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The potential of Endophytic Fungal Extract Isolated from Cinnamon (*Cinnamomum burmannii*) as Antidiabetic and Antioxidant

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Abstract

An increase in blood glucose levels in people with diabetes can cause an increase in free radicals, which can worsen the disease. Thus, drugs that have antidiabetic and antioxidant activities are needed. The cinnamon plant is high in antioxidants and has long been used as a source for a diabetes drug. The utilization of endophytic fungi isolated from cinnamon plants as antidiabetic and antioxidant has never been reported. This study aims to investigate the antidiabetic as well as antioxidant activity from the extract of endophytic fungi from the cinnamon plant. The antidiabetic activity was tested using the α-glucosidase enzyme inhibition method, while antioxidant activity was tested using the DPPH free radical scavenging method. Total phenol content was measured based on the Follin-Ciocalteu reagent reaction. All endophytic fungal extracts from the cinnamon leaves, twigs, flowers, and fruit have antidiabetic and antioxidant activity as well as high total phenol content. The three parameters measured showed a positive correlation. Endophytic fungal extract of Cb.D6 isolate derived from the leaf had the highest antidiabetic and antioxidant activity among the other isolates amounting to 92.41% and 90.28%, respectively. In addition, the total phenol content of Cb.D6 isolates was also the highest with 357.83 mg equivalent to gallic acid/g extract. Therefore, the endophytic fungal extract of Cb.D6 isolate has the potential to be developed as a source of the antidiabetic and antioxidant ingredients.

1. Introduction

Diabetes mellitus is a condition of increasing blood sugar levels to be above normal (hyperglycemia) caused by decreased insulin secretion and activity [1]. This is due to damage to the metabolic system that disrupts the metabolism of carbohydrates, fats, and proteins. This disease is one of the five main causes of death and affects more than 100 million people worldwide [2]. According to a report from the ministry of health written in Riskesdas, in 2018 as many as 16.5 million Indonesians aged over 15 years have been diagnosed with diabetes [3]. Diabetes has a close relationship with an increase in free radicals in the patient's body due to the existence of a glucose auto-oxidation process [4].

The pathogenicity of diabetes can be triggered by increased free radicals in the patient's body. Within normal limits, free radicals can be overcome by the body's natural defense system with the help of antioxidant compounds. However, in conditions of high blood glucose levels, endothelial cells will trigger an increase in the number of superoxide compounds so that the body's natural antioxidants are unable to reduce those radicals and can worsen diabetes itself [5]. Therefore, the search for natural ingredients that have antioxidant activity is a noteworthy topic at the current time because of its ability to protect the human body from attacks by several diseases caused by free radical reactions.

Search for chemical compositions that have the potential to be used as antidiabetic and antioxidants from

nature continue. Medicinal plants are a promising alternative because they are safer than synthetic drugs. Some medicinal plants have been used to cure diabetes and contain antioxidants. Water and ethanol extracts of cinnamon bark possess strong antioxidant activity by reducing free radicals with IC_{50} values of 3.03 µg/mL and 8.36 µg/mL, respectively [6]. These extracts hold antidiabetic activity through the inhibition of the α-glucosidase enzymes, each at 94.51% and 90.30%, respectively, with extract concentrations of 1.5% [7].

The efficacy of medicinal plants to cure a particular disease is closely related to the chemical compounds it contains. The presence of active compounds in medicinal plants is related to the existence of endophytic microbes in them. Endophytic fungi are part of the endophyte group microbes that live in the tissues of a host plant and are widely studied for their biological activity, including the inhibition of the α-glucosidase enzyme activity and their antioxidant content. Some endophytic fungi are reported to produce compounds that can inhibit the activity of the α-glucosidase enzyme, namely *Xylariaceae* sp. which can be isolated from the *Quercus gilva* plant [8]. In addition, *Pseudocercospora* sp. isolated from the *Elaeocarpus sylvestris* plant is reported to have antioxidant activity [9]. There are also endophytic fungi that have antidiabetic and antioxidant activity, namely *Penicillium pimiteouiense*, isolated from Simarouba glauca plant [10].

