Trop J Nat Prod Res, July 2024; 8(7): 7734-7739 ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Short Communication

Antioxidant and Tyrosinase Inhibitory Activities of Unripe and Ripe Fruit and Seed Extracts of *Eugenia uniflora*

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ARTICLE INFO

Article history:
Received : 01 June 2024
Revised : 04 June 2024
Accepted : 12 July 2024
Published online 01 August 2024

ABSTRACT

Eugenia uniflora is widely distributed in Java island and used in traditional medicine for the treatment of many ailments. This study aims to compare the bioactivity of the seeds and pericarp of the unripe and ripe fruits of *E. uniflora*. The plant parts were extracted by maceration in ethanol. Qualitative and quantitative phytochemical analysis of the extracts was carried out according to standard methods. The antioxidant activity was performed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging and ferric reducing antioxidant power (FRAP) assays. The antiaging potential was assessed using the tyrosinase inhibitory assay. Phytochemical screening show the presence of alkaloids, flavonoids, and terpenoids in the seed and fruit extracts of *E. uniflora*. The seeds contain more total phenolic and flavonoid contents, as well as a higher antioxidant activity than the fruits. The unripe seeds had the highest antioxidant activity in the DPPH assay with IC_{50} value of 4.70 µg/mL, which was comparable to that of quercetin (IC_{50} = 4.76 µg/mL). In the FRAP assay, the ripe fruits exhibited the highest antioxidant activity with a FRAP value of 4245.32 µMFSE/100 g extract. The anti-tyrosinase activity assay showed that the unripe and ripe seeds of *E. uniflora* have weak tyrosinase inhibitory activity with IC_{50} of 309.96 and 539.67 µg/mL, respectively compared to kojic acid with IC_{50} of 27.54 µg/mL. In contrast, the unripe and ripe fruits showed no anti-tyrosinase activity. Therefore, *E. uniflora* seeds possesses higher potential as antioxidant and antiaging agent than *E. uniflora* fruits.

***Keywords*:** Antioxidant, Anti-tyrosinase, *Eugenia uniflora*, Seed extract, Fruit extract

Introduction

The skin is the most superficial layer of the human body. It is the largest organ that protects the body from harmful substances. However, the skin can experience complex biological phenomena involving persistent and unavoidable physiological changes that result in skin aging.[1–3] The skin tissue gradually loses its capacity to replenish or regenerate itself, retain its structure, and carry out its usual functions as it ages. Some people age with age, while others experience faster aging, referred to as early aging.[4–7] Skin aging is a result of intrinsic aging, which is subject to genetic factors associated with chronological age, and extrinsic aging, influenced by environmental factors such as UV exposure, smoking, chemicals, and gravity. One of the most essential factors in extrinsic aging is ultraviolet radiation (UV), which occurs in photoaging, where repeated exposure to sunlight can lead to reactive oxygen species (ROS) formation.[8–10]

Reactive oxygen species (ROS) contribute significantly to the dermal extracellular matrix changes brought about by intrinsic aging and photoaging from a molecular perspective.[11]

Biochemically, in the body, reactive oxygen species (ROS) can be generated through several mechanisms, encompassing the electron transport chain within mitochondria, proteins located in the endoplasmic reticulum, and enzymatic processes.[12,13] Under normal conditions without ligand, receptor tyrosine kinase (RTK) activity on the cellular membrane is suppressed by receptor protein tyrosine phosphatase (RPTP), which catalyzes the dephosphorylation of RTK. However, the cellular chromophore absorbs energy under UV radiation and becomes excited, generating oxidation products and ROS. In addition, the photoaging effect activates an increase in neutrophil elastase influx as a result of induction of the occurring angiogenesis so that the elastin network is degraded and triggers the appearance of wrinkles on the skin.[14,15] Another effect caused by photoaging is the activation of tyrosinase, which causes the formation of eumelanin and hyperpigmentation clinically associated with aging.[16–18]

Cosmetic products that contain antioxidants can reduce signs of skin aging. Recently, the back-to-nature trend has aroused consumer interest in products made from natural ingredients. Moreover, the antioxidants and other biological activities of natural ingredients can improve skin condition. The discovery and development of natural anti-aging ingredients, as well as efforts to find new active compounds that are efficacious, such as tyrosinase inhibitors and free radical scavengers, are good prospects. Therefore, there is the need to search for natural ingredients that could be developed into cosmetic products that would be of great benefits to the community.

