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(RESEARCH ARTICLE)



Antioxidant and antidiabetic activities of cinnamon bark (*Cinnamomum burmannii*) Extracts *In Vitro*

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Abstract

Cinnamomum burmannii belongs to the Lauraceae family. This herbal plant can potentially lower blood glucose and ward off free radicals. It can be a candidate for diabetes mellitus therapy. This study aims to determine the phytochemical profiles and antioxidant and antidiabetic activities in vitro of *C. burmanii* powder as a basis for pharmacological testing of the bioactive compounds contained therein. This study took place in several stages, namely the manufacture of simplicia, extraction, phytochemical screening, and *in vitro* antioxidant and antidiabetic activity tests. The results showed that the yield of water solvent is $7.57 \pm 1.18\%$ and ethanol solvent is $6.67 \pm 0.39\%$. Phytochemical screening revealed the presence of flavonoids, saponins, tannins, terpenoids, alkaloids, and steroids. The cinnamon extract contained phenolic compounds of $116,79 \pm 2,84$ mg GAE/g extract in a higher amount than the Aqueous extract $69,37 \pm 2,84$ mg GAE/g extract. The antioxidant activity with Aqueous extracts was 68.86 ± 10.89 and $114,98 \pm 7.89$ ppm, and the inhibition of α -glucosidase activity of $478 \pm 17,78$ ppm of aqueous extract, $271 \pm 14,22$ ppm of Ethanol extract and acarbose $223 \pm 8,54$. Conclusion: *C. Burmanii* has a sweet taste, distinctive odor, and yellowish color. It contains flavonoid compounds, saponin, tannin, and terpenoid, and has antioxidant and antidiabetic activity.

Keywords: *Cinnamomum burmanii*; Phytochemistry; Phenol; Antioxidant activity; Inhibition α -glucosidase

1 Introduction

Diabetes is a degenerative disease characterized by high blood sugar levels (hyperglycemia) due to abnormalities in insulin secretion, insulin action, or both (1). Hyperglycemia is indicated by blood glucose levels above 200 mg/dl and fasting blood glucose above 126 mg/dl (2). An addition to increased blood sugar levels, diabetes due to oxidative stress. Oxidative stress is an imbalance between antioxidants and oxidants in the body (3). The incidence of diabetes continues to increase every year. Patients with diabetes mellitus 463 million, and will continue to grow to 700 people in 2045 Indonesia ranks seventh (4). With the high number of cases of Diabetes Mellitus in the world, the development of disease treatment has shifted to using natural or herbal ingredients. The choice of using herbal medicine is preferred over using synthetic drugs because herbal drug therapy has few side effects (5). Herbal plants that function to lower blood glucose are cinnamon (*Cinnamomum burmannii* Ness) of the genus *Cinnamomum*, Lauraceae family. Compounds in cinnamon that have potential as antidiabetics as Methylhydroxy Calcone Polymer (MHCP), cinnamaldehyde, and polymer procyanidin type-ApolymID Fers (6,7,8). Cinnamaldehyde in cinnamon can reduce the effects of oxidative stress in people with Diabetes Mellitus by counteracting free radicals (9). Antioxidants are antidotes to the adverse effects of oxidants, the mechanism of replenishing lost oxidant electrons so that they are stable and do not harm the body (10). In addition to antioxidants, α -glucosidase enzyme inhibitors also have the potential to reduce glucose in the blood (11). The enzyme can lessen carbohydrate hydrolysis and α -D glucose absorption so that postprandial blood glucose levels

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drop (12). Some studies show that ethanol extracts and water extracts can inhibit α -glucosidase enzymes by 94.88% and 94.51%, respectively (13). In addition, the results of a mixture of cinnamon (28%) with red betel leaves (42%), red ginger (15%), and lime (15%) have the potential as an α -glucosidase inhibitor of 88.7% (11). Based on this information, cinnamon extract has potential as a raw material for antidiabetic traditional medicine, so this study aims to determine the yield, total phenolic, DPPH free radical silencing, and α -glucosidase enzyme inhibitory activity in ethanol extract of cinnamon bark (*Cinnamomum burmannii*).