Thus far, the efficacy of cinnamon as a diabetes drug that has a high antioxidant activity is not accompanied by studies that report the ability of its endophytic fungi with the same activity. Moreover, research on endophytic fungi from cinnamon plants obtained from parts other than its bark has also not been widely reported. Hence, this study aims to determine antidiabetic activity through measuring the inhibitory ability of the α-glucosidase enzyme, antioxidant activity through the reduction of free radicals, and total phenol content from endophytic fungal extracts of some parts of the cinnamon plant originated from Bogor.

2. Research Methodology

2.1. Equipment and Materials

The tools used were Shaker incubator (Thermolyne, ROSI 1000), Autoclave (Tomy, ES-315), UV-VIS spectrophotometer (Hitachi, U-3900H), analytical balance (Precisa, 204A), Rotavator (Stuart, RE300DB), incubator (Grant, SE15), micropipette (Axygen), and Laminar Air Flow (ESCO, Airstream E-Series).

The material used in this study were samples of endophytic fungal isolates, namely Cb.Bn3, Cb.Bn4 isolated from flowers; Cb.Bh1, Cb.Bh2, Cb.Bh3, Cb.Bh5 and Cb.Bh6 isolated from fruits; Cb.D1, Cb.D3, Cb.D6, Cb.D7, and Cb.D8 isolated from leaves; and Cb.B1, Cb.B2, Cb.B5, Cb.B6, and Cb.B9 which were isolated from twigs of the cinnamon plant from Bogor, West Java, Indonesia. Chemicals such as technical ethyl acetate (Brataco), technical 70% ethanol (Brataco), Dimethyl sulfoxide 99% (Merck), *Potato Dextrose Agar* (BD-Difco), *Potato Dextrose Broth* (BD-Difco), α-glucosidase enzymes from *Saccharomyces cerevisiae* (Sigma), phosphate buffer pH 7 (Sigma), ρ-nitrophenyl-α-D-glucopyranoside (Sigma), sodium carbonate (Merck), 2,2-Diphenyl-1-picrylhydrazyl (Sigma), acarbose Glucobay (Bayer) , ascorbic acid (Sigma), and 99% methanol pro analysis (Merck) were also used.

2.2. Fermentation and Extraction of Endophytic Fungi

All endophytic fungi colonies were re-grown on Potato Dextrose Agar (PDA) media and incubated for seven days at room temperature. After seven days, two pieces of endophytic fungi isolates were taken from perforation with a sterile hole puncher 6 mm in diameter to be planted into 100 mL of Potato Dextrose Broth (PDB) fermentation media. Fermentation was carried out on a shaker at a speed of 120 rpm for 14 days at room temperature. After 14 days, the fermentation results were filtered with sterile filter paper in a vacuum Buchner funnel to separate the filtrate and biomass. The filtrate was then put into a separating funnel to be extracted with ethyl acetate three times. The ethyl acetate extract obtained was then concentrated using a rotary evaporator to obtain a dry extract of endophytic fungi ethyl acetate [11].

2.3. Antidiabetic Activity Test

A total of 1 mg of the α-glucosidase enzyme (9.8 units/mg) was dissolved in 1 mL of 0.01 M phosphate buffer (pH 7) as an enzyme stock solution (9.8 units/mL). A total of 0.02 mL of the enzyme stock solution was diluted by dissolving it into 5 mL in a 0.01 M phosphate buffer (pH 7) to obtain an enzyme working solution of 0.04 units/mL. The concentration of the test solution for ethyl acetate extract of endophytic fungi in DMSO was 100 µg/mL.

