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Research Article

Ultrasound-Assisted Extraction, Quality Assessment, and DPP-IV Inhibitory Activity of *Cissus verticillata* Hydroalcoholic Extract

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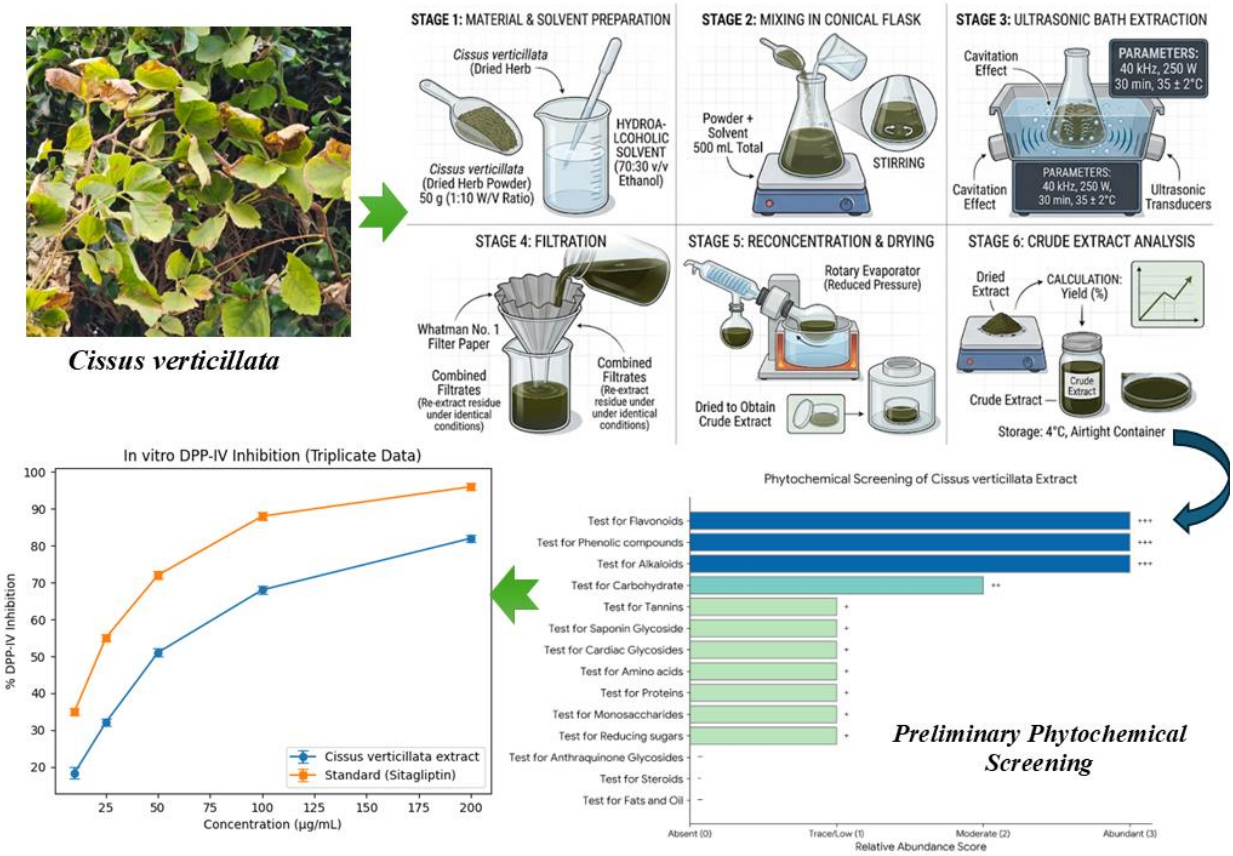
Keywords:
Cissus verticillata; UAE; Phytochemical screening; Physicochemical evaluation; DPP-IV inhibition; Antidiabetic activity.

