Est. 1984

ORIENTAL JOURNAL OF CHEMISTRY
An International Open Access, Peer Reviewed Research Journal

www.orientjchem.org

ISSN: 0970-020 X
CODEN: OJCHEG
2023, Vol. 39, No.(1):
Pg. 202-211

In vitro Anti-diabetic and Antioxidant Potential of the Sprout of *Borassus flabellifer* L. Extract

ANITHA P.[1,2]*, NARGIS BEGUM T.[3] and SENTHIL KUMAR R.[4]

[1]Department of Biotechnology, Jamal Mohamed College (Autonomous), Affiliated to Bharathidasan University Tiruchirapalli-620020, Tamilnadu, India.
[2]Department of Biochemistry, Shrimati Indira Gandhi College, Affiliated to Bharathidasan University, Tiruchirappalli-620002, Tamilnadu, India.
[3]Department of Biotechnology, Jamal Mohamed College (Autonomous), Affiliated to Bharathidasan University Tiruchirapalli-620020, Tamilnadu, India.
[4]Department of Biochemistry, Thanthai Periyar Government Arts and Science College (Autonomous), Affiliated to Bharathidasan University, Tiruchirappalli–620023, Tamilnadu, India.
*Corresponding author E-mail: anithapeter.sigc.@gmail.com

http://dx.doi.org/10.13005/ojc/390125

(Received: September 10, 2022; Accepted: January 06, 2023)

ABSTRACT

This study aimed to evaluate *In vitro* free radical scavenging activity and *In vitro* anti-diabetic properties of extracts from the sprout of *Borassus fabellifier*. Antioxidant activity was performed using DPPH, ABTS, FRAP, SO and NO inhibition methods. The enzymes (α-amylase and β-Glucosidase inhibitory activities were investigated. Moreover, glucose adsorption and diffusion properties were also studied to confirm the potential effect of the sprout of *B. fabellifier* as an anti-diabetic drug. The ethanolic extracts of *B. flabellifer* exhibited high antioxidant activity towards DPPH, ABTS, FRAP, SO, and NO. However, the ethanolic extracts possessed the strongest inhibitory effect towards α-amylase and β-Glucosidase. The ethanolic extract showed excellent anti-diabetic activity when we examined using glucose adsorption and Glucose diffusion methods. This study suggested that the extracts from *Borassus fabellifier* sprout may act as a potential medicinal plant to treat diabetic complications.

Keywords: *Borassus fabellifier*, *In vitro* anti-diabetic activity, Free radical scavenging activity, α-amylase and β-glucosidase.

INTRODUCTION

Diabetes mellitus is a metabolic endocrine disorder considered by hyperglycemia, metabolic changes in fat, carbohydrates and proteins and elevated risk of cardiovascular complications[1]. Differences in obesity, age, and insulin resistance are just a few of the physiological differences between type 1 and type 2 DM. However, hyperglycemia and microvascular and macrovascular complications are common features of both types of DM. Furthermore, changes in lipoprotein metabolism play a similar role in the DM and type 2 DM's shared pathogenesis of cardiovascular disease[2]. DM is also associated with

increased free radical production or a reduction in antioxidant defences. Oxidative stress is associated with the onset, progression and complications of diabetes mellitus[3].

Complications from DM have a major impact on patient's health, quality of life, and longevity and thus pose serious challenges to healthcare systems. Oral hypoglycemic agents and insulin are two of the few drugs currently available for managing DM, but both have drawbacks. Diabetes Mellitus has traditionally been treated with a wide variety of herbal remedies and medicinal plants[4]. Medicinal plants' diverse phytoconstituents are believed to act via distinct modes and mechanisms, making them useful for treating various conditions. This means that DM and its complications might be treatable with the help of plants[5]. Given that medicinal plants contain a wide range of phytoconstituents that could provide new drug leads that are effective and safe in DM, screening these plants is an alternative and valid approach to the drug development process[6]. In India, various plants have traditionally been used to treat diabetic conditions, and their active principles have been isolated, but only a few have been scientifically studied[7].

Antioxidants are highly effective radical scavengers[8]. The antioxidant action consists of radical scavenging, preventing lipid peroxidation, chelating metal ions, and reducing free radicals. Antioxidants are beneficial components that neutralise free radicals before they can attack cells, thus protecting cell proteins, lipids and carbohydrates from damage[9]. Antioxidants are chemicals that inactivate free radicals, either endogenously or exogenously. Among these substances are lipid-soluble vitamins, ascorbic acid, sulfhydryl-containing chemicals and serum proteins. Antioxidants have been proposed for use in the treatment of a variety of human ailments[10].

The leaves of *Borassus flabellifer* L. (Arecaceae) are 0.9-1.5 m in diameter and palmately fan-shaped, while the petiole edges are covered in hard horny spinescent serratures; the flowers are unisexual; the male spadix is branched while the female spadix is simple and the fruits are large, subglobose drupes on the greatly enlarged perian[11]. The herb was historically used for its stimulating, anti-laprotic, diuretic, and antiphlogistic properties. The fruit can be used as an aphrodisiac, laxative, sedative, or for upset stomachs. Roots and juice from the plant have anti-inflammatory effects[12]. The methanolic extract of *B. flabellifer* male flowers contains steroid saponins of the spirostane type, which have been shown to reduce the rise in serum glucose levels in rats fed sucrose. It has also been demonstrated to possess immunosuppressive properties. *Borassus flabellifer* Linn. has been used as an antidote, anti-inflammatory, wound healing, anthelmintic action, analgesic, and antipyretic, according to a review of the literature[13]. Due to a lack of information about the antioxidant and anti-diabetic effects of an ethanolic extract of *Borassus fabellifier* L. (EtS-Bf), this study was undertaken.