Table 1. Enzyme reaction system for a sample with a total volume of 2 mL

	Blank Sample (µL)	Control Sample (µL)	S0 (µL)	S1 (µL)
Sample	-	-	25	25
DMSO	25	25	-	-
Phosphate buffer (0,1 M)	475	475	475	475
Substrate	250	250	250	250
Phosphate buffer (0,01 M)	250	-	250	-
Enzyme	-	250	-	250
Na_2CO_3	1000	1000	1000	1000

A total of 475 µL of 0.1 M phosphate buffer (pH 7), 250 µL of 0.2 M ρ-nitrophenyl-α-D-glucopyranoside (ρNPG) substrate and 25 µL of each ethyl acetate extracted endophytic fungi samples were put into test tubes. The mixture in the test tube was then incubated at 37°C for 5 minutes, added with 250 µL of 0.04 unit/mL enzyme solution, and re-incubated at 37°C for 30 minutes. The enzyme reaction was stopped by adding 1000 µL of 0.2 M sodium carbonate solution (Table 1). The α-glucosidase inhibitory was determined by measuring the absorption of ρ-nitrophenol released from the ρNPG substrate at a wavelength of 400 nm using a UV-Vis spectrophotometer [12], which can be seen in equation (1).

$$\% \text{ inhibition} = [(C - S)/ C] \times 100\% \quad (1)$$

Descriptions:
C = control absorbance (DMSO) without sample (control with enzyme – blank without enzyme)
S = sample absorbance (S1 with enzyme – S0 without enzyme)

2.4. Antioxidant Activity Test

The antioxidant activity test was carried out using the DPPH free radical scavenging method [13] with wavelength modification from 515 nm to 517 nm. The concentration of endophytic fungi ethyl acetate extracts in methanol, as the test solution, was 100 µg/mL, and the 0.4 mM DPPH functioned as a blank sample. All samples of the test solutions and blank were incubated at 37°C for 30 minutes. The absorptions of all samples and blanks were then measured at a wavelength of 517 nm. Antioxidant activity is calculated using equation (2).

$$\% \text{ Inhibition} = (A-B)/ A \times 100\ \% \quad (2)$$

Descriptions:
A = blank absorbance
B = sample absorbance

2.5. Total Phenol Content Test

The endophytic fungal ethyl acetate extract concentration in ethanol was 1000 µg/mL. To test the total phenol content, gallic acid in distilled water used as a standard was made into a series of concentrations of 5; 10; 20; 40; and 80 µg/mL to create a standard curve. A total of 1 mL of extract (test sample) and gallic acid (standard) were put into separate test tubes, and each was added with 0.1 mL of Follin-Ciocalteu reagent and 0.9 mL of distilled water. Each mixture was then incubated in a dark place at room temperature for 5 minutes. To the mixture was added with 1 mL of 7% sodium carbonate and 0.4 mL of distilled water. It was incubated in a dark place at room temperature for 30 minutes. The mixture absorbance was measured at a wavelength of 765 nm. Total phenol content is expressed as mg gallic acid equivalent per gram of dry extract [14].

2.6. Data Analysis

Statistical tests were performed by one-way ANOVA ($P < 0.05$) and continued with Duncan's multiple range test ($P < 0.05$) to determine the significance between treatments. The relationship between variables, namely antidiabetic activity, antioxidants, and also total phenol content, were tested with Pearson correlation ($P \geq 0.05$).

3. Results and Discussion

3.1. Fermentation and extraction of endophytic fungi

The procurement of dry extracts from endophytic fungi filtrate from the cinnamon plant obtained the lowest value of 5.5 mg, i.e., the Cb.Bn3 endophytic fungi isolate, and the highest value of 84.6 mg of the Cb.Bh6 endophytic fungi isolate (Figure 1). Fermentation was carried out by the shake flask method so that aeration and agitation can be maintained. Aeration is needed to supply oxygen to endophytic fungi while agitation or stirring aims to increase oxygen supply in the medium, and increasing temperature homogeneity [15]. The harvesting of endophytic fungi was carried out during the stationary growth phase because, in general, fungi produce secondary metabolites in the stationary growth phase [16]. Secondary metabolites are released into the fermentation media, so endophytic fungi extract contains more secondary metabolite compounds, in terms of weight, than mycelia or biomass [17].

