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Antioxidant Activity Test Combination Inclusion of Curcumin-β-Cyclodextrin Complex and *Aloe vera*

Uji Aktivitas Antioksidan Kombinasi Kompleks Inklusi *Curcumin-β-Cyclodextrin* dan *Aloe vera*

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ABSTRACT

Free radicals occur from regular cellular metabolism. Free radicals are atoms or molecules with one or more unpaired electrons in their valence shell or outer orbit. Antioxidants are substances that are capable of neutralizing free radicals by contributing electrons to them. Curcumin is a polyphenolic molecule with numerous health benefits, including the capacity to combat free radicals as an antioxidant. *Aloe vera* is another plant known to contain a high concentration of bioactive compounds with antioxidant potential. Using the DPPH method, we evaluated the antioxidant capacity of the Curcumin-β-Cyclodextrin Complex and *Aloe vera* combination. According to the results of our investigation, the combination of curcumin-β-cyclodextrin and *Aloe vera* inclusion complexes has greater antioxidant capacity than the use of either component alone. However, additional research employing a variety of methodologies is required to determine the antioxidant potential of this combination.

Keywords: *Aloe vera*, antioxidant, curcumin

Introduction

Free radicals occur from regular cellular metabolism. Atoms or molecules with one or more unpaired electrons in their valence shell or outer orbit are considered free radicals. The odd number of free radicals is unstable, has a short lifespan, and is highly reactive. Due to their high reactivity, free radicals are known to abstract electrons from other compounds to achieve stability. Free radicals are quite dangerous because they can damage living cells (Phaniendra et al., 2015).

Antioxidants are substances that are capable of neutralizing free radicals by contributing electrons to them. Free radicals and antioxidant molecules can

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react immediately (Lü et al., 2010). Curcumin is a polyphenolic compound known to have many health benefits; one of its benefits is its ability as an antioxidant to ward off free radicals (Bassolino et al., 2022). Although it has good antioxidant activity, curcumin is known to have problems with its solubility. It is known that a complex inclusion method employing β-Cyclodextrin can solve this issue (Mangolim et al., 2014).

Aloe vera is a plant known to contain a large number of bioactive compounds with antioxidant potential. *Aloe vera* is known to contain compounds such as flavonoids, lectins, terpenoids, fatty acids, tannins, anthraquinones and sterols, as well as other compounds with biological activity that is beneficial to humans (Hęś et al., 2019). On the basis of the above information, we are interested in testing the antioxidant capacity of the combination of Curcumin-β-Cyclodextrin Complex and *Aloe vera*; we expect that in the future, this combination will be a good antioxidant option.

Research Method

Instruments and Materials

Instruments used in this study were aluminum foil (Klin pak), drop pipette (OneMed), electron microscopes (Hitachi High-Tech Group), freeze dryer BK-FD10S (Biobase), measuring cup 50 ml (Iwaki), micropipette 100-1000ul (Eppendorf), UV-Vis Spectrometer (Shimadzu 1800, Shimadzu).

Materials used in this study were *Aloe vera* (Sciyu Biotech., LTD, China), β-Cyclodextrin (Sciyu Biotech., LTD, China), Curcumin (Sciyu Biotech., LTD, Chin.), 0.45 µl filter paper (Whatman).

Complex Inclusion Method

The curcumin complex was included with cyclodextrin using the freeze-drying method. Examination of inclusion results was carried out with SEM (Scanning Electron Microscope). Freeze-drying was chosen because this method is a widely used complexation procedure and is well known for heating unstable guests. The inclusion complexes we performed used (1:1) in a 1:1 molar proportion. 1.38 g of -cyclodextrin is dissolved in 50 mL of water. 0.3680 grams of curcumin were added to the solution, which was then agitated. Following filtration with 0.45 m filter paper, the mixture was freeze-dried (Tanfil et al., 2021).

Antioxidant Test with DPPH Method

Using the DPPH technique, the radical-scavenging antioxidant activity of methanol extract was determined. Samples were diluted to 100 ppm, 110 ppm, 120 ppm, 130 ppm, and 140 ppm for each ratio (1:1). 0.1 mM DPPH was added to 2 mL of each sample. Following homogenization, the solution was incubated for 30 minutes at room temperature in the dark. Using a UV-Vis Spectrophotometer, the absorbance of this solution was then measured at a maximum wavelength of 517 nm. The same procedure was followed for the

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blank solution (DPPH solution without test material) and the Ascorbic Acid control with concentrations of 50 ppm, 100 ppm, 150 ppm, 200 ppm, and 250 ppm. 2 mL of 0.1 mM DPPH and 1 mL of methanol p.a. constituted the blank solution (Molyneux P, 2004).

Using the following equation, the absorbance measurement data were evaluated to determine the percentage of antioxidant activity (1).

$$\%\ Inhibition = \frac{\text{Absorbance Blank} - \text{Sample Absorbance}}{\text{Absorbance Blank}}\ x\ 100\% \quad (1)$$

Quercetin was used as a comparison with the same working method using concentrations of 50 ppm, 60 ppm, 70 ppm, 80 ppm, and 90 ppm.