2 Material and methods

A *Cinnamomum burmannii*, methanol, Folin-Ciocalteu reagent 10%, Na CO₂ 10%, gallic acid, DPPH solution, ascorbic lacid, phosphate buffer 0.1 M pH 7.0, an α -glucosidase enzyme from *Bacillus stearothermophilus* (SIGMA), p-NPG powder, acarbose.

2.1 Extract preparation

Extraction of *Cinnamomum burmannii* by maceration method using water and ethanol solvents. A *Cinnamomum burmannii* (500 mg) was added solvent in a ratio of 1:4 (b:v). the solution was soaked for thirty minutes at a sonicator. The solution was shaken using a shaker incubator at room temperature for two hours. The solution was centrifuged, and the dissolved phase was filtered using Whatman paper no 1. The extract was calculated. as the yields.

$$\% \text{ Yield} = (\text{gram of extract} / \text{gram of cinnamon simplesia}) \times 100$$

2.2 Phytochemical Analysis

Phytochemical screening that includes qualitative analysis of tannins, alkaloids, flavonoids, saponins, and triterpenoids was carried out according to standard methods contained in *Materia Medika Indonesia*.

2.3 Determination of total phenolic content

Water extracts and ethanol extracts were each dissolved in water and ethanol solvents (1000 ppm). A sample (1 ml) was mixed with 4 mL of sodium carbonate solution (75 g/L) in a 10 mL volumetric flask and then shaken. Add Folin-Ciocalteu reagent (2mL) and distilled water. The mixture was allowed to stand in a dark room at room temperature for one hour. The absorbance was measured at a wavelength of 760 nm using a Shimadzu UV-160 spectrophotometer. A standard was using a gallic acid solution. Total phenolics obtained were expressed as gallic acid equivalents (GAE) in mg per gram of dry extract.

2.4 Antioxidant capacity analysis of the DPPH method

The free radical scavenging test was using the DPPH method. The several-concentration extract (2mL) was added to 1.0 mL of 0.5 mM DPPH in ethanol. The mixture was shaken and left at 25°C in the dark for 25 min. A blank solution for each sample solution by mixing 2 mL of sample solution and 1.0 mL of ethanol. As a negative control, 1.0 mL of 0.5 mM DPPH solution was added to 2.0 mL of ethanol. Absorbance (Abs) was measured at a wavelength of 518 nm using a spectrophotometer.

$$\% \text{ inhibition} = [1 - (\text{Abs sample} / \text{Abs control})] \times 100$$

The IC₅₀ value was calculated at the regression equation from the curve between percent inhibition versus concentrations.

2.5 Assay of α -glucosidase inhibitor activity

The reagent mixture used contained 50 μ L phosphate buffer 0.1 M (pH 7.0), 25 μ L 4-nitrophenyl α -D-glucopyranoside 0.5 mM, 10 μ L test samples at various concentrations (50-1000 ppm) and 25 μ L α -glucosidase solution (0.2 units/mL), and incubated at 37°C for 30 minutes. The reaction stopped by 100 μ L of 0.2 M sodium carbonate solution. Enzymatic hydrolysis of the substrate was calculated based on the amount of p-nitrophenol produced. The result for absorbance at 410 nm wavelength. Blank by replacing the enzyme with buffer. Controls used solvents (water or ethanol). Acarbose as a positive control. Percentage inhibition α -glucosidase calculated by the formula.

$$\% \text{ inhibition} = [1 - (\text{Abs sample} / \text{Abs control})] \times 100$$

The IC₅₀ value was calculated at the regression equation from the curve between percent inhibition versus concentrations.

2.6 Statistical analysis

All analyses are in triplicate. All results as mean \pm SD. Data analysis was performed using descriptive analysis and comparative analysis of mean values. Comparative analysis of mean values was performed using one sample t-test.

