Trop J Nat Prod Res, July 2025; 9(7): 3007 - 3012 **ISSN 2616-0684 (Print)**
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

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

Original Research Article

Alpha-Glucosidase Inhibitory Activity and Analysis of Eleuthoside B from *Eleutherine bulbosa* Urb. Bulbs Purified Extract from Lampo Donggala, Central Sulawesi, Indonesia

Syariful Anam[1*], Yonelian Yuyun[1], Siti Nurhasana Tanaijo[1], Friska R. Langkoda[1], Yusriadi[1], Asriana Sultan[1], David Pakaya[2]

[1]*Department of Pharmacy, Faculty of Mathematics and Natural Sciences, Tadulako University, Jalan Soekarno Hatta Km. 9, Tondo, Mantikulore, Palu 94119, Indonesia*
[2]*Department of Histology, Faculty of Medicine, Tadulako University, Jalan Soekarno Hatta Km. 9, Tondo, Mantikulore, Palu 94119, Indonesia*

ARTICLE INFO

Article history:
Received 10 February 2025
Revised 07 June 2025
Accepted 18 June 2025
Published online 01 August 2025

ABSTRACT

Diabetes mellitus is becoming a severe health problem with associated economic and societal impacts. Type 2 diabetes (T2D) is the most common, accounting for about 90% of all diabetes cases in the world, and the prevalence is projected to increase, reaching 643 and 783 million by 2030 and 2045, respectively. This disease is characterized by a diminished responsiveness of the liver and muscle cells to insulin, causing fluctuations in blood glucose levels. Bawang Dayak (*Eleutherine bulbosa* Urb.) is a traditional Indonesian medicinal plant proven to have many pharmacological properties, including antioxidant, and antidiabetic properties. Therefore, this study aimed to determine the *α*-glucosidase inhibitory activity, and phytochemical analysis of *E. bulbosa* Urb. Bulbs purified extract from Lampo Donggala, Central Sulawesi, Indonesia. The bulb extract of *E. bulbosa* Urb was assessed for its antidiabetic activity *in vitro* using the *α*-glucosidase inhibitory activity assay. Thin-layer chromatography (TLC) and high-performance liquid chromatography photodiode array (HPLC-PDA) detection were used to characterize the phytochemical composition and quantify the analytes, with Eleuthoside B as a marker compound. The purified extract exhibited potent *α*-glucosidase inhibitory activity with IC_{50} value of 5.29 mg/mL. Eleuthoside B was detected in the purified extract by TLC with Rf value of 0.71. A quantitative determination using HPLC-PDA yielded Eleuthoside B content of 34.7916 ± 0.16 mg/g extract. The study concluded that the purified extract from the bulb of *E. bulbosa*, sourced from Lampo Donggala, Central Sulawesi, possess *α*-glucosidase inhibitory activity, suggesting its potential as an antidiabetic agent.

Keywords: *α*-glucosidase inhibitors, Phytochemical analysis, Eleuthoside B, *Eleutherine bulbosa* Urb., Lampo Donggala.

Introduction

Diabetes mellitus is a metabolic disease characterized by elevated blood glucose levels. This can be due to insufficient insulin production or insensitivity of insulin receptors or both.[1] Diabetes has become a severe health problem worldwide, with a prevalence of 536.6 million individuals in 2021. This number was projected to increase to 783.2 million by 2045.[2] Type-2 diabetes (T2D), the most common type of diabetes, comprises approximately 90% of all cases worldwide. T2D is characterized by decreased sensitivity to insulin within liver and muscle cells.[3, 4] This disease may result in severe complications such as cardiovascular problems, retinopathy, nephropathy, kidney failure, and peripheral neuropathy.[5] These serious life-threatening complications underscore the need for selective and effective therapeutic strategies. A suitable management strategy for T2D is inhibiting the α-glucosidase enzyme found in the brush border of the small intestine.[6] The enzyme hydrolyzes carbohydrates into glucose, which is transported to the bloodstream.

*Corresponding author. Email: syariful.anam@untad.ac.id
Tel: +62-81354971776

Citation: Anam S, Yuyun Y, Tanaijo SN, Langkoda FR, Yusriadi, Sultan A, Pakaya D. Alpha-glucosidase inhibitory activity and analysis of eleuthoside B from *Eleutherine bulbosa* Urb. bulbs purified extract from Lampo Donggala, Central Sulawesi, Indonesia. Trop J Nat Prod Res. 2025; 9(7): 3007 – 3012 https://doi.org/10.26538/tjnpr/v9i7.7

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

Alpha-glucosidase inhibitors prevent the conversion of dietary carbohydrates into glucose in the small intestine, reducing postprandial blood glucose levels. The inhibition of *α*-glucosidase is a well-established method for managing T2D.[7,8] However, the adverse side effects and limited selectivity of the existing clinical drugs targeting this enzyme (i.e., acarbose),[9] necessitate the urgent need for new *α*-glucosidase inhibitors.

