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Evaluation of α-Glucosidase Inhibitory and Antioxidant Activities of Tasmannia insipida Stem Extracts

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ABSTRACT : The rising prevalence of type 2 diabetes mellitus (T2DM) has prompted the search for safer and more effective antidiabetic agents, particularly from natural sources. Medicinal plants offer a rich reservoir of bioactive compounds with potential to reduce blood glucose levels and oxidative stress—two key factors in diabetes pathogenesis. This study aimed to evaluate the antidiabetic and antioxidant activities of water and ethanol extracts of *Tasmannia insipida* stems. The antidiabetic potential was assessed by measuring the inhibition on α-glucosidase. Antioxidant activity was evaluated using a phosphomolybdenum assay by an ascorbic acid standard curve. Both extracts inhibited α-glucosidase activity, with the ethanol extract (IC_{50} 3.14 µg/mL) showing significantly stronger inhibition than the water extract (IC_{50} 9.75 µg/mL). Similarly, the ethanol extract demonstrated higher total antioxidant capacity (56.29 mgAAE/g extract) in the phosphomolybdenum assay compared to water extract 7.31 mgAAE/g extract. *T. insipida* stems, particularly its ethanol extract, exhibits promising α-glucosidase inhibitory and antioxidant activities, supporting its potential as a natural source of antidiabetic agents. Further phytochemical characterization and *in vivo* studies are recommended to explore its therapeutic value.

KEYWORDS : Ascorbic acid, ethanol extract, phosphomolydenum, stem, water extract.

I. INTRODUCTION

Type 2 diabetes mellitus (T2DM) is a progressive metabolic disorder characterized by chronic hyperglycemia resulting from insulin resistance and/or impaired insulin secretion. The global prevalence of T2DM has increased dramatically over the past decades, with 822 million people living with T2DM [1], making it a major public health concern [2]. Despite the availability of various pharmacological treatments, current antidiabetic drugs are often associated with side effects such as gastrointestinal disturbances, hypoglycemia, and long-term toxicity [3]. Moreover, some patients exhibit poor adherence or resistance to conventional therapies, highlighting the need for alternative, safer, and more accessible treatment options. Medicinal plants have long been used in traditional medicine systems for managing diabetes, particularly in rural and resource-limited settings. These plants are rich in bioactive compounds such as flavonoids, phenolics, and alkaloids, which may act through various mechanisms—including inhibition of carbohydrate-digesting enzymes, enhancement of insulin sensitivity, and reduction of oxidative stress. Investigating the antidiabetic activity of plant extracts not only supports the validation of traditional knowledge but also contributes to the discovery of novel therapeutic agents.

One promising target for antidiabetic screening is the enzyme α-glucosidase, which plays a key role in carbohydrate digestion by breaking down disaccharides into glucose in the small intestine [4]. Inhibiting this enzyme slows glucose absorption, thereby reducing postprandial blood glucose spikes. Currently used α-glucosidase inhibitors, such as acarbose, can cause adverse gastrointestinal effects, which has driven the search for natural inhibitors from plant sources [4].

In addition to enzyme inhibition, oxidative stress is closely linked to the pathogenesis of diabetes [5]. Excessive production of reactive oxygen species (ROS) can impair insulin secretion, damage pancreatic β-cells, and contribute to diabetic complications [5]. Thus, evaluating the antioxidant activity of plant extracts is relevant to their potential antidiabetic effect. *Tasmannia insipida*, a plant native to Australia and belonging to the Winteraceae family, is known for its aromatic and medicinal properties [6]. While various parts of related *Tasmannia* species have been studied, the pharmacological activity of the stems of *T. insipida* remains largely unexplored. This study aimed to evaluate the antidiabetic potential of water and ethanol extracts of *T. insipida* stems through α-glucosidase inhibition, alongside their total antioxidant capacity as measured by the phosphomolybdenum assay. The findings may contribute to identifying natural compounds with dual antidiabetic and antioxidant activity.