3 Results and discussion

3.1 Yield Extract *Cinnamomum burmanii*

Extraction of *Cinnamomum burmanii* with water solvent gives a yield of $7.57 \pm 1.18\%$ while for ethanol solvent gives a yield of $6.67 \pm 0.39\%$. Based on the one-sample t-test statistical test, there is a significant difference between the two types of solvents where extraction using water solvents provides a greater extract yield compared to ethanol solvents. The yield is the ratio between the dry weight of the extract of a sample and the weight of the raw material (simplicia) of a sample (14). Calculation of extract yield can determine how many bioactive components are found in an extract (9,15). Other studies show different results from this study. Research with the infusa method gave a yield of 18.83% (16,17). Another method using soxhlet with methanol solvent gave a yield of 9.8% (18). The difference in results is influenced by several factors such as temperature, extraction time, extraction method, and solvents used during extraction.

3.2 Phytochemical screening

The phytochemicals compounds of aqueous and ethanol extracts of *Cinnamomum burmanii* (Table 1). The phytochemical compounds in ethanol extracts alkaloids, tannins, phenolics, flavonoids, and triterpenoids. The aqueous extracts such as alkaloids, phenolics, and triterpenoids. All types of compounds are known to have antioxidant activity.

Table 1 Screening phytochemical of ethanol extract and aqueous of *Cinnamomum burmanii*

No	Test	Extract		Information
		Aqueous	Ethanol	
1	Alkaloid	+++	++	++++ very strong positive
2	Saponin	+	+	+++ strong positive
3	Tanin	+	++	++ positive
4	Phenolic	++	+++	+ weak positive
5	Flavonoid	-	+++	
6	Terpenoid	++++	++++	
7	Steroid	+	+	

3.3 Total phenolics

The results showed that the ethanol extract of the cinnamon extract contained phenolic compounds of $116,79 \pm 2,84$ mg GAE/g extract in a higher amount than the Aqueous extract $69,37 \pm 2,84$ mg GAE/g extract (Figure 1A). The polyphenolic compounds in plants are multifunctional in reducing and donating hydrogen atoms as antioxidants and absorbers of singlet oxygen formation. The nature of the polyphenol group has its ability as an antioxidant. A total phenolic in this study using Folin-Ciocalteu reagent. This method of the phenolic hydroxy group. All phenolic compounds can react with the Folin Ciocalteu reagent. The presence of aromatic in phenolic compounds (phenolic hydroxy groups) can reduce phosphomolybdate phosphotungstate to molybdenum are blue. Polyphenolic compounds have antioxidant activity. The antioxidant activity of polyphenolic compounds is the high activity as hydrogen or electron donors and the ability to stabilize or move unpaired electrons and chelate transition metals (19,20). Phenolic compounds are secondary metabolites in plants consisting of aromatic rings and one or more hydroxyl groups (21). The phenolic compounds have a function on the free radicals (22). The test to determine the total phenolic content in this study used the Folin-Ciocalteu method. Other studies have shown that the phenolic-hydroxyl group in the extract reacts with the Folin-Ciocalteu reagent to form a blue phosphotungstate-phosphomolybdate complex (23). The phenolic of the aqueous extract of *C. burmannii* with yields of 4.61%. The total phenolics in this study, namely 93-100 mg GAE g⁻¹(16), cinnamon methanol

extract (*C. burmannii*) results are 119 mg GAE / g extract (24). The difference is due to the use of different solvents. Phenolic levels in methanol extracts are higher than in aqueous extracts because methanol has hydroxyl groups (polar) and methyl groups (non-polar) (25). Research on *Eleutherine palmifolia* plants belongs to the genus Iridaceae. Species of the genus *Iris* (Iridaceae) have a long history in traditional medicine in the world and have long been known to contain many secondary metabolites, especially phenolic compounds such as flavonoids (26). In a study of ethanol extracts of *Eleutherine americana* grown in Thailand, the total phenol of the extract was 4.56 μmol gallic acid equivalent/g dry extract and could inhibit DPPH free radical activity with an IC₅₀ value of 8.4 $\mu\text{g/mL}$ (27). The relationship between total phenol content and antioxidant activity (28). The antioxidant activity determined by DPPH and ABTS methods of the 10 Croatian wines studied with their total phenol content with correlation coefficients of 0.9926 and 0.0938.