Nature is a veritable treasure trove of novel bioactive compounds, with over half of the approved pharmaceuticals originating from natural sources.[10] Acarbose and Voglibose, two compounds recently used as *α*-glucosidase inhibitors, are exemplary naturally derived medicines.[11, 12] Plants are traditionally rich in bioactive compounds and scientific literature has documented over 400 plant species with hypoglycemic properties.[13-16] Indonesia is renowned for its exceptional biodiversity, including approximately 11% of the world's recognized flowering plant species. According to the results of previous studies, approximately half of these species are endemic to Indonesia.[17, 18]

Eleutherine bulbosa Urb. is a species of Iridaceae, commonly known as Bawang Dayak by the Dayak people residing in Kalimantan. This species, a native of South America, also flourishes in Africa, Malaysia, Indonesia, and the Philippines. The bulbs of *E. bulbosa* are commonly used in traditional preparation, where about seven bulbs are boiled and consumed.[19] *E. bulbosa* shows remarkable adaptability to various climatic and soil conditions. The Dayak tribe also uses this species to treat various ailments, including cancer, hypertension, diabetes mellitus, cholesterol, and ulcers.[20-22]

Several studies have investigated the bioactivities and chemical constituents of *E. bulbosa*. A study by Shibuya identified naphthalene

compounds in the extract of *E. bulbosa*, including eleuthoside, eleutherol, eleutherin, and isoeleutherin.[23] The isolation of fifteen naphthalene derivatives from *Eleutherine palmifolia* has been reported.[24] These derivatives exhibited inhibitory activity on the Wnt/beta-catenin protein, a vital system in regulating cell development.[24] According to Kusuma *et al*. (2010),[21] *E. bulbosa* inhibits melanogenesis in melanoma cells through *in vitro* tests, suggesting that the species is a safe skin toning agent suitable for cosmetic use.[21] Three naphthalene derivatives from *Eleutherine americana* extract were reported to inhibit α-glucosidase, with Eleutherinoside A showing the most promising activity, with an IC_{50} of 0.5 mM.[25]

E. bulbosa, also known as Bawang Dayak, is cultivated in Lampo Donggala and is used by society as traditional tea. The Indonesian government promotes the traditional plants to become standardized herbal medicine. An example is *E. bulbosa* from Lampo Donggala. In this study, α-glucosidase inhibitory activity assays and phytochemical analysis, including the concentration of eleuthoside B as a marker compound, were conducted to ascertain the purity and quality of a standardized herbal medicine derived from *E. bulbosa* from Lampo Donggala, Central Sulawesi, Indonesia. This is the first study that investigated the marker compound of *E. bulbosa* from Lampo Donggala, Central Sulawesi. These investigations are part of our effort to develop a standardized herbal-based medicine with antidiabetic properties from Indonesian medicinal plants and provide valuable insights into developing *E. bulbosa* in the future, mainly cultivated in Lampo Donggala.

Materials and Methods

Plant Collection and Identification

E. bulbosa plant was obtained from LPHD Lampo (6PVG+6V) Lumbudolo, Donggala, Central Sulawesi, Indonesia, on 19th October 2023. The plant taxonomy was determined as *E. bulbosa* Urb. (L.) in The Plant Biosystematics Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Tadulako University. Herbarium specimen voucher 668 was kept in the Department of Pharmacy, Faculty of Mathematics and Natural Sciences, Tadulako University, Indonesia.

Plant Material Extraction and Purification

The bulbs section was extracted and purified using the Ieyama *et al*. (2011) procedure with minor modifications.[25] A 500-gram dried sample was macerated in 2000 mL of 50% methanol (Brataco Chem., Indonesia) at room temperature for 24 h. The liquid extract was collected, filtered, and concentrated at 40°C using a rotary evaporator at 40 rpm to yield a crude extract. The crude extract was purified using column chromatography with Diaion HP-20 (Merck, Germany) as the stationary phase, and water and methanol as the mobile phase. Furthermore, the methanol-eluted fraction was partitioned between ethyl acetate and water, and finally, the water-soluble component was evaporated to obtain the purified extract.

Alpha-glucosidase Inhibition Assay

The α-glucosidase inhibition assay was conducted using the purified extract according to methods previously described with minor modifications.[26-30] In summary, the assays were carried out on a 96-well microtiter plate. The reaction mixture consisted of 50 µL of 0.1 M phosphate buffer (Merck, Germany) (pH 7.0), 25 µL of 0.5 mM 4-nitrophenyl α-D-glucopyranoside (Sigma, Germany) (dissolved in 0.1 M phosphate buffer (Merck, Germany), pH 7.0) and 10 µL of the test sample (concentration: 1000, 2000, 4000, 6000, and 8000 µg/mL). A total of 25 µL of α-glucosidase (α-glucosidase from *Bacillus stearothermophilus*, Sigma, Germany) solution (a stock solution of 1 mg/mL in 0.01 M phosphate buffer (Merck, Germany), pH 7.0 was diluted 0.04 Units/mL with the same buffer, pH 7.0 just before assay) were also contained in the mixture. The reaction mixture was incubated at 37°C for 30 minutes. The reaction was terminated by adding 100 µL of 0.2 M sodium carbonate (Sigma, Germany). Enzymatic hydrolysis was monitored by measuring p-nitrophenol (Sigma, Germany) at 410 nm using a microplate reader (BioTek Epoch Microplate Spectrophotometer, Agilent, United States). All experiments were carried out in triplicate.