II. METHODS

2.1. Chemicals and plant materials : The substrate *p*-nitrophenyl-α-D glucopyranoside and α-glucosidase (EC 3.2.1.20) from *Saccharomyces cerevisiae* were bought from Sigma-Aldrich (St. Louis, MO, USA). Sodium phosphate and ammonium molybdate were bought from Merck.

Stems of *T. insipida* was bought from a local shop in Jakarta, Indonesia. Dried stem was pulverized into course powder and used for extraction.

2.2. Plant extraction : Ethanol extract of *T. insipida* was prepared by maceration of pulverized stem (10 g) in 100 mL ethanol overnight at ambient temperature. The filtrate was filtered through a Whatman no 1 and the residue was re-macerated twice. The filtrate was pooled and evaporated by a rotary evaporator to obtain dried extract. Test solution was prepared by dissolving dried extract in ethanol. Water extract was prepared by decoction procedure. Pulverized stem (10 g) was soaked in 100 mL boiled water. The mixture was left to cool at room temperature. The residue was filtered. The filtrate was freeze-dried to obtain dried extract. Test solution was prepared by dissolving dried extract in water.

2.3. Determination of α-glucosidase inhibition activity : The α-glucosidase inhibition activity of *T. insipida* extracts was determined based on our method [7]. Extracts of different concentration (50 µL) was added with 50 µL of phosphate buffer (50 mM, pH 6.8) and α-glucosidase (0.5 U/mL, 50 µL). The mixture was incubated at 37 °C for five minutes. Thereafter, 100 µL of 1 mM substrate (p-nitrophenyl-α-D glucopyranoside) was added and the reaction mixture was further incubated for 20 minutes. The reaction was terminated by the addition of Na_2CO_3 (100 mM, 750 µL). The absorbance was read at 405 nm. Inhibition percentage was plotted against extract concentrations to obtain the linier regression equation. The activity was presented as IC_{50} values, which were calculated based on the regression equation. Acarbose was tested as a positive control.

2.4. Determination of antioxidant activity by phosphomolybdenum assay : The antioxidant activity of *T. insipida* extracts was determined based on a phosphomolybdenum assay by our method [8]. Phosphomolybdenum reagent was prepared by mixing 100 mL of each solution of 0.6 M H_2SO_4, 0.028 M Na_3PO_4, and 0.004 M ammonium molybdate. In a capped tube, extracts (0.3 mL) was mixed with phosphomolybdenum reagent (3 mL). The mixture was incubated in a hot water bath (95 °C) for 90 minutes. The absorbance of the bluish green solution was read at 695 nm. A calibration curve was generated using ascorbic acid (12.5 – 400 µg/mL). Activity was presented as mg ascorbic acid equivalent (mgAAE/g dried extract).

III. RESULTS AND DISCUSSION

3.1. α-Glucosidase Inhibitory Activity : The α-glucosidase inhibitory activity of *T. insipida* stem extracts was evaluated to assess their potential antidiabetic properties. Intestinal α-glucosidase which is produced at the brush border of the small intestine plays a key role in carbohydrate digestion. The enzyme helps break down complex polysaccharides into glucose [4]. Thus, inhibition on this enzyme delays glucose absorption in the intestine, and in consequence, lower post prandial blood glucose levels.

Table 1 Inhibition on the α-glucosidase by *T. insipida* extracts.

T. insipida extracts	Concentration (µg/mL)	Inhibition (%)	IC_{50} (µg/mL)
Ethanol extract	6.25	91.61±3.05	3.14±0.13
	3.13	59.93±7.09	
	1.56	23.58±1.57	
	0.78	8.62±1.09	
Water extract	29.29	89.86±1.89	9.75±0.19
	14.65	71.39±4.08	
	3.66	32.18±2.74	
	1.83	36.85±1.56	
Acarbose			99.79±2.75

Results can be seen in Table 1. Both ethanol and water extracts demonstrated the ability to inhibit α-glucosidase, with the ethanol extract (IC_{50} 3.14±0.13 µg/mL) exhibiting notably stronger inhibition than the water extract (IC_{50} 9.75±0.19 µg/mL). In both extracts, a dose dependent trend was observed.