Abstract:
Cissus verticillata is a medicinal plant rich in bioactive phytoconstituents with promising therapeutic potential, particularly against diabetes. The present study aimed to evaluate the physicochemical characteristics, phytochemical composition, microbial quality, and in vitro dipeptidyl peptidase-IV (DPP-IV) inhibitory activity of the hydroalcoholic extract of *Cissus verticillata*. The whole herb was extracted using Ultrasound-assisted extraction (UAE) with a hydroalcoholic solvent (ethanol, 70:30 v/v). The obtained extract was characterized for organoleptic properties, physicochemical parameters, microbial quality, and qualitative phytochemical constituents using standard pharmacopeial methods. The antidiabetic potential was assessed by a chromogenic DPP-IV inhibition assay using sitagliptin as the reference standard. The hydroalcoholic extract exhibited a yield of 9.2% with satisfactory organoleptic characteristics. Physicochemical analysis demonstrated acceptable pH (7.2 and 6.1), low moisture content (4.9%), minimal foreign matter (0.5%), appropriate ash values, higher water-soluble extractive value (25.25%) than alcohol-soluble extractive value (19.35%), and the absence of heavy metals and pesticide residues. Microbiological evaluation confirmed the absence of *Escherichia coli*, *Salmonella* spp., *Shigella* spp., *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. Preliminary phytochemical screening revealed abundant alkaloids, phenolic compounds, and flavonoids, along with carbohydrates, proteins, amino acids, glycosides, saponins, and tannins. The extract exhibited concentration-dependent DPP-IV inhibition, increasing from 18.33 ± 1.52% at 10 µg/mL to 82.00 ± 1.00% at 200 µg/mL, with an IC_{50} of approximately 70 µg/mL. Overall, the hydroalcoholic extract of *Cissus verticillata* demonstrated satisfactory quality, safety, a rich phytochemical profile, and promising antidiabetic potential, supporting its further investigation as a natural source for the development of phytopharmaceuticals against diabetes.

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Graphical Abstract



1. Introduction

Diabetes mellitus is one of the most prevalent chronic metabolic disorders worldwide and is characterized by persistent hyperglycemia resulting from impaired insulin secretion, insulin resistance, or both. The global burden of diabetes continues to rise, posing a major public health challenge owing to its association with severe complications, including cardiovascular disease, nephropathy, neuropathy, and retinopathy. Although several synthetic antidiabetic agents are available, their long-term use is often associated with adverse effects, such as hypoglycemia, gastrointestinal disturbances, weight gain, and reduced patient compliance. Consequently, increasing attention has been directed toward medicinal plants as potential sources of safer and more effective therapeutic agents possessing multiple pharmacological activities [1–3]).

Among the emerging therapeutic targets for the management of type 2 diabetes mellitus, dipeptidyl peptidase-IV (DPP-IV) has attracted considerable interest. DPP-IV is a serine protease responsible for the rapid degradation of incretin hormones, particularly glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide (GIP), which regulate glucose homeostasis by enhancing glucose-dependent insulin secretion. Inhibition of DPP-IV prolongs the biological activity of these incretin hormones, resulting in

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improved insulin release, suppression of glucagon secretion, and better postprandial glycemic control. Although synthetic DPP-IV inhibitors, such as sitagliptin, have demonstrated clinical efficacy, the exploration of naturally occurring DPP-IV inhibitors from medicinal plants has become an important area of research because of their potential for improved safety and additional antioxidant and anti-inflammatory benefits [4–7].

Cissus verticillata (L.) Nicolson and C.E. Jarvis, belonging to the family Vitaceae, is a perennial climbing medicinal plant widely distributed in tropical and subtropical regions (8–10). Traditionally, the plant has been used in various systems of medicine for the management of diabetes, inflammation, microbial infections, wound healing, gastrointestinal disorders, and oxidative stress-related diseases. Phytochemical investigations have revealed that *Cissus verticillata* contains a diverse array of bioactive constituents, including flavonoids, phenolic compounds, alkaloids, tannins, saponins, glycosides, and terpenoids. These phytochemicals possess well-documented antioxidant, anti-inflammatory, antimicrobial, and antihyperglycemic activities, suggesting that the plant may serve as a promising natural source of multifunctional therapeutic agents [8–10].

Efficient extraction of phytoconstituents is a critical step in the standardization and quality evaluation of herbal medicines. Ultrasound-assisted extraction (UAE) has emerged as an advanced green extraction technique that enhances mass transfer through acoustic cavitation, leading to improved extraction efficiency while reducing solvent consumption, extraction time, and thermal degradation of heat-sensitive compounds (11,12). Hydroalcoholic solvents, particularly ethanol-water mixtures, are widely employed because of their ability to extract a broad spectrum of both polar and moderately polar phytochemicals, thereby maximizing the recovery of biologically active constituents. Following extraction, comprehensive quality assessment through organoleptic, physicochemical, microbiological, and phytochemical evaluations is essential to establish the identity, purity, safety, and consistency of herbal extracts before their pharmaceutical application.

Despite the traditional medicinal importance of *Cissus verticillata*, comprehensive studies integrating UAE, systematic quality assessment, phytochemical characterization, and DPP-IV inhibitory evaluation remain limited. The novelty of the present study lies in the integrated application of UAE with comprehensive organoleptic, physicochemical, microbiological, and qualitative phytochemical evaluation, followed by in vitro assessment of DPP-IV inhibitory activity of the resulting hydroalcoholic extract. This combined approach provides a systematic assessment of both the quality attributes and antidiabetic potential of *Cissus verticillata* extract. Therefore, the present study was undertaken to prepare a hydroalcoholic extract of *Cissus verticillata* using UAE and evaluate its quality characteristics and DPP-IV inhibitory activity. The findings provide a basis for the scientific validation and quality standardization of *Cissus verticillata* as a potential source for future phytopharmaceutical development.