Data Analysis

Using the linear regression method in Microsoft Excel 365, an analysis of the data was conducted.

Results and Discussion

Complex Inclusion

Results Our inclusion complex showed success after being observed using SEM (Figure 1). β-cyclodextrin has a structure that looks like crystal flakes in the sample; Curcumin shows the presence of spherical crystals that are very rod-like. However, neither crystalline flakes nor spherical crystals could be found in the inclusions of the Curcumin-β-cyclodextrin complex. These results are in agreement with the inclusion complexes we performed previously (Tanfil et al., 2021).

Antioxidant Activity

The DPPH method was used to evaluate the antioxidant activity of complex inclusions of curcumin-β-cyclodextrin using UV-Vis spectrophotometry with a maximum wavelength of 517 nm. The IC_{50} value, which is the concentration of sample solution required to inhibit 50% of DPPH free radicals, indicates the antioxidant activity.

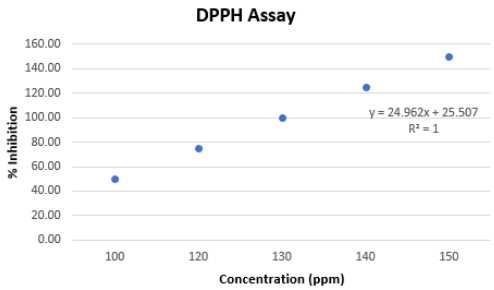

Figure 2. Curve of the correlation between the concentration of curcumin-β-cyclodextrin and the percentage of inhibition.

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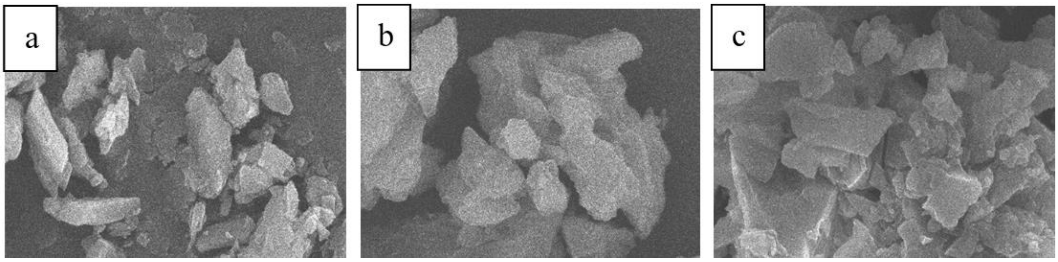

Figure 1. The scanning electron microscope photographs of (a) β-cyclodextrin, (b) curcumin, (c) complex inclusion of curcumin-β-cyclodextrin.

It was determined that curcumin-β-cyclodextrin and *Aloe vera*'s IC_{50} values were derived from a linear regression equation with the equation Y = ax + b, sample concentration (ppm) as the X axis and percentage inhibition value as the Y axis, from the relationship curve of sample concentration to percent inhibition. As depicted in Figure 2, the linear regression equation for the relationship between the curcumin-β-cyclodextrin combination with *Aloe vera* concentration and the inhibition percentage was used to calculate IC_{50} values of 2.98 ppm, 3.79 ppm; 3.79 ppm; 3.79 ppm; 4.19 ppm; 4.59 ppm; and 4.99 ppm.

Activation of antioxidants ranges from extremely strong to moderate to weak to very weak. More than half of all antioxidants have IC_{50} values of less than 50 ppm, while the rest have IC_{50} values of between 50 and 100 ppm, between 100 and 150 ppm, and more than 200 ppm for the weakest antioxidants, respectively (Molyneux P, 2004). When comparing the IC_{50} value of the Quercetin ratio with the IC_{50} value of curcumin-β-cyclodextrin obtained, it is known that the IC_{50} value of quercetin is higher.

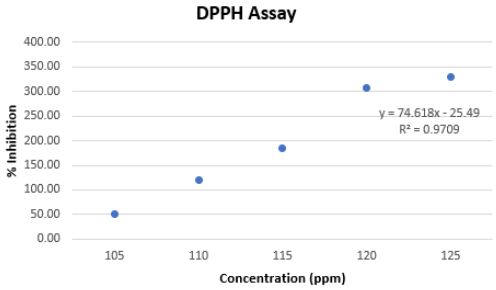

Figure 3. The correlation curve between the concentration of Quercetin and the percentage of inhibition.

This indicates that the antioxidant properties of curcumin-β-cyclodextrin are comparable when compared to quercetin. The concentrations used in the antioxidant quercetin test were 50 ppm, 60 ppm, 70 ppm, 80 ppm, and 90 ppm. Based on the linear regression equation from (Figure 3), which depicts the relationship between quercetin concentration and percentage of inhibition, the IC_{50} values obtained are 1.07 ppm, 1.27 ppm, 2.08 ppm, 3.71 ppm, and 4.07 ppm, respectively.

Conclusion

According to the results of our study, the combination of curcumin-β-cyclodextrin and *Aloe vera* inclusion complexes has greater antioxidant potential than the use of either component alone. However, additional research employing a variety of methodologies is required to determine the antioxidant potential of this combination.

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