VOLUME 11 ISSUE 1 2025

ISSN 2454 – 3055

INTERNATIONAL JOURNAL OF ZOOLOGICAL INVESTIGATIONS

Forum for Biological and Environmental Sciences

Published by Saran Publications, India

International Journal of Zoological Investigations Vol. 11, No. 1, 57-68 (2025)

International Journal of Zoological Investigations

Contents available at Journals Home Page: www.ijzi.net

Editor-in-Chief: Prof. Ajai Kumar Srivastav

Published by: Saran Publications, Gorakhpur, India

ISSN: 2454-3055

In Vitro Study on Antidiabetic Activity of *Benincasa hispida* Leaf Aqueous Extract

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Received: 22nd June, 2024; **Accepted:** 11th October, 2024; **Published online:** 13th January, 2025

https://doi.org/10.33745/ijzi.2025.v11i01.006

Abstract: Diabetes mellitus is a growing public health concern of the twenty-first century. During the last two decades, anti-diabetic properties of medicinal plants have been continuously studied for the identification of several classes of safe and effective bioactive molecules. The current study examined the antidiabetic properties of *Benincasa hispida* aqueous leaf extract. *In vitro* alpha amylase and glucosidase activity proved that *B. hispida* leaves exhibit inhibitory effects on the breakdown of starch. The study found that the highest level of amylase inhibition activity was present in neutral extract that had been kept in cold environment. Increased glucose uptake is reported in the yeast cells treated with the aqueous extract of *B. hispida* indicating elevated glucose metabolism. Phytochemical and GC-MS analysis revealed the presence of potential bioactive compounds in the aqueous extract. The anti-diabetic properties of the present extract may be due to the individual or synergic effects of these compounds.The present study provides a strong basis to augment the use of *B. hispida* leaves in the current day systems of medicine in order to develop potent antidiabetic medicines.

Keywords: Alpha amylase, Alpha glucosidase, Bioactive compounds, Diabetes mellitus, GC-MS analysis, Phytochemical analysis

Citation: Anu B.L., Neethu C.B. and Honey S.: *In vitro* study on antidiabetic activity of *Benincasa hispida* leaf aqueous extract. Intern. J. Zool. Invest. 11(1): 57-68, 2025.

https://doi.org/10.33745/ijzi.2025.v11i01.006

Introduction

Diabetes mellitus is a major global health concern and one of the top five causes of mortality worldwide. According to the statistics of the International Diabetes Mellitus Federation, 463 million people worldwide are diagnosed with diabetes mellitus in 2019, accounting for 9.3% of the adult population, and this number is expected to increase to 700 million by 2045 (Saeedi *et al.*, 2019). Diabetes mellitus is an 'iceberg' disease. According to World Health Organization (WHO) the prevalence of diabetes worldwide is 180 million and will reach the 300 million in 2025

(Joselin *et al.*, 1998). In India, WHO estimated that there were 19.4 million diabetics in the year 1995 and it is likely to increase to 57.2 million by the year 2025. The revised figure is 80.9 million by the year 2030. The prevalence rates have been increasing steadily since the ICMR study in 1970 which had reported the prevalence rate of 2.3% in the urban population and 1.5 % in the rural population (Ahuja *et al.*, 1979). Current prevalence rates are 10% to 18% in adult urban Indian population and there is evidence that the prevalence of type-2 diabetes is increasing in rural population also. The prevalence of diabetes is rising because of increase in the life expectancy as well as a substantial increase in obesity and sedentary life style. The factors for this steep rise include genetic predisposition, urbanization, ethnicity, insulin resistance and central obesity. Every 5th diabetic in the world is an Indian and every 5th and 10th Indian in metro city like Mumbai is diabetic (Gan *et al.*, 2001).