2. Materials and Methods

2.1 Collection of Plant Material and Authentication

The whole herb of *Cissus verticillata* was collected from Buldhana, Maharashtra, India, in December 2024. The plant was authenticated by P. Suresh Babu, Assistant Professor, Department of Botany, Government Degree College, Kukatpally, Telangana, India. A voucher specimen (No. 0937) was deposited for future reference, and the authentication certificate was issued on March 13, 2026. The authenticated plant material was subsequently used for extraction studies. Representative plant images are shown in Figure 1.

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Figure 1. The *Cissus verticillata* plant

2.2 Ultrasound-assisted extraction (UAE)

The dried and powdered whole herb of *Cissus verticillata* was subjected to UAE using a hydroalcoholic solvent (ethanol, 70:30 v/v). Briefly, 50 g of plant powder was mixed with 500 mL of extraction solvent in a conical flask and sonicated in an ultrasonic bath operating at 40 kHz frequency and 250 W power for 30 min at 35 ± 2°C. After sonication, the extract was filtered through Whatman No. 1 filter paper, and the residue was re-extracted under identical conditions to ensure maximum recovery of phytoconstituents. The combined filtrates were concentrated under reduced pressure using a rotary evaporator and further dried to obtain the crude extract. The dried extract was weighed, percentage yield was calculated, and the extract was stored in an airtight container at 4°C until further analysis [11-13].

2.3 Physicochemical Analysis

The hydroalcoholic extract of *Cissus verticillata* was subjected to physicochemical evaluation according to standard pharmacopeial procedures. The percentage yield of the extract was calculated based on the weight of the dried extract obtained from the initial plant material. The moisture content was determined as loss on drying by heating the sample at 105°C until a constant weight was achieved. The total, acid-insoluble, and water-soluble ash values were determined to assess the inorganic content and purity of the extract. The water-soluble and alcohol-soluble extractive values were estimated to evaluate the number of active constituents extracted in different solvents. The pH of 1% and 10% (w/v) extract solutions was measured using a calibrated digital pH meter at room temperature. All determinations were performed in triplicate, and the results were expressed as mean ± standard deviation [14,15].

2.4 Qualitative Phytochemical Screening

Preliminary phytochemical screening of the hydroalcoholic extract was carried out using established qualitative chemical tests to identify the major classes of phytoconstituents. Alkaloids were detected using Dragendorff's, Wagner's, and Mayer's reagents. Flavonoids were identified using alkaline reagent and Shinoda tests, while phenolic compounds and tannins were evaluated using ferric chloride and lead acetate tests. Saponins were detected by the frothing test, and glycosides were screened using Keller–Killiani and Borntrager's tests. Carbohydrates were assessed using Molisch's, Benedict's, and Fehling's tests, whereas proteins and amino acids were identified using Biuret and Ninhydrin tests, respectively. Terpenoids and steroids were examined using the Salkowski and Liebermann–Burchard reactions. The presence or absence of each phytochemical group was recorded based on the characteristic color changes, precipitate formation, or foam production observed during the tests [16–18].

2.5 Microbial Content Determination

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The microbial quality of the hydroalcoholic extract of *Cissus verticillata* was evaluated using the standard plate count method. Briefly, 1 g of extract was dispersed in 9 mL of sterile distilled water, followed by serial dilution and pour plate inoculation. The plates were incubated at 37°C for 24 h, and the microbial load was expressed as colony-forming units per gram (CFU/g). The total fungal count was determined on Sabouraud dextrose agar after incubation at 27°C for 72 h. The presence of specified microorganisms, including *Escherichia coli*, *Salmonella* spp., *Pseudomonas aeruginosa*, and *Staphylococcus aureus*, was assessed using selective enrichment media, selective agar plates, and standard biochemical confirmation tests. All analyses were performed in duplicate according to pharmacopeial guidelines [19–21].

2.6 *In Vitro* Analysis

2.6.1 Preparation of Extract

The hydroalcoholic extract of *Cissus verticillata* was prepared and concentrated under reduced pressure. The dried extract was reconstituted in phosphate buffer (pH 7.4) for the enzyme assay.

2.6.2 DPP-IV Inhibition Assay

The DPP-IV inhibitory activity of the extract was determined using a chromogenic assay based on the cleavage of Gly-Pro-p-nitroanilide substrate. The assay was carried out in Tris-HCl buffer (pH 8.0) using DPP-IV enzyme, with sitagliptin as the reference standard. The release of p-nitroaniline was monitored spectrophotometrically at 405 nm [22].