3.4 Antioxidant activity

The free radical scavenging was obtained with the standard antioxidant ascorbic acid (Table 2 and Figure 1B). The activity of the ethanol extract was greater than that of the water extract. The IC₅₀ is the concentration of a compound that causes 50% inhibition of a given system, and the DPPH free radical scavenging activity, the IC₅₀ value can be interpreted as the concentration of the extract that causes a 50% decrease in DPPH concentration from the initial DPPH concentration (29). The IC₅₀ for ethanol and Aqueous extracts were 68.86 \pm 10.89 and 114,98 \pm 7.89 ppm, respectively (Figure 1B). The effectiveness of phenolic compounds as antioxidants depends on their structure, hydrophobicity, biological activity, and oxidative activity. The ability of radical chain reactions by phenol compounds. The presence of two o-hydroxyl groups on the B ring. The formation of intramolecular hydrogen bonds between hydroxyl groups the stability of phenoxyl radicals (30). The Phenol compounds are a 3',4'-dihydroxy group on the B ring and/or have a 3-OH group next to the 4-keto have antiradical activity (31). In addition to phenol and polyphenol compounds, triterpenoid compounds have antioxidant activity (32). In diabetes mellitus, a strengthening of the production of Radical Oxygen Species results in the body experiencing oxidative stress (33). Oxidative stress is a condition in which the content of oxidants or free radicals in the body is more than antioxidants. Antioxidants play a role in preventing complications in DM patients. Non-enzymatic protection systems against oxidative stress can be vitamin C, vitamin E, carotenoids, and polyphenols. The Carotenoids such as free radical quenchers. Polyphenols can donate hydrogen atoms, free radical scavengers, and metal ion chelators(34).

Tabel 2 Antioxidant of standart, extract etanol, and aqueous of ekxtract etanolic *C. burmanii*

Antioksidan Activity(equivalent/gram extracts	Aqueous Extract	Etanol Extract
Mg ascorbic acid equivalent/g extract	68.86 \pm 10.89	114,98 \pm 7.89
Mg BHT equivalent/gram extract	114 41 25,57	215.32 20,82
Mg trolox equivalent/g extract	113.87 20.04	187.09 13.14

3.5 Inhibition of α -glucosidase activity

The inhibition of α -glucosidase activity of aqueous extract is 478 \pm 17,78, ethanol extract is 271 \pm 14,22 Ethanol extract, and acarbose is 223 \pm 8,54. The hypoglycemic potential of cinnamon bark extract was tested in vitro by α -glucosidase enzyme activity. The Acarbose is used as a comparator. The α -glucosidase enzyme inhibition is a concentration-dependent activity (Figure 1C). Determination of IC₅₀ values of aqueous extract, ethanol extract, and acarbose in inhibiting α -glucosidase enzyme activity (Figure 1D). Acarbose is an oligosaccharide compound that works at the brush border to break down dextrin, maltose, and sucrose to produce digestible monosaccharides (35). The α -glucosidase is an enzyme that catalyzes the cutting of glycosidic bonds in oligosaccharides (36). The phytochemical compounds can inhibit the work of α -glucosidase enzymes, such as compounds from the alkaloid group (37), triterpenes (38), and flavonoids (26). Phenolic compounds such as flavonols can inhibit α -glucosidase (34). Luteolin, myricetin, and quercetin (39). The α -glucosidase inhibitors work by mimicking the transition position of the pyranosidic unit of the natural glucosidase substrate (40), The higher activity of α -glucosidase enzyme inhibition in ethanol extracts is related to the content of phenolic compounds.