MATERIAL AND METHODS

Plant material

The fresh sprouts of *Borassus fabellifier* L. were collected from the local region of Tiruchirappalli, Tamil Nadu, India, in November 2021.

Plant extract

About 500 g of the sprout powder was put into a soxhlet extractor, and different solvents (Hexane, Chloroform, Ethylacetate, Ethanol and Water) were used to extract the substance. The crude extract was dried up in a rotary flash evaporator by concentrating it under low pressure and temperature control. The extract was put in vacuum desiccators to be used in future research.

DPPH radical scavenging assay

The extract was dissolved in alcohol at 20 and 100 g/mL concentrations and added to a methanolic DPPH (1mM). The combined volume was exactly 1 mL. The alcohol dose given to the placebo group was the same as the real one. The absorbance at 517nm was determined after 20 minutes. There were three sets of experimental conditions[14].

ABTS radical scavenging assay

The reaction mixture with 0.3 mL of ABTS radical, 1.7 mL of phosphate buffer, and 0.5 mL of extract at different concentrations was mixed with 20–100g/mL of extract. Blank had finished his work without using drugs. Absorption was measured to be 734nm. The experiment happened three times[15].

FRAP Assay

Add 3.6 mL of FRAP solution to 0.4 mL of distilled water and let it sit for 5 min at 37°C. The solution was then mixed with a certain concentration of plant extract (20-100g/mL) and kept at 37°C for 10 minutes. The reaction mixture's absorbance was measured at 593nm. Five concentrations of $FeSO_4$, $7H_2O$ were used to make the calibration curve, and absorbance values for sample solutions were found[16].

Superoxide scavenging

Alkaline DMSO was used as a superoxide-making system. From 20g/mL to 100g/mL of the test compound, 1 mL of alkaline DMSO and 0.2 mL of 20mM NBT in phosphate buffer with a pH of 7.4 were added. Three times, the same experiment was done[17].

Nitric oxide radical scavenging

In a phosphate buffer with a pH of 7.4, 5mM of sodium nitroprusside was made. 0.3 mL of sodium nitroprusside was added to different concentrations of test compound that ranged from 20 to 100 g/mL. After 5 h at 25°C, 0.5 mL of Griess reagent was added to the test tubes. The chromophore's absorbance was measured to be 546nm. Three times, the experiment was done[18].

Inhibition of α-amylase enzyme

The standard medication and test samples were incubated at 25°C for 10 min in 500 l of 0.20mM phosphate buffer (pH 6.9) with α-amylase (0.5 mg/mL). Each tube received 500L of 1% starch in 0.02 M sodium phosphate buffer (pH 6.9) for 10 min at 25°C incubated reaction mixes. 3, 5 di-nitro salicylic acid colour reagent inhibited the process. After 5 min in boiling water, test tubes were cooled to room temperature. 540nm absorbance was measured after dilution with 10 mL of pure water[19].

Inhibition of α-glucosidase enzyme

For 5 min at 37°C, a solution of the starch substrate (2%w/v maltose or sucrose) was mixed with 0.2M Tris buffer at pH 8.0 and different concentrations of plant extract. This was done to figure out the inhibitory activity. To start the reaction, 1 mL of α-glucosidase enzyme (1U/mL) was added, and the mixture was left to sit for 40 min at 35°C. Adding 2 mL of 6N HCl finally stopped the reaction. The brightness of the colour was then measured to be 540nm[20].

Determination of glucose adsorption capacity

25 mL of glucose solution was mixed with different amounts of 1% plant extracts (5, 10, 20, 50 and 100mM). The mixture was stirred well, put in a shaker water bath at 37°C for six hours, centrifuged at 4,000 g for twenty minutes, and the glucose concentration in the supernatant was measured[21].

Glucose diffusion Test

Dialysis bags (MWCO 500-1000) with a pore size of 6.4 centimeters were used. Each extract was dissolved in 15M NaCl and 22 M D-glucose, and 2 mL of each solution was added to dialysis bags before they were sealed and placed in a 50 mL tube containing 45 mL of 15M NaCl. Blood glucose levels were monitored for 24 h and compared to baseline levels every two hours[22].

Statistical analysis

The data is presented in Mean SEM form. We used linear regression analysis to determine the IC_{50} values (MS Excel).

RESULT AND DISCUSSION

The existence of bioactive chemicals in the Borassus genus may be responsible for the anti-diabetic and antioxidant activities of EtS-*Bf*. As a result, one of these chemicals in EtS-*Bf* extract could aid glucose absorption in the current investigation. The buildup of functional glucose transporter molecules in the cell membrane is responsible for glucose uptake by skeletal muscles. To date, one of the most crucial aspects of DM care has been researched into the effect of medications on lowering postprandial hyperglycemia.

DPPH radical scavenging assay

The antioxidant potential of foods and plants is commonly measured by their ability to scavenge the DPPH free radical. The antioxidant activity of medicinal herbs is often measured by their ability to quench DPPH radicals[23]. *In vitro* antioxidant investigations of five *B. flabellifer* extracts revealed the degree of DPPH radical scavenging at various doses (25, 50, 75 and 100 g/mL). This was determined using ascorbic acid as a reference DPPH assay demonstrated that antioxidants could catalyse the conversion of the stable radical DPPH into the positively fluorescent diphenyl-picrylhydrazine[24].