The inhibition of the α-glucosidase enzyme was calculated as follows (equation 1):

$$\text{Percentage inhibition} = \left(\frac{Slope\ blank - Slope\ sample}{Slope\ blank}\right) x\ 100\% \quad (1)$$

Phytochemical Analysis

Thin Layer Chromatography (TLC) analysis

TLC analysis was carried out according to the previously described procedure.[19, 31, 32] The TLC elution process was performed in a chromatography chamber containing hexane (Brataco Chem., Indonesia) and ethyl acetate (Brataco Chem., Indonesia) (1:2) as mobile phase. Purified extract and Eleuthoside B (≥95%, Merck, Germany) were spotted on the silica gel GF_{254} TLC plate of size 8.5 x 1.5 cm (Merck, Germany). After the elution was completed, the plate was observed under a UV light (Camag, Germany) at 254 nm and 366 nm.

High-Performance Liquid Chromatography (HPLC) analysis

HPLC (Waters, Arc HPLC, United States) analysis was carried out according to the procedure previously described, with slight modifications.[33, 34] A mixed standard solution with concentrations of 250.0, 125.0, 100.0, 50.0, 25.0, and 12.5 µg/mL, each with six appropriate concentrations, was injected into the HPLC and replicated three times. The measurements were made using HPLC, and a calibration curve was produced to represent the relationship between concentration and area, resulting in the equation; $y = bx + a$. The limit of detection (LOD) and limit of quantification (LOQ) were calculated using the linear equation of the calibration curve. The 1 mg/mL purified extract was prepared by dissolving 25 mg in 25 mL of methanol and sonicated for 20 minutes. Subsequently, the content of the extract was determined using HPLC-PDA, equipped with a Waters C18 column (Reliant, 5µm, 150x4.6 mm). The mobile phase consisted of two components, namely, (A) water (Ikapharmindo Putramas, Indonesia) and (B) acetonitrile (Merck, Germany), which flowed through a membrane filter at a flow rate of 1 mL/min. The mobile phase (B) composition range was set at 20% from 0.0 to 8.0 minutes, then increased to 100% between 8.0 and 12.0 minutes, and returned to 20% from 12.0 to 17.0 minutes. The column was maintained at a temperature of 30°C, with a mobile phase flow rate of 1 mL/min. The detector was set at a wavelength of 210 nm. The UV-Vis detector detected the compounds based on the elution time of the sample. The data obtained from the detector were then analyzed using chromatogram software (Waters, Empower Software Solutions, United States).

Data Analysis

Primary data were collected from the area under the curve (AUC) and the concentration of Eleuthoside B. These data were then used to develop a calibration curve by plotting standard concentration on the X-axis versus the Y-axis's peak area (AUC). The eleuthoside B content was estimated from the linear equation of the calibration curve.

Results and Discussion

The present study used a 50% methanol solution to extract *E. bulbosa* bulb, followed by column chromatography with resin Diaion HP-20 as the stationary phase, and water and methanol as the mobile phases. The methanol column fraction was subjected to liquid-liquid extraction with ethyl acetate and water, and subsequent concentration of the water resulted in a purified extract. The purified extract exhibited antidiabetic activity through inhibition of the α-glucosidase enzyme. The α-glucosidase inhibitory activity of the purified extract of *E. bulbosa* is presented in Table 1. The result showed that the extract demonstrated a concentration-dependent inhibition of α-glucosidase with the highest percentage inhibition of 61.014% at 8 mg/mL. The half-maximal inhibitory concentration (IC_{50}) was obtained from the logarithmic equation of concentration versus percentage inhibition (Figure 1). From the equation, the IC_{50} was obtained as 5.29 mg/mL, which indicated that the purified extract of *E. bulbosa* possesses strong α-glucosidase inhibitory activity. Therefore, the IC_{50} value can be used as a standard

concentration for further investigation. These results were supported by the findings from previous studies, which showed that *E. bulbosa* exhibits *α*-glucosidase inhibitory activity,[25, 35] hence could be a potential source of antidiabetic agents. This investigation is the first report of the *α*-glucosidase inhibitory activity of *E. bulbosa* bulbs extract from Lampo Donggala, confirming the purified extract's potential antidiabetic activity.

Table 1: Alpha-glucosidase inhibitory activity of purified extract of *E. bulbosa*

Concentration (mg/mL)	Percent inhibition (%)*
1	9.409 ± 0.179
2	20.350 ± 0.179
4	43.217 ± 0.236
6	54.121 ± 0.136
8	61.014 ± 0.186

*Values are mean ± standard deviation (SD), (n = 3).

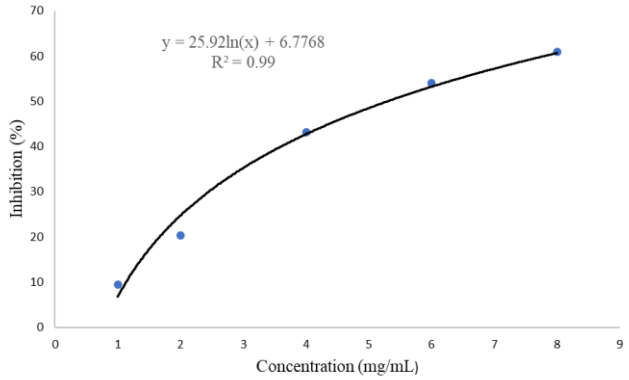

Figure 1: Plots of α-glucosidase inhibitory activity versus concentration of purified extract of *E. bulbosa*