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Citation: Wenas, DM, Elya, B, Sutriyo and Setiawan, H. Antioxidant and Tyrosinase Inhibitory Activities of Unripe and Ripe Fruit and Seed Extracts of *Eugenia uniflora*. Trop J Nat Prod Res. 2024; 8(7): 7734-7739 https://doi.org/10.26538/tjnpr/v8i7.16

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Eugenia uniflora is a medicinal plant that belongs to the Myrtaceae family. It originated from Brazil, but widely distributed in Indonesia, especially in the Java province. The multiple-branched shrub is known for its folkloric use in the treatment of stomach disorders, and inflammation.[19] The polyphenols and flavonoids found in *E. uniflora* have been shown to have antioxidant activity that prevents the breakdown of collagen due to free radicals. ROS, promoted by UV radiation, can damage DNA molecules and promote the production of tyrosine, making pigmentation faster than in normal aging.[20] Red colour fruits possess high antioxidant activity due to their abundance of secondary metabolites such as anthocyanins and carotenoids.[21] Anthocyanins have the potential to inhibit tyrosinase enzyme which plays important roles in melanin production and is usually responsible for skin pigmentation. Flavonoids also have potential skin-brightening activity as a tyrosinase inhibitor. The polyhydroxyphenolic structure of flavonoids enables them to function as metal chelators, specifically binding to copper ions in tyrosinase active site.[14] According to various researches, unripe fruits usually contained more phenolics and flavonoids than ripe fruits, and flavonoids in fruit seeds are more than the those in fruits.[15,22,23] Therefore, the fruits and seeds of *Eugenia uniflora* may be potential sources of skin-brightening agents that can be used in cosmetic products. The purpose of this study was to conduct a comparative analysis of the bioactivity of unripe fruit, ripe fruit, unripe seed, and ripe seed of *Eugenia uniflora* with respect to their antioxidant and tyrosinase inhibitory activities, and their total phenolics and flavonoids contents.

Materials and Methods

Chemicals and reagents

The reagents used were Trizma base (Sigma Aldrich No.T1503), L-DOPA (Sigma Aldrich SLBR 7591V), gallic acid, kojic acid, quercetin (Sigma Aldrich, India), Folin Ciocalteu Reagent (Merck, Germany), DPPH (2,2-diphenyl-1-picrylhydrazyl), TPTZ (2,4,6-tris(2-pyridyl)-s-triazine), ethanol 96%, acetic acid, dimethyl sulfoxide, ferric chloride hexahydrate, hydrochloric acid, aluminum chloride, potassium dihydrogen phosphate, methanol, sodium acetate, sodium hydroxide, and sodium carbonate.

Sample Preparation

The ripe and unripe fruits of *Eugenia uniflora* were harvested in September 2021, from Jarkata, West Java province, Indonesia (6° 21', 58.4903" S, 106° 51' 48.974" E). The plant samples were identified at the Herbarium Bogoriense, Botany Research Centre-BRIN Cibinong, with voucher number B-126/V/DI.05.07/9/2021. Green and yellow fruits were the unripe fruits, while orange and red fruits were the ripe fruits. The unripe and ripe fruit seeds were separated from the fruit pericarp. The seeds were air-dried at room temperature for 7 days. The fruits (peel and pulp) were freeze-dried and stored in a freezer. The plant materials (unripe fruit seed, ripe fruit seed, unripe fruit pericarp, and ripe fruit pericarp) were powdered, and 5-10 g each of the powdered sample were macerated seperately with 70% ethanol at a sample to solvent ratio of 1:10 for 18 hours. The extracts were removed by decantation, and the pulp were remacerated twice. The extracts were concentrated using a rotary evaporator (Büchi Rotavapor R-205, Germany). The concentrated extracts were thereafter kept in the refregirator at -4ºC until ready for use.