The strategy involves reacting an alcoholic DPPH solution with a hydrogen-donating antioxidant to produce the non-radical form DPPH-H[25]. As shown in Fig. 1, the scavenging ability of *B. flabellifer* extracts increases with its concentration, increasing the effective suppression of DPPH radical concentration. The ethanol extract of *B. flabellifer* (94.08±1.74%) had a stronger scavenging effect than other extracts such as AqS-*Bf* (88.40±1.15%), EaS-*Bf* (81.38±0.58%), ChS-*Bf* (78.41±0.58%), HeS-*Bf* (73.19±2.33%), and ascorbic acid (97.44±0.63%). Using MS Excel, the IC_{50} value of several extracts against DDPH scavenging activity was determined. The results suggested that the ethanol extract from the plant's leaves may have the strongest free radical scavenging activity against DPPH, as measured by a decrease in DPPH concentration[26]. The results of various extracts of *B. flabellifer* show that ethanol extracts with high total phenolic contents possess potent radical scavenging actions, which would be connected to the inherent character of phenolic compounds, which contributes to their ability to transfer electrons or donate hydrogen[27]. It has also been found that the type of solvent utilized[28] strongly influences the antioxidant potential of compounds with varying polarities. This investigation used ethanol and a water-ethanol mixture as solvents to extract low molecular weight and moderate polar compounds[29]. Fig. 1 depicts the DPPH radical scavenging ability of the various extracts. According to this diagram, all of the extracts had an inhibitory effect on the DPPH free radical. The percentages of inhibition range from 73.19±2.33% for the EtS-*Bf* hexane extract to 97.44±0.63% for vitamin C. EtS-*Bf* had the strongest and most significant inhibitory potential among the extract samples tested at various doses compared to the other extracts. Because of their high abundance of phenolic chemicals, plants operate as electron donors. This could explain the DPPH radical scavenging ability shown in the extracts studied. This finding supports a prior study that found that the DPPH-scavenging effects of plant extracts increase with concentration[30,31]. HeS-*Bf*, ChS-*Bf*, EaS-*Bf*, AqS-*Bf*, EtS-*Bf*, EtS-*Bf*, and Ascorbic Acid have IC_{50} values of 83.58, 72.21, 64.57, 46.41, 57.64, and 46.89g/mL, respectively.

ABTS radical scavenging assay

Screening samples and cultivars for high levels of natural antioxidants may benefit from the ABTS method, which is a rapid method for determining antioxidant activity[32]. Fig. 2 illustrates the ABTS+ scavenging activity. This data shows that the EtS-*Bf* had a stronger inhibitory potential than the other samples at all concentrations, with a maximum percentage of inhibition of 90.29±2.48% at 100 g/mL. Plant flavonoid has been shown to break the chain reaction of free radicals by donating a hydrogen atom, as was reported in a previous study[33].

The extract efficiently removed the light-scavenging radical scavenger ABTS (2,2'-azinobis-3-ethylbenzothiazoline-6-sulfonic acid)[34]. Fig. 2 shows that EtS-*Bf* has the highest activity (90.29±2.48%), followed by AqS-*Bf* (79.31±0.60%), ChS-*Bf* (82.78±0.34%), EaS-*Bf* (85.08±1.87%) and HeS-*Bf* (79.99±3.14%). The control activity (ascorbic acid) is 95.17±1.51% and the IC_{50} values for five extracts of *B. flabellifer* were computed using MS Excel and reported in Fig. 2. A protonated radical, ABTS, exhibits a distinctive absorbance peak at 734nm that lowers when the proton radicals are scavenged[35]. HeS-*Bf*, ChS-*Bf*, EaS-*Bf*, AqS-*Bf*, EtS-*Bf*, and Ascorbic Acid have IC_{50} values of 70.66, 67.16, 65.42, 62.57, 68.95, and 60.12 g/mL. Therefore, the EtS-*Bf* can scavenge free radicals, leading to lipid oxidation via a chain-breaking reaction, as evidenced by its ABTS radical scavenging activity.

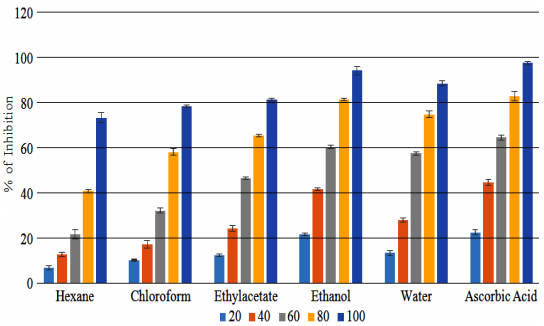

Fig. 1. DPPH Scavenging Potential of the Different Extracts of *B. flabellifer*

Values are expressed as mean ± SEM of three replicates.

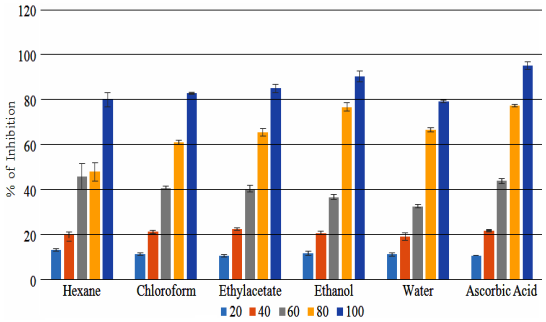

Fig. 2. ABTS radical scavenging potential of the Different Extracts of *B. flabellifer*

Values are expressed as mean±SEM of three replicates.