Inhibition percentage (%) increased linearly by the increase of extract concentrations, which was confirmed by R^2 values closed to whole number *i.e*. 0.9568 and 0.9226 for ethanol and water extracts, respectively. With regards to the sensitivity of inhibition, the ethanol extract obtained steeper slope value of the regression equation than water extract, which suggests higher susceptibility to inhibition of the enzyme to the ethanol extract than water extract.

To date, there is no report yet on the α-glucosidase inhibition by *T. insipida* extract. However, leaf extract of other species *i.e. T. lanceolata* with IC_{50} 0.83 mg/mL [9]. Results in the present study suggests that the ethanol extract contains a higher concentration or broader range of bioactive compounds capable of interfering with the enzyme's activity. The IC_{50} value of the ethanol extract was significantly lower than that of the water extract, indicating greater potency. Acarbose, which is the first line drug of antidiabetic agent, under the same experimental conditions obtained an IC_{50} of 99.79±2.75 µg/mL. Studies reported the effective effect of acarbose in inhibiting α-glucosidase [4]. In addition, acarbose also enhances insulin sensitivity and promotes glucagon-like peptide 1 (GLP-1), which aids in the management of obesity [10].

The higher activity of the ethanol extract may be attributed to its ability to dissolve more lipophilic phenolic compounds, flavonoids, or other secondary metabolites that are less soluble in water [11]. These compounds are known to interact with the active site of α-glucosidase, inhibiting carbohydrate breakdown and potentially reducing postprandial blood glucose spikes. This result is consistent with previous studies reporting stronger α-glucosidase inhibitory activity in ethanol or methanol extracts of other medicinal plants, as compared to their water extract [11, 12].

3.2. Antioxidant activity : In this study, antioxidant activity of *T. insipida* extract was evaluated using a phosphomolybdenum assay, which measures the total antioxidant capacity of the sample based on its ability to reduce Mo(VI) to Mo(V). This assay has been used to determine antioxidant activity of plant extracts [13]. Assessment of antioxidant activity of lipophilic extract was specifically reported and its comparison with β-carotene linoleate assay [14]. It has been reported that results by phosphomolybdenum assay was consistent with those of by DPPH and ABTS assays [13]. All these assays are particularly suitable for initial antioxidant screening, especially when the extract contains a complex mixture of phytochemicals.

For the determination of antioxidant activity by the phosphomolybdenum assay, a calibration curve using ascorbic acid as a standard antioxidant was generated (Figure 1). The curve obtained good linearity between ascorbic acid concentrations and antioxidant response, with R^2 = 0.9997 within the range of ascorbic acid concentrations of 12.5 – 400 µg/mL (Figure 1). Results are expressed as milligrams of ascorbic acid equivalent (AAE) per gram extract. Among the two extracts tested, the ethanol extract exhibited significantly higher total antioxidant activity with 56.29±1.85 mgAAE/g extract than the water extract with 7.31±0.06 mgAAE/g extract. The observed activity suggests the presence of compounds in *T. insipida* stem extracts capable of contributing to redox balance. In the previous study, *T. insipida* leaf extract was reported to have DPPH radical scavenging activity [6]. Crude plant extracts, such as what were prepared in the current study, may contain diverse phytochemicals including phenolics, flavonoids, saponin, which may act through multiple antioxidant mechanisms. This suggests that ethanol, as an organic solvent with intermediate polarity, was more effective in extracting antioxidant compounds such as flavonoids, phenolic acids, and other polyphenolic constituents. These compounds are known for their reducing power and ability to donate electrons in redox reactions, which directly contributes to the reduction of Mo(VI) to Mo(V) in the assay.