2.6.3 Procedure

The DPP-IV inhibitory activity of *Cissus verticillata* extract was evaluated using a chromogenic assay under controlled experimental conditions. The extract and the standard drug, sitagliptin, were prepared at five different concentrations (10, 25, 50, 100, and 200 µg/mL) using an appropriate buffer system. For each concentration, the assay was performed in triplicate (n = 3) to ensure reproducibility and statistical reliability of the results. The reaction mixture, consisting of DPP-IV enzyme solution, test sample or standard, and Tris-HCl buffer (pH 8.0), was pre-incubated at 37°C to allow proper interaction between the enzyme and inhibitor. Following this, the chromogenic substrate Gly-Pro-p-nitroanilide was added to initiate the enzymatic reaction. The mixture was then incubated at 37°C for 30 min to facilitate substrate cleavage. After incubation, the reaction progress was monitored by measuring the absorbance at 405 nm using a UV–visible spectrophotometer. The decrease in absorbance relative to the control was used to calculate the percentage inhibition of DPP-IV enzyme activity [23,24].

2.6.4 Calculation

The percentage inhibition of DPP-IV enzyme activity was calculated using the following equation:

$$\%\ Inhibition = \frac{(A_{control} - A_{sample})}{A_{control}} \times 100$$

where $A_{control}$ represents the absorbance of the control (without inhibitor) and A_{sample} represents the absorbance in the presence of the extract or standard.

3. Results and Discussion

3.1 Organoleptic and Physicochemical Analysis of Extracts

The hydroalcoholic extract of *Cissus verticillata* was successfully obtained by UAE, producing a dark brown, semi-solid mass with a characteristic herbal aroma and slightly bitter taste (Table 1). The dark brown color and bitter taste suggest the presence of polyphenols, flavonoids, tannins, and other secondary metabolites. The extract exhibited a yield of 9.2% (Table 1), indicating efficient recovery of phytoconstituents using the hydroalcoholic solvent system.

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The physicochemical evaluation confirmed the quality and purity of the extract (Table 2). The pH values of the 1% and 10% solutions were 7.2 and 6.1, respectively, indicating a near-neutral nature suitable for pharmaceutical applications. The foreign matter content (0.5%) reflected proper cleaning and processing of the plant material, while the low loss on drying (4.9%) suggested minimal moisture content, thereby reducing the risk of microbial growth and improving storage stability.

The ash values, including total ash (9.1%), acid-insoluble ash (2.9%), water-soluble ash (5.6%), and sulfated ash (10.2%), were within acceptable limits, indicating minimal inorganic impurities and good quality of the crude drug. The water-soluble extractive value (25.25%) was higher than the alcohol-soluble extractive value (19.35%), suggesting that the extract is rich in polar phytoconstituents such as phenolics, flavonoids, glycosides, and sugars, while the hydroalcoholic solvent efficiently extracted both polar and moderately polar compounds.

Furthermore, the absence of heavy metals and pesticide residues confirmed the safety and purity of the extract. Overall, the organoleptic and physicochemical characteristics demonstrated that the hydroalcoholic extract of *Cissus verticillata* possessed satisfactory quality, purity, and stability, supporting its suitability for subsequent phytochemical characterization and pharmaceutical formulation development.

Table 1. The organoleptic properties of *Cissus verticillata* extract

Parameters	Observation
Colour	Dark brown
Odour	Herbal and bitter aroma.
Taste	Slightly Bitter
Texture	Semi-solid mass
% yield	9.2%

Table 2. The physicochemical analysis of *Cissus verticillata* extract

Parameters	Observation
pH	
1% Solution	7.2
10% Solution	6.1
Foreign content	0.5%
LOD	4.9%
Ash values	
Total Ash value	9.1%
Acid insoluble ash value	2.9%
Water soluble ash value	5.6%
Sulphated ash value	10.2%
Extractive values	
Alcohol-soluble extractive	19.35%
Water-soluble extractive	25.25%
Heavy metals estimation (present)	Absent
Pesticide residues	Absent

3.2 Microbial Content Determination

Microbial quality evaluation of the hydroalcoholic extract of *Cissus verticillata* demonstrated the absence of pathogenic microorganisms, including Escherichia coli, Salmonella spp., Shigella spp., Pseudomonas aeruginosa, and Staphylococcus aureus (Table 3). The absence of these microorganisms indicates that the plant material was collected, processed, and stored under hygienic conditions, minimizing the risk of microbial contamination.

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Microbial contamination in herbal extracts can adversely affect product quality, stability, and patient safety by causing spoilage and increasing the risk of infection. The complete absence of these pathogenic organisms confirms that the extract complies with acceptable microbiological quality requirements for herbal raw materials and is suitable for pharmaceutical applications. These findings also suggest that the extraction process and handling procedures effectively maintained the microbiological integrity of the extract, supporting its further use in phytochemical characterization and formulation development.