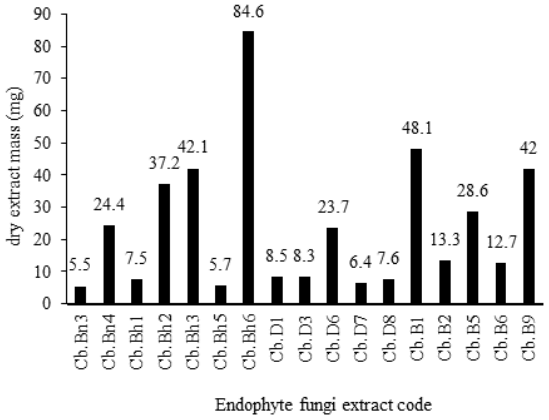

Figure 1. Dry mass (mg) of endophytic fungal extract from the cinnamon plant with Cb.Bn = endophytic fungi from flowers, Cb.Bh = endophytic fungi from fruits, Cb.D = endophytic fungi from leaves, Cb.B = endophytic fungi from bark

3.2. Antidiabetic activity test

All extracts at a concentration of 100 µg/mL showed the ability to inhibit the activity of the α-glucosidase enzyme. The percentage of inhibition varied between 1.03-92.41%, as shown in Table 2. The endophytic fungal extract of Cb.Bn4 has the lowest percentage inhibition value while Cb.D6 has the highest. On average, endophytic fungal extracts isolated from the barks of the cinnamon plant have higher antidiabetic activity.

Table 2. Inhibitory activity of α-glucosidase enzymes by endophytic fungal extracts of cinnamon plant

No.	Extract code	Percent Inhibition ±SD (%)
1	Cb.Bn3	$10,04^{f}$±0,35
2	Cb.Bn4	$1,03^{a}$±0,36
3	Cb.Bh1	$3,47^{c}$±0,18
4	Cb.Bh2	$13,13^{h}$±0,39
5	Cb.Bh3	$1,80^{b}$±0,01
6	Cb.Bh5	$11,84^{g}$±0,34
7	Cb.Bh6	$5,02^{d}$±0,19
8	Cb.D1	$12,10^{g}$±0,34
9	Cb.D3	$15,19^{i}$±0,03
10	Cb.D6	$92,41^{o}$±0,17
11	Cb.D7	$7,46^{e}$±0,01
12	Cb.D8	$15,96^{i}$±0,03
13	Cb.B1	$22,71^{j}$±0,55
14	Cb.B2	$72,82^{m}$±0,24
15	Cb.B5	$60,81^{l}$±0,03
16	Cb.B6	$24,89^{k}$±0,23
17	Cb.B9	$77,62^{n}$±0,53

Descriptions: Cb.Bn = endophytic fungi from flowers, Cb.Bh = endophytic fungi from fruits, Cb.D = endophytic fungi from leaves, Cb.B = endophytic fungi from bark. The numbers in the column followed by the same letter are not significantly different at the 0.05 level Duncan's multiple range test.

The inhibitory power on the α-glucosidase enzyme by the extracts can be grouped into three: strong, if it has a percentage inhibition of >90%; moderate, if it has a percentage inhibition of 60-90%; and low, if it has a percentage inhibition of <60% [18]. The inhibitions of the α-glucosidase enzyme by the endophytic fungal extracts from the cinnamon plant are distributed into all three categories. Endophytic fungal extract coded Cb.D6 is the only isolate in the strong category. The extracts Cb.B2, Cb.B5, and Cb.B9 are included in the category of moderate antidiabetic while the rest fall into the category of weak antidiabetic.