The chemical composition of the purified extract of *E. bulbosa* was determined by thin-layer chromatography (TLC) using silica gel 60 F254 plate and n-hexane:ethyl acetate (1:2) as eluting solvent. Eleuthoside B, a naphthalene derivative, was identified as a major component of *E. bulbosa* bulb; it was detected as a single spot with an Rf value of 0.71 (Figure 2). The biological activities of extracts are usually attributed to their chemical components. *E. bulbosa* majorly contains naphthalene components, including eleutherol, eleutherinoside A, and eleuthoside B.[35] Naphthalene, derived from *E. bulbosa* plant, exhibits diverse pharmacological properties, including anti-cancer, anti-diabetic, anti-bacterial, anti-fungal, anti-viral, anti-inflammatory, and dermatological activities. Furthermore, its antioxidant and anti-fertility effects have been substantiated through a previous study.[35] Three naphthalenes, namely eleutherol, eleutherinoside A, and eleuthoside B have been shown to inhibit the α-glucosidase enzyme.[25] The potent inhibition of the α-glucosidase enzyme in the purified extract can be related to the amount of naphthalene derivatives, specifically eleuthoside B. Further quantitative standardization using high-performance liquid chromatography and photodiode array detection (HPLC-PDA) with Eleuthoside B as the standard compound revealed a distinct peak for the purified extract, closely resembling the standard Eleuthoside B (retention time: 8.0) (Figure 3). The HPLC-PDA method was selected due to its reputation for reproducibility and accuracy in analyzing organic acids extracted from plants and edible mushrooms.[40] Therefore, eleuthoside B was used as a chemical marker to standardize bulbs from *E. bulbosa*.

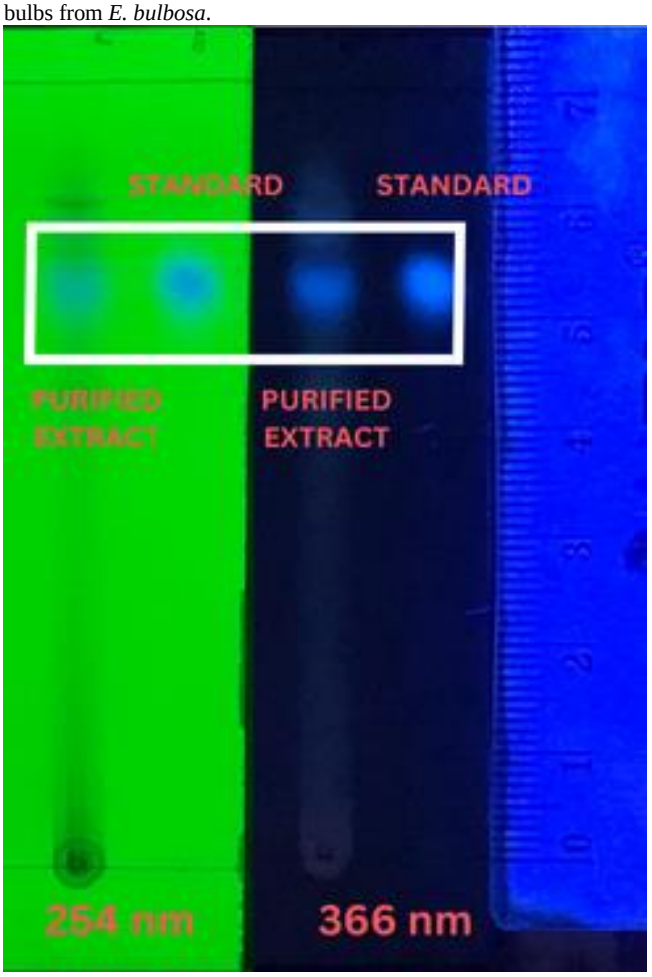

Figure 2: TLC chromatogram of purified extract of *E. bulbosa* from Lampo Donggala with Eleuthoside B standard

Figure 3 shows a satisfactory separation of Eleuthoside B, with sufficient peak resolution, suggesting that the employed method is highly selective for identifying Eleuthoside B. The standard curve, constructed by analyzing solutions at six different concentrations of Eleuthoside B standard, as depicted in Figure 4, exhibited a strong linear relationship within the specified concentration range (12.5 – 250 µg/mL). The correlation coefficient ($R^2 = 0.999$) is remarkable, showing an excellent fit. Furthermore, Eleuthoside B content was determined to be 3.47916 ± 0.16% (w/w). Table 2 shows the Limit of Detection (LOD) and Limit of Quantitation (LOQ) values of Eleuthoside B. The LOD quantifies the lowest analyte concentration in a sample that can still be detected, generating a distinct response from the blank or background noise. The present study has established a detection limit (LOD) for eleuthoside B at 0.0853 µg/L. The LOQ quantifies the lowest concentration of an analyte that can be accurately and precisely measured using a specific analytical method. Furthermore, LOQ serves as a measure of the method's sensitivity. In this study, the LOQ of eleuthoside B was determined to be 0.2585 µg/L.