Table 3. The microbial content determinations of *Cissus verticillata* extract

Parameters	Observation
Escherichia coli	Absent
Salmonella spp	
Shigella spp	
Pseudomonas aeruginosa	
Staphylococcus aureus	

3.3 Preliminary Phytochemical Screening of Extract

Preliminary phytochemical screening of the hydroalcoholic extract of *Cissus verticillata* revealed the presence of several bioactive constituents (Figure 2). The extract showed abundant presence (+++) of alkaloids, phenolic compounds, and flavonoids, indicating that these phytochemicals are the major constituents of the extract. These compounds are well known for their antioxidant, anti-inflammatory, antimicrobial, and wound-healing properties, which may contribute to the therapeutic potential of *Cissus verticillata*.

Moderate amounts (++) of carbohydrates were also detected, while reducing sugars, monosaccharides, proteins, amino acids, cardiac glycosides, saponin glycosides, and tannins were present in trace to low amounts (+) (Figure 2). These phytoconstituents may collectively contribute to the pharmacological activities of the plant through synergistic mechanisms. In contrast, fats and oils, steroids, and anthraquinone glycosides were absent (–), suggesting that these classes of compounds are either absent or present below the detectable limits of the qualitative screening methods employed.

Overall, the phytochemical profile confirms that the hydroalcoholic extract is rich in phenolic and flavonoid constituents, supporting the effectiveness of the selected extraction solvent for isolating polar bioactive compounds. These findings are consistent with previous reports on Cissus species and provide a scientific basis for the subsequent phytochemical characterization and biological evaluation of the extract.

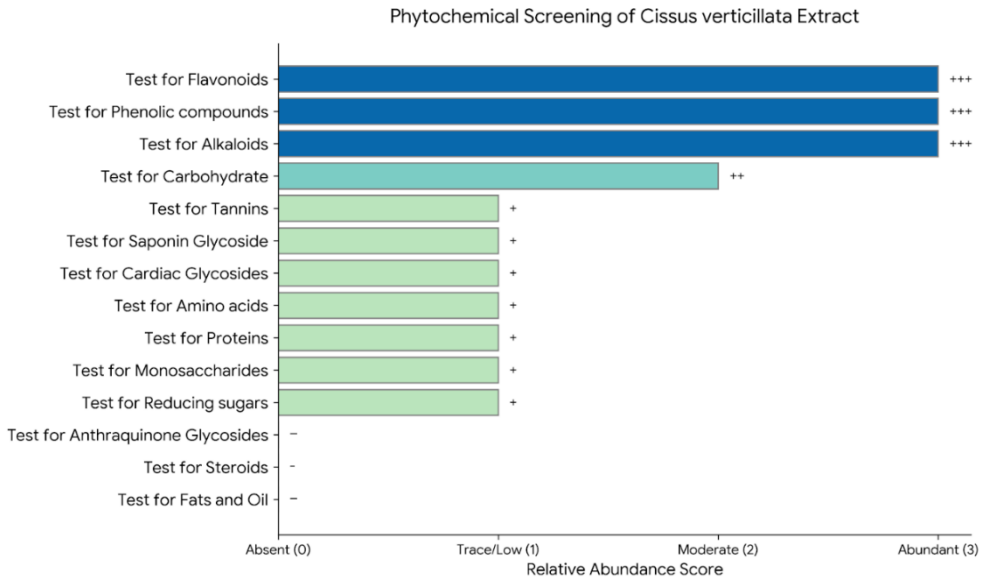


Figure 2. The results of preliminary phytochemical screening of *Cissus verticillata* extract

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3.4 *In Vitro* Analysis

Cissus verticillata extract exhibited a concentration-dependent inhibitory effect on DPP-IV enzyme activity. The inhibition increased from 18.33 ± 1.52% at 10 µg/mL to 82.00 ± 1.00% at 200 µg/mL. The standard drug sitagliptin showed significantly higher inhibition, reaching 96.00 ± 1.00% at 200 µg/mL. The low standard deviation values indicate good reproducibility of the assay. The IC_{50} value of the extract (~70 µg/mL) suggests moderate inhibitory activity compared to sitagliptin. The results are tabulated in Table 4 and the graph is illustrated in Figure 3.