Traditional medicinal practices across various cultures have long utilized plant-derived remedies for managing diabetes. With the growing prevalence of diabetes and the need for effective prevention and management strategies, the study of plant extracts provides a pathway towards developing complementary and alternative therapies that address the evolving needs of patients worldwide. In essence, research into the antidiabetic activity of plant extracts not only enriches our understanding of traditional medicine but also holds immense potential for shaping the future of diabetes care, offering safer, more accessible, and holistic approaches to disease management (Mamun-or-Rashid *et al.* 2014). *Benincasa hispida* (generally called ash gourd, winter melon, winter gourd, wax gourdetc) belongs to the family Cucurbitaceae. It is a popular vegetable harvest, especially among Asian communities both for nutritive and medicinal purposes. The Cucurbitaceae family is basically distributed around the tropical regions. Index of Nutritional Quality (INQ) data shows that the *Benincasa hispida* is valued as a high-quality vegetable. In Ayurvedic system of drug, ash gourd pulp is employed as a main component, which has implicit application for neuro degenerative diseases and to upgrade immunity. It is also used medicinally in various ailments such as gastrointestinal problems, respiratory conditions, heart conditions, diabetes mellitus and urinary conditions. Fruits of this plant are traditionally used as a laxative, diuretic, tonic, aphrodisiac, cardiotonic, and also in cases of hostility, dyspepsia, urinary calculi, blood complaint, insanity, epilepsy, fever, and menstrual diseases (Uma *et al.*, 2023). Also, the leaves of the plant contain anti diabetic exertion. The present study mainly focused on the anti-diabetic properties of aqueous extract of *Benincasa hispida* leaves.

Materials and Methods

Collection of plants

The plant species *Benicasa hispida* was selected for the current study based on its ethnopharmacological relevance, verified medicinal properties, availability, and leaf adaptability. The leaves were collected from local areas of Thrissur, Kerala, India.

Preparation of plant extract

Fresh and healthy *B. hispida* leaves were air-dried and made into powder by electrical blender. The crude aqueous extract was prepared by decoction method using 5 g of the leaf powder by mixing it with 100 ml of distilled water. Boiled the mixture at 100°C for 20-30 min and allowed it to cool. Then the sample was centrifuged at 6000 rpm for 15 min. Discarded the residue and collected the supernatant, stored it in the refrigerator for further use.

Phytochemical screening

Standard techniques were followed to perform a preliminary phytochemical screening of the plant extract in order to determine the presence or absence of secondary metabolites such as carbohydrate, aldehyde, reducing sugar, ketose/ aldose, amino acid, protein, starch, alkaloid, terpenoids, flavonoids, phenol, tannins, saponins, cardiac glycosides, phlobatannins, coumarins and

anthraquinones (Trease and Evans, 1989).

Alpha-amylase inhibition assay

The Alpha-amylase inhibitory assay of aqueous extracts of *B. hispida* leaves was performed according to a previously described method by Shettar *et al.* (2017). Briefly, the reaction mixture contained 0.5 ml of extract with different concentrations (75, 100, 125 and 150 mg/ml), added 0.5 ml of α-amylase solution and 0.02 M sodium phosphate buffer, which was incubated at room temperature for 10 min. Then, 0.5 ml of starch solution in 0.02 M sodium phosphate buffer was added and the mixture was further incubated at room temperature for 10 min. Finally, the reaction was terminated using 1 ml of dinitrosalicylic acid and the test tubes were placed in a water bath (100 °C for 5 min) and cooled until room temperature was attained. Distilled water was used as negative control. The absorbance of the mixture was measured at 540 nm.

% inhibition of alpha amylase =

$$(Abs_{Control} - Abs_{Sample})/Abs_{Control} \times 100$$

Optimization of limiting factors in Alpha-amylase inhibition assay

(i) *Effect of different pH*

The effect of pH on Alpha-amylase inhibitory of aqueous extract was studied by varying the pH 4, 7 and 10 in a constant concentration (100 mg/ml) of *B. hispida* leaves extract. pH was maintained by three different buffers: acetate (3.6-5.6), phosphate (5.8-7.4) and bicarbonate (9.2-10.6). The influence of pH was analyzed by measuring the enzymatic activity (Shettar *et al.*, 2017).