Plant-based compounds have been extensively recognized as a significant source of antidiabetic agents. The primary objectives of evaluating antidiabetic activity from plant extracts are to identify bioactive compounds with potential as lead molecules for antidiabetic drug development and to develop standardized herbal medicine for use as antidiabetic agents.[19,36,37] Precise identification, quantification, and rigorous purity assessment of active principles of herbal medicines and comprehensive quality evaluation of initial materials are essential for ensuring their safety and efficacy.[38,39]

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

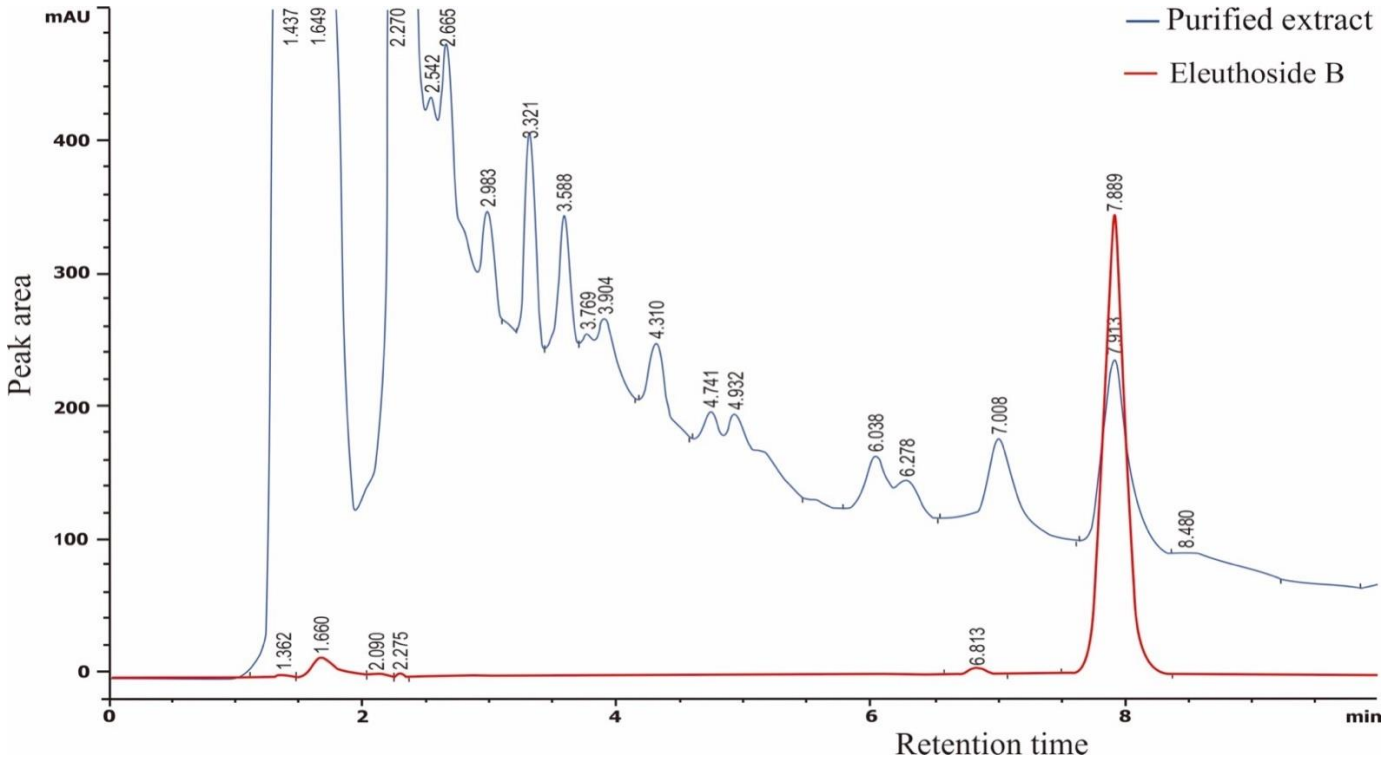

Figure 3: HPLC chromatogram of purified extract of *E. bulbosa* from Lampo Donggala showing the presence of Eleuthoside B