Table 4. DPP-IV enzyme inhibition assay (data was expressed as mean ± SD where n=3)

Concentration (µg/mL)	Extract (% Inhibition)	IC_{50} value (µg/mL)	Standard (% Inhibition)	IC_{50} value (µg/mL)
10	18.33 ± 1.52	~70	35.00 ± 1.00	~27
25	32.00 ± 1.00		55.00 ± 1.00	
50	51.00 ± 1.00		72.00 ± 1.00	
100	68.00 ± 1.00		88.00 ± 1.00	
200	82.00 ± 1.00		96.00 ± 1.00	

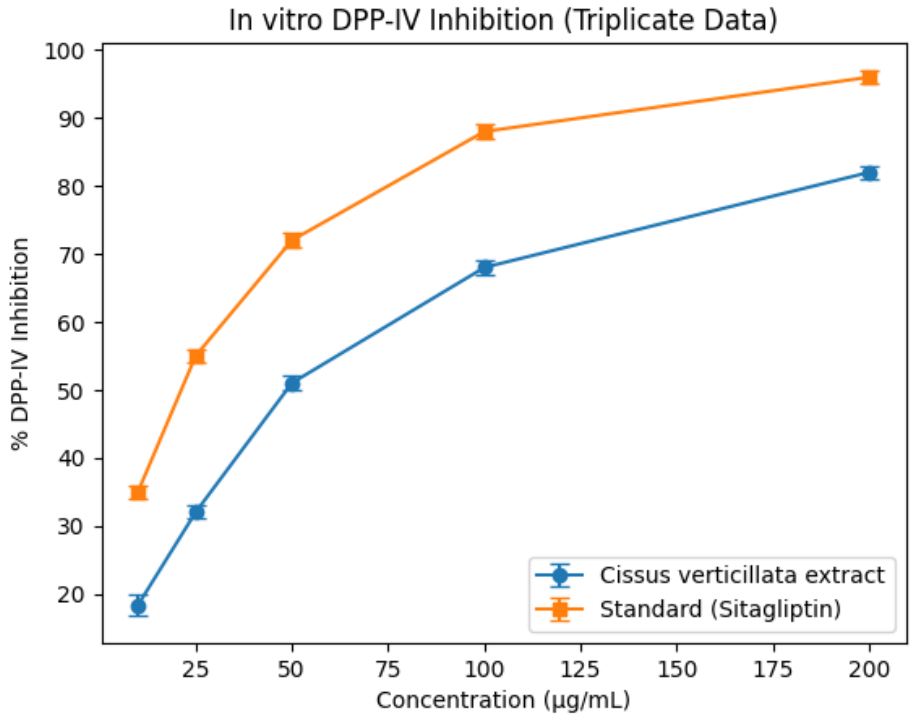


Figure 3. DPP-IV inhibition by Cissus verticillata extract and standard.

The results demonstrate that *Cissus verticillata* possesses significant DPP-IV inhibitory potential in a dose-dependent manner. The presence of phenolic compounds, flavonoids, and triterpenoids likely contributes to this activity.

The observed DPP-IV inhibitory activity of *Cissus verticillata* extract can be mechanistically attributed to the interaction of its bioactive phytoconstituents with the catalytic site of the DPP-IV enzyme. DPP-IV is a serine protease responsible for the rapid degradation of incretin hormones, particularly glucagon-like peptide-1 (GLP-1), which plays a crucial role in glucose homeostasis by stimulating insulin secretion in a glucose-dependent manner. Phytochemicals such as flavonoids and phenolic compounds present in the extract are known to possess suitable structural features—such as hydroxyl groups and planar aromatic rings—that enable them to bind effectively within the active site of DPP-IV through hydrogen bonding, π–π stacking, and

hydrophobic interactions. This binding leads to inhibition of enzymatic activity, thereby preventing the cleavage of GLP-1 and prolonging its physiological action. As a result, enhanced GLP-1 levels contribute to improved insulin secretion from pancreatic β-cells and better regulation of postprandial blood glucose levels.

Although the inhibitory potency of the extract is lower than that of the standard drug sitagliptin, which is a highly selective synthetic DPP-IV inhibitor, the extract offers several pharmacological advantages. Being of natural origin, it contains a diverse array of bioactive compounds that can exert synergistic effects through multiple pathways, including antioxidant and anti-inflammatory mechanisms, which are beneficial in managing diabetes and its complications. Additionally, plant-based extracts are generally associated with a lower risk of adverse effects compared to synthetic drugs, making them promising candidates for long-term therapeutic use. Thus, the moderate yet significant DPP-IV inhibition observed for *Cissus verticillata* supports its potential as a multifunctional antidiabetic agent. The low variability (SD ≤ 1.52) confirms the reliability of the experimental model.

