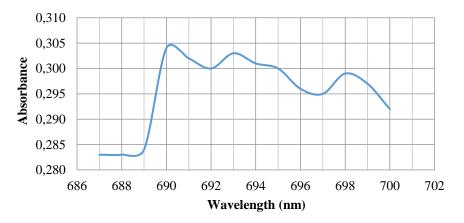


Figure 1. Graph of determining the maximum wavelength.

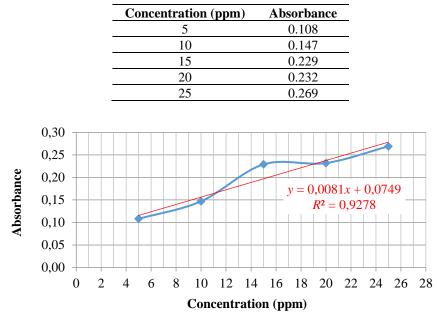


 Table 1. Standard solution absorbance measurement results.

Figure 2. Graph of the absorbance measurement of the buffer solution.

4. Conclusion

Grapes and broccoli contain vitamins that are good for the body, such as vitamin C, vitamin E, minerals and antioxidant effects. The antioxidants in these fruits and vegetables are essential for the body because they can eliminate free radical molecules in the body to prevent disease and improve the immune system. In the study of the antioxidant activity of Grecoli Candy using the FRAP method with a spectrometer, it was found that sample 2 extract was 25.01 mgAAE/mL, which was better for increasing antioxidant properties than sample 1, which was only 22.88 mgAAE/mL.

Acknowledgment

This research was supported financially by the PNBP Universitas Negeri Malang 2021.

References

- [1] World Health Organization, "WHO Coronavirus (COVID-19) Dashboard," World Health Organization. https://COVID19.who.int/ (Accessed on 27 November 2020).
- [2] L. Amalia and F. Hiola, "Analisis gejala klinis dan peningkatan kekebalan tubuh untuk mencegah penyakit COVID-19", *Jambura J. Health Sci. Res.*, vol. 2, no. 2, pp. 71 – 76, 2020, doi: 10.35971/jjhsr.v2i2.6134.
- [3] A. Adawiyah, T. Cahyanto, M. A. Salim, and D. Suparman, "Bioprospek microgreens sebagai agen antivirus dalam menghambat penyebaran coronavirus disease 2019 (COVID-19)," *Journal LP2M-Penulisan Karya Tulis Ilmiah Dosen selama WFH*, 2020. [Online]. Available: http://digilib.uinsgd.ac.id/id/eprint/30689
- [4] Y. Guan *et al.*, "Influence of cut type on quality, antioxidant substances and antioxidant activity of fresh-cut broccoli," *Int. J. Food Sci. Technol.*, vol. 55, no. 8, pp. 3019 3030, 2020, doi: 10.1111/ijfs.14567.
- [5] A. N. Azhar, M. Panirselvam, N. A. Amran, M. S. Ruslan, and S. Samsuri, "Retention of total phenolic content and antioxidant activity in the concentration of broccoli extract by progressive freeze concentration," *Int. J. Food Eng.*, vol. 16, no. 10, p. 20190237, 2020, doi: 10.1515/ijfe-2019-0237.
- [6] F. Cosme, T. Pinto, and A. Vilela, "Phenolic compounds and antioxidant activity in grape juices: A chemical and sensory view," *Beverages*, vol. 4, no. 1, p. 4010022, 2018, doi: 10.3390/beverages4010022.