FRAP Assay

The ferric-reducing antioxidant power activities of the HeS-*Bf*, ChS-*Bf*, EaS-*Bf*, EtS-*Bf*, and AqS-*Bf* were also determined in this investigation. In general, the results demonstrated a concentration-dependent increase in absorbance measurements of reaction mixtures in the UV-Vis spectrum at 900nm.[36]

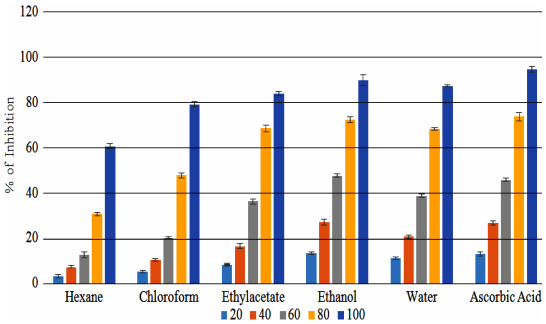

Fig. 3. Ferric Reducing Antioxidant Power (FRAP) Activities of the Different Extracts of *B. flabellifer*

Values are expressed as mean±SEM of three replicates.

The enhanced absorbance of several extracts indicated an increase in reductive capacity. In vitro antioxidant investigations of five *B. flabellifer* extracts revealed the degree of FRAP activity at varying doses of *B. flabellifer* (20, 40, 60, 80, and 100 g/mL). At maximal concentration (100 g/mL), the FRAP of EtS-*Bf* was determined to be 89.81±2.31%, followed by AqS-*Bf* (87.34±0.55), EaS-*Bf* (83.84±0.96), ChS-*Bf* (79.13±1.15%), and HeS-*Bf* (60.65±1.25%). The control (ascorbic acid) was found to have 94.68±1.38% activity, and IC_{50} values for five extracts of *B. flabellifer* were calculated using MS Excel and displayed in Fig. 3. The ferric-reducing assay revealed that water extract had all the extracts' highest reducing power. This indicates the plant's reduction potential is the consistent reduction of Fe^{3+} to Fe^{2+} [37]. It appears that the FRAP activity of the ethanol sprout extract of *B. flabellifer* has increased, which could be attributed to the high amounts of total phenolics and flavonoids in the extract[38]. The action of FRAP was found to be more correlated with total phenols and total flavonoids, which is an interesting finding[39]. Our findings support the role of flavonoids and phenolics as antioxidant agents in EtS-*Bf*, which contribute significantly to total antioxidant capacity. HeS-*Bf*, ChS-*Bf*, EaS-*Bf*, AqS-*Bf*, EtS-*Bf*, and Ascorbic acid have IC_{50} values of 99.14, 78.79, 67.19, 59.88, 64.68, and 59.23 µg/mL, respectively. However, the efficacy of all solvent extracts of *B. flabellifer* sprouts to reduce power was much higher than that of the synthetic antioxidant ascorbic acid. This could be because water has a higher reactive concentration of bioactive components (particularly phenols and flavonoids) than any other extract.

Superoxide scavenging

Superoxide, a reactive oxygen species, has some harmful qualities that can be imposed on cells and DNA, resulting in various disorders[40]. As a result, a proposal has been made to evaluate the antioxidant extracts' comparative interceptive ability to scavenge the superoxide radical[41]. Fig. 4 shows the results of superoxide anion scavenging activities of various *B. flabellifer* extracts. From 25 to 100 g/mL, the different extracts have good superoxide anion radical scavenging activity. At 100 g/mL, the SOD scavenging activities of EtS-*Bf*, AqS-*Bf*, EaS-*Bf*, ChS-*Bf*, and HeS-*Bf* were found to be 97.37±0.59, 90.85±0.80, 87.16±1.05, 79.45±0.48 and 72.15±0.95%, respectively. Fig. 4 shows the IC_{50} values for five *B. flabellifer* extracts calculated using MS Excel. On the other hand, the standard (ascorbic acid) showed 96.25±1.18% inhibition at the same concentration. This could be because the extract contains a reactive concentration of bioactive constituents and a mixture of other nutrients[42]. HeS-*Bf*, ChS-*Bf*, EaS-*Bf*, AqS-*Bf*, EtS-*Bf*, and Ascorbic acid have IC_{50} values of 75.28, 67.54, 60.64, 50.73, 53.67, and 48.54 g/mL. All of the fractions had superoxide radical scavenging activities in a dose-dependent manner[43].

Nonetheless, the extracts' superoxide scavenging activities were found to be significantly lower when compared to ascorbic acid. This could be because the extract contains flavonoids and other antioxidants.

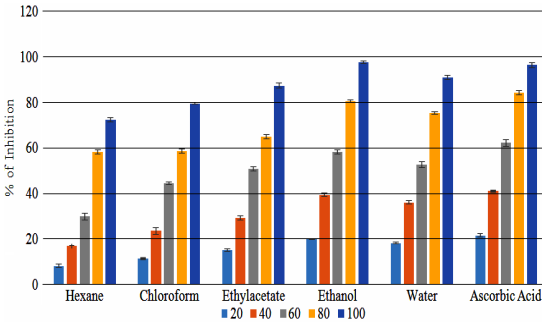

Fig. 4. Superoxide anion Activities of the Different Extracts of *B. flabellifer*

Values are expressed as mean±SEM of three replicates.

Nitric oxide radical scavenging

The antioxidant activity of plant extracts and pure phytochemicals can be evaluated using several different complementary methods[44]. It is common practice to test for free radical scavenging abilities *In vitro* by inactivating radicals like hydroxyl (OH) and nitric oxide (NO) radicals[45]. The results of the extracts' OH radical scavenging activities are shown in Fig. 5. The scavenging properties of AqS-*Bf* (61.25±0.55%), ChS-*Bf*(87.69±1.34%), EaS-*Bf* (72.59±1.44%), EtS-*Bf*(93.65±1.19%), HeS-*Bf* (87.08±1.32%), and ascorbic acid (91.15±0.75%) were tested at the lowest concentration of extract (100 g/mL). The inhibitory potential of the extracts studied increases as concentration is increased. As indicated in earlier research, the phenolic chemicals detected in the plant extracts may be implicated in the scavenging action of the samples[46]. Also, the ability of polyphenols to get rid of free radicals depends on their molecular structure, hydroxyl group substitution pattern, availability of phenolic hydrogen, and ability to stabilise the HO and NO radicals made as a result through hydrogen donation or expansion electron delocalization[47]. HeS-*Bf*, ChS-*Bf*, EaS-*Bf*, AqS-*Bf*, EtS-*Bf*, and Ascorbic acid have IC_{50} values of 95.31, 64.96, 76.84, 52.40, 57.93 and 50.36 g/mL.