(ii) *Effect of different temperature*

The effect of temperature on Alpha-amylase inhibitory of aqueous extract was estimated by incubating reaction mixture at various temperature such as 4°C, 37°C and 60°C. After incubation, the enzymatic activity was measured at 540 nm (Shettar *et al.*, 2017).

Glucose uptake assay

Glucose uptake assay by yeast cells was carried out using the method of Cirillo *et al.* (1963). For the assay, the yeast *Saccharomyces cerevisiae* was suspended in the distilled water and repeatedly centrifuged (3000 x g, 10 min) to produce clear supernatant fluids and then 10% (v/v) of the suspension was made in distilled water. Optimum concentration of plant extract (100 mg/ml) was combined with 1 ml of glucose solution (5 mM) and incubated for 10 min at 37°C.The reaction was initiated by adding 100 µl of yeast suspension, vortexing, and then continuing to incubate for an additional 60 min at 37°C. The tubes were centrifuged (2500 x g, 5 min) after 60 min, and the amount of glucose in the supernatant was measured at 500 nm.

Increase in glucose uptake (%) = $\frac{Abs_{Sample} - Abs_{Control}}{Abs_{Sample}} \times 100$

Alpha Glucosidase Inhibitory Assay

The inhibitory assay for α-glucosidase was performed by method of Li *et al.* (2010). For the Glucosidase Inhibitory Assay, 2 ml plant extract (100 mg/ml), 2 ml alpha- glucosidase (0.5 U/ml) and 1 ml of 10 mM potassium phosphate buffer (pH 6.8) were pre-incubated at 37° C for 15 min before adding 2 ml of 5 mM P-nitrophenol a-D-glucopyranoside substrate. After the potassium phosphate buffer addition, the mixture was then incubated further at 37° C for 15 min. After 15 min, 2 ml of stop solution containing 200 mM sodium carbonate was added. Then the absorbance at 520 nm was recorded. The enzyme and substrate combination, free of inhibitors, served as the positive control sample. The inhibition (%) of the test sample on analpha-glucosidase was calculated same way as with alpha-amylase assay.

Protein Glycation Assay

Bovine Serum Albumin (BSA)-Glucose Assay was done based on the method of McPherson *et al.* (1988). BSA (10 mg/ml) was incubated with glucose (500 mM) in phosphate buffered- saline (PBS) (pH 7.4), 2 ml plant extract (100 mg/ml) and 0.02% sodium azide at 37 °C for 24 h.

Table 1: Phytochemical analysis of *B. hispida*

TEST	PHYTOCHEMICAL	INFERENCE
Molish's test	Carbohydrate	-
Fehling's test	Aldehyde	+
Benedict's test	Reducing sugar	+
Seliwanoff's test	Ketose/aldose	+
Ninhydrin test	Amino acid	-
Millon test	Amino acid	-
Biurete test	Protein	-
Xanthoprotein test	Protein	-
KI test	Starch	-
Mayers test	Alkaloid	-
Copper acetate test	Terpenoids	+
NaOH test	Flavonoids	-
H_2SO_4 test	Flavonoids	-
$FeCl_3$ test	Phenol	-
$FeCl_3$ test	Tannins	-
Alkaline reagent test	Tannins	-
Foam test	Saponins	-
Froth test	Saponins	-
Keller Killani test	Cardiac glycosides	-
HCl test	Phlobatannins	-
Test for coumarins	Coumarins	-
Test for Anthraquinones	Anthraquinones	-

Aminoguanidine was used as an inhibitor positive control and reactions without any inhibitor was blank control. Percentage inhibition was calculated as follows:

$$\text{Inhibition \%} = 1 - \frac{Aa - Ab}{Ac - Ab} \times 100$$

Here, Aa = fluorescence of the incubated mixture with sample, Ac= The fluorescence of the incubated mixture without sample as a positive control, Ab= The fluorescence of incubated mixture without sample as a blank control.