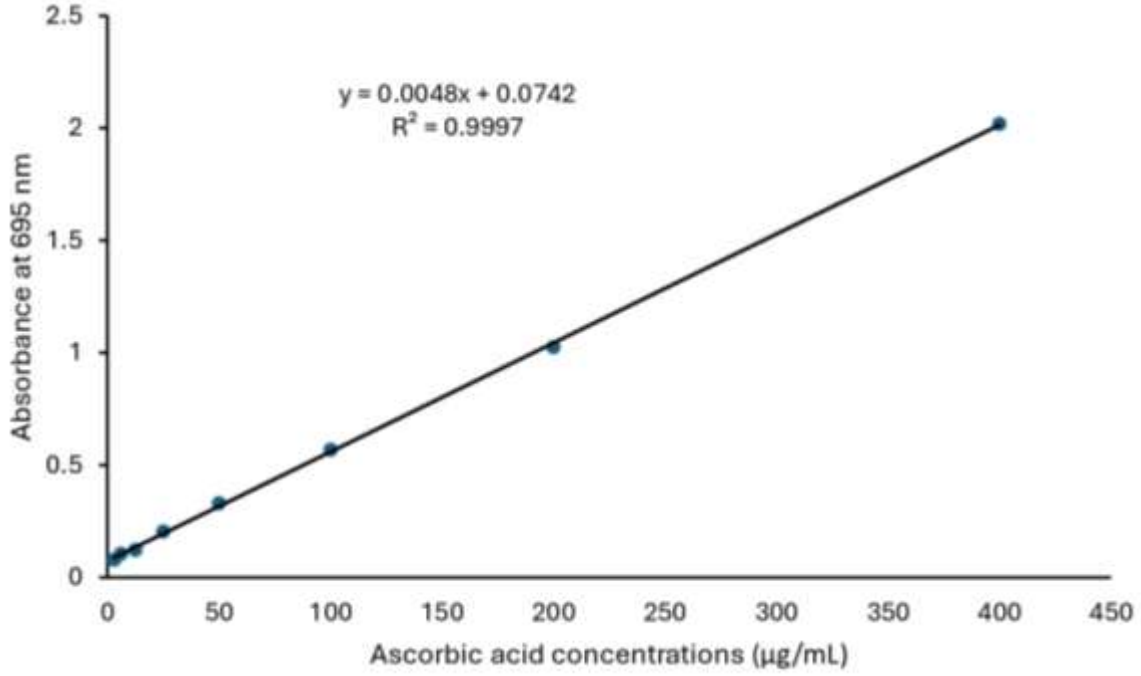

Figure 1 Ascorbic acid calibration curve of the Phosphomolybdenum assay.

The antioxidant activity of the ethanol extract, as revealed by the phosphomolybdenum assay, may contribute to its antidiabetic potential. Since oxidative stress is a key contributor to pancreatic β-cell dysfunction and insulin resistance [15, 16], antioxidants play a protective role in diabetes management. In addition, the antioxidant activity may help protect tissue from oxidative damage. Thus, the dual activity of *T. insipida* ethanol extract—both as an α-glucosidase inhibitor and as an antioxidant—supports its potential as a potential candidate for preventing or managing type 2 diabetes. These findings underscore the value of ethanol extraction for isolating bioactive compounds from *T. insipida* stems and highlight the need for further phytochemical characterization to identify the specific compounds responsible for these effects.

IV. CONCLUSION

The present study demonstrates that the stem extracts of *T. insipida*, particularly the ethanol extract, possess promising α-glucosidase inhibitory and antioxidant activities. The ethanol extract exhibited stronger enzyme inhibition and higher total antioxidant capacity compared to the water extract, likely due to the presence of more bioactive compounds soluble in ethanol. These findings suggest that *T. insipida* stem could serve as a potential natural source of antidiabetic agents, with added antioxidant benefits that may help mitigate oxidative stress associated with diabetes. Further studies, including phytochemical characterization and in vivo evaluations, are recommended to confirm these in vitro findings and explore the therapeutic potential of this plant.

onclusion section must be included and should indicate clearly the advantages, limitations, and possible applications of the paper. Although a conclusion may review the main points of the paper, do not replicate the abstract as the conclusion. A conclusion might elaborate on the importance of the work or suggest applications and extensions.

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Biography

Adelina Simamora is a researcher in the center for enzymes in health and diseases studies and a lecturer in biochemistry at the Faculty of Medicine and Health Sciences, Krida Wacana Christian University. At present, her research focus on the bioactive potential of medicinal plants, with special interest in green extraction. Currently, her work involves various Indonesian medicinal plants and

inhibition properties on various enzymes. Her interest also spans to transition and rare earth metals and their application in living system.

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