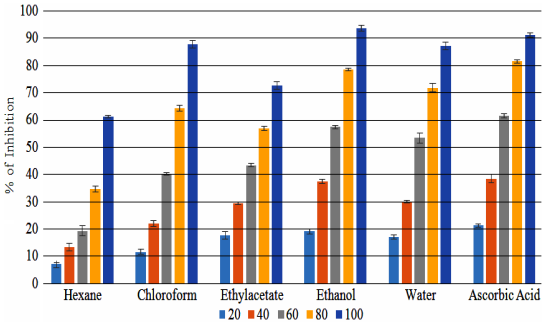

Fig. 5. Nitric oxide (NO) radical scavenging potential of the Different Extracts of *B. flabellifer*

Values are expressed as mean±SEM of three replicates.

Inhibition of α-amylase enzyme

According to our research, numerous studies have been done on the anti-diabetic effect of *B. flabellifer* sprouts, but few have been done on the sprout of *B. flabellifer*. Controlling the amount of small-intestinal human pancreatic-amylase (HPA) activity is important in treating type 2 diabetes because it prevents an increase in postprandial glucose levels[48].

The results of αα-amylase and αα-glucosidase inhibition of AqS-*Bf* and EtS-*Bf* in this work are shown in Fig. 6 for the first time. We can see from Fig. 6 that AqS-*Bf* is less active than EtS-*Bf* in the α-amylase inhibition assay. The most effective extract was the one made by infusing the plant for the AqS-*Bf*, with an IC_{50} of around 117.46 g/mL. The macerated ethanol had the best α-amylase inhibitory action in the EtS-*Bf*, with an IC_{50} of around 78.58 g/mL. Acarbose has an IC_{50} in the 82.65 g/mL range, much higher than the AqS-*Bf* and EtS-*Bf* compared to the reference standard.

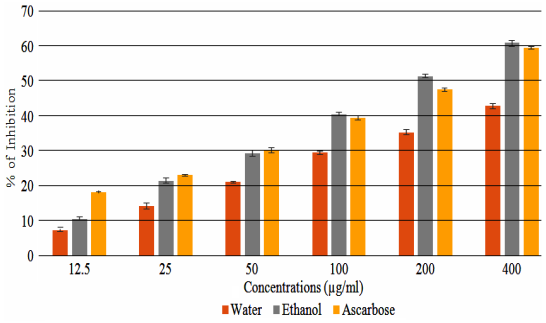

Fig. 6. Effect of *B. flabellifer* on α-Amylase activity

Values are expressed as mean±SEM of three replicates.

Inhibition of α-glucosidase enzyme

Figure 7 shows the results of the α-glucosidase inhibitory activity of the AqS-*Bf* and EtS-*Bf* extracts generated by Soxhlet extraction and heat maceration by water (AqS-*Bf* IC_{50}=33.33 g/mL). EtS-*Bf* made through cold maceration had an intriguing hypoglycemic effect with an IC_{50} of about 30.92 g/mL. In contrast to the AqS-*Bf* and EtS-*Bf*, the IC_{50} value for the reference standard, acarbose, was 57.74 g/mL.

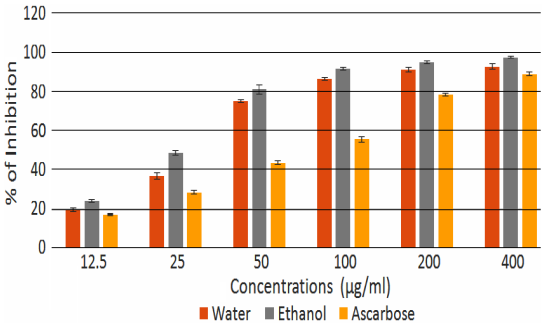

Fig. 7. Effect of *B. flabellifer* on α–Glucosidase activity

Values are expressed as mean ± SEM of three replicates.

All AqS-*Bf* and EtS-*Bf* were found to have some inhibitory action against α-amylase and α-glucosidase enzymes. This activity differs between enzymes and between the AqS-*Bf* and EtS-*Bf* systems. Our study used two different extraction methods, likely explaining the discrepancy in secondary metabolite concentrations between AqS-*Bf* and the control group. Similarly, researchers could classify the extracts under study by their mechanisms of action after selecting multiple assays to evaluate anti-diabetic effectiveness, thereby altering the results[49].

In our prior work, we described how the presence of different flavonoids, tannins, and phenolic compounds in aqueous and organic extracts accounts for the observed differences in activity between the two[50]. The improved outcomes can be traced back to the fact that the extracts used here come from a different botanical family and thus contain chemicals not found in those obtained from *B. flabellifer* sprouts. To prevent type 2 diabetes, limiting intestinal blood glucose uptake by consuming foods rich in phenolic compounds has been shown to improve postprandial glycemic levels, fasting blood glucose, insulin secretion, and insulin sensitivity[51-54].