4. Conclusion

In conclusion, the present study demonstrated that UAE using a hydroalcoholic solvent effectively yielded a high-quality extract of *Cissus verticillata* with a satisfactory extraction yield (9.2%) and desirable organoleptic characteristics. Physicochemical evaluation confirmed the purity, stability, and quality of the extract, as evidenced by acceptable pH, low moisture content, minimal foreign matter, appropriate ash values, higher water-soluble extractive value than alcohol-soluble extractive value, and the absence of heavy metals and pesticide residues. Microbiological analysis further established the safety of the extract by confirming the absence of pathogenic microorganisms, including Escherichia coli, Salmonella spp., Shigella spp., Pseudomonas aeruginosa, and Staphylococcus aureus. Preliminary phytochemical screening revealed abundant alkaloids, phenolic compounds, and flavonoids along with the presence of carbohydrates, glycosides, saponins, tannins, proteins, amino acids, and reducing sugars, indicating a rich phytochemical composition. Furthermore, the extract exhibited concentration-dependent DPP-IV inhibitory activity with an IC_{50} of approximately 70 µg/mL, demonstrating moderate antidiabetic potential. Overall, these findings suggest that *Cissus verticillata* is a promising source of bioactive phytoconstituents and may serve as a potential natural candidate for the development of antidiabetic phytopharmaceutical formulations. Further studies involving compound isolation, mechanistic investigations, and in vivo evaluation are warranted to validate its therapeutic efficacy.

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None

Conflict of Interest

The authors declare no conflict of interest.

Authors' Contribution

Conceptualization; methodology; software; validation; formal analysis; investigation; resources; data curation; writing—original draft preparation; writing—review and editing; visualization; supervision; project administration by K. K. Ganjare, S. D. Tayade, A. S. Tamboli, I. A. Shaikh, V. S. Borkar, and J. B. Khedekar. All authors have read and agreed to the published version of the manuscript.

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References

1. Tamboli AS, D. Tayade S. In-Depth Investigation of Berberine and Tropane through Computational Screening as Possible DPP-IV inhibitors for the Treatment of T2DM. J Pharm Sci Comput Chem. 2025;1(1):1–11. doi: 10.48309/jpscc.2025.515525.1000.
2. Elotla SF, Fouad AM, Mohamed SF, Joudeh AI, Mostafa M, Hayek S El, et al. Association between diabetes-

related distress and glycemic control in primary care patients with Type 2 diabetes during the coronavirus disease 2019 (COVID-19) pandemic in Egypt. J Fam Community Med. 2023;30(1):42–50. doi: 10.4103/jfcm.jfcm_238_22.