Gas Chromatography

The bioactive constituents present in leaf extract was determined by Gas Chromatography (Agilent 6890 series) equipped with HP-SMS column mass spectrometer operated at initial column temperature of 30 °C and heated up to 300 °C at 10 °C /5 min. Chromatographic conditions were: flow of 1.0 ml/min of high purity helium as carrier gas in split mode. The identification of the compounds in spectra was done based on retention time and integral area of peaks. The similarity of compounds matched with > 70% are listed based on NIST library search.

Results

Phytochemical screening

The results of the qualitative phytochemical analysis showed that Fehling's test, Benedict's test, Seliwanoff's test and copper acetate test are positive, which indicated that the extract contains aldehyde, reducing sugar, aldose, and terpenoids. All other tests showed negative inference (Table 1).

Alpha-amylase inhibition assay

The Alpha-amylase inhibition assay was carried

Table 2: Alpha-amylase inhibition in different concentration of extract

Concentration (mg/ml)	Percentage of inhibition (%)
75	69.23
100	74.35
125	71.79
150	61.53

Table 3: Effect of pH on alpha-amylase inhibition

pH	Percentage of inhibition (%)
4	No activity
7	57.14
10	No activity

Table 4: Effect of temperature on alpha-amylase inhibition

Temperature	Percentage of inhibition (%)
4	50
37	21.42
95	No activity

out in *B. hispida* leaf aqueous extract at various concentration (75 mg/ml, 100 mg/ml, 125 mg/ml, 150 mg/ml). The results showed that 100 mg/ml concentration was the optimum concentration for the alpha-amylase inhibition (Table 2). The findings highlighted that the percentage of inhibition decreases with increasing extract concentration.

Effect of different pH on alpha-amylase inhibition

The reaction mixture was incubated at various pH level ranging from 4 to 10 by using different buffers. The extract exhibited no inhibitory effect at both basic and acidic pH. The results exhibited that activity was maximum at neutral pH that is 57.14% (Table 3).

Effect of temperature on alpha-amylase inhibition

The results showed that at 4℃ the enzyme activity was 50% and at 37℃ it had 21.42% activity. But at 95℃ there was no activity. Hence, the results revealed that extract showed maximum activity in a cold temperature (Table 4).

Glucose uptake assay, Alpha glucosidase inhibitory assay and Protein glycation assay

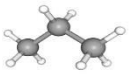
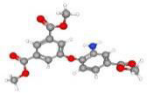
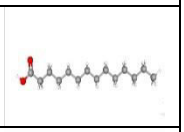
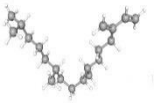
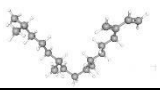
B. hispida leaf extract showed 99.34% activity in glucose uptake assay, 80% activity in alpha-glucosidase inhibitory assay and 67% activity in protein glycation assay (Table 5).

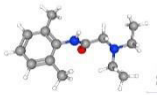
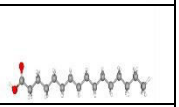
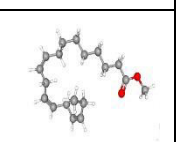
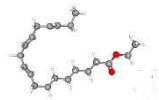
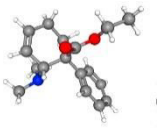
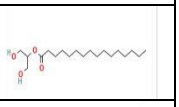
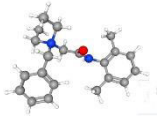
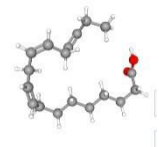
The components of the plant extract were separated using gas chromatography. A total of twenty three different components were separated using Gas chromatography (Table 6; Fig. 1). Table 7 depicts the nature and activity of