Determination of glucose adsorption capacity

The physical process of glucose molecules adhering to a solid surface is known as glucose adsorption[55]. The effect of glucose adsorption is a decrease in free glucose in the solution, which might limit glucose diffusion and glucose uptake into the blood[56].

Figure 8 displays the difference *In vitro* glucose adsorption efficiency between AqS-*Bf* and EtS-*Bf*. This study found that the extract had a notable capacity for glucose adsorption at all tested concentrations. Glucose adsorption by the test sample was also found to be proportional to glucose concentration when the sample weight was held constant[57].

Adsorption was measured and found to be greatest at 30mM glucose and least at 5mM. Also, it was demonstrated that the test extract could bind glucose at concentrations as low as 0.058. As expected, the glucose binding capacity increased with increasing glucose concentration, and both AqS-*Bf* and EtS-*Bf* showed good glucose binding. AqS-*Bf* and EtS-*Bf* were effective in glucose adsorption at concentrations as low as 5 mmol/L and as high as 100 mmol/L. EtS-*Bf* had the highest activity of the extracts tested, which could be attributed to both insoluble and soluble components and EtS-*Bf*. Higher glucose concentrations bound the increasing concentration of sugar molecules like glucose[59]. Because glucose molecule adsorption occurs at a lower concentration, the amount of glucose accessible for transport through the intestinal lumen is reduced. As a result, postprandial hyperglycemia was reduced[60].

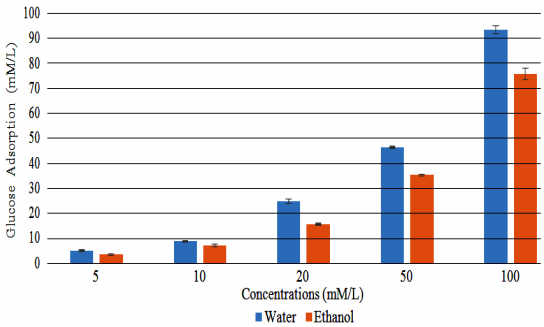

Fig. 8. Effect of *B. flabellifer* on Glucose adsorption

Values are expressed as mean ± SEM of three replicates.

Additionally, it was demonstrated in this study that the sample's glucose adsorption capacity is proportional to the molar concentration of glucose. The AqS-*Bf* and EtS-*Bf* extracts contain dietary fibers, some of which are soluble and some insoluble, which may account for their adsorption properties. Intestinal glucose absorption by extract can slow the rise in blood sugar after a meal[57]. There are three putative methods by which dietary fibre can aid in lowering postprandial hyperglycemia[61]. First, they may increase the viscosity of the small intestinal fluids, which makes the transport of glucose from the lumen into the blood slower and less efficient. Second, the concentration of these fibres in the intestine's lumen may drop because glucose binds to them[63]. Finally, dietary fibre may contain inhibitors of α-amylase (a starch-digesting enzyme), preventing starch digestion and lowering postprandial hyperglycemia[64].

Glucose diffusion Test

At controlled time intervals of 30, 60, 120, and 180 min, the glucose transfer from a closed dialysis tube into an external solution was observed. More effectively blocking glucose efflux across the dialysis membrane than the AqS-*Bf* was the EtS-*Bf* extract.

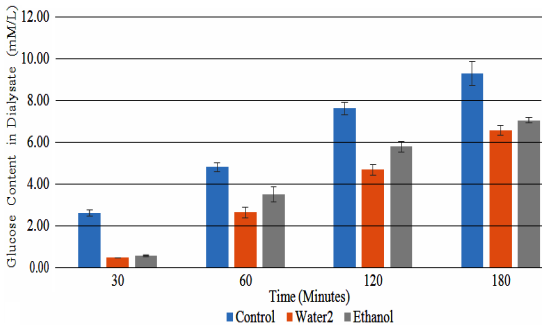

Fig. 9. Effect of *B. flabellifer* on Glucose diffusion

Values are expressed as mean ± SEM of three replicates.

The effect of AqS-*Bf* and EtS-*Bf* on glucose retardation activity across the dialysis tube was determined using the glucose diffusion inhibition test. At different times, the glucose entrapment abilities of the AqS-*Bf* and EtS-*Bf* were discovered to be considerably different. Among these, the EtS-*Bf* showed a lot of glucose entrapment, which slowed the flow of glucose into the external solution compared to the control at 180 minutes. EtS-*Bf* showed the biggest drop in glucose transport because it has the most insoluble fiber particles, which trap glucose molecules[65]. The dialysis tube method is a simple way to determine how AqS-*Bf* and EtS-*Bf* might affect glucose diffusion through the normal dialysis membrane. In contrast, glucose transporters that work with other molecules and intestinal contractions help move glucose through the intestinal tract[66]. As a result, more *In vivo* research is needed to evaluate the true effect of AqS-*Bf* and EtS-*Bf* on glucose diffusion.

CONCLUSION

In conclusion, the current study showed that the EtS-*Bf* has the potential to be an antioxidant and an anti-diabetic. These traditional medicinal plant extracts also demonstrated high α-amylase, α-glycosidase inhibitory, glucose adsorption, glucose diffusion, and antioxidant activity, indicating that the polyphenols present in the extracts have the potential to inhibit α-amylase and α-glycosidase activities, glucose adsorption, glucose diffusion studies, and DPPH, ABTS, FRAP SO and NO. This study shows that it is safe for traditional healers to use Borassus flabellifer to treat DM. Given that they have a lot of secondary metabolites and are very good at getting rid of free radicals, can lower blood sugar, and can prevent oxidative stress, these plants should be studied more.