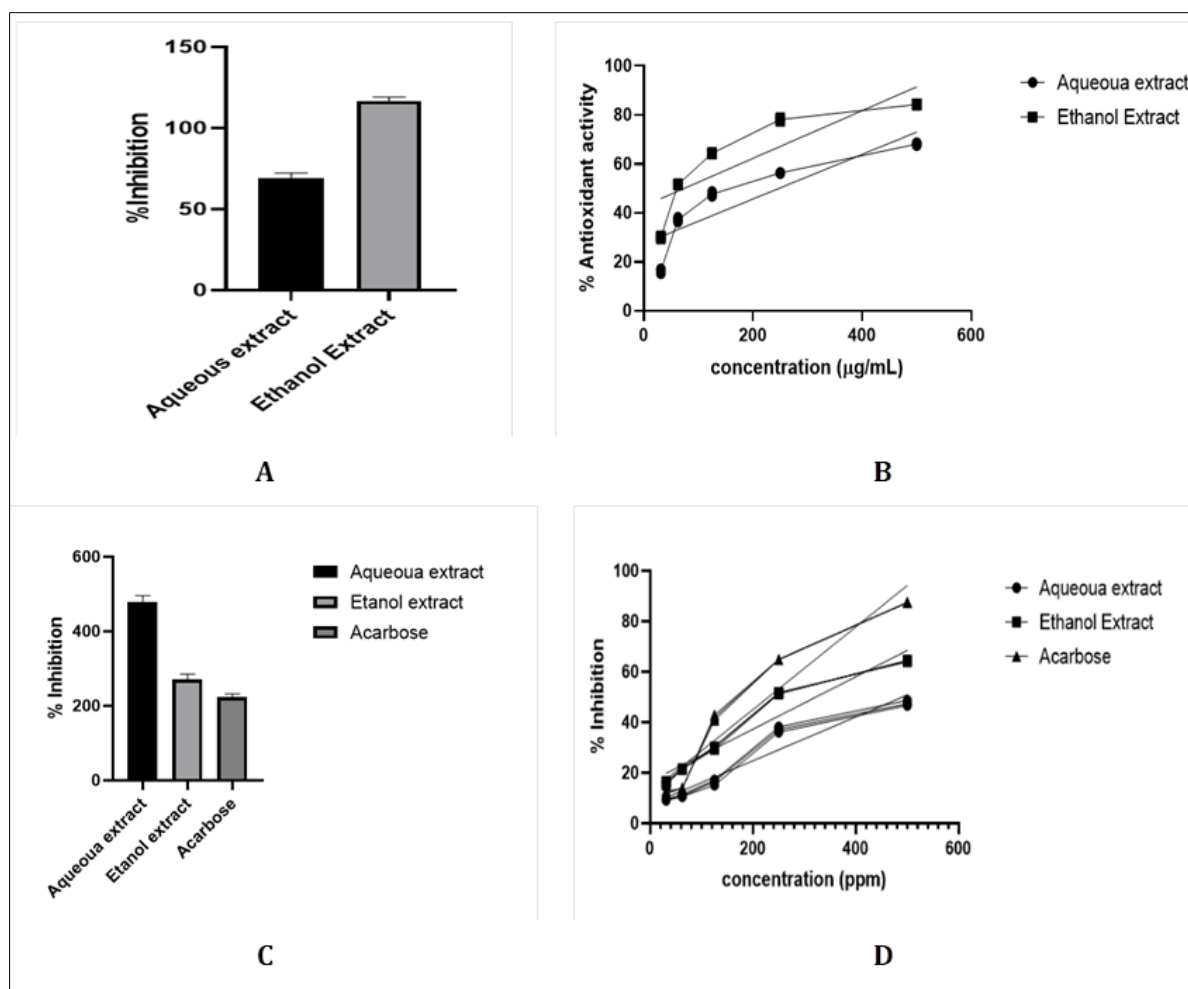


Figure 1 The Total Fenol, antioxidant activity n inhibition α -glukosidase of Ektrakt Aqueous and extract ethanol of *Cinnamomum burmanii*. A. Persen total henol, B, % antioxidant activity n % Inhibition α -glukosidase

4 Conclusion

Cinnamomum burmanii contains flavonoids, phenols, saponins, tannins, terpenoids, alkaloids, and steroids with total phenols in the aqueous extract of 69.37 ± 2.84 mg GAE/g extract. The antioxidant activity with Aqueous extracts is 68.86 ± 10.89 and 114.98 ± 7.89 ppm and has antioxidant and antidiabetic activity.

Compliance with ethical standards

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Disclosure of conflict of interest

All authors declare that they have no conflict of interest.

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