- [7] K. Maesaroh, D. Kurnia, and J. Al Anshori, "Perbandingan metode uji aktivitas antioksidan DPPH, FRAP dan FIC terhadap asam askorbat, asam galat dan kuersetin," *Chimica et natura acta*, vol. 6, no. 2, pp. 93 – 100, 2018, doi: 10.24198/cna.v6.n2.19049.
- [8] P. Zimmermann and N. Curtis, "Factors that influence the immune response to vaccination," *Clin. Microbiol. Rev.*, vol. 32, no. 2, p. e00084-18, 2019, doi: 10.1128/CMR.00084-18.
- [9] D. Wu, E. D. Lewis, M. Pae, and S. N. Meydani, "Nutritional modulation of immune function: analysis of evidence, mechanisms, and clinical relevance," *Front. Immunol.*, vol. 9, p. 3160, 2019, doi: 10.3389/fimmu.2018.03160.
- [10] A. Maulana, T. Naid, D. T. Dharmawat, and M. Pratama, "Analisa aktivitas antioksidan ekstrak biji nangka (Artocarpus heterophyllus Lam) dengan metode FRAP (ferric reducing antioxidant power)," *bionature*, vol. 20, no. 1, 2019, doi: 10.35580/bionature.v20i1.9757.
- [11] W. Widyawati, U. A. Putri, and D. Lestari, "Daun Kopasanda sebagai tanaman alternatif penangkal radikal bebas," *J. Kesehatan*, vol. 14, no. 1, pp. 1–5, 2021, doi: 10.24252/kesehatan.v14i1.13365.
- [12] R. A. Wabula, S. Seniwati, and H. Widiastuti, "Aktivitas antioksidan ekstrak etanol buah merah (Pandanus conoideus Lam.) dengan metode FRAP," *Window of Health: Jurnal Kesehatan*, vol. 2, no. 4, pp. 329-337, 2019.
- [13] N. Awaluddin and S. Wahyuningsih, "Uji aktivitas antioksidan ekstrak metanol klika anak dara (Croton oblongus Burm) menggunakan metode DPPH," *J. Farmasi UIN Alauddin Makassar*, vol. 7, no. 2, pp. 38–45, 2019, doi: 10.24252/jurfar.v7i2.11578.
- [14] J. Sukweenadhi, O. Yunita, F. Setiawan, M. T. Siagian, and C. Avanti, "Antioxidant activity screening of seven Indonesian herbal extract," *Biodiversitas J. Biol. Diversity*, vol. 21, no. 5, pp. 2062–2067, 2020, doi: 10.13057/biodiv/d210532.
- [15] M. Pratama, A. Muflihunna, and N. Octaviani, "Analisis aktivitas antioksidan sediaan propolis yang beredar di kota Makassar dengan metode FRAP (ferric reducing antioxidant power)," J. Ilm. As-Syifaa, vol. 10, no. 1, pp. 11–18, 2018, doi: 10.33096/jifa.v10i1.312.
- [16] D. Raharjo and H. Haryoto, "Antioxidant activity of mangrove Sonneratia caseolaris L using the FRAP method," in *Proc. Int. Conf. Sci. Technol. Humanit.*, Universitas Muhammadiyah Surakarta, Surakarta, Indonesia, 2019, pp. 623–629.
- [17] F. Y. Halim, Y. Marsono, and M. M. Suprijono, "Identifikasi potensi antioksidan dalam minuman coklat dari kakao lindak (Theobroma Cacao L.) dengan berbagai cara preparasi: Metode ferric reducing antioxidant power (FRAP)," *J. Teknol. Pangan dan Gizi*, vol. 12, no. 1 pp. 10–16, 2017, doi: 10.33508/jtpg.v12i1.1476.
- [18] A. L. Puspita and S. Susilowati, "Aktivitas antioksidan fraksi daun pegagan (Centella asiatica (L) Urb.) dengan Metode FRAP," *IJMS-Indones. J. Med. Sci.*, vol. 8, no. 2, pp. 154–159, 2021.
- [19] K. Kusmiati, I. G. A. K. Wijaya, and Y. Yadi, "Potency test of antioxidant lutein of marigold flower (Tagetes erecta) extract yellow and orange color with FRAF and DPPH methods," In *Pros. Sem. Nas. Masyarakat Biodiversitas Indones.*, vol. 4, no. 2, 2018, pp. 274-279, doi: 10.13057/psnmbi/m040231.
- [20] R. Sroynak, P. Srikalong, and P. Raviyan, "Radical scavenging capacity and antioxidant activity of the vitamin E extracted from palm fatty acid distillate by sequential cooling hexane," J. Agric. Sci., vol. 5, no. 4, p. 224, 2013.