Phytochemical Screening

Qualitative phytochemical screening of the extracts were carried out to check for the presence or absence of secondary metabolites including alkaloids, flavonoids, saponins, tannins, and terpenoids according to standard methods. Terpenoids were identified by Liebermann Burchard's tests, alkaloids by Dragendorff's tests, tannins by the ferric chloride test, and flavonoids by aluminum chloride test.[24]

Determination of Total Phenolic Content

The total phenolic contents in the extracts of *E. uniflora* fruits and seeds were quantified by colorimetric method using Folin-Ciocalteu reagent, according to procedures previously described with slight modification.[25] In a 10 mL volumetric flask was mixed 1 mL of each extract solution, 5 mL of 7.5% Folin-Ciocalteu reagent. The mixture was incubated at room temperature for 8 min. A 4 mL volume of 1% sodium hydroxide solution was added to the mixture, and then incubated at room temperature for 1 h while protected from light. The absorbance of the resulting solution was measured at 730 nm using a UV-Vis Spectrometer (Model T80+, PG Instruments Ltd, UK). The experiment was done in triplicates. Gallic acid was used as the reference standard to produce gallic acid calibration curve. The total phenolic contents of the extracts was calculated from the linear regression equation of the gallic acid calibation curve. The total phenolic content for each extract was expressed as milligrams gallic acid equivalent per gram of dried extract (mg GAE/g extract).

Determination of Total Flavonoid Content

The total flavonoid content of each extract was determined using the Aluminium chloride colorimetric method as previously described.[26] To 0.5 mL of each extract solution was added 2.8 mL distilled water, 1.5 mL ethanol, 0.1 mL $AlCl_3$ (10%), and 0.1 mL Na_2CO_3 (1 M). The mixtures were vortexed, and then incubated at room temperature for 30 min while protected from light. The absorbance of the resulting solution was measured at 437 nm using a UV-Vis spectrophotometer (Model T80+,PG Instruments Ltd, UK). The experiment was conducted in triplicates. A standard calibration curve was constructed using quercetin at concentration range of 5-100 mg/L. The total flavonoid content was expressed as milligrams quercetin equivalent per gram of dried extract (mg QE/g extract).[26]

Determination of Antioxidant Activity

2,2-Diphenyl-1-picryl hydrazyl (DPPH) Radical Scavenging Assay

A methanol solution of DPPH (0.3 mM) was produced and combined with 5 mL of each extract. The mixtures were incubated at 25°C for 30 min in the dark. Following incubation, the absorbance of the solutions was measured at 517 nm. Gallic acid was used as the standard. The experiments was done in triplicates both for the sample and standard. The percentage inhibition of DPPH radical for each sample was computed using the equation (1), below.[26]

$$\%\ Inhibition = \frac{(control\ absorbance - extract\ absorbance)}{control\ absorbance}\ x\ 100 \quad [1]$$

The equation y = a + bx was derived from a linear regression curve with x denoting the extract concentration in µg/mL unit and y denoting the percentage inhibition (%). The IC_{50} value, which corresponds to the concentration needed to cause 50% inhibition of DPPH radical, was obtained using the equation (2) below.

$$IC50 = \frac{(50 - a)}{b} \quad [2]$$

Ferric Reducing Antioxidant Power (FRAP) Assay

The sample solution was mixed with 1 mL of FRAP reagent, and the volume was made up to 5 mL with methanol. The reaction mixture was incubated at room temperature in the dark for 30 min. The absorbance of the resulting solution was measured at 596 nm using a UV-Vis spectrophotometer. Iron (II) sulphate heptahydrate solution (10, 15, 20, 25, and 30 μM) was used to prepare the calibration curve. Quercetin was used as the positive control. The FRAP value is the amount of Fe^{3+} reduced to Fe^{2+} by the sample in the presence of $FeSO_4$. The FRAP value of the extracts and the positive control was determined using the equation (3) below.[27,28]

$$\text{FRAP value}\left(\mu\text{mol}\frac{\text{FeEAC}}{\text{g}}\right) = \frac{\text{C x V x Fp}}{\text{m}} \quad [3]$$

Where;
C = Sample concentration (µM)
V = Sample volume (mL)
Fp = Dilution factor
M = Sample Weight (mg)

The FRAP value was expressed as micromolar Ferrous sulphate equivalent per hundred grams of dried extract (µM FSE/100 g extract).