REFERENCES

1. Poznyak, A.; Grechko, A. V.; Poggio, P.; Myasoedova, V. A.; Alfieri, V.; Orekhov, A. N. *Int. J. Mol. Sci.*, **2020**, *21*(5),1835.
2. Petrie, J. R.; Guzik, T. J.; Touyz, R. M. *Can. J. Cardiol.*, **2018**, *34*(5), 575–84.
3. Ceriello, A.; Testa, R.; Genovese, S. *Nutr. Metab. Cardiovasc. Dis.*, **2016**, *26*(4), 285–92.
4. Farzaei, F.; Morovati, M. R.; Farjadmand, F.; Farzaei, M. H. *J. evidence-based Complement. & Altern. Med.*, **2017**, *2*(4), 944–55.
5. Zhou, P.; Xie, W.; He, S.; Sun, Y.; Meng, X.; Sun, G.; *Cells.*, **2019**, *8*(3), 204.
6. Saad, B.; Zaid, H.; Shanak, S.; Kadan, S. Anti-Diabetes. Anti-Obes. *Med. Plants Phytochem.*, **2017**, 59–93.
7. Tran, N.; Pham, B.; Le, L. *Biology (Basel).,* **2020**, *9*(9), 252.
8. Dose, J.; Matsugo, S.; Yokokawa, H.; Koshida, Y.; Okazaki, S.; Seidel, U. *Int. J. Mol. Sci.,* **2016**, *17*(1), 103.

9. Ozougwu, J. C. *Int. J. Res.*, **2016**, *1*(8).
10. Ezhilarasan, D. *Arab. J. Gastroenterol.*, **2018**, *19*(2), 56–64.
11. Thi Le, D.H.; Chiu, C. S.; Chan, Y. J.; Wang, C. C. R.; Liang, Z. C.; Hsieh, C. W.; *Syrup. Biology. (Basel).,* **2021**, *10*(10), 1028.
12. Job, J. T.; Rajagopal, R.; Alfarhan, A.; Narayanankutty, A. *Drug. Chem. Toxicol.*, **2021**, 1–7.
13. Jerry, A. *J. Acad. Ind. Res.*, **2018**, *7*(7), 93–7.
14. Pkekal, A.; Pyrzynska, K. *Int. J. Food. Sci. Nutr.*, **2015**, *66*(1), 58–62.
15. Zheng, L.; Zhao, M.; Xiao, C.; Zhao, Q.; Su, G. *Food. Chem.*, **2016**, *192*, 288–94.
16. Long, Q.; Qi, J.; Li, W.; Zhou, Y.; Chen, K.; Wu, H. *Star Protoc.,* **2021**, *2*(3), 100706.
17. Jampafuang, Y.; Tongta, A.; Waiprib, Y. *Polymers (Basel).,* **2019**, *11*(12), 2010.
18. Ahmadinejad, F.; Geir Møller, S.; Hashemzadeh-Chaleshtori, M.; Bidkhori, G.; Jami, M. S. *Antioxidants.*, **2017**, *6*(3), 51.
19. Agarwal, P.; Gupta, R. *Res. Rev. J. Med. Heal. Sci.*, **2016**, *5*(4), 1–8.
20. Assefa, S. T.; Yang, E. Y.; Chae, S. Y.; Song, M.; Lee, J.; Cho, M. C. *Plants.*, **2019**, *9*(1), 2.
21. Zheng, Y.; Wang, Q.; Huang, J.; Fang, D.; Zhuang, W.; Luo, X.; *Food. Chem. Toxicol.*, **2019**, *127*, 120–6.
22. Ouassou, H.; Bouhrim, M.; Bencheikh, N.; Addi, M.; Hano, C.; Mekhfi, H. Antioxidants., **2021**, *10*(11), 1747.
23. Galketiya, C.; Weerarathna, T.S.; Punchihewa, J. C.; Wickramaratne, M. N.; Wickramaratne, D. B. M. *J. Med. Plants. Stud.*, **2017**, *5*, 91–5.
24. Mokrani, A.; Cluzet, S.; Madani, K.; Pakina, E.; Gadzhikurbanov, A.; Mesnil, M. Int. J. Mass. Spectrom., 2019, 445, 116192.
25. Shon, M. Y.; Kim, T. H.; Sung, N. *J. Food. Chem.*, **2003**, *82*(4), 593–7.
26. Maisuthisakul, P.; Suttajit, M.; Pongsawatmanit, R. *Food. Chem.*, **2007**, *100*(4), 1409–18.
27. de Velde, F.; Grace, M. H.; Esposito, D.; Pirovani, M. É.; Lila, M. A. *J. Food. Compos. Anal.,* **2016**, *47*, 82–91.
28. Bae, H.; Jayaprakasha, G.K.; Crosby, K.; Jifon, J.L.; Patil, B. S. *Plant. Foods. Hum. Nutr.*, **2012**, *67*(2), 120–8.
29. Zhao, B.; Wang, H.; Xu, S.; Qian, L.; Li, H.; Gao, *J. Fuel.*, **2022**, *307*, 121930.
30. Ismail, A.; Hong, T.S. *Malays. J. Nutr.*, **2002**, *8*(2), 167–77.
31. Kalaivani, T.; Mathew, L. Food. Chem. Toxicol., **2010**, *48*(1), 298–305.
32. Souli, I.; Jemni, M.; Rodriguez-Verástegui, L.L.; Chaira, N.; Artés, F.; Ferchichi, A. *J. Food. Biochem.*, **2018**, *42*(6), e12634.
33. Santos-Sánchez, N. F.; Salas-Coronado, R.; Villanueva-Cañongo, C.; Hernández-Carlos, B. *Antioxidants.*, **2019**, *10*, 1–29.
34. Lalremruati, M.; Lalmuansangi, C.; Siama, Z. *J. Appl. Pharm. Sci.*, **2019**, *9*(12), 94–102.
35. Pu, S.; Li, J.; Sun, L.; Zhong, L.; Ma, Q. *Carbohydr. Polym.*, **2019**, *211*, 161–72.
36. Bähring, S.; Root, H.D.; Sessler, J.L.; Jeppesen, J. O. *Org. & Biomol. Chem.*, **2019**, *17*(10), 2594–2613.
37. Eslami, S.; Ebrahimzadeh, M.A.; Biparva, P., *R. S. C. Adv.*, **2018**, *8*(46), 26144–26155.
38. Tusskorn, O.; Pansuksan, K.; Machana, K.; *Asian. Pac. J. Trop. Biomed.*, **2021**, *11*(2), 81.
39. Aryal, S.; Baniya, M.K.; Danekhu, K.; Kunwar, P.; Gurung, R.; Koirala, N. *Plants.*, **2019**, *8*(4), 96.
40. Ozel, R.E.; Bulbul, G.; Perez, J.; Pourmand, N. *A.C.S. Sensors.,* **2018**, *3*(7), 1316–21.
41. Sowmya, B.; Usha Anandhi, D. *Indian. J. Tradit. Knowl.*, **2021**, *20*(3), 716–22.
42. Szabo, K.; Cuatoi, A.F.; Vodnar, D.C. *Plant. Foods. Hum. Nutr.*, **2018**, *73*(4), 268–77.
43. Guo, H.; Saravanakumar, K.; Wang, M. H. *Biocatal. Agric. Biotechnol.*, **2018**, *15*, 235–9.
44. Kusznierewicz, B.; Mróz, M.; Koss-Mikołajczyk, I.; Namieœnik, *J. Food. Chem.*, **2021**, *362*, 130132.
45. Radi, R. *Proc. Natl. Acad. Sci.*, **2018**, *115*(23), 5839–48.
46. Hijaz, F.; Al-Rimawi, F.; Manthey, J. A.; Killiny, N. *Plant. Signal. & Behav.*, **2020**, *15*(5), 1752447.
47. Belli, S.; Rossi, M.; Molasky, N.; Middleton, L.; Caldwell, C.; Bartow-McKenney, C.; *Antioxidants.*, **2019**, *8*(1), 14.
48. Roden, M.; Shulman, G. I. *Nature.*, **2019**, *576* (7785), 51–60.
49. Boulfia, M.; Lamchouri, F.; Toufik, H. *Biomed. Res. Int.*, **2021**, 2021.
50. Hong, S.; Pangloli, P.; Perumal, R.; Cox, S.; Noronha, L.E.; Dia, V.P. *Antioxidants.*, **2020**, *9*(12), 1297.