Table 5: Percentage of activity in Glucose uptake assay, Alpha glucosidase inhibitory assay and Protein glycation assay

Test	Percentage of activity (%)
Glucose uptake assay	99.34
Alpha- Glucosidase inhibitory assay	80
Protein Glycation assay	67

Table 6: Recorded phytoconstituents of aqueous extract of *B. hispida* by GC-MS

S.No.	COMPOUND NAME	RETENTION TIME (min)	MOLECULAR FORMULA	MOLECULAR WEIGHT	STRUCTURE
1	Benzene	7.607.	C_6H_6	78.11g/mol	
2	Propane	7.753	C_3H_8	44.09g/mol	
3	Phthalic acid Ethyl 3-methoxy-4-nitrobenzyl ester	14.726	$C_{18}H_{17}NO_7$	359.33g/mol	
4	Phthalic acid 5-methyl-2-yl ethyl ester	16.081	$C_{17}H_{24}O_4$	292.4g/mol	
5	Tetradecanoic acid	16.685	$C_{14}H_{28}O_2$	228.37g/mol	
6	6-Hydroxy-4,4,7a-tetrahydrobenzofuran-2(4H)-one	17.013	$C_{11}H_{16}O_3$	196.24g/mol	
7	Neophytadiene	17.519	$C_{20}H_{38}$	278.51g/mol	
8	3,7,11,15-Tetra methyl hexadec-2-ene	17.585	$C_{20}H_{40}$	280.5g/mol	
9	Neophytadiene	17.767	$C_{20}H_{38}$	278.51g/mol	

10	Neophytadiene	17.953	$C_{20}H_{38}$	278.51g/mol	
11	Lidocaine	18.309	$C_{14}H_{22}N_2O$	234.34g/mol	
12	n-Hexadecanoic acid	18.823	$C_{16}H_{32}O_2$	256.42g/mol	
13	Hexadecanoic acid ethyl ester	19.038	$C_{18}H_{36}O_2$	284.48g/mol	
14	9,12,15-Octadecatrienoic acid, methyl ester (Z,Z,Z)	20.087	$C_{19}H_{32}O_2$	292.5g/mol	
15	Phytol	20.210	$C_{20}H_{40}O$	296.5g/mol	
16	9,12,15-Octadecatrienoic acid(Z,Z,Z)	20.564	$C_{18}H_{30}O_2$	278.43g/mol	
17	Octadecanoic acid	20.658	$C_{18}H_{26}O_2$	284.48g/mol	
18	9,12,15-Octadeca-trienoic acid, ethyl ester(Z,Z,Z)	20.706	$C_{20}H_{34}O_2$	306.5g/mol	
19	Carbonic acid, 2-dimethylaminoethyl ethyl ester	23.189	$C_{17}H_{24}ClNO_2$	309.8g/mol	
20	Hexadecanoic acid, ethyl ester	23.553	$C_{19}H_{38}O_4$	330.50g/mol	
21	Benzyldiethyl-(2,6-xylylcarbamoylmethyl) ammonium benzoate	23.859	$C_{28}H_{34}N_2O_3$	446.6g/mol	
22	Linolenic acid	25.010	$C_{18}H_{32}O_2$	278.4g/mol	
23	6,6-Diethylhoctadecane	26.357	$C_{22}H_{46}$	310.6g/mol	