Tyrosinase Inhibitory Assay

The antiaging properties of *E. uniflora* extracts were assessed by enzymatic colorimetric method by measuring Tyrosinase inhibitory activity. The extract solution was mixed with 40 µL of tyrosinase (75 units/mL) in a 96-well plate containing 50 mM phosphate buffer pH 6.8 and 40 µL of L-dihydroxyphenylalanine substrate (4 mM). The reaction mixture was incubated at 25°C for 30 min. After incubation, the absorbance of the solution was read at 515 nm using a microplate reader Biotek Epoch 2 (Model No EPOCH2NSC, Agilent, US). The experiment was done in triplicates, and kojic acid was used as standard. The tyrosinase inhibitory activity of the extract was determined using the formula shown in equations 4 below.[29]

$$\text{Tyrosinase Inhibition (\%)} = \frac{(A-B)-(C-D)}{(A-B)} \times 100 \quad [4]$$

Where;
A = Absorbance of control with enzyme
B = Absorbance of control without enzyme
C = Absorbance of sample with enzyme
D = Absorbance of sample without enzyme

IC_{50} values were determined using the formula presented in equation 2.

Statistical analysis

All the assays were done in triplicates, and data was expressed as mean ± standard deviation (SD).
The data were analysed using Microsoft Excel 2018 software.

Results and Discussion

Extract yield

The colour of the fruits differentiate between the ripe and unripe fruits. Green and yellow colour indicate unripe fruits, while orange and red colour indicate ripe fruits (Figure 1). The percentage yields of the ethanol extracts of the unripe fruits and ripe fruits were 40.93% (w/w) and 37.39% (w/w), respectively, while that of the seeds of unripe fruits and the seeds of ripe fruits, were 16.72% (w/w) and 15.14% (w/w), respectively.

A B C

Figure 1: Fruits and seeds of *Eugenia uniflora*. A: Ripe Fruit (red, orange). B: Unripe Fruit (yellow, green). C: Seeds.

Phytochemical constituents of Eugenia uniflora extracts

The qualitative phytochemical analysis of *Eugenia uniflora* seed and fruit extracts showed a variation in the phytochemicals present in the different plant parts. Alkaloids, flavonoids, and terpenoids were present in both seeds and fruits, whereas, saponins was detected in the fruit only (Table 1).

Table 1: Phytochemical constituents of *Eugenia uniflora* extracts

Extract	Alkaloids	Flavonoids	Terpenoids	Saponins
Unripe seed	+	+	+	-
Ripe seed	+	+	+	-
Unripe fruit	+	+	+	+
Ripe fruit	+	+	+	+

+ indicate presence of phytochemical; - indicate absence of phytochemical.

Total phenolic and flavonoid contents of Eugenia uniflora extracts

The seed and fruit pericarp extracts of *Eugenia uniflora* were assessed for their total phenolic content (TPC) using gallic acid as standard. The linear regression equation derived from the gallic acid calibration curve was given as y = 0.0706x – 0.0145 with R-squared value of 0,9971 (Figure 2).