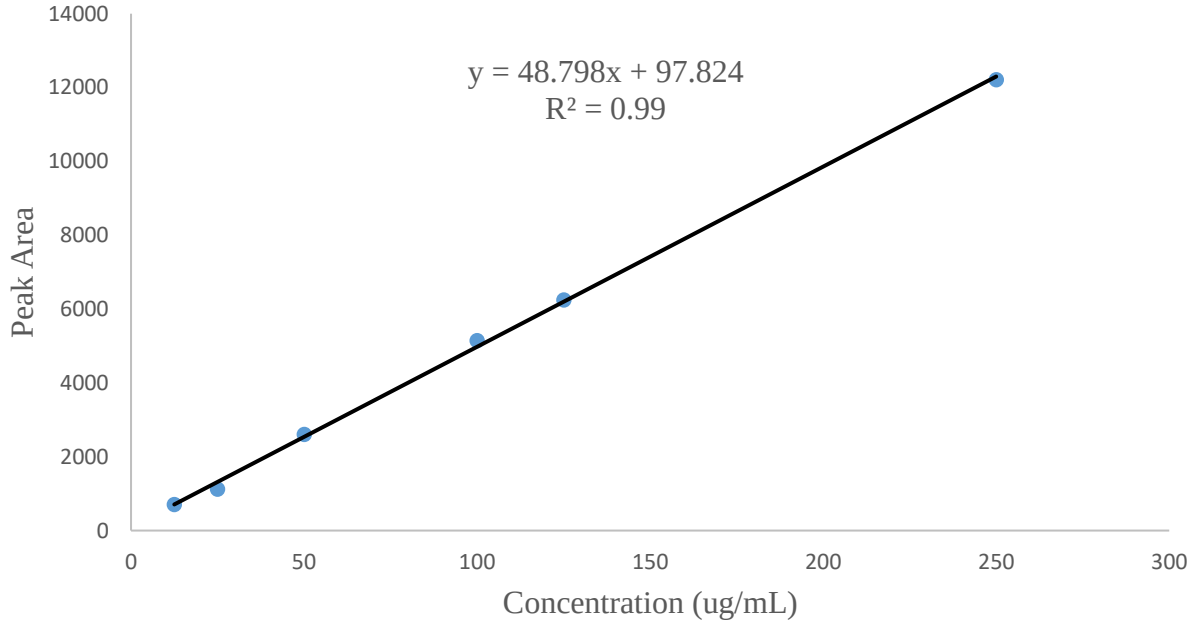

Figure 4: Calibration curve of standard Eleuthoside B

Table 2: Data obtained from the HPLC analysis of the purified extract of *E. bulbosa* from Lampo Donggala

Sample	Standard	Detector	Content (mg/g)	Content (%)	LoD (µg/mL)	LoQ (µg/mL)	Linear Regression equation (r^2)
Purified extract	Eleuthoside B	UV max 210 nm	34.7916 ± 0.16	3.47916 ± 0.16	0.0853	0.2585	y = 48.798x + 97.824 (0.99)

Conclusion

The purified extract of *E. bulbosa* bulbs exhibited inhibitory effects against the *α*-glucosidase enzyme, likely attributed to the presence of eleuthoside B. However, the purification process encountered limitations, resulting in low yields. This study provides valuable preliminary information for assessing the purity and quality of *E. bulbosa* extract as a potential starting material for herbal medicine formulation. Furthermore, the findings from this study pave the way for transforming *E. bulbosa* bulb extract into functional foods, supplements, and standardized herbal formulations with anti-diabetic properties.

Conflict of Interest

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.

Acknowledgements

The authors are grateful to the Ministry of Education, Culture, Research, and Technology of the Republic of Indonesia for funding the study through the study grant (Contract numbers 095/E5/PG.02.00.PL/2024 and 1739/UN28.16/AL.04/2024). The authors are also grateful to Professor Ramadhanil, Moh. Iqbal and Sahlan for their contributions to the identification of the plant.