Carbohydrates that enter the digestive tract are converted into simpler sugars, which are then absorbed by the small intestine. In the in vitro antidiabetic test, the α-glucosidase enzyme was used. This is because the enzyme α-glucosidase is an enzyme found in the small intestine that is responsible for converting disaccharide carbohydrates into simpler monosaccharide carbohydrates [19]. Inhibition of the activity of α-glucosidase enzymes in the small intestine can reduce the breakdown of complex carbohydrates into glucose so that glucose level in the blood will decrease [20]. In in vitro testing, the α-glucosidase enzyme hydrolyzes ρ-nitrophenyl-α-D-glucopyranoside as a substrate to yellow ρ-nitrophenyl and glucose [21]. In vitro test results show that all endophytic fungal extracts have the ability to inhibit the activity of the α-glucosidase enzyme.

The inhibitory activity of the endophytic fungal extract with cinnamon bark as its host on the α-glucosidase enzyme activity is higher than the activity of the cinnamon bark extract alone. In this study, endophytic fungal extracts obtained from cinnamon bark (Cb.B) have inhibitory activity on α-glucosidase enzymes ranging from 22.71 to 77.62% with a test material concentration of only 0.01%. This can be considered to be higher when compared to water and 30% ethanol extracts of *Cinnamomum burmannii* bark with a concentration of 1.5%, which are reported to have the ability to inhibit the activity of α-glucosidase enzymes at 94.51 and 94.88% [7]. The α-glucosidase enzyme inhibition activity by endophytic fungal extracts of Cb.B2, Cb.B5, and Cb.B9 are better than the *Colletrotricum sp*. TSC13 endophytic fungal ethyl acetate extract isolated from other antidiabetic medicinal, *Taxus sumatrana*, which has an α-glucosidase enzyme inhibitory activity of 41.1% [22].

3.3. Antioxidant activity test

The results of antioxidant activity testing show that all extracts of cinnamon endophytic fungi at a concentration of 100 µg/mL have DPPH free radical scavenging activity (Table 3). The percentage of inhibition varies between 8.35-90.28%. The Cb.B1 extract has the lowest inhibitory activity, while the Cb.D6 extract has the highest.

Table 3. DPPH free radical scavenging activity by cinnamon endophytic fungal extracts

No.	Extract code	Percent inhibition ±SD (%)
1	Cb.Bn3	$43,87^{f}$±0,03
2	Cb.Bn4	$33,67^{g}$±0,03
3	Cb.Bh1	$35,58^{i}$±0,03
4	Cb.Bh2	$22,52^{d}$±0,02
5	Cb.Bh3	$27,77^{e}$±0,10
6	Cb.Bh5	$34,41^{h}$±0,03
7	Cb.Bh6	$21,88^{c}$±0,02
8	Cb.D1	$38,61^{j}$±0,10
9	Cb.D3	$42,91^{k}$±0,03
10	Cb.D6	$90,28^{q}$±0,07
11	Cb.D7	$32,40^{f}$±0,02
12	Cb.D8	$54,59^{n}$±0,04
13	Cb.B1	$8,35^{a}$±0,34
14	Cb.B2	$49,49^{m}$±0,13
15	Cb.B5	$82,88^{o}$±0,10
16	Cb.B6	$18,51^{b}$±0,16
17	Cb.B9	$88,87^{p}$±0,18

Descriptions: Cb.Bn = endophytic fungi from flowers, Cb.Bh = endophytic fungi from fruits, Cb.D = endophytic fungi from leaves, Cb.B = endophytic fungi from bark. The numbers in the column followed by the same letter are not significantly different at the 0.05 level Duncan's multiple range test.