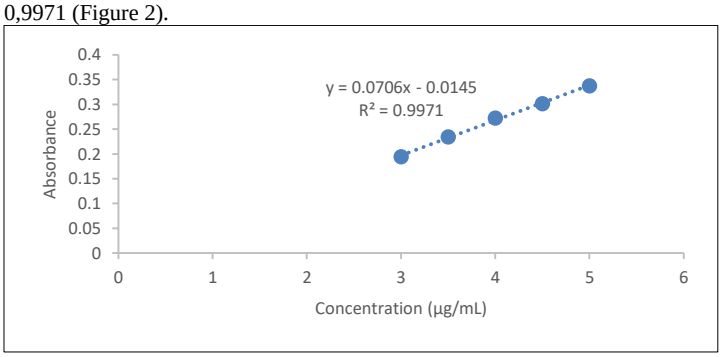

Figure 2: Gallic acid calibration curve for estimation of total phenolic content

Plant polyphenols reacts with Folin-Ciocalteu reagent to generate a complex with a blue colour the intensity of which is measured by UV-VIS spectrophotometer. The fundamental principle behind determining phenol content using the Folin Ciocalteu reagent involves the generation of a blue molybdenum compound by reducing phosphomolybdate phosphotungstate by phenolic compounds. The spectrophotometric measurement of the intensity of the blue colour allows for the quantification of the phenolic content. Phenolic content is usually expressed in terms of Gallic Acid Equivalent (GAE), which represents the amount of gallic acid in milligrams present in one gram of the test substance. The result of this study shows that the unripe seeds of *Eugenia uniflora* has the highest concentration of phenolic compounds (127.67 ± 16.76 mgGAE/g extract), followed by the ripe seed, unripe fruit, and ripe fruit (Table 2). The unripe fruit TPC was about two times that of the ripe fruit extract. This result is similar to that of Celli *et al*. (2011) which found that the green unripe fruit extract of *Eugenia uniflora* has more amount of total phenols than the mature fruit extract.[22]

The seed and fruit extracts of *E. uniflora* were assessed for their total flavonoid content using aluminum chloride colorimetric test. The aluminum chloride solution may attach to the carbonyl group (or hydroxyl group) and form a stable complex. Furthermore, it is plausible that labile acid complexes may also be formed with hydroxyl groups located in the ortho position within the B ring of flavonoids.[14] A calibration curve of quercetin standard was used to estimate the total flavonoid content in *E. uniflora* extract. The resulting curve has a linear regression equation of y = 0.0082x – 0.0402 with a coefficient of determination (R^2) value of 0,9997 (Figure 3).

The result from this study shows that the highest concentration of flavonoids (3.21 ± 0.09 mg QE/g extract) was found in the unripe seed. This was followed by ripe seed, unripe fruit, and ripe fruit (Table 2). This happens because flavonoids accumulate in the seeds of most plants, including *Eugenia uniflora*. This finding is also supported by that of Celli *et al.* (2011), who found that the total phenolic content of the immature green fruit extract of *Eugenia uniflora* was higher than that of the ripe red fruit extract.[22]

Table 2: Total phenolic and total flavonoid contents of *Eugenia uniflora* extracts

Extract	Total phenols (mgGAE/g dry extract)	Total flavonoids (mgQE/g dry extract)
Unripe seed	127.67 ± 16.76	3.21 ± 0.09
Ripe seed	115.46 ± 11.73	2.72 ± 0.04
Unripe fruit	23.96 ± 7.99	0.70 ± 0.04
Ripe fruit	10.48 ± 0.04	0.44 ± 0.01