References

1. Antar SA, Ashour NA, Sharaky M, Khattab M, Ashour NA, Zaid RT, Roh EJ, Elkamhawy A, Al-Karmalawy AA. Diabetes Mellitus: Classification, Mediators, and Complications; A Gate to Identify Potential Targets for the Development of New Effective Treatments. Biomed Pharmacother. 2023; 168:115734. doi:10.1016/j.biopha.2023.115734
2. Sun H, Saeedi P, Karuranga S, Pinkepank M, Ogurtsova K, Duncan BB, Stein C, Basit A, Chan JCN, Mbanya JC, Pavkov ME, Ramachandaran A, Wild SH, James S, Herman WH, Zhang P, Bommer C, Kuo S, Boyko EJ, Magliano DJ. IDF Diabetes Atlas: Global, Regional and Country-Level Diabetes Prevalence Estimates for 2021 and Projections for 2045. Diabetes Res Clin Pract. 2022; 183:109119. doi:10.1016/j.diabres.2021.109119
3. Galicia-Garcia U, Benito-Vicente A, Jebari S, Larrea-Sebal A, Siddiqi H, Uribe KB, Ostolaza H, Martín C. Pathophysiology of Type 2 Diabetes Mellitus. Int J Mol Sci. 2020; 21(17):6275. doi:10.3390/ijms21176275
4. Dilworth L, Facey A, Omoruyi F. Diabetes Mellitus and Its Metabolic Complications: The Role of Adipose Tissues. Int J Mol Sci. 2021; 22(14):7644. doi:10.3390/ijms22147644
5. Wu H, Norton V, Cui K, Zhu B, Bhattacharjee S, Lu YW, Wang B, Shan D, Wong S, Dong Y, Chan SL, Cowan D, Xu J, Bielenberg DR, Zhou C, Chen H. Diabetes and Its Cardiovascular Complications: Comprehensive Network and Systematic Analyses. Front Cardiovasc Med. 2022; 9:841928. doi:10.3389/fcvm.2022.841928
6. Martiz RM, Kumari V B C, Huligere SS, Khan MS, Alafaleq NO, Ahmad S, Akhter F, Sreepathi N, P A, Ramu R. Inhibition of Carbohydrate Hydrolyzing Enzymes by a Potential Probiotic *Levilactobacillus brevis* RAMULAB49 Isolated from Fermented *Ananas comosus*. Front Microbiol. 2023; 14:1190105. doi:10.3389/fmicb.2023.1190105
7. Rangel-Galván M, Pacheco-Hernández Y, Lozoya-Gloria E, Villa-Ruano N. Dietary Natural Products as Inhibitors of α-Amylase and α-Glucosidase: An Updated Review of Ligand-Receptor Correlations Validated by Docking Studies. Food Biosci. 2024; 62:105456. doi:10.1016/j.fbio.2024.105456
8. Lu H, Xie T, Wu Q, Hu Z, Luo Y, Luo F. Alpha-Glucosidase Inhibitory Peptides: Sources, Preparations, Identifications, and Action Mechanisms. Nutrients. 2023; 15(19):4267. doi:10.3390/nu15194267
9. Zamani M, Nikbaf-Shandiz M, Aali Y, Rasaei N, Zarei M, Shiraseb F, Asbaghi O. The Effects of Acarbose Treatment on Cardiovascular Risk Factors in Impaired Glucose Tolerance and Diabetic Patients: A Systematic Review and Dose–Response Meta-Analysis of Randomized Clinical Trials. Front Nutr. 2023; 10:1084084. doi:10.3389/fnut.2023.1084084
10. Atanasov AG, Zotchev SB, Dirsch VM. International Natural Product Sciences Taskforce. Natural Products in Drug Discovery: Advances and Opportunities. Nat Rev Drug Discov. 2021; 20(3):200-216. doi:10.1038/s41573-020-00114-z
11. Dirir AM, Daou M, Yousef AF, Yousef LF. A Review of α-Glucosidase Inhibitors from Plants as Potential Candidates for the Treatment of Type-2 Diabetes. Phytochem Rev. 2022; 21(4):1049-1079. doi:10.1007/s11101-021-09773-1
12. Yin Z, Zhang W, Feng F, Zhang Y, Kang W. α-Glucosidase Inhibitors Isolated from Medicinal Plants. Food Sci Hum Wellness. 2014; 3(3-4):136-174. doi:10.1016/j.fshw.2014.11.003
13. Bisht D, Owais M, Venkatesan K. Potential of Plant-Derived Products in the Treatment of Mycobacterial Infections. In: Ahmad I, Aqil F, Owais M, eds. Modern Phytomedicine. Wiley-VCH Verlag GmbH & Co. KGaA; 2006:293-311p.
14. Choudhury H, Pandey M, Hua CK, Mun CS, Jing JK, Kong L, Ern LY, Ashraf NA, Kit SW, Yee TS, Pichika MR, Gorain B, Kesharwani P. An Update on Natural Compounds in the Remedy of Diabetes Mellitus: A Systematic Review. J Tradit Complement Med. 2018; 8(3):361-376. doi:10.1016/j.jtcme.2017.08.012
15. Malviya N, Jain S, Malviya S. Antidiabetic Potential of Medicinal Plants. Acta Pol Pharm. 2010; 67(2):113-118.
16. Rahman MM, Dhar PS, Sumaia, Anika F, Ahmed L, Islam MR, Sultana NA, Cavalu S, Pop O, Rauf A. Exploring the Plant-Derived Bioactive Substances as Antidiabetic Agent: An Extensive Review. Biomed Pharmacother. 2022; 152:113217. doi:10.1016/j.biopha.2022.113217
17. Fathurahman F, Nursanto J, Madjid A, Ramadanil R. Ethnobotanical Study of "Kaili Inde" Tribe in Central Sulawesi Indonesia. Emir J Food Agric. 2016; 28(5):337-347. doi:10.9755/ejfa.2015-06-463
18. Sun J, Liu B, Rustiami H, Xiao H, Shen X, Ma K. Mapping Asia Plants: Plant Diversity and a Checklist of Vascular Plants in Indonesia. Plants (Basel). 2024; 13(16):2281. doi:10.3390/plants13162281
19. Anam S, Akhmad K, Nurdinah, Nur'afia, Yonelian Y. Standardization of *Eleutherine bulbosa* Urb. Bulbs Extract from Lampo, Donggala, Central Sulawesi. JST (Jurnal Sains dan Teknologi). 2023; 12(1):29-34. doi:10.22487/25411969.2023.v12.i1.16146
20. Kuntorini E, Astuti M, Nugroho LH. Anatomical Structure and Antioxidant Activity of Dayak Onion Bulbs (*Eleutherine americana* Merr.) From South Kalimantan. Berk Penel Hayati (J Biol Res). 2010; 16(1):1-6. doi:10.23869/bphjbr.16.1.20101
21. Kusuma IW, Arung ET, Rosamah E, Purwatiningsih S, Kuspradini H, Syafrizal, Astuti J, Kim YU, Shimizu K. Antidermatophyte and Antimelanogenesis Compound from *Eleutherine americana* Grown in Indonesia. J Nat Med. 2010; 64(2):223-226. doi:10.1007/s11418-010-0396-7
22. Fatiha R, Az Z, Laras N, Sari W, Saputry R, Dwi Nugroho G, Sunarto, Pribadi T, Setyawan A. Review: Traditional Knowledge of the Dayak Tribe (Borneo) in the Use of Medicinal Plants. Biodiversitas. 2021; 22(10):4633-4647. doi:10.13057/biodiv/d221057
23. Shibuya H, Fukushima T, Ohashi K, Nakamura A, Riswan S, Kitagawa I. Indonesian Medicinal Plants. XX. Chemical