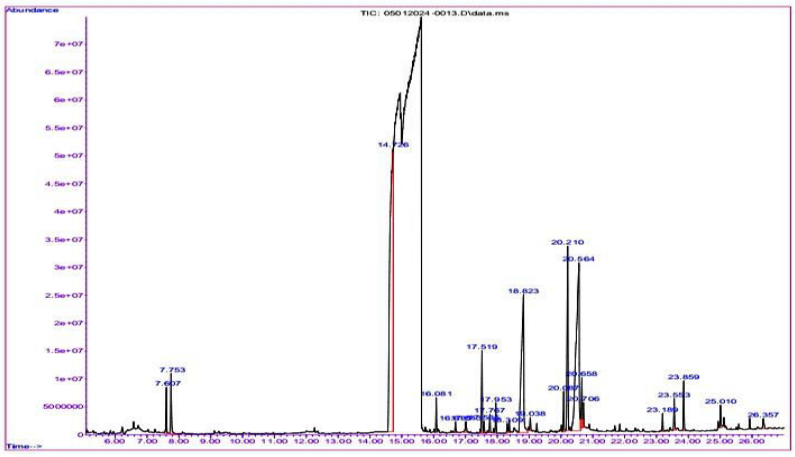


Fig. 1: GC-MS chromatogram of *B. hispida.*

Table 7: Nature and activity of GC MS compounds from aqueous extract of *B. hispida*

S. No.	COMPOUND	NATURE	ACTIVITIES
1	Benzene	Colorless liquid	Solvent in chemical and pharmaceutical industries.
2	Propane	Colorless, odorless liquid	Water heating and fuel for engine applications
3	Phthalic acid Ethyl 3-methoxy-4-nitrobenzyl ester	Aromatic colorless organic compound	Allelopathic, anti- microbial, insecticidal
4	Phthalic acid 5-methyl-2-yl ethyl ester	Colorless liquid	Plasticizers in PVC and also fixative in perfumes
5	Tetradecanoic acid	White to yellowish white crystalline solid	Used as fragrance in soap and cosmetics
6	6-Hydroxy-4,4,7a-tetrahydrobenzofuran-2(4H)-one	Aromatic compound	Have anti tumour, anti -bacterial, anti -oxidant activities
7	Neophytadiene	Colorless to light yellow liquid	Have analgesic, anti-inflammatory properties also possess anti-diabetic activity
8	3,7,11,15-Tetra methyl hexadic-2-ene	20-carbon branched chain fatty acid	Increase cell membrane fluidity, modify various protienes and expressing several genes
9	Neophytadiene	Colorless to light yellow liquid	Have analgesic, anti-inflammatory properties also possess anti-diabetic activity
10	Neophytadiene	Colorless to light yellow liquid	Have analgesic, anti-inflammatory properties also possess anti-diabetic activity
11	Lidocaine	Viscous thick liquid	Local anesthetic agent used for local anesthesia also used for treat minor burns and insect bites
12	n-Hexadecanoic acid	White solid or colorless liquid	Plant metabolite, algal metabolite
13	Hexadecanoic acid ethyl ester	Colorless crystal or liquid	Serve as plant metabolite
14	9,12,15-Octadecatrienoic acid, methyl ester (Z,Z,Z)	Liquid	Possess anti-inflammatory, analgesic properties
15	Phytol	Diterpene alcohol	Used as a precursor for manufacture of synthetic vitamin E and vitamin K1
16	9,12,15-Octadecatrienoic acid(Z,Z,Z)	liquid	Possess anti-inflammatory, analgesic properties

17	Octadecanoic acid	White solid	Hardening soaps, softening plastics making cosmetics and candles
18	9,12,15-Octadeca-trienoic acid, ethyl ester(Z,Z,Z)	Clear colorless liquid	Anti-bacterial, anti-inflammatory properties also a effective anti-acne agent
19	Carbonic acid, 2-dimethyl-aminoethyl ethyl ester	Colorless gas	Used in preparation of carbonated water and other aerated drinks
20	Hexadecanoic acid, ethyl ester	Colorless solid with a wax like odour	Anti-oxidant, anti-inflammatory, anti-helminthic properties
21	Benzyldiethyl-(2,6-xylylcarbamoyl-methyl) ammonium benzoate	White crystalline	Alcohol denaturant
22	Linolenic acid	Colorless to straw colored liquid	Structural component to maintain certain level of membrane fluidity of epidermis
23	6,6-Diethylhoctadecane	liquid	Solvent lubricant and anti-corrosion agent

GC MS compounds from aqueous extract of *B. hispida*.