Values are mean ± standard deviation (SD) of triplicates determination

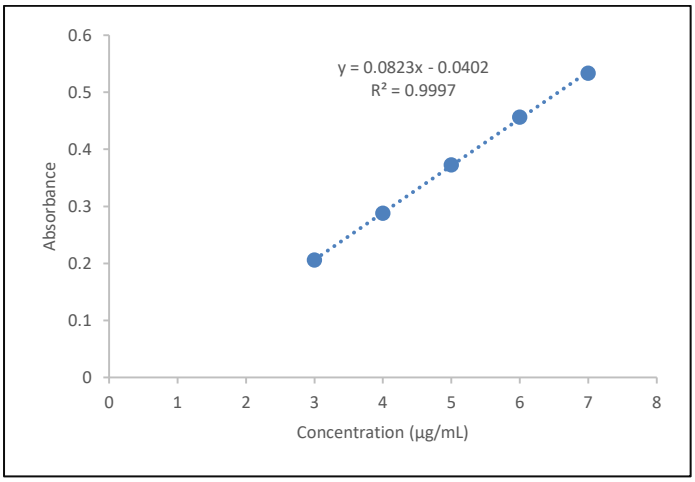

Figure 3: Quercetin calibration curve for estimation of total flavonoid content

Antioxidant Activity of Eugenia uniflora Extracts

The antioxidant activity of *Eugenia uniflora* extracts was assessed using DPPH radical scavenging activity and the ferric reducing antioxidant power (FRAP). Antioxidant activity assessment using the DPPH radical scavenging activity is based on the concept that DPPH concentration decreases linearly with a corresponding decrease in the intensity of the purple DPPH colour in the presence of antioxidant compounds. DPPH will receive hydrogen from the test antioxidant substance, forming a yellow reduced DPPH-H.[30] This approach is also based on the capacity of antioxidants to transfer electrons and counteract the unpaired electrons on the DPPH free radical. This process results in a change in colour from deep violet to yellow, accompanied by a decrease in absorbance value. The antioxidant activity of the test sample, specifically the ability to scavenge DPPH free radical, improves as the absorbance decreases.[31] DPPH free radical is present as a monomer and can dissolve in ethanol or methanol, but it is insoluble in water. In this study, the DPPH radical scavenging assay employs various concentrations of the test sample to determine the percentage of DPPH scavenging activity of each sample concentration. The result of the DPPH radical scavenging activity shows that the unripe seed extract has the highest antioxidant activity compared to the other extracts. In addition, the seed extracts have higher antioxidant activity than the fruit extracts, based on the IC_{50} values obtained. The IC_{50} value is the concentration of a test substance required to inhibit 50% of the radical activity under the experimental conditions. Antioxidant capacity can be classified into five levels: very high, high, medium, low, and very low. Antioxidants possess high potency when their IC_{50} value is below 50 µg/mL. Antioxidants are classified as strong if their IC_{50} value ranges from 50-100 µg/mL, moderate if the IC_{50} value falls within 100-150 µg/mL, and weak if the IC_{50} value ranges from 150-200 µg/mL. An IC_{50} value exceeding 200 µg/mL designates an antioxidant belonging to the category of extremely weak potency.[32]

For the FRAP assay, the antioxidant activity of *Eugenia uniflora* extracts was assessed using the FRAP value. the higher the FRAP value, the higher the antioxidant activity. The FRAP value was obtained from the equation of the calibration curve of ferrous sulfate heptahydrate, which was y = 0.0196x + 0.0225 with coefficient of determination (R^2) value of 0.9943 (Figure 4). The FRAP values for *Eugenia uniflora* unripe seed, ripe seed, unripe fruit, ripe fruit, and quercetin were 3207.91 ± 33.75, 4245.32 ± 19.48, 197.39 ± 1.28, 106.19 ± 3.98, and 1177.99 ± 39.11 µMFSE/100 g extract, respectively (Table 3).

Therefore, the unripe and ripe seed extracts of *Eugenia uniflora* have very strong antioxidant activity, while the unripe and ripe fruit extracts have moderate antioxidant activity. Phenolic compounds are known to be a major determants of the free radical scavenging ability of plants. Polyphenolic compounds have predominantly polar groups, rendering them more soluble in organic solvents than aqueous solvents. Therefore, the selection of the extraction solvent holds significant importance in the antioxidant activity of the extract.[33]

Flavonoids and phenolic compounds in the ethanol extracts of *Eugenia uniflora* seeds and fruits could contribute significantly to their strong antioxidant activity. Plants with a high concentration of phenolic compounds are recognized for their inherent ability to act as natural antioxidants. The high phenolic content may be attributed to the existence of flavonoids, which are prominent components of biologically potent medicinal plants. These findings are consistent with the results of the qualitative phytochemical screening. Flavonoids have various pharmacological benefits, such as antioxidant, antidiabetic, neuroprotective, antibacterial, and anti-inflammatory activities.[34] This result is supported by the study of Girardelo *et al*. (2020)[35], which proved that seed extract of *Eugenia involucrata* have stronger *in vitro* antioxidant capacity than the fruit extract. The seed extract of *Eugenia involucrata* was also found to have a higher phenolic content than the fruit extract.[35]