Structures of Eleuthosides A, B, and C, Three New Aromatic Glucosides from the Bulbs of *Eleutherine palmifolia* (Iridaceae). Chem Pharm Bull (Tokyo). 1997; 45(7):1130-1134. doi:10.1248/cpb.45.1130
24. Li X, Ohtsuki T, Koyano T, Kowithayakorn T, Ishibashi M. New Wnt/β-Catenin Signaling Inhibitors Isolated from *Eleutherine palmifolia*. Chem Asian J. 2009; 4(3):540-547. doi:10.1002/asia.200800354
25. Ieyama T, Gunawan-Puteri MD, Kawabata J. α-Glucosidase Inhibitors from the Bulb of *Eleutherine americana*. Food Chem. 2011; 128(2):308-311. doi:10.1016/j.foodchem.2011.03.021
26. Schmidt JS, Lauridsen MB, Dragsted LO, Nielsen J, Staerk D. Development of a Bioassay-Coupled HPLC-SPE-ttNMR Platform for Identification of Alpha-Glucosidase Inhibitors in Apple Peel (*Malus xdomestica* Borkh.). Food Chem. 2012; 135(3):1692-1699. doi:10.1016/j.foodchem.2012.05.075
27. Sancheti S, Sancheti S, Seo S-Y. *Chaenomeles Sinensis*: A Potent α-and β-Glucosidase Inhibitor. Am J Pharmacol Toxicol. 2009; 4(1):8-11. doi:10.3844/ajptsp.2009.8.11
28. Dej-Adisai S, Rais IR, Wattanapiromsakul C, Pitakbut T. Alpha-Glucosidase Inhibitory Assay-Screened Isolation and Molecular Docking Model from *Bauhinia pulla* Active Compounds. Molecules. 2021; 26(19):5970. doi:10.3390/molecules26195970
29. Osiako FH, Samuel BB, Oluyemi WM. Effects of Selected *Terminalia* and *Ficus* Species in the Inhibition of α-Amylase and α-Glucosidase Enzymes. Trop J Nat Prod Res. 2023; 7(8):3775-3780. doi:10.26538/tjnpr/v7i8.31
30. Malik A, Ardalani H, Anam S, McNair LM, Kromphardt KJK, Frandsen RJN, Franzyk H, Staerk D, Kongstad KT. Antidiabetic Xanthones with α-Glucosidase Inhibitory Activities from an Endophytic *Penicillium canescens*. Fitoterapia. 2020; 147:104522. doi:10.1016/j.fitote.2020.104522
31. Kowalska T and Sajewicz M. Thin-Layer Chromatography (TLC) in the Screening of Botanicals—Its Versatile Potential and Selected Applications. Molecules. 2022; 27(19):6607. - doi:10.3390/molecules27196607
32. Gauthier L, Wagner BD, Boyetchko S, Kirby CW. Bioassay-Guided Identification of Natural Products for Biocontrol by Thin Layer Chromatography-Direct Bioautography. J Vis Exp. 2024; 209:e66967. doi:10.3791/66967
33. Zubair MS, Anam S, Lallo S. Cytotoxic Activity and Phytochemical Standardization of *Lunasia amara* Blanco Wood Extract. Asian Pac J Trop Biomed. 2016; 6(11):962-966. doi:10.1016/j.apjtb.2016.04.014
34. Nahid S, Saha T, Amin R, Rahman MA, Thousif AM, Chowdhury AT, Sadiq MZA, Faroque ABM. The Altered Efficacy of Traditional Antidiabetic Formulations in Chittagong Division: Metformin Admixing. Trop J Nat Prod Res. 2021; 5(3):453-459. doi:10.26538/tjnpr/v5i3.7
35. Kamarudin AA, Sayuti NH, Saad N, Razak NAA, Esa NM. *Eleutherine bulbosa* (Mill.) Urb. Bulb: Review of the Pharmacological Activities and Its Prospects for Application. Int J Mol Sci. 2021; 22(13):6747. doi:10.3390/ijms22136747
36. Anam S, Muhammad Y, Alfred T, Nurlina I, Ahmad K, Ramadanil, Muhammad SZ. Standarisasi Ekstralk Etil Asetat Kayu Sanrego (*Lunasia amara* Blanco). JST (Jurnal Sains dan Teknologi). 2013; 2(3):1-8. doi:10.22487/25411969.2013.v2.i3.1861
37. Salehi B, Ata A, V Anil Kumar N, Sharopov F, Ramírez-Alarcón K, Ruiz-Ortega A, Abdulmajid Ayatollahi S, Tsouh Fokou PV, Kobarfard F, Amiruddin Zakaria Z, Iriti M, Taheri Y, Martorell M, Sureda A, Setzer WN, Durazzo A, Lucarini M, Santini A, Capasso R, Ostrander EA; Atta-ur-Rahman, Choudhary MI, Cho WC, Sharifi-Rad J. Antidiabetic Potential of Medicinal Plants and Their Active Components. Biomolecules. 2019; 9(10):551. doi:10.3390/biom9100551
38. Prasad DS, Laloo DD, Kumar M, Hemalatha S. Quality Control Standardization and Antioxidant Activity of Roots from *Eriosema chinense*. Pharmacogn J. 2013; 5(4):149-155. doi:10.1016/j.phcgj.2013.07.007
39. Murdifin M, Wahyudin E, Lao G, Subehan S, Manggau M, Alam G. Phytochemical Analysis and Antioxidant Activity of *Mezzetia parviflora* Becc. Woodbark Extract. Pharmacogn J. 2012; 4(30):18-21. doi:10.5530/pj.2012.34.3
40. Pereira C, Barros L, Carvalho AM, Ferreira ICFR. Use of UFLC-PDA for the Analysis of Organic Acids in Thirty-Five Species of Food and Medicinal Plants. Food Anal Methods. 2013; 6(5):1337-1344. doi:10.1007/s12161-012-9548-6