Discussion

Diabetes mellitus is a non-communicable, primarily genetically based disease while lifestyle factors can also contribute to the disease's development. Insulin and different oral hypoglycemic medications, such as sulfonylureas, metformin, glucosidase inhibitors, troglitazone, etc., are currently the main treatments for diabetes. Antidiabetic medicinal plants can be a valuable resource for discovering more affordable and safer anti-diabetic medications. Phytochemical screening of *B. hispida* leaf extract has been elaborated. It was observed that there was presence of aldehydes, reducing sugar, aldose and terpenoids. In plants, carbohydrates produced by photosynthesis are well known for their essential role as vital sources of energy and carbon skeletons for organic compounds and storage components (Trouvelot *et al.*, 2014). An aldose is defined as a monosaccharide whose carbon skeleton has an aldehyde group. They are primarily found in plants (Murzin *et al.*, 2017). Terpenoids, also known as isoprenoids, are the most numerous and structurally diverse natural products found in many plants. Several studies, *in vitro*, preclinical, and clinical have confirmed that this class of compounds display a wide array of very important pharmacological properties (Ludwiczuk *et al.*, 2017).

The current study investigated the anti-diabetic potential of *B. hispida* aqueous extract. The intestinal enzyme alpha-amylase is crucial for the breakdown of carbohydrates and the uptake of glucose. The α-amylase enzyme inhibiting activity of *B. hispida* aqueous extract showed that the reducing sugar production was probably decreased due to the inhibiting effect of the α-amylase enzyme. The enzymes α-Amylases are responsible for breaking down internal α-1,4-glycosidic bonds in starch to produce low molecular weight products like glucose, maltose, etc. (De Souza and de Oliveira, 2010). Inhibiting the α-amylase enzyme is a treatment for hyperglycemia because it slows down the breakdown of starch and oligosaccharides. Hence, *B. hispida* aqueous extract has good α-amylase enzyme inhibition capacity. Inhibiting α-amylase enzymes with medicinal plants has been the subject of many investigations. Optimization of variables such as temperature and pH were analysed to maximize the amylase inhibition activity of *B. hispida* aqueous extract. The study assumes that neutral extract which stored in a cold condition has maximum activity. Determining the best temperature for α-amylase inhibition activity in plant extracts can vary depending on the specific plant species, the type of α-amylase enzyme being inhibited, and the composition of the extract.

The management of diabetes is further aided by the inhibition of the alpha glucosidase enzyme,

which delays the digestion and absorption of carbohydrates. The optimum concentration of the *B. hispida* was taken for alpha glucosidase assay and found that it had 80% of alpha glucosidase inhibitory activity. According to Derosa *et al.* (2012), standard drug acarbose proved to reverse impaired glucose tolerance to normal glucose tolerance. But long-term use of this drug had some side effects like renal tumor. Thus, from the present result it can be concluded that the plant extract has alpha glucosidase inhibitory activity with less side effects because it is a plant derived product. *B. hispida* leaves aqueous extract was subjected to *in vitro* glucose uptake assay employing yeast as a model. Transport of glucose across yeast membrane may involve facilitated diffusion rather mediation of a phosphotrans-ferase enzyme system or any other unknown process. The glucose uptake by the yeast cells may be affected by several variables, such as glucose concentration inside the cells or the subsequent metabolism of glucose. If most of the internal sugar is converted readily into other metabolites, the internal glucose concentration will get low and high uptake of glucose into the cell will be favored (Rehman *et al.*, 2018). Likewise, there are possibilities that the glucose uptake by yeast cells in the presence of *B. hispida* leaf aqueous extract is due to both facilitated diffusion and elevated glucose metabolism.