The strengths of antioxidant activity can be classified as high if it has a percent inhibition of > 90%, medium if it has a 60-90% percent inhibition, and a low if it has a <60% percent inhibition [18]. The antioxidant activities of endophytic fungal extracts from the cinnamon plant fall into all classifications. The Cb.D6 extract is the only isolate that can be classified to have high antioxidant activity. Extracts Cb.B5 and Cb.B9 are classified as moderate antioxidants while the rest are included in the category of weak antioxidants. The antioxidant activity of endophytic fungal extract from cinnamon leaves with the highest DPPH free radical inhibition activity is 90.28%, higher than methanol extract of adult cinnamon leaves with inhibition of 74.49% at a concentration of 100 µg/mL. Different results are obtained from the endophytic fungal extract from the cinnamon bark. The highest free radical scavenging activity is 88.87%, lower than the methanol extract of cinnamon bark, which has inhibition of 91.49% at a concentration of 100 µg/mL [23].

The DPPH scavenging method is an in vitro antioxidant test method that is commonly used. The antioxidant effect observed in the DPPH free radical scavenging method is caused by the ability of a compound to donate hydrogen [24]. The DPPH free radical scavenging activity of Cb.D6 endophytic fungal ethyl acetate extract is better than the endophytic fungal extract of *Pestaloptiopsis sp.* isolated from another antioxidant medicinal plant, *Hugonia mystax*, which has a highest DPPH free radical scavenging activity of 88.53% [25].

3.4. Test of Total Phenol Content

The test results show that the total phenol contents of cinnamon endophytic fungal extracts vary between 56.00-357.83 mg equivalent to gallic acid/g extract (Table 4). The Cb.Bh6 endophytic fungal extract is the extract with the lowest total phenol content, while Cb.D6 is the extract with the highest total phenol content compared to other extracts. The ability of an endophytic fungus to give positive results on the test of antioxidant activity is correlated to the chemical compounds contained therein. One group of chemical compounds contained in endophytic fungi that is active as an antioxidant is the phenol group. Phenol compounds contained in some endophytic fungi from *Calotropis procera* plant are compounds that act as antioxidants [26].

The phenol content of endophytic fungal extract of Cb.D6 isolate is the highest compared to other isolates. This is directly proportional to its antioxidant ability, by reducing DPPH free radicals, and antidiabetic activity, by inhibiting the activity of the α-glucosidase enzyme. The phenol content (in mg equivalent gallic acid/g extract) in an endophytic fungal extract can be divided into several categories. Endophytic fungi with total phenol content <20 mg/g is categorized into the low category, 20-40 mg/g in the medium category, and > 40 mg/g in the high category [27]. Therefore, the total phenol contents of all isolates in this study are included in the high category because the phenol contents are > 40 mg/g.

Table 4. Total phenol content of cinnamon endophytic fungi extracts

No.	Extract Code	Total Phenol Content ± SD (mg gallic acid equivalent/g extract)
1	Cb.Bn3	$79,98^{e}$±0,71
2	Cb.Bn4	$87,17^{f}$±0,20
3	Cb.Bh1	$95,50^{h}$±0,71
4	Cb.Bh2	$69,36^{c}$±0,20
5	Cb.Bh3	$66,85^{b}$±0,59
6	Cb.Bh5	$86,94^{f}$±0,20
7	Cb.Bh6	$56,00^{a}$±0,20
8	Cb.D1	$100,30^{i}$±0,40
9	Cb.D3	$152,58^{m}$±0,71
10	Cb.D6	$357,83^{p}$±0,40
11	Cb.D7	$77,24^{d}$±0,40
12	Cb.D8	$104,63^{k}$±0,52
13	Cb.B1	$89,79^{g}$±1,23
14	Cb.B2	$110,11^{l}$±0,71
15	Cb.B5	$155,89^{n}$±0,59
16	Cb.B6	$102,92^{j}$±1,05
17	Cb.B9	$180,09^{o}$±1,58

Descriptions: Cb.Bn = endophytic fungi from flowers, Cb.Bh = endophytic fungi from fruits, Cb.D = endophytic fungi from leaves, Cb.B = endophytic fungi from bark. The numbers in the column followed by the same letter are not significantly different at the 0.05 level Duncan's multiple range test.