51. Costabile, G.; Vitale, M.; Luongo, D.; Naviglio, D.; Vetrani, C.; Ciciola, P.; *Clin. Nutr.*, **2019**, *38*(6), 2727–34.
52. Vetrani, C.; Vitale, M.; Bozzetto, L.; Della Pepa, G.; Cocozza, S.; Costabile G.; *Acta. Diabetol.*, **2018**, *55*(2), 149–53.
53. Cianciosi, D.; Forbes-Hernández, T.Y.; Afrin, S.; Gasparrini, M.; Reboredo-Rodriguez, P.; Manna, P. P. *Molecules.*, **2018**, *23*(9), 2322.
54. Eseberri, I.; Laurens, C.; Miranda, J.; Louche, K.; Lasa, A.; Moro, C. *Int. J. Mol. Sci.*, **2021**, *22*(3), 1384.
55. Villena Gonzales, W.; Mobashsher, A. T.; Abbosh, A. *Sensors.*, **2019**, *19*(4), 800.
56. Müller, U.; Stübl, F.; Schwarzinger, B.; Sandner, G.; Iken, M.; Himmelsbach, M.; *Mol. Nutr. & food Res.*, **2018**, *62*(11), 1701012.
57. Rehman, G.; Hamayun, M.; Iqbal, A.; Ul Islam, S.; Arshad, S.; Zaman, K.; *Biomed. Res. Int.,* **2018**, 2018.
58. Van Enter, B. J.; Von Hauff, E. *Chem Commun.,* **2018**, *54*(40), 5032–45.
59. Xiao, Y.; Hu, Y.; Du, *J. Mater Horizons.*, **2019**, *6*(10), 2047–55.
60. Koepsell, H. Pflügers. *Arch. J. Physiol.,* **2020**, *472*(9), 1207–48.
61. Vitale, M.; Hanhineva, K.; Koistinen, V.; Auriola, S.; Paananen, J.; Costabile, G. *Nutr. Metab. Cardiovasc. Dis.*, **2021**, *31*(4), 1156–65.
62. Ayua, E.O.; Nkhata, S.G.; Namaumbo, S. J.; Kamau, E. H.; Ngoma, T.N.; Aduol, K.O. *Heliyon.*, **2021**, *7*(2), e06245.
63. Wu, Q.; Min, Y.; Xiao, J.; Feng, N.; Chen, Y.; Luo, Q.; *Food. & Funct.*, **2019**, *10*(8), 4877–87.
64. Rana, Z.H.; Alam, M.K.; Akhtaruzzaman, M. *Antioxidants.*, **2019**, *8*(7), 203.
65. Jarrett, S.; Ashworth, C. J. *J Anim Sci Biotechnol.*, **2018**, *9*(1), 1–11.
66. Wang, J.; Wang, Z.; Yu, J.; Kahkoska, A. R.; Buse, J. B.; Gu, Z. *Adv. Mater.*, **2020**, *32*(13), 1902004.