Increased glycation during hyperglycaemia can cause intra or intermolecular cross-linking of proteins as they accumulate advanced glycation end-products. Numerous studies have shown that build-up of cross-linked advanced glycation end-products on long-lived proteins may underlie the development of complications affecting diabetes and ageing. This leads to plaque formation, basement membrane thickening, and loss of vascular elasticity. Furthermore, the levels of serum advanced glycation end products reflect the severity of these complications whereas therapeutic interventions aimed at reducing advanced glycation end-products can inhibit or delay their progression. Aminoguanidine (AG) inhibits advanced glycation end products (AGEs) and advanced oxidation protein products (AOPP) accumulated as a result of excessive oxidative stress in diabetes. According to the study of Magdaleno *et al.* (2019), aminoguanidine is a highly effective drug in reduction of blood glucose level. The present study also evaluated 67% protein glycation in the extract of *B. hispida*.

Gas chromatography was performed to isolate the components of plant extract. Twenty three different components was separated using Gas chromatography which include hexadecanoic acid, phytol, phthalic acid etc. The amylase inhibition action of *B. hispida* leaves may be due to the presence of some specific compounds. These α-amylase inhibitors are also called as starch blockers as they prevent or slows the absorption of starch into the body mainly by blocking the hydrolysis of 1,4-glycosidic linkages of starch and other oligosaccharides into maltose, maltriose and other simple sugars. Rajalakshmi (2018) identified the phytochemicals present in the methanolic extract of the leaves of *B. hispida* by GC MS analysis. From the GC MS analysis eighteen compounds were identified. But till date there are few reports on GC MS analysis of leaves of *B. hispida*. Thus, this type of GC MS analysis is the first step towards understanding the nature of active compounds in this plant and this is helpful for further detailed study. An extensive research and development work should be undertaken on the plant and its products for better economic and therapeutic utilization. So, present study demonstrates the antidiabetic property of *B. hispida* aqueous leaf extract Therefore, this plant has high potential for further research into antidiabetic medical applications.

Conclusion

Diabetes with cardiovascular complications impose a major threat on human health claiming death in every 10s. Diabetes cases are exponentially increasing in India because of societal influence and lifestyles. Since ancient times, people have used plants as medicines as they can provide drugs to widen the therapeutic index. The development of plant extract-based

antidiabetic agents holds significant promise in the field of diabetes management. The present study concluded that *B. hispida* leaf aqueous extract has antidiabetic property. It inhibits the activity of certain enzymes like amylase, glucosidase which aid in the development of diabetes. Also, after the treatment of the yeast cells with the aqueous extract of *B. hispida*, the glucose uptake had increased. The extracts offer a rich source of diverse bioactive compounds which have the potential to modulate glucose uptake, improve insulin sensitivity, and mitigate complications associated with diabetes. The most commonly used drugs of modern medicine such as aspirin, anti-malarials, anti-cancers, digitalis, etc. have originated from plant sources. Out of an estimated 250000 higher plants, less than 1% have been screened pharmacologically and very few in regard to diabetic mellitus (DM). Therefore, it is prudent to look for options in herbal medicine for diabetes as well. Collaborative efforts between researchers, clinicians, and traditional healers are imperative to harness the full therapeutic potential of plant extract-based antidiabetic agents, ultimately paving the way for the development of novel and effective treatments for diabetes.

Acknowledgements

The authors are thankful to the Principal and Management of Vimala College (Autonomous), Thrissur for providing the infrastructure and support for conducting the study and the Marian Centre for Advanced Research, St. Mary's College, Thrissur for their guidance and valuable suggestions throughout the course of this work. The financial support provided under DBT- Star College Scheme, Department of Science and Technology, Govt of India, DST –FIST and DST-CURIE (DST/CURIE-PG/2022/33 dated 30-03-2022) for the research facilities provided at Vimala College (Autonomous), Thrissur is gratefully acknowledged.

Conflict of Interest

The authors declare no conflicts of interest.

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