The highest phenol content of endophytic fungi extract from cinnamon (Cb.D6) in this study is still lower than the ethanol extract of *Cinnamomum zeylanicum* leaves, which had an average total phenol content of 3.326 mg equivalent to gallic acid/g extract [28]. In this study, each endophytic fungal extract has different antioxidant and antidiabetic abilities. The difference in the ability of endophytic fungal extracts from different plant parts in one plant is because each endophyte can produce compounds with different functions and amounts adapted to the role of endophytic fungi in the interactions with host plants [29].

3.5. Relationship between antidiabetic activity, antioxidant activity, and total phenol content

The relationships between antidiabetic activity with antioxidant activity and total phenol levels were analyzed using the Pearson correlation. The relationship between antidiabetic activity with antioxidant activity shows a positive correlation (Figure 2), where the higher the antidiabetic activity, the higher the antioxidant activity. The correlation between antidiabetic and antioxidant activities is strong with a value of r = 0.792, and it is significant with P <0.05.

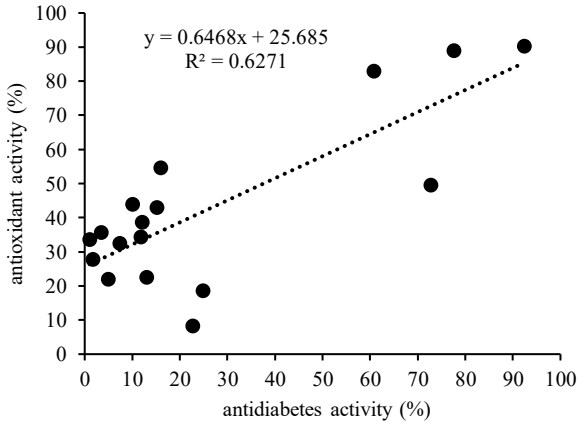

Figure 2. Relationship between antidiabetic activity and antioxidant activity

Diabetes type two is the most common case of diabetes found, with about 90% of the total cases of diabetes mellitus [30]. Diabetes is closely related to the presence of free radicals. Glucose and its metabolic products are known to undergo an auto-oxidation process and become free radicals after reacting with hydrogen peroxide [27]. A common thing that occurs in patients suffering from metabolic disorders, such as diabetes, is the occurrence of oxidative stress due to an imbalance between the formation of free radicals with the body's natural antioxidant defense mechanism [31]. Oxidative stress can also cause damage to blood vessels, especially in patients with acute diabetes. Further impacts that occur are β cell dysfunction and reduced insulin production [32]. Hence, comprehensive treatment by reducing oxidative stress and blood vessel damage can help in the prevention of complications due to type 2 diabetes [27].

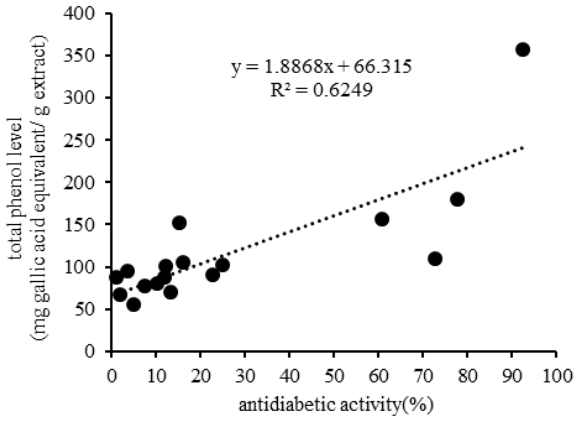

Figure 3. The relationship between antidiabetic activity with total phenol contents

The relationship between antidiabetic and antioxidant activities to the total phenol content both show a positive correlation (Figures 3 and 4), where the higher the antidiabetic and antioxidant activities, the higher the total phenol contents. The correlation between antidiabetic activity and antioxidant activity with total phenol content are both strong with r=0.791 and r=0.772, respectively. Both correlations are significant, with a P-value <0.05.