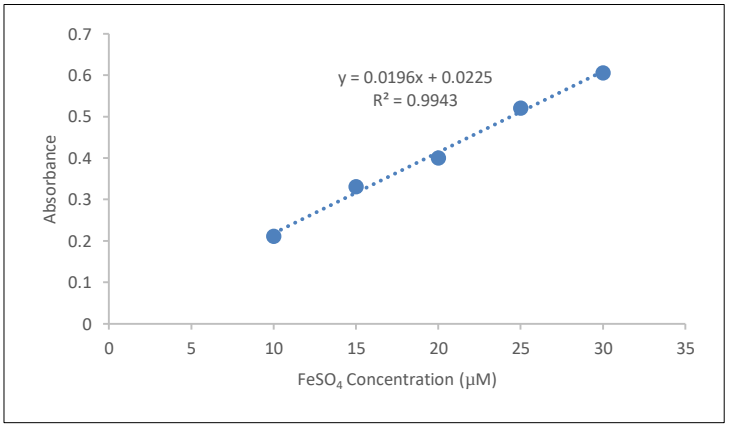

Figure 4: $FeSO_4$ calibration curve for FRAP antioxidant activity

Tyrosinase Inhibitory Activity of Eugenia uniflora Extracts

The tyrosinase inhibitory activity assay aims to evaluate the efficacy of *E. uniflora* extracts as antiaging agent. The potential of cosmetic antiaging agents can be assessed by evaluating their ability to inhibit tyrosinase during eumelanin synthesis. In tyrosinase inhibitory activity assay, mushroom tyrosinase is used as the enzyme, L-dihydroxyphenyl-alanine is used as the substrate, while kojic acid is used as a standard to determine tyrosinase inhibition.[29] Melanin synthesis involves the conversion of L-tyrosine to dopaquinone through hydroxylation and oxidation reactions facilitated by a tyrosinase enzyme that contains copper. This multifunctional tyrosinase enzyme is crucial in the process of melanin production. The enzyme plays significant role in the mechanism of melanin synthesis by facilitating two phases that determine the overall rate of the reaction. These steps include monophenolase activity which catalyzes the process of hydroxylation of L-tyrosine to generate L-dopa, followed by the subsequent oxidation of L-dopa to yield dopaquinone.[36]

The tyrosinase inhibitory activity assay results show that the IC_{50} of *E. uniflora* unripe seed extract was 309.96 ± 1.94 µg/mL, while that of the ripe seed extract was 539.67 ± 11.85 µg/mL (Table 3). The IC_{50} value is the concentration of an inhibitor required to inhibit 50% of the

enzyme activity under the experimental conditions. The tyrosinase inhibitory activity of the extract is considered significantly high when it exhibits an IC_{50} value less than 100 µg/mL. The tyrosinase inhibitory activity is categorized as weak if the IC_{50} value is 100 - 450 µg/mL, and 450 - 700 µg/mL is an extremely weak category of anti-tyrosinase activity.[37] Therefore, the unripe and ripe seed extracts of *Eugenia uniflora* are considered as weak tyrosinase inhibitor, while the unripe and ripe fruit extracts do not have tyrosinase inhibitory activity. The tyrosinase inhibitory activity of *E. uniflora* fruit and seed extracts has not been investigated in any previous study. However, some other *Eugenia* genera are able to inhibit the activity of tyrosinase enzyme, for example, the clove bud oil of *Eugenia caryophyllata*, and the leaf extract of *Eugenia dysenterica* have tyrosinase inhibitory activity with IC_{50} values of 8.2 µg/mL, and 11.88 µg/mL, respectively.[38,39]

Table 3: Antioxidant and tyrosinase inhibitory activities of *Eugenia uniflora* extracts

Sample	DPPH RSA (IC_{50} in µg/mL)	FRAP (FSE/100g Extract)	Tyrosinase Inhibitory Activity (IC_{50} in µg/mL)
Unripe seed	4.70 ± 0.16	3207.91 ± 33.75	309.96 ± 1.94
Ripe seed	5.23 ± 0.04	4245.32 ± 19.48	539.67 ± 11.85
Unripe fruit	78.58 ± 1.73	197.39 ± 1.28	> 1000
Ripe fruit	148.06 ± 2.33	106.19 ± 3.98	> 1000
Positive control	Q: 4.76 ± 0.03	Q: 1177.99 ± 39.11	KA : 27.54 ± 1.09

Values are mean ± standard deviation (SD) of triplicates determination. Q: Quercetin, KA: kojic acid

Conclusion

The seed extracts of *Eugenia uniflora* have been shown to have higher total phenolic and flavonoid contents than the fruit extract. Similarly, the antioxidant activity was higher in the seeds compared to the fruits. The tyrosinase inhibitory activity assay showed that *Eugenia uniflora* unripe and ripe seed extracts have weak tyrosinase inhibitory activity. In contrast, the unripe and ripe fruit extracts showed no anti-tyrosinase activity. These findings suggest that the seed extract of *E. uniflora* have better capacity as antioxidant and antiaging agent than the fruit extract.

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.

Acknowledgments

The authors are grateful to Head of Pharmacognosy Laboratory in Pharmacy Faculty University Indonesia for research facilities

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