3. Antal B, McMahon LP, Sultan SF, Lithen A, Wexler DJ, Dickerson B, et al. Type 2 diabetes mellitus accelerates brain aging and cognitive decline: Complementary findings from UK Biobank and meta-analyses. Elife. 2022 May 24;11. doi: 10.7554/eLife.73138.
4. Deacon CF. Physiology and Pharmacology of DPP-4 in Glucose Homeostasis and the Treatment of Type 2 Diabetes. Front Endocrinol (Lausanne). 2019;10. doi: 10.3389/fendo.2019.00080.
5. Baliyan S, Mukherjee R, Priyadarshini A, Vibhuti A, Gupta A, Pandey RP, et al. Determination of Antioxidants by DPPH Radical Scavenging Activity and Quantitative Phytochemical Analysis of Ficus religiosa. Molecules. 2022;27(4). doi: 10.3390/molecules27041326.
6. Nakagawa S, Ohmura R, Toshima S, Park H, Narasako Y, Hirano T, et al. Changes in polyphenols, anthocyanins, and DPPH radical-scavenging activities in sweetpotato (Ipomoea batatas L.) during tuber growth. Sci Hortic (Amsterdam). 2021;284. doi: 10.1016/j.scienta.2021.110100.
7. Ganjare KK, Tayade SD, Tamboli AS, Haq I, Shaikh A, Borkar VS, et al. Integrated Phytochemical and Computational Investigation of *Cissus verticillata* as a Natural DPP-IV Inhibitor. Adv Biores. 2026;17(4):90–115. doi: 10.15515/abr.0976-4585.17.4.90115.
8. Saichandana C, Prasad PD. Exploring *Cissus verticillata* as a Source of HER2 Inhibitors: Phytochemical Profiling, Molecular Docking, and ADMET Analysis. Int J Drug Deliv Technol. 2025;15(4):1514–32. doi: 10.25258/ijddt.15.4.8.
9. Valderrama-Landeros L, Camacho-Cervantes M, Velázquez-Salazar S, Villeda-Chávez E, Flores-Verdugo F, Flores-de-Santiago F. Detection of an invasive plant (*Cissus verticillata*) in the largest mangrove system on the eastern Pacific coast—a remote sensing approach. Wetl Ecol Manag. 2025;33(1). doi: 10.1007/s11273-024-10028-x.
10. Cardoso FN, dos Santos Nunes EV, Figueiredo ID, Rodrigues WD, Assis RP, dos Santos AG, et al. *Cissus verticillata* Leaf Extract Decreases the Production of AGEs and ROS In Vitro. Molecules. 2026 Feb;31(4):697. doi: 10.3390/molecules31040697.
11. Bonilla-Loor M, Solórzano-Zambrano L, Pazmiño A, Segovia-Cedeño D, Delgado-Mera J, Tuárez-Párraga M, et al. Phytochemical characterization and antioxidant potential of black mangrove (Avicennia germinans L.) from the northern coast of Ecuador using aqueous and ethanolic extraction. South African J Chem Eng. 2025;54:451–60. doi: 10.1016/j.sajce.2025.08.015.
12. Mrkalić EM, Sovrlić MM, Šaćirović EM, Petrović AM, Milosavljević MN, Zvezdanović JB, et al. UHPLC-DAD-MS/MS phytochemical profiling of Geranium robertianum L. herba extract obtained by UAE. In 2025. doi: 10.46793/ICCBIKG25.507M.
13. Feizi P, Jafari SM. Optimization of astaxanthin extraction from red (Gracilaria corticata) and brown (Sargassum polycystum) macroalgae through ultrasonication and microwave processing. Ultrason Sonochem. 2025;121. doi: 10.1016/j.ultsonch.2025.107556.
14. Tamboli AS, Tayade SD, Haq I, Shaikh A, Ganjare KK, Borkar VS, et al. Phytochemical Characterization and Antidiabetic Evaluation of Rhynchosia Rothii using LC – HRMS and In Silico Approaches. Adv Biores. 2026;17(4):198–224. doi: 10.15515/abr.0976-4585.17.4.198224.
15. Shaikh IHA, Tayade SD, Tamboli AS, Ganjare KK, Borkar VS, Khedekar JB. Exploration of Bioactive Constituents of Feronia elephantum Through Phytochemical Evaluation , GC – MS Analysis , and Computational Approaches. Adv Biores. 2026;17(04):170–97. doi: 10.15515/abr.0976-4585.17.4.170197.
16. Kawade M, Ingole V, Shinde A, Katade S. Qualitative Phytochemical Screening And Metal Analysis Of Some Selected Medicinal Plants. South East Eur J Public Heal. 2025;1313–7. doi: 10.70135/seejph.vi.6394.
17. Khan SL, Bakshi V. Physicochemical characterization , GC – MS profiling , and computational evaluation of ailanthus excelsa hydroalcoholic extract against mutant EGFR. Chem Rev Lett. 2026;9:358–76. doi: 10.22034/crl.2026.571902.1771.
18. Khan SL, Bakshi V. LC-HRMS and In Silico Analysis of Ailanthus excelsa Metabolites Targeting EGFR Triple Mutation (L858R/T790M/C797S). Adv J Chem Sect A. 2026;9(2):292–311. doi: 10.48309/ajca.2026.533867.1879.

19. Hashimi A, Siddiqui A, Ahmed Y, Siraj MB, Khatoon R. Quality Control and Phytochemical Validation of Saussurea lappa (Costus/Qust). Int J Green Pharm. 2020;14(1):38–43. doi: 10.22377/ijgp.v14i1.2768.
20. Kumatia EK, Ofosu-Koranteng F, Appiah AA, Barimah KB. Standardization and Quality Control of the Herbal Medicine Mist Nibima, Employed to Treat Malaria and COVID-19, Using Physicochemical and Organoleptic Parameters and Quantification of Chemical Markers via UHPLC-MS/MS. Int J Anal Chem. 2021;2021. doi: 10.1155/2021/6390481.
21. Sarraf M, Sani AM, Atash MMS. Physicochemical, Organoleptic Characteristics and Image Analysis of the Doughnut Enriched with Oleaster Flour. J Food Process Preserv. 2017;41(4). doi: 10.1111/jfpp.13021.
22. Vawhal PK, Jadhav SB, Kaushik S, Panigrahi KC, Nayak C, Urmee H, et al. Coumarin-Based Sulfonamide Derivatives as Potential DPP-IV Inhibitors: Pre-ADME Analysis, Toxicity Profile, Computational Analysis, and In Vitro Enzyme Assay. Molecules. 2023;28(3). doi: 10.3390/molecules28031004.
23. Hansen KB, Balsells J, Dreher S, Hsiao Y, Kubryk M, Palucki M, et al. First generation process for the preparation of the DPP-IV inhibitor sitagliptin. Org Process Res Dev. 2005;9(5):634–9. doi: 10.1021/op0500786.
24. Savage SA, Jones GS, Kolotuchin S, Ramrattan SA, Vu T, Waltermire RE. Preparation of saxagliptin, a novel DPP-IV inhibitor. Org Process Res Dev. 2009;13(6):1169–76. doi: 10.1021/op900226j.