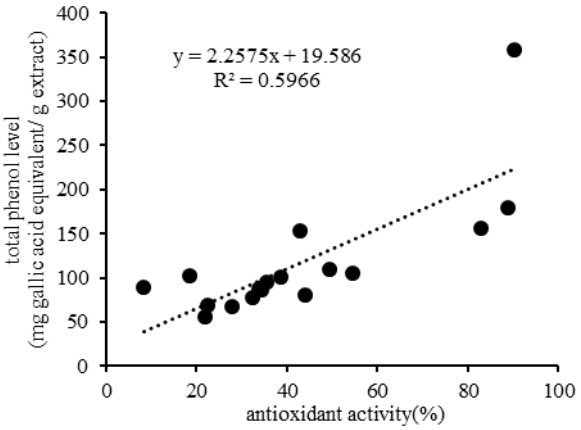

Figure 4. Graph of the relationship between antioxidant activity and total phenol contents

The results of this study are in line with previous studies, which stated that antioxidant and antidiabetic activities have a very close relationship with total phenol contents. The higher the total phenol content, the higher the antioxidant activity [30] and antidiabetic ability [33]. Phenol compounds are compounds that have long been known to have antioxidant and antidiabetic abilities. This ability is related to the structure of phenolic compounds, which are composed of hydroxyl groups that bind to aromatic hydrocarbon groups. The hydroxyl group in phenolic compounds has been reported to have the ability to significantly reduce free radicals by donating hydrogen atoms and an electron to a free radical hydroxyl group [34].

Additionally, the hydroxyl group in the phenolic compound can be effectively bound to the active site of the α-glucosidase enzyme. This bond allows the phenolic compound to donate hydrogen atoms to form hydrogen bonds with the active site of the enzyme in the mechanism of inhibiting the activity of the α-glucosidase enzyme [35]. Phenolic compounds have many types of compounds, such as flavonoids, phenolic acids, and so forth. The mechanism of phenolic compounds in reducing free radicals and inhibiting the activity of the α-glucosidase enzyme is relatively the same, namely by donating hydrogen atoms to radical compounds and the active site of the enzyme. Therefore, the higher the content of total phenolic compounds contained in the extract, the antioxidant and antidiabetic activities may be higher too.

In this study, each extract of endophytic fungi showcased different abilities in inhibiting the activity of α-glucosidase enzymes and antioxidant through reducing free radicals. This difference is due to the fact that each endophytic fungi can produce compounds with different functions and quantities adjusted to the role of endophytic fungi in the interactions with host plants [29]. Cb.D6 endophytic fungi extract exhibit a linear activity between the inhibition of α-glucosidase enzyme activity, reduction of DPPH free radicals, and also the best total phenol content among all extracts tested.

Although the extract of endophytic fungi isolate Cb.D6 sequestered from the leaves of cinnamon plant has the highest antidiabetic and antioxidant activities along with total phenol content, on average, the extract of endophytic fungi obtained from the barks have better activity and contents. This is possibly the reason that grounds the use of the barks and twigs for the treatment of diabetes. It is suspected that antidiabetic chemicals and antioxidants are more active in the bark than in other parts of the cinnamon plant. Purification of the active chemical compound produced by the best isolate of Cb.D6 is needed to increase the inhibitory activity of α-glucosidase enzymes and antioxidant to be developed as a source of new antidiabetic drugs.

4. Conclusion

All endophytic fungi extracts isolated from some parts of the cinnamon plant have antidiabetic and antioxidant activities related to their total phenol contents. Endophytic fungi extract of Cb.D6 isolates obtained from the leaves of cinnamon plant has the highest antidiabetic activity, antioxidant activity, and total phenol content among all endophytic fungi